

ISSN 0494-9846
UDK 664.9:614.31: 637.5(05)

tehnologija mesa

meat technology

MEĐUNARODNO 55. SAVETOVANJE INDUSTRIJE MESA
Tara 15-17. jun 2009.

INTERNATIONAL 55th MEAT INDUSTRY CONFERENCE
Tara 15-17. june 2009

God.

50

Vol.

Br.

1-2

No.

Beograd,

2009

Belgrade,

Osnivač i izdavač – **FOUNDER AND PUBLISHER**
INSTITUT ZA HIGIJENU I TEHNOLOGIJU MESA, BEOGRAD
INSTITUTE OF MEAT HYGIENE AND TECHNOLOGY

TEHNOLOGIJA MESA JE ČASOPIS ZA UNAPREĐENJE INDUSTRIJE MESA
Meat technology is journal for the improvement of meat industry

Tehnologija mesa objavljuje rezultate osnovnih i primenjenih istraživanja u oblasti nauke o mesu
Meat technology publishes the results of fundamental and applied research in meat science

UREĐIVAČKI ODBOR – EDITORIAL BOARD

Prof. dr Milan Ž. Baltić

Fakultet veterinarske medicine, Beograd, RS
Faculty of Veterinary Medicine, Belgrade, Republic of Serbia

Ph. D. Andrzej Borys

Institut za istraživanje mesa i masti, Varšava, Poljska
Meat and Fat Research Institute, Warszawa, Poland

Prof. dr Sava Bunčić

Poljoprivredni fakultet, Katedra za veterinarsku medicinu,
Novi Sad, RS
Faculty of Agriculture, Department for Veterinary Medicine,
Novi Sad, RS

Prof. dr Luca Cocolin

Poljoprivredni fakultet, Katedra za eksploataciju i zaštitu
agrikulturalnih i šumskih resursa, Sektor za mikrobiologiju,
Torino, Italija
Faculty of Agriculture, DIVAPRA, Torin, Italy

Prof. dr Radoslav Grujić

Tehnološki fakultet, Banja Luka, Bosna i Hercegovina
Faculty of Technology, Banja Luka, Bosnia and Herzegovina

Prof. dr Andrej B. Lisicin

Sveruski istraživački institut za meso, Moskva,
Rusija
The All-Russian Meat Research Institute, Moscow,
Russia

Dr Vesna Matekalo-Sverak

Institut za higijenu i tehnologiju mesa, Beograd, RS
Institute of Meat Hygiene and Technology, Belgrade,
Republic of Serbia

Prof. dr Dragojlo Obradović

Poljoprivredni fakultet, Institut za prehrambenu tehnologiju i
biohemiju, Beograd, RS
Faculty of Agriculture, Institute for Food Technology and
Biochemistry, Belgrade, Republic of Serbia

Prof. dr Radomir Radovanović

Poljoprivredni fakultet, Katedra za tehnologiju animalnih
proizvoda, Beograd, RS
Faculty of Agriculture, Department for Technology of Animal
Products, Belgrade, Republic of Serbia

Dr Apostolos Rantsios

Konsultant EBTE, Ltd; Marousi, Grčka
EBTE Consultants, Ltd; Marousi, Greece

Dr Aurelija Spirić

Institut za higijenu i tehnologiju mesa, Beograd, RS
Institute of Meat Hygiene and Technology, Belgrade,
Republic of Serbia

Dr Aleksandra Stjepanović

Institut za higijenu i tehnologiju mesa, Beograd, RS
Institute of Meat Hygiene and Technology, Belgrade,
Republic of Serbia

Prof. dr Mitre Stojanovski

Fakultet za biotehničke nauke, Bitolj, RM
Faculty of Biotechnical Sciences, Bitola,
Republic of Macedonia

Prof. dr Marija Škrinjar

Tehnološki fakultet, Novi Sad, RS
Faculty of Technology, Novi Sad, Republic of Serbia

Prof. dr Klaus Troeger

Institut za tehnologiju, Savezni istraživački zavod za ishranu i
životne namirnice, Kulmbach, Nemačka
Institute of Technology, Federal Research Centre for Food and
Nutrition, Kulmbach, Germany

Dr Lazar Turubatović

Institut za higijenu i tehnologiju mesa, Beograd, RS
Institute of Meat Hygiene and Technology, Belgrade,
Republic of Serbia

Prof. dr Ilija K. Vuković

Fakultet veterinarske medicine, Beograd, RS
Faculty of Veterinary Medicine, Belgrade,
Republic of Serbia

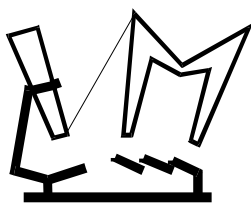
Prof. dr Božidar Žlender

Biotehnički fakultet, Katedra za hranu, istraživanja i tehnologiju,
Ljubljana, Republika Slovenija
Faculty of Biotechnology, Department of Food, Science and
Technology, Ljubljana, Republic of Slovenia

Rukopisi prispeli za štampanje obavezno podležu recenziji. Redakcija časopisa „Tehnologija mesa“ zadržava pravo da rukopise prilagodi usvojenom stilu časopisa ili da ih vrati autorima radi ispravke. Institut ne preuzima nikakvu odgovornost za postavke iznesene u člancima „Tehnologije mesa“. Rukopisi se ne vraćaju. Časopis se objavljuje u tri dvobroja godišnje. Tiraž je 250 primeraka. Reprodukovanje časopisa nije dozvoljeno.

Manuscripts submitted for publishing are subject to reviewing. The Editorial staff of the journal „Tehnologija mesa“ reserves privilege of editing manuscripts to mark them conform with the adoted style of the journal or returning them to authors for revision. The Institute is not responsible for the statements and opinions expressed in the articles published in the „Tehnologija mesa“. The manuscripts are not sent back. journal is published in three double issues. Circulation 250 copies. Reprinting of the Journal is not permitted.

Abstracced or indexed in FSTA (Food Science Techology Abstracts), Chemical Abstract, AGRIS (Agricultural Information Servis)



tehnologija mesa

naučni časopis

Tehnologija mesa God. 50 Br. 1-2 Str. 1-178 Beograd 2009

OSNIVAČ I IZDAVAČ

**Institut za higijenu i
tehnologiju mesa**

11000 Beograd, Kačanskog 13
P. fah 33-49
Tel (011) 2650-655
Telefax 011/ 2651-825
E-mail: meatinst@beotel.yu

DIREKTOR
Dr Lazar Turubatović

GLAVNI I ODGOVORNI
UREDNIK
Dr Aurelija Spirić

LEKTOR ZA SRPSKI JEZIK
Vlada Janković

LEKTOR ZA ENGLJSKI JEZIK
Srđan Stefanović

TEHNIČKO UREĐENJE
Danijela Šarčević
Radmila Zdravković

Na osnovu mišljenja Ministarstva za nauku i tehnologije Republike Srbije (br. 413-00-00416/2000-01), ova publikacija je od posebnog interesa za nauku.

Cena godišnje pretplate za časopis za Republiku Srbiju iznosi 4500,00 din. Uplate se mogu vršiti na tekući račun Instituta broj 205-7803-56 kod Komercijalne banke AD Beograd, sa naznakom «pretplata na časopis». O izvršenoj uplati obavestiti Institut i navesti adresu na koju treba poslati časopis.

Cena godišnje pretplate za časopis za inostranstvo iznosi: 50 EUR za evropske zemlje i 90 EUR za van-evropske zemlje. Naručuje se kod: Institut za higijenu i tehnologiju mesa, P.O. Box 33-49, Kačanskog 13, 11000 Beograd, R. Srbija.

SADRŽAJ

Raspor P., Jevšnik Mojca

- *Novi koncepti bezbednosti hrane za dobijanje zdravstveno ispravnih proizvoda: industrija prerade mesa* 1

Schwegele F., Andree Sabine

- *Praćenje i sledljivost u oblasti proizvodnje mesa*..... 11

Lisitsyn A. B.

- *Savremena ispitivanja na polju kvaliteta i kontrole bezbednosti mesnih sirovina i proizvoda od mesa u Rusiji* 21

Alvseike O.

- *Meso i proizvodi od mesa – opasnosti i rizik – strategije i iskustva Norveške* 28

Vesna Matekalo-Sverak, Turubatović L., Petronijević R.

- *Postupci unapređenja kontrole kvaliteta proizvoda od mesa – strategija zaštite potrošača* 31

Tröeger K.

- *Novo tehnologije tokom klanja, grubog i finog rasecanja – uticaj na bezbednost i kvalitet mesa* 37

Zamaratskaia Galia

- *Uzrok i mogućnosti eliminacije polnog mirisa u svinjskom mesu* 43

Irina Dederer

- *Tretiranje proizvoda od mesa visokim pritiskom* 48

Compasada J. Arnau, Garriga Margarita, Ferring...

- *Brzo sušenje suvih i salamurenih proizvoda od mesa: tehnologija brzog sušenja odrezaka* 54

Slavica Vesković-Moračanin, Obradović D.

- *Mikrobiološki ekosistem tradicionalnih fermentisanih kobasica u Srbiji – mogućnosti stvaranja sopstvenih starter kultura* 60

Vuković I., Saičić Snežana, Vasilev D., Tubić M., Vasiljević Nađa, Milanović-Stevanović Mirjana

- *Neki parametri kvaliteta i nutritivna vrednost funkcionalnih fermentisanih kobasica* 68

Nastasijević I.

- *Integrirani monitoring zoonotskih alimentarnih patogena u lancu mesa* 75

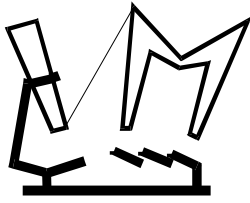
Lilić S., Tamara Ilić, Sanda Dimitrijević

- *Kokcidioza u proizvodnji živine* 90

Miličević D.

- *Mikotoksini u lancu ishrane – stari problemi i nova rešenja* 99

Smole Možina Sonja, Kurinčić Marija, Kramar Ana, Uršič Simona, Katalinić Višnja	
▪ <i>Zastupljenost Campylobacter spp. i rezistencija na različita antimikrobna jedinjenja u živinskom mesu iz prometa</i>	112
Ljiljana Petrović, Tomović V., Džinić Natalija, Tasić Tatjana, Ikonić P.	
▪ <i>Parametri i kriterijumi za ocenu kvaliteta polutki i mesa svinja</i>	121
Okanović Đ., Mastilović Jasna, Ristić M.	
▪ <i>Održivost lanca proizvodnje hrane</i>	140
Ristić M.	
▪ <i>Značaj senzorne ocene kao kriterijuma kvaliteta mesa – poređenje između različitih vrsta mesa i proizvoda od mesa</i>	148
Irina Černuha	
▪ <i>Senzorni sistem elektronskog nosa za kontrolu kvaliteta mesa</i>	159
Baltić Ž. M., Kilibarda Nataša, Dimitrijević Mirjana	
▪ <i>Činioci od značaja za održivost ribe i odabranih proizvoda od ribe u prometu</i>	166
<i>UPUTSTVO AUTORIMA ZA PISANJE RADOVA</i>	177



meat technology scientific journal

Meat Technology Vol. 50 No. 1-2 P. 1-178 Belgrade 2009

FOUNDER AND PUBLISHER

**Institute of Meat Hygiene and
Technology**

11000 Belgrade, Kačanskog 13
P.O. Box 33-49
Phone (011) 2650-655
Fax 011/ 2651-825
E-mail: meatinst@beotel.yu

DIRECTOR

Lazar Turubatović, PhD

EDITOR IN CHIEF

Aurelija Spirić, PhD

PROOFREADER FOR
SERBIAN LANGUAGE

Vlada Janković

PROOFREADER FOR
ENGLISH LANGUAGE

Srdan Stefanović

TECHNICAL EDITION

Danijela Šarčević

Radmila Zdravković

Based on the opinion issued by
Ministry of Science and Technology
Republic of Serbia (No. 413-00-
00416/2000-01), this publication is
of special interest for the science.

Subscription

Annual subscription rate is: 50 EUR
in Europe, 90 EUR outside Europe-
including postage. Orders should be
Institute for Meat Hygiene and
Technology, P.O. Box 33-49,
Kačanskog 13, 11000 Belgrade, R.
Serbia.

CONTENTS

Raspor P., Jevsnik Mojca

▪ *Novel Food Safety Concepts for Safe Food: Case Meat Processing Industry* 1

Schwegele F., Andree Sabine

▪ *Tracking and Tracing in Meat Area*..... 11

Lisitsyn A. B.

▪ *State of the Art of the Investigation in the Field of Quality and Safety Control
of Meat Raw Materials and Meat Products in Russia* 21

Alvseike O.

▪ *Meat and Meat Products – Hazards and Risks – Norwegian Strategies and
Experiences* 28

Vesna Matekalo-Sverak, Turubatovic L., Petronijevic R.

▪ *Procedures in Improvement of the Control of the Quality of Meat Products –
Consumer Protection Strategy*..... 31

Tröger K.

▪ *New Technologies in Slaughtering, Pre-Cutting and Cutting – Influence on
Safety and Quality of Meat* 37

Zamaratskaia Galia

▪ *Cause and Possible Ways to Eliminate Board Taint in Pork* 43

Irina Dederer

▪ *High Pressure Treatment with Meat Products* 48

Compasada J. Arnau, Garriga Margarita, Ferring...

▪ *Fast Drying of Dry-Cured Meat Products Quick-Dry-Slice (QDS) Process
Technology* 54

Slavica Veskovc-Moracanin, Obradovic D.

▪ *The Microbiological Ecosystem of Traditional Fermented Sausages in Serbia –
Possibility to Create Our Own Starter Cultures* 60

**Vukovic I., Saicic Snezana, Vasilev D., Tubic M., Vasiljevic Nadja,
Milanovic-Stevanovic Mirjana**

▪ *Some Quality Parameters and Nutritional Value of Functional Fermented
Sausages* 67

Nastasijevic I.

▪ *Integrated Monitoring of Zoonotic Foodborne Pathogens in the Meat Chain* 75

Lilic S., Tamara Ilic, Sanda Dimitrijevic

▪ *Coccidiosis in Poultry Industry* 90

Milicevic D.

▪ *Mycotoxins in the Food Chain – Old Problems and New Solution* 99

Smole Mozina Sonja, Kurincic Marija, Kramar Ana, Ursic Simona, Katalinic Visnja	
▪ <i>Prevalence and Resistance Against Different Antimicrobial Compounds of Campylobacter spp. in/from Retail Poultry Meat</i>	112
Ljiljana Petrovic, Tomovic V., Dzinic Natalija, Tasic Tatjana, Ikonc P.	
▪ <i>Parameters and Criteria for Quality Evaluation of Pork Carcass Halves</i>	121
Okanovic Dj., Mastilovic Jasna, Ristic M.	
▪ <i>Sustainability of Food Production Chain</i>	140
Ristic M.	
▪ <i>The Meaning of Sensory Evaluation as a Criterion for Meat Quality – A Comparison of Different Meat (Products)</i>	148
Irina Cernuha	
▪ <i>Electronic Nose Sensory Systems for Meat Quality Control</i>	159
Baltic Z. M., Kilibarda Natasa, Dimitrijevic Mirjana	
▪ <i>Factors Significant for the Shelf-Life of Fish and Selected Fish Products in Retail</i>	166
<i>GUIDES FOR THE SUBMISSION OF PAPERS</i>	177

NOVEL FOOD SAFETY CONCEPTS FOR SAFE FOOD: CASE MEAT PROCESSING INDUSTRY*

Raspor P., Jevšnik Mojca

Abstract: Consumers' concern about dangers associated with food is high. Due to recent food crises in Europe, food quality and food safety have become a hot topic in the media. Meat, as one of the most sensitive industries regarding microbial contamination in food supply chain, deserves all this attention and we need to bring new skills to practice to manage food safety from the farm to the fork. The aim of this short review was to evaluate and compare the few food safety issues which are relevant for meat industry, namely food safety knowledge in practice, employees' attitude toward food safety and employees' work satisfaction and diversification of the systems connected to meat processing industry.

Today we master food safety through good practices at different levels of food production, distribution and consumption. The novelties which enter food supply chain through new substrates, new processes and technologies and new nutrition practices are key factor for building up a new dimension in food safety, which should be handled holistically. All these elements are very complex and closely connected to social factors, e.g. employees' knowledge, awareness and attitude. Based on the research results on this field it is determined that food safety education and individual awareness are the most important tools for food safety assurance, that's why every food handler requires a complex and individual dealing. The human factor must be discussed equally like all the other risk factors, such as hygiene, technical and technological factors. For food safety it is essential that every link in food supply chain understands and fulfils his responsibilities and relies upon the previous and the next step in a chain.

Key words: food safety, safe food, meat industry

Novi koncepti bezbednosti hrane za dobijanje zdravstveno ispravnih proizvoda: industrija prerade mesa

Sadržaj: Zabrinutost potrošača za opasnosti povezane sa hranom je velika. Zbog skorašnje krize sa hranom u Evropi, kvalitet i bezbednost hrane su postali „vruće“ teme u medijima. Industrija mesa, kao jedna od najosetljivijih oblasti u snabdevanju hranom sa aspekta mikrobiološke kontaminacije zaslužuje svu moguću pažnju i zbog toga moraju da se uvedu u praksu nove veštine u upravljanju bezbednošću hrane „od njive do trpeze“. Cilj rada je ocena i poređenje nekoliko problema iz oblasti bezbednosti hrane relevantnih za industriju mesa – konkretno, saznanja o aspektima bezbednosti hrane u praksi, stav zaposlenih prema bezbednosti hrane kao i zadovoljstvo radom i razgranatost sistema povezanih sa industrijom prerade mesa.

Danas ovladavamo poljem bezbednosti hrane kroz dobru praksu na različitim nivoima proizvodnje, distribucije i potrošnje. Noviteti koji ulaze u lanac snabdevanja hranom, kao što su novi supstrati, novi procesi i tehnologije kao i novi načini predstavljanja, su ključni faktori za izgradnju nove dimenzije u oblasti bezbednosti hrane sa kojima se mora upravljati holistički. Svi ovi elementi su veoma kompleksni i tesno povezani sa socijalnom faktorima, na primer: znanja zaposlenih, svest i stav. Na osnovu rezultata istraživanja u ovoj oblasti utvrđeno je da su edukacija o bezbednosti hrane i individualna svest najvažniji alati za osiguranje bezbednog proizvoda – zato svako ko rukuje hranom zahteva da mu se posveti kompleksna pažnja na individualnom nivou. Ljudski faktor mora da se obradi jednako kao i ostali faktori rizika, kao što su higijena, tehnički i tehnološki faktori. Esencijalno je, sa aspekta bezbednosti hrane, da svaka karika u lancu snabdevanja razume i ispunjava svoje odgovornosti kao i da može da se osloni na prethodni i sledeći korak u lancu.

Ključne reči: bezbednost hrane, zdravstveno ispravni proizvodi, industrija mesa

Novi koncepti bezbednosti hrane za Introduction

Since April 2004 when the European Parliament adopted Regulation (EU) No 852/2004 on the hygiene of foodstuffs it focused strongly on the sys-

tem of food safety management until 1st of January 2006 when it has to be applied to all food operators. The main change in the law relates to food safety management systems, i.e. risk based methodologies to ensure the safety of food. Successful implementations of the procedures based on the HACCP prin-

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTHORS: Peter Raspor, peter.raspor@bf.uni-lj.si, Biotechnical Faculty, Food Science and Technology Department, Jamnikarjeva 10; Mojca, Jevšnik, Faculty of Health Sciences, Department of Sanitary Engineering, Poljanska 26a, University of Ljubljana, 1000 Ljubljana, Slovenia.

ciples are requiring the full cooperation and commitment of food business employees. To this end, employees should undergo training.

A major problem that still remains is the employees' fully acceptance of prerequisite programs (PRP) and HACCP system, especially in small and medium-sized (SMEs) food businesses. Many authors discuss about barriers or hindrances which have impact to the effective implementation of HACCP in SMEs (Vela and Fernandez, 2003; Walker et al., 2003; Taylor and Taylor, 2004a; Taylor and Taylor, 2004b; Hennroid and Sneed, 2004; Azanza and Zamora-Luna, 2005; Baš et al., 2005; Hielm et al., 2006). Among the key ones, Walker et al. (2003) mentioned lack of expertise and perception of benefits, absence of legal requirements, various attitude barriers and financial constrains. According to Hielm et al. (2006) most difficulties were established in devising the own-checking plan/HACCP plan the most common answers were choosing the critical control points, committing the firm's entire workforce and organising the documentation of monitored results. One of the major problems is that the food workers often lack interest and they often have a negative attitude toward food safety programs (Griffith, 2000).

It is obvious that the food represents one of major problems in current world, beside health and environmental problems. We can expect this trend to continue in the future. Development of new techniques and methods will definitely help us to reduce (avoid) certain hazards and maintain the quality of life, but we should not forget basic principles of nature (Raspor & Jevšnik, 2008).

Food safety management and personnel

The acceptance of food safety systems has put employee training under the microscope (Collis, 2002). Under the personnel programme of HACCP, employees must be trained in such areas as food safety, manufacturing controls and personnel hygiene. Once HACCP plans have been established, employees must be trained to manage any critical control points (CCPs). Though numerous companies have developed documented and implemented training programmes, few understand why employee training is important, what their training requirements are, or how to assess the effectiveness of in-house training programmes. So far most publications about HACCP training have described what should be done, but little has been written about effectiveness of such training and how to motivate employees to follow all food safety requirements. Food business operators have to engage with these issues in their own way, as every

company has its own specific means of ensuring safety. HACCP has been described as a philosophy in theory and a tool in practice (Gilling et al., 2001) and cited by Bryan (1981) »It should therefore come as no surprise that there can be different opinions on how it should be applied« HACCP problems are a complex mix of managerial, technical and behavioral issues requiring specific remedies (Gilling, 2001). By taking a psychological approach and utilizing practical experience and a theoretical knowledge of HACCP, Gilling et al. (2001) identified 11 key barriers and organized them around knowledge, attitude and behavior framework. The proposed Behavioral Adherence Model therefore acts as a diagnostic tool, identifying progressive stages to successful HACCP guideline adherence. They emphasized that the model should be of significant help to those offering advice and guidance to food operators undertaking HACCP implementation; a problem which has considerable influence on acceptance of introduced "new" food safety system especially when it begun were the way of presenting HACCP and qualification of trainers. Mortimore and Smith (1998) mentioned that many trainers had been willing to provide HACCP training without considering the scope (what had to be taught and what need not) and the depth of coverage. They also described that there was a wide disparity in content and quality between courses. Moreover, several authors suggested that most managers in food industry have limited understanding of the global food safety strategy (Ehiri et al., 1995; Mortimore and Smith, 1998; Khandke and Mayes, 1998; Williams et al., 2003). MacAuslan (2003) cited Aston (2001) who wrote that the majority of food businesses do not have satisfactory training policies for all their staff. He emphasized that too much reliance is being placed upon attaining a certificate rather than attention is paid to achieving competency in food hygiene practice. He suggested that more emphasis and resources need to be diverted towards assisting managers to become highly motivated food hygiene managers who develop and maintain a food safety culture within their business. A small business owner may be tempted to place the burden of training responsibility on an external employer and not shoulder any responsibility towards themselves. Upon MacAuslan (2003) the problem can have two sides; firstly, the employer lacks key management skills in leadership, motivation, training and evaluation and secondly, going for a certificate course as it is the "done thing".

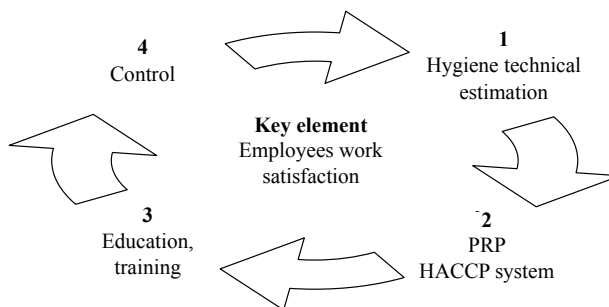
Personal as main food safety factor

Factors, which have a significant impact on employers' behavior, are correlated with organisa-

tional climate in the company, level of job satisfaction and labor conditions and with relations between employees. *Marolt and Gomišček* (2005) described a new management approach to employees, which stimulates employees to be initiative, to learn, to devote to company, to self-confidence, to higher efficiency and better team-work; that all contribute to higher successfulness and effectiveness of the organisation. They emphasized a function of leadership, which plays a key role in realisation of the new principles into practical work and thus can significantly contribute to better usage of existing resources. A leader should, with his leadership function, persuade the employees to fulfill their needs and desires by effective working and should enable them to use their potentials and by doing so, to contribute and to achieve the goals of a team and an organisation. It would be ideal if people would be motivated to such level, so they would not work just because they have to, but would work with eagerness and with trust. Skills of a successful leader motivation, communication, improvement and introduction of modifications are also mentioned (*Černetič*, 2001; *Marolt and Gomišček*, 2005). In review on history of motivational research and theory *Latham and Ernst* (2006) summarised that psychologists now know the importance of (1) taking into account a person's needs (Maslow's need hierarchy theory, Hackman and Oldham's job characteristics theory), (2) creating a job environment that is likely to facilitate self-motivation (Herzberg's job enrichment theory, Hackman and Oldham's job characteristics theory), and (3) ways to directly modify, that is to directly increase or decrease another person's behavior by administering environmental reinforces and punishers contingent upon a person's response (Skinner's contingency theory). They also stress the importance on attaining employees' goals, then they not only feel satisfied, they generalise their positive affect to the task (*Locke and Latham*, 1990). *Jannadi* (1995) emphasized that workers are the ones who carry out the work in a company, and they can be an important factor in making the company profitable or bankrupt. Human behavior is very important, and it is difficult to control, so handling people requires situational leadership. Hazards can not be solved and eliminated just through engineering control. They also need to be recognized by employees who will minimize their effects (*Jannadi*, 1995).

For efficient food safety management, *Jevšnik et al.* (2007) suggested that food business operators follow the model of "Four elements analysis" for efficient hygiene-technical situation management in food-processing plants. The model includes equally important elements, where every individual element

requires competent and trained person's involvement. Model's benefit is exposure of human factor in food safety assurance. The first element includes current hygiene-technical estimation in food-processing plant. Hygiene-technical deficiencies and/or irregularities have to be analyzed and plan of improvements has to be made. The second element includes establishing of hygiene basics, so called prerequisite programs, which are the basic for HACCP system establishment – a tool for food safety management. The third element includes planning and execution of periodical training and education, adapted to specific work tasks, for employees of all the food hygiene levels. The fourth element rests on employees' knowledge during food handling checking and on responsible person's opinion regarding involvement of individual worker in specific work task. This demands professionally trained, competent person, who possess adequate technical and pedagogical knowledge, practical experiences and knowledge from human resource management. The various techniques and methods of training involvement and control of work process performance are required as well. By last, fourth, element, a human factor as risk for food safety assurance has been pointed out. In the future an equal discussion for human risk factor as for the other risk factors in production processes (biological, chemical and physical) is suggested. Based on the results of the research it is determined that hygiene education and individual awareness are the most important tools for food safety assurance, that's why every food handler requires a complex and individual dealing. The human factor must be discussed equally like all the other risk factors such as hygiene, technical and technological factors. For food safety it is essential that every link in food supply chain understands and fulfils his responsibilities and relies upon the previous and the next step in a chain (*Jevšnik et al.*, 2007).



Scheme 1. "Four elements analysis" model for HACCP system effectiveness
Shema 1. Model "Četiri elementa" za efektivnost HACCP sistema

Personnel management and education

Human resource management and education of food safety managers in food premises has not captured the strong attention of researchers until recently (Jevšnik *et al.*, 2008). Strict performance of working procedures in accordance with HACCP system principles and food hygiene is essential for food related diseases prevention and efficient safe food assurance. To achieve this purpose two basic conditions: (1) suitable working environment from the hygienic – technical point of view and (2) motivated, satisfied and qualified personnel must be assured. It is interesting that many understand HACCP system as a novelty, when in fact it is about more complete approach to food safety assurance as stated by Ehiri *et al.* (1995). HACCP system assures more structured surveillance over determined hazards as was the case with the usual classical type of surveillance. Hazards and corrective actions are not something new. What is new is how separate activities and procedures are logically ranged. The approach is multidisciplinary. It requires personal responsibility, document and record control and rapid action when non-conformities are discovered. It enables traceability as well. Its greatest ability lies in responding to changes as well as in enabling continuous checking and efficiency confirmation. It brings changes in thinking, organising, managing, education and training at all levels, from employers to employees (Likar *et al.*, 2001; Likar and Jevšnik, 2004). The system becomes efficient when understandable to employees and when the responsible ones perform their duties. Then the requirements of the system are not considered as irrational, unnecessary and additional burden, but as desire for continuous improvement of one's own work. That is why the training from top management to all employees is crucial for food safety. Bryan (1988) predicted that in the future the number of HACCP principles would increase from seven to ten or more. The ninth HACCP principle, according to him, would be education and training, which is now being incorporated into the existing principles or other related guidelines. If routine-work employees do not understand the significance of hazards associated with food safety well enough, this may hinder a successful implementation of preventive and control actions.

Legislative changes in 2004 demand that now all food premises must provide food hygiene training appropriate for the work activities of their staff (Regulation, 2004). The results of our study showed as well that training carried out by company experts and by supervisors directly in working place is the

most efficient one. Mortlock *et al.* (2000) suggested that it is also important to recognize that whilst formal training might ensure greater consistency and quality (Manning, 1994), improper training could present a greater risk to food safety than no training at all. In a study by Cohen *et al.* (2001) they analyzed the impact of an in-house food sanitation training program on the performance of a catering company. They concluded that for fully effective sanitation program, it must be taken into consideration the different environments and circumstances in which the departments operate. It is very important that those performing a training have suitable food safety knowledge as well as skills in pedagogical – andragogical field. Those people have to be competent experts in their field so that adequate knowledge and skills can be passed on to the employees. A problem lies in SMEs, where owners of a company are usually at the same time responsible persons for food safety programs, which includes training as well. Because lack of time or poor knowledge such trainings are not carried out as intended by the Law. The results of our study show poor knowledge about microbiological hazards and their control among employees in retail, catering and food production units. MacAuslan (2003) stressed the importance on helping managers to understand what is expected of them and giving them a support in managing effective food hygiene. He pointed out that too much reliance has been placed upon certificates and not enough on the competence. According to his opinion this is defined as the ability of an individual to demonstrate the activities within their workplace, or to function to the standards expected in a food business.

The purpose of internal surveillance is to identify specific hazards, in particular company and then to establish a strategy of efficient control or successive elimination of hazards as stated by Jevšnik *et al.* (2008).

Owners or managers must, besides equal economic growth of a company, take care of human resource management as well. A positive motivational atmosphere in working environment significantly contributes to higher productivity, employees' loyalty and to general good feeling in workplace. The results of work satisfaction elements carry important messages for companies' management. In the three studied food units food production employees are the least satisfied in workplace and the most satisfied ones are employees in catering. A low score of employees in food production units regarding their opinion and suggestion consideration, rewarding for good work, wages, work conditions and promotion possibility must be stressed out. All that weakens

motivation and satisfaction in workplace as well as reduce a number of those, who perform their work well. Food safety assurance stands between two strong poles, which have to be balanced to achieve global food safety. The first pole is system requirements, namely flexible, faultless, which requires in forms of strategies, not directives. The second pole is work performance and a person in all his uniqueness his knowledge, qualification, working in a group and consciousness. A company's management should be aware that a quality and safe products is a result of an immediate performer, who should be paid full of many-sided attention to (Jevšnik *et al.*, 2008).

Strict performance of working procedures in accordance with HACCP system principles and food hygiene is essential for food related diseases prevention and efficient safe food assurance. A novel food safety concepts for safe food separate activities and procedures in logically ranged. The approach is multidisciplinary. It requires personal responsibility, document and record control and rapid action when non-conformities are discovered. It enables traceability as well. Its greatest ability lies in responding to changes as well as in enabling continuous checking and efficiency confirmation. It brings changes in thinking, organizing, managing, education and training at all levels, from employers to employees (Likar *et al.*, 2001; Likar and Jevšnik, 2004; Jevšnik *et al.*, 2008).

Current limitations in food safety management

In most Small Enterprises (SE) there are area limitations and they are not constructive-technical suitable for performing food related activities (Baš *et al.*, 2006, Jevšnik *et al.*, 2007). In small plants technical and hygiene conditions for hand washing were estimated as inadequate and worrying. Un-negligible share of (14%) small plants does not meet even minimal hygiene-technical requirements for food handling (e.g. wash-hand basin is missing or is not installed properly – enables cross contamination between high and low risk area; unsuitable and worn out materials do not enable efficient sanitation and maintenance etc.). Aarnisalo *et al.* (2006) summarize the results of many studies which have shown that food processing equipment could be a source of contamination, e.g. *Listeria monocytogenes*. Hygiene problems in equipment are caused when microorganisms become attached to the surfaces and survive on them and later become detached from them contaminating the product (Aarnisalo *et al.*, 2006). In some of Medium Enterprises (MEs) as

well as in some of small sized ones the wash-hand basins installation does not prevent cross contamination between high and low risk areas. Hygienic equipment of basins is inadequate mainly in SEs, since in more than a third of (39%) plants necessary hygienic equipment by the basins was missing (e.g. liquid soap, paper towels). In regulation (EC) No 852/2004 it is stated that an adequate number of hand-wash basins is to be available, suitably located and designated for cleaning hands. Washbasins for cleaning hands are to be provided with hot and cold running water, materials for cleaning hands and for hygienic drying. Where necessary, the facilities for washing food are to be separated from the hand washing facility (Regulation, 2004).

By observing employees during their work, the fact that most of workers in both groups do not wash their hands after performing any dirty work (e.g. when changing between high and low risk phase of work, after packaging handling etc.) or do not wash hands properly (e.g. they do not use liquid soap, negligent hand washing technique etc.), was determined. It was concluded that employees do not understand the meaning of proper hand washing and are not aware of microbiological hazards that can occur due to dirty hands. The causes for the latter can be found among insufficient hygiene training, negligent, insufficient employees' knowledge and/or inefficient control by supervisors. (Jevšnik *et al.*, 2007)

Microorganisms are always present on hands, because they are a part of normal microflora, but nevertheless in food production and trade the presence of some of bacteria is not allowed. In the research for bacteriological analyses of hands a blood agar plates were used, which enable quick estimation of hygiene condition in the selected plants. In further analyses selective growth medium would be used only for not allowed bacteria, which show hygienic status of food-processing plants. Bacteria from employees' hands have grown from some to 100 and more (on an individual hand). It was determined that on right hands there were less microorganisms than on left hands. If studying an individual person in the most of the cases can be seen that in the same person has either low or high bacteria count on both hands. Therefore it may be wise to take swabs from workers hands more frequently and communicate the results. That could be a motivation for better hand hygiene at work. However, as shown in previous studies of food handlers' beliefs and self-reported practices (Clayton *et al.*, 2002), food handlers were aware of the food safety behaviors they should be carrying out, but 63% of respondents admitted that they did not always carry out these behaviors. Food handlers

also reported carrying out food safety practices, particularly hand washing, much more frequently than they actually implemented them (Manning and Snider, 1993). This suggests that food handlers could be carrying out food safety practices less frequently than the self-reported data implies (Clayton et al., 2002). Shojaei et al. (2006) cited that many authors emphasized that hands of food handlers are an important vehicle of food cross-contamination and that improved personal hygiene and scrupulous hand washing would lead to the basic control of faces-to-hand-to-mouth spread of potentially pathogenic transient micro-organisms. Lues and Van Tonder (2007) summarize results of several studies where it was established that various bacteria, among others *Staphylococcus aureus*, *Escherichia coli* and *Salmonella* sp., survive on hands and surfaces for hours or even days after initial contact with the micro-organisms.

Every person working in a food-handling area is to maintain a high degree of personal cleanliness and is to wear suitable, clean and, where necessary, protective clothing (Regulation, 2004). It was determined that personal hygiene is significantly poorer in SEs than in MEs. More than a third (36%) of workers in SEs did not wear clean and suitable overalls, more than half (52%) performed work with no head-covering. The cause of the problem contributing to the stated results in SEs is lack of control by trained and responsible persons. Workers are to a large extent left on their own, beside that the owners do not provide necessary means for the safe food handling. In MEs situation regarding personal hygiene is better (Jevšnik et al., 2007). In most of MEs there is responsible person authorized by management, who is responsible for hygiene and has required professional education. A periodical training for workers is performed in accordance with a plan and work performance us checked daily. The main problem identified among food handlers in Ss is related to the fact that they receive no specific or insufficient knowledge about food hygiene.

Knowledge and training for working according to HACCP system were estimated by prior designed questions. By asking a question: "How do you record temperatures in cooling appliances and during heat treatment?" it was determined that in 12% SEs and in 20% of MEs temperatures were registered in advance and for the past (Jevšnik et al., 2007). From the results it is concluded that the majority of workers follow work instructions, but are not familiar with or do not understand why that is necessary and are not aware of hazards in case of hygiene violations and un-fulfillment of the requirements. This finding was consistent with the findings of Panisello et al.,

1999, Ramirez Vela and Martin Fernández, 2003, Yapp and Fairman, 2006, where they established that smaller companies may lack knowledge and expertise in HACCP and appropriate resources to obtain knowledge, both resulting in insufficient understanding of functions of HACCP principles. It was established that education and training is not efficient mainly in SEs, since it is carried out by incompetent persons without suitable professional and pedagogical knowledge. Yapp and Fairman (2006) pointed out that in some cases SMEs do not realize that they are breaking the law and often do not understand what is required of them. It is particularly evident when recording parameters according to HACCP plan. It was determined that documentation regarding prerequisite programs in both types of food enterprises is incomplete, but in SEs the situation is worse. Mitchell (1998) stated that the HACCP plan is sometimes a »paper exercise« that overburdens the need of SMEs and it is not implemented in practice.

With regulation (EC) No 852/2004 the responsibilities for food safety lays entirely on food business operators, which means that operators are responsible for education and training of their employees as well (Regulation, 2004).

It is still a question which training type will prove to be more effective in the future. Irrespective of that, the most important fact, according to Seaman and Eves (2007), is that the training will only lead to an improvement in food safety if the knowledge imparted leads to desired changes in behavior in the workplace. For conscientious hygiene it is not important in which enterprise people work, but depends upon hygiene awareness and education of an individual person.

Novel Solutions in food safety management

As Raspor stated in 2008, food safety is a result of several factors: legislation should lay down minimum hygiene requirements; official controls should be in place to check food business operators' compliance and food business operators should establish and operate food safety programmes and procedures. In theory it seems that we manage food safety completely, but practical experiences show some deviations. For that reason we have to proceed to new solutions which are based on synthesis of all relevant key factors included in food supply chain. One of possibility is to link all relevant Good practices in good nutritional practice (Fig 1.), as it was published recently (Raspor, 2008; Raspor and Jevšnik, 2008).

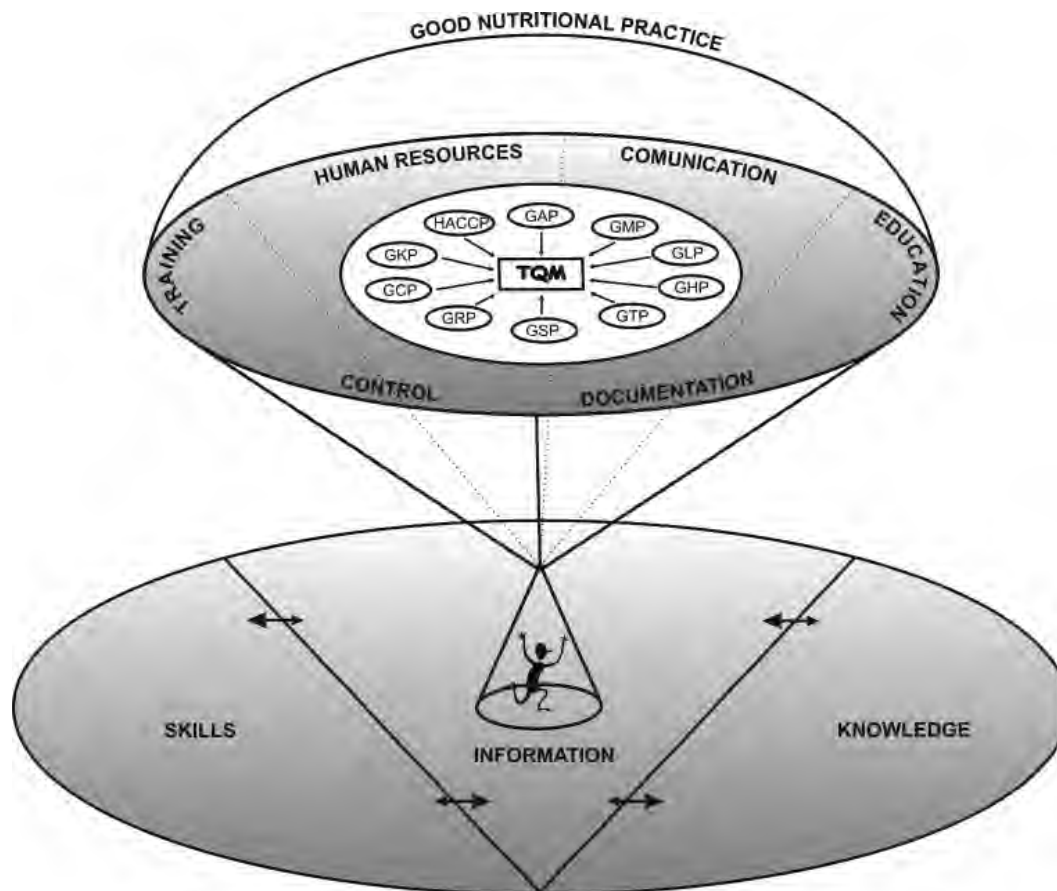


Figure 1. Food safety platform: balanced model for ensuring food safety from Good Nutritional Practice viewpoint (*Raspor and Jevšnik, 2008; with Permission of CRC*)

Slika 1. Platforma bezbednosti hrane: balansirani model za osiguranje bezbednosti hrane sa gledišta dobre nutricionističke prakse (*Raspor i Jevšnik, 2008; sa dozvolom CRC*)

Today we master food safety with different good practices which are the consequence of human culture, history and lifestyle. If we analyse good practices in the broad spectre of food area we could arrange them in three categories. First category of good practices is directly connected with food technology (i.e. Good Manufacturing Practice - GMP). Second category is indirectly connected with food issues (i.e. Good Research Practice - GRP, Good Educational Practice - GEP, Good Training Practice - GTrP). Third category deals with all the activities regarding consumers' food handling (Good Housekeeping Practice - GHKP). Consumers are not connected to food supply chain according to chain principles.

However, it has been shown that present maintenance of food safety in food supply chain can be easily broken down, because of different kind of barriers or simple misunderstanding. Therefore a new approach called "Good Nutritional Practice" (GNP) was coined to manage food safety (*Raspor,*

2008, Raspor and Jevšnik, 2008). It is important to reconstruct the existent food safety system with GNP, which includes consumers, and is based on a model that covers subsystems from other good practices.

New techniques for reducing pathogen contamination in meat and poultry are entering meat processing field every day. It is hard to cope with all novelties since is not always totally clear what is really new and what is just improvement of existing technique or protocol. The compilations done by different author or authorities around the globe are trying to solve this issue. However such information can provide a reference for processors worldwide searching for better ways to improve food safety in their plants. The new technologies have to bring significant improvements to the safety of meat and poultry. In recent years new technology has been defined as new, or new applications of equipment, substances, methods, processes, or procedures affecting the slaughter of livestock and poultry or processing of meat, poultry, or egg products.

General believes that increased public and industry awareness of the new technologies being used could further promote their use, by small and very small plants in particular, towards improving the safety of meat, poultry, and egg products. The new technologies listed should be viewed as information of current state of the art.

master of its particular area and will trust in activity of both previous and following link in the food safety circle »from farm to table«, not ignoring consumer as the one who should be aware of potential risks, proper handling and preparation of food for safe and balanced everyday meal (Raspor and Jevšnik, 2008).

Table 1: Selection of new/ novel technologies and Protocols to improve meat safety

Tabela 1. Odabir novih tehnologija i Protokola za unapređenje bezbednosti mesa

Application of Sodium Metasilicate on Raw Beef Carcasses as an Anti-microbial Processing Aid.	Chemical
Hyperchlorinated (≤ 200 ppm) solution applied to beef hide surfaces utilizing a washing/rinsing cabinet.	Chemical
Use of, a bromine-based biocide, as an effective poultry carcass antimicrobial when used in poultry chiller water in poultry processing plants at a level up to 100 ppm available bromine in the supply water.	Chemical
Use of up to 5% lactic acids on hot beef carcasses.	Chemical
Use of acidified sodium chlorite antimicrobial solutions as processing aids on i) pre- or post-chill poultry or red meat carcasses, carcass parts, trim or organs, or; ii) on processed, comminuted or formed meat products, in meat and poultry establishments pre-chill for COP (continuous-online-processing) in poultry processing.	Chemical
Ozone wash system using aqueous ozone on ready-to-eat (RTE) meat and poultry products for control of Listeria monocytogenes .	Chemical
Use of a bromine-based biocide, as an effective poultry carcass antimicrobial when used in poultry chillers and/or inside-outside bird washers (IOBW) at a level up to 100 ppm available bromine in the supply water.	Chemical
Cryovac Barrier Foam Tray/ LID551P Tray/Lid peelable barrier package with carbon monoxide as a component of a low oxygen modified atmosphere package (MAP) system.	Combination
High Pressure Processing (HPP) as a post-lethality, post-packaging intervention method for Listeria monocytogenes contamination in ready-to-eat foods such as deli sliced meats. HPP uses pressures up to 87,000 psi to inactivate pathogens and spoilage organisms throughout the product package.	Physical
Germicidal UVC light systems and equipment for surface decontamination of food products and food contact surfaces.	Physical
Infra-Red Grill is a radiant oven used as a pre-package surface pasteurization for the control of Listeria in RTE products.	Physical
Aquaflow Water Pasteurizer used as a post-package surface pasteurization system either alone or in combination with the Infra-Red Grill system (radiant oven used for pre-package surface pasteurization)for the control of Listeria in RTE products.	Physical
Video Food Safety Technology is a non-intrusive imaging system, which identifies organic contamination on meat and other surfaces utilizing a portable device similar in size and weight to a video camera.	Video
Carcass Inspection System (CIS) is a non-intrusive imaging system which identifies organic contamination in real-time on full carcass (beef) sides on the rail within a slaughter plant.	Video

Also we can ignore effort of ISO 22000 which is planed to harmonise various standards which we have today in different supply chains today and they have few aim.

Global food safety will be achieved only than, when every single link in the food chain will entirely (in its indoor and outdoor environment) become

Conclusion

Meat, as one of the most sensitive industries regarding microbial contamination in food supply chain, deserves all this attention and we need to bring new skills to practice to manage food safety from farm to the fork. The aim of this short review

was to evaluate and compare the few food safety issues which are relevant for meat industry, namely food safety knowledge in practice, employee attitude toward food safety and employee work satisfaction and diversification of the systems connected to meat processing industry. It has to be stressed that all this elements are very complex, in particular when one understand high fluctuation of workers in meat industry. Their knowledge and awareness is constantly unenriched, due to fast regulatory

changes in the area, but also due to social factors which were mentioned before. It looks that the system for food safety assurance is not the weakest at the technological level, as we get impression, but it is the weakest at workers level, which is not always respected as it should be, neither in Good practices nor in HACCP realization. ISO 22000 try to compensate few of this shortcomings, but far the best would be the concept of GMP. The future will ask for its realization.

References

- Aarnisalo, K., Tallavaara, K., Wirtanen, G., Maiala, R., Raaska, L., 2006. The hygienic working practices of maintenance personnel and equipment hygiene in the Finnish food industry. *Food Control*, 17, 1001–1011;
- Azanza, M. P. V., Zamora-Luna, M. B. V., 2005. Barriers of HACCP team members to guideline adherence. *Food Control*, 16, 1, 15–22;
- Baş, M., Şafak, A., Kıvanç, G., 2006. The evaluation of food hygiene knowledge, attitudes, and practices of food handlers' in food businesses in Turkey. *Food Control*, 17, 4, 317–322;
- Bryan F., 1988. Risks of practices, procedures, and processes that lead to outbreaks of foodborne diseases. *Journal of Food Protection*, 51, 663–673;
- Černetič, M., 2001. Vrednotenje dela in motivacija – ravnanje z ljudmi pri delu, Kranj, 2001;
- Clayton, D. A., Griffith, C. J., Price, P., Peters, A. C., 2002. Food handlers' beliefs and self-reported practices. *International Journal of Environmental Health Research*, 12, 1, 25–39;
- Cohen, E., Reichel, A., Schwartz, Z., 2001. On the efficacy of an in-house food sanitation training program: statistical measurements and practical conclusions. *Journal of Hospitality & Tourism Research*, 25, 1, 5–16;
- Collis, B., Winnips, K., 2002. Two scenarios for productive learning environments in the workplace. *British Journal of Educational Technology*, 33, 2, 133–148;
- Ehiri, J. E., Morris, G. P., McEwen, J., 1995. Implementation of HACCP in food businesses: the way ahead. *Food Control*, 6, 6, 341–345;
- Gilling, S. J., Taylor, E. A., Kane, K., Taylor, J. Z., 2001. Successful hazard analysis critical control point implementation in the United Kingdom: understanding the barriers through the use of a behavioral adherence model. *Journal of Food Protection*, 64, 5, 710–715;
- Gilling, S., 2001. *Food Science & Technology Today*, 15, 3, 44–47;
- Griffith, C. J., 2000. *Food safety in catering establishments - Safe Handling of Foods*, Marcel Dekker, New York;
- Henroid, D., Sneed, J., 2004. Readiness to implement hazard analysis and critical control point (HACCP) systems in Iowa schools. *Journal of the American Dietetic Association*, 104, 2, 180–185;
- Hielm, S., Tuominen, P., Aarnisalo, K., Raaska, L., Maijala, R., 2006. Attitudes towards own-checking and HACCP plans among Finnish food industry employees. *Food Control*, 17, 5, 402–407;
- Jannadi, M. O., 1995. Impact of human relations on the safety of construction workers. *International Journal of Project Management*, 13, 6, 383–386;
- Jevšnik, M., Bauer, M., Zorc, A., Raspor, P., 2007. Hygienic status of small and medium sized food enterprises during adoption of HACCP System, *International Journal of Food Science, Technology & Nutrition*, 1, 1, 95–113;
- Jevšnik, M., Hlebec, V., Raspor, P., 2008. Food safety knowledge and practices among food handlers in Slovenia, *Food Control*, 19, 1107–1118;
- Khandke, S. S., Mayes, T., 1998. HACCP implementation: a practical guide to the HACCP plan. *Food Control*, 9, 2-3, 103–109;
- Likar, K., Bauer, M., Jevšnik, M., 2001. Postopek sanitarnega nadzorstva po uveljavitvi HACCP. V: *Praktični pristopi vzpostavljanja in uvajanja HACCP v prehranske obrate*, ur. B. Juteršek, A. Krulec. Ljubljana: Inštitut za sanitarno inženirstvo, pg. 14–20;
- Likar, K., Jevšnik, M., 2004. Pogoji za vzpostavitev učinkovitega notranjega nadzora. V: *Obvladovanje higienskih procesov v vrtcih in domovih za starejše*, ur. N. Ferfila, M. Jevšnik. Ljubljana: Inštitut za sanitarno inženirstvo, pg. 69–78;
- Locke, E. A., Latham, G. P., 1990. *A theory of goal setting and task performance*. Englewood Cliffs, NJ Prentice-Hall;
- Lues, J. F. R., Van Tonder, I., 2007. The occurrence of indicator bacteria on hands and aprons of food handlers in the delicatessen sections of a retail group. *Food Control*, 18, 4, 326–332;
- MacAuslan, E., 2003. The boss, the owner, the proprietor... the food hygiene manager? *The Journal of the Royal Society for the Promotion of Health*, 123, 4, 229–232;
- Manning, C. K., 1994. Food safety knowledge and attitudes of worker's from institutional and temporary food service operations. *Journal of the American Dietetic Association*, 94, 8, 895–897;
- Manning, C. K., Snider, S., 1993. Temporary public eating places: food safety knowledge, attitudes and practices. *Journal of Environmental Health*, 56, 24–28;
- Marolt, J., Gomišček B., 2005. *Management kakovosti*. Kranj: Moderna organizacija, 574 pp.
- Mitchell, R. T., 1998. Why HACCP fails. *Food Control*, 9, 101;
- Mortimore, S., Smith, R. A., 1998. Standardized HACCP training: assurance for food authorities. *Food Control*, 9, 2, 141–145.
- Mortlock, M. P., Peters, A. C., Griffith, C. J., 2000. A national survey of food hygiene training and qualification levels in the UK food industry. *International Journal of Environmental Health Research*, 10, 111–123;
- Panisello, P. J., Quantick, P. C., 2001. Technical barriers to hazard analysis critical control point (HACCP). *Food Control*, 12, 165–173;
- Ramirez-Vela, A., Martin-Fernandez, J., 2003. Barriers for the developing and implementation of HACCP plans: results from a Spanish regional survey. *Food Control*, 14, 5, 333–337;

- Raspor, P., Jevšnik, M., 2008.** Good nutritional practice from producer to consumer. *Critical Reviews in Food Science and Nutrition*, 48, 276–292;
- Raspor, P., 2008.** Total food chain safety: how good practices can contribute? *Trends of Food Science and Technology*, 19, 405–412;
- Regulation EC (2004).** No 852/2004 of the European Parliament and of the Council of 29 April 2004 on the Hygiene of Foodstuffs. *Official Journal of the European Communities*, 18 pp;
- Seaman, P., Eves, A., 2007.** The management of food safety - the role of food hygiene training in the UK service sector. *International Journal of Hospitality Management*, 25, 278–296;
- Shojaei, H., Shooshtaripoor, J., Amiri, M., 2006.** Efficacy of simple hand-washing in reduction of microbial hand contamination of Iranian food handlers. *Food Research International*, 39, 5, 525–529;
- Taylor, E. A., Taylor, J. Z., 2004a.** Perceptions of the »bureaucratic nightmare« of HACCP. A case study. *British Food Journal*, 106, 1, 65–72;
- Taylor, E. A., Taylor, J. Z., 2004b.** Using qualitative psychology to investigate HACCP implementation barriers. *International Journal of Environmental Health Research*, 14, 1, 53–63;
- Vela, A. R., Fernández, J. M., 2003.** Barriers for the developing and implementation of HACCP plans: results from a Spanish regional survey. *Food Control*, 14, 5, 333–337;
- Walker, E., Pritchard, C., Forsythe, S., 2003.** Food handlers' hygiene knowledge in small food businesses. *Food Control* 14, 5, 339–343;
- Williams, A. P., Smith, R. A., Gaze, R., Mortimore, S. E., Motarjemi, Y., Wallace, C. A., 2003.** An international future for standards of HACCP training. *Food Control*, 14, 111–121;
- Yapp, C., Fairman, R., 2006.** Factors affecting food safety compliance within small and medium-sized enterprises: implications for regulatory and enforcement strategies. *Food Control*, 17, 42–51.

Paper received: 14.04.2009.

TRACKING AND TRACING IN THE MEAT AREA*

Schwägele F., Andréé Sabine

Abstract: At pan-European level there is a need for traceability systems that gives information on origin, processing, retailing and final destination of foodstuffs. Such systems shall enhance consumer confidence in food, enable the regulatory authorities to identify and to withdraw health hazardous and non-consumable foodstuffs from the market. Animal feed is an element in this "food-to-farm" approach to public health. Such feedstuffs are preliminary elements of some foods for human consumption, and hence are an inherent element of the food chain.

A harmonised pan-European food traceability protocol would greatly assist authorities in detecting fraud as well as dangerous substances. The food chain comprises a range of sequential and parallel stages bridging the full spectrum from primary production to the consumable foodstuffs. EU legislation on traceability and the technologies needed to implement this system for poultry and poultry products are the focus of this paper.

Key words: tracking, tracing, food, feed, meat, meat products, poultry, poultry meat

Praćenje i sledljivost u oblasti proizvodnje mesa

S a d r Ź a j: Postoji potreba na evropskom nivou za uvođenje sistema sledljivosti koji pruža informacije o poreklu, preradi, maloprodaji i konačnom odredištu namirnica. Ovakvi sistemi bi pojačali poverenje potrošača u hranu, omogućili nadležnima da identifikuju i odstrane sa tržišta namirnice koje su neupotrebljive i štetne po zdravlje. Hrana za životinje je takođe jedan od elemenata u ovom „od njive do trpeze“ pristupu javnom zdravlju. Ona je preliminarni element određene vrste hrane za ljudsku upotrebu i kao takva je inherentni element lanca ishrane.

Harmonizovani, panevropski protokol za sledljivost hrane bi mnogo pomogao nadležnima u otkrivanju prevara, kao i opasnih supstancija. Lanac ishrane obuhvata niz sekvencijalnih i paralelnih nivoa koji povezuju ceo spektar počevši od primarne proizvodnje do namirnica namenjenih ishrani. U fokusu ovog rada je zakonodavstvo EU o sledljivosti i tehnologijama neophodnim kako bi se ovaj sistem implementirao u oblasti uzgoja živine i živinskih proizvoda.

ključne reči: sledljivost, praćenje, hrana, hrana za životinje, meso, proizvodi od mesa, živina, živinsko meso.

1. Introduction

Until the end of the year 2004 food and feed business operators had to conform to the traceability directives demanded by their customers along the entire chain. Large retailers in Europe like Aldi, Lidl, Real, Metro, and Marks and Spencer were very rigorous in their criteria for traceability. But, from 1 January 2005, the new EU regulations mandate that all food and feed business operators be legally bound to have traceability systems, even when their customers do not require it.

The General Food Law, i.e. Regulation (EC) 178 (2002) of the European Parliament and the Council published on 28 January 2002:

- i) outlines the general principles and requirements of food law,
- ii) establishes the European Food Safety Authority and

- iii) provides procedures in matter of food safety, i.e. among other things the implementation of traceability systems in the food and feed supply chains in Europe.

Article 18 of the regulation referring to traceability is effective since 1 January 2005. The following describes the details of the EU legislation on traceability and summarises possibilities for tracing and tracking of poultry and poultry products.

2. European legislation on traceability

Article 18 of Regulation (EC) 178 (2002) refers to traceability and consists of five major points:

1. The traceability of food, feed, food-producing animals, and any other substance intended to be, or expected to be, incorporated into a food or feed shall be established at all stages of production, processing and distribution.

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mauntain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

2. Food and feed business operators shall be able to identify any person from whom they have been supplied with a food, a feed, a food-producing animal, or any substance intended to be, or expected to be, incorporated into a food or feed. To this end, such operators shall have in place systems and procedures, which allow for this information to be made available to the competent authorities on demand.

3. Food and feed business operators shall have in place systems and procedures to identify the other businesses to which their products have been supplied. This information shall be made available to the competent authorities on demand.

4. Food or feed which is placed on the market or is likely to be placed on the market in the Community shall be adequately labelled or identified to facilitate its traceability, through relevant documentation or information in accordance with the relevant requirements of more specific provisions.

5. Provisions for the purpose of applying the requirements of this Article in respect of specific sectors may be adopted in accordance with the procedure laid down in Article 58, paragraph 2, referring to *Committee and Mediation Procedures*.

In particular, Article 58, paragraph 2 of the above Regulation (EC) 178 (2002) says: Where

will immediately initiate procedures to withdraw the food/feed in question from the market where the food/feed has left the immediate control of that initial food/feed business operator and inform the competent authorities thereof.

2.1 Traceability along the full supply chain

The General Food Law covers the entire supply chain [Regulation (EC) 178 (2002), Article 18, paragraph 1]. In order to be able to trace products and retrieve related information, producers must collect information and keep track of products during all stages of production (primary production, processing, distribution, retailing, and consumption). Therefore, traceability can be divided into two key functions, tracking and tracing (Fig. 1). Tracking can be defined as the ability to follow the path of an item as it moves downstream through the supply chain from the beginning to the end. Tracing is the ability to identify the origin of an item or group of items, through records, upstream in the supply chain (Schwägele, 2005). Methodologies for the analyses of the food and feed materials combined with information technology systems are essential to deliver a working tracking and tracing system.

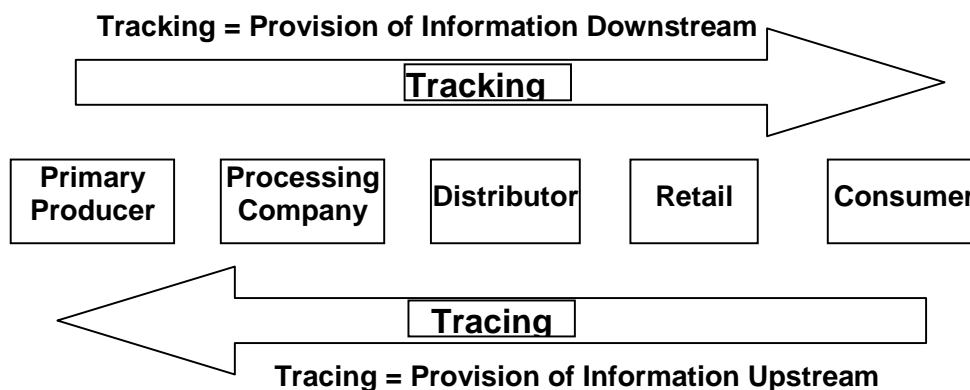


Figure 1. Tracking and tracing along the food chain

Slika 1. Praćenje i sledljivost u lancu ishrane

reference is made to this paragraph, the procedure laid down in Article 5 of Decision (EC) 468 (1999) dealing with regulatory measures shall apply, in compliance with Articles 7 and 8 thereof.

Articles 19 and 20 of Regulation (EC) 178 (2002) cover the responsibilities of food and feed business operators respectively and state that, if an operator considers, or has reason to believe that a food/feed which they have imported, produced, processed, manufactured or distributed is not in compliance with the food/feed safety requirement, they

Previously it was sufficient for a processor to be able to identify the source of an ingredient; now the processor is obliged to ensure that the food products meet the requirements of the Food Law. This implies that the source of all ingredients can be traced and a processor must therefore be able to prove that his supplier can provide full traceability.

If any problem is suspected, tracking must go as far as the consumer. Traceability applies to everything that contributes to food safety, including packaging, closures, seals, jars, etc. Traceability also

covers everything that happens to the products before, during and after the manufacturing, packaging, and distribution. This involves ingredients, processes, test and test results, environment (temperature, time, humidity), resources used (people, machines, knives), transport methods, timescales, etc.

2.2 Implications for food processors

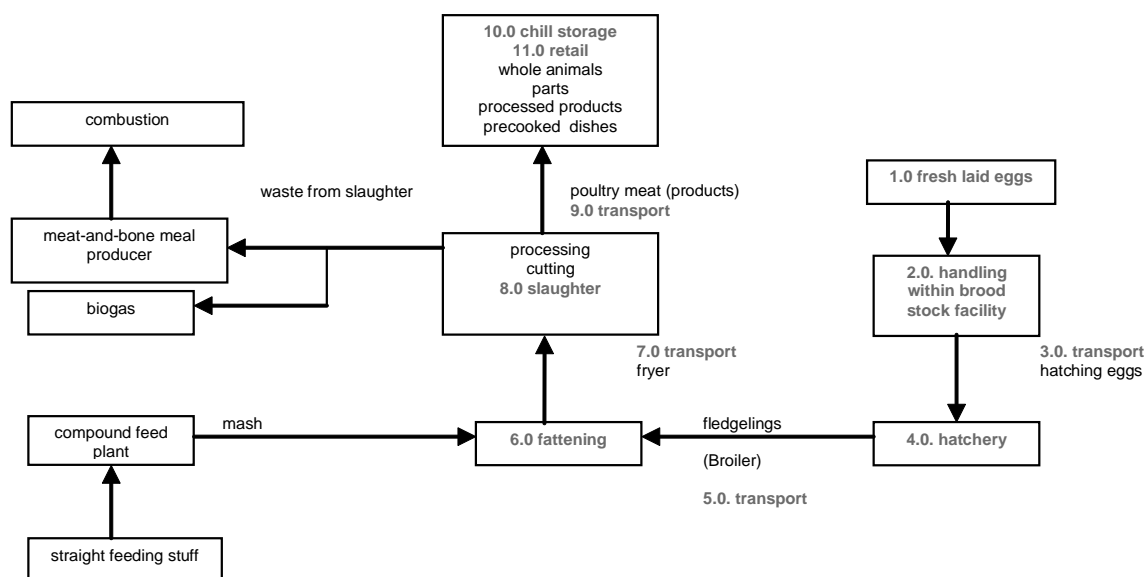
A number of implications exist for food processors, which they will ignore on their peril: more data will have to be recorded on different levels. Who will do this and how will this be done? Data have to be kept for extended periods of time. Therefore, storage and accessibility have to be taken into consideration. Gathered data have to be linked for traceability and have to be highly accurate, as a data error could result in a whole consignment of products being recalled unnecessarily or even lead to a factory shutdown. Data have to be collected and stored quickly. Food processors cannot afford to let data

enquiries through highly integrated traceable data will be required. Food processors must have thoroughly tested proven, infallible systems.

3. Tracking and tracing in the meat area

There are several technologies available that can detect certain characteristics of (or elements in) foodstuffs derived from animal tissue. Some of these technologies can be used to make definite inferences regarding the foodstuff's origin or history, while others can only be used to confirm the presence of specific components.

With respect to tracking and tracing along the full supply chain, for instance in the case of poultry and poultry products (Fig. 2.), the following aspects are of importance. They shall, if possible, give information on poultry species, origin, authenticity, age, composition and production systems (including feed).



According to: Geflügelmast in Deutschland; Georg von Bittner und Hans-Wilhelm Windhorst; Weiße Reihe; Band 24; 2005

Figure 2. Generic poultry production chain (according to: von Bittner, Windhorst, 2005)

Slika 2. Opšta šema lanca živinske proizvodnje (prema Fon Bittner, Windhorst, 2005)

collecting affect their production costs. All of this has to be achieved at the lowest cost possible. Food processors cannot rely on paper records, systems that are not linked together or manual data entry. Automated data logging is the only possible option. Food processors will need integrated traceability data through production, storage, selling and quality control. Systems designed to provide instant trace

3.1. Species identification – protein, fatty acids and DNA based methods

It is necessary to have reliable methods, which allow a fast and unequivocal identification of poultry species. Potential analytical targets proved to be proteins, DNA or lipids.

3.1.1 Protein based methods

Proteins (enzymes, myoglobin, etc.) have been widely used as animal species markers. Applicable techniques include separation of water-soluble proteins by starch, polyacrylamide and agarose gel electrophoresis (Cowie, 1968; Mackie, 1980) or isoelectric focusing (IEF) (Hofmann, 1986; Jemmi and Schlosser, 1993). Highly resolved water-soluble protein patterns can be used to differentiate genetically close-related animal species (Hofmann and Blüchel, 1986). However, the mentioned gel electrophoretic methods proved to be not practical and reliable for species identification in composed poultry products consisting of mixtures of different poultry species (Hofmann, 1997).

Immunological techniques like Western-Blotting (Schwägele, 2001) and a specific type of enzyme immuno assay (EIA), the so called "enzyme linked immuno sorbent assay" (ELISA) (Schwägele, 2001) performed on the solid surface of microwell plates are using suitable target proteins for analysis. A qualitative detection of animal species is possible and the limit of detection depends upon their content in meat products (pork $\leq 1\%$; poultry (in general) and beef $\leq 2\%$; sheep $\leq 5\%$).

Proteomics can be used to try to differentiate species, breeds, and varieties by their specific protein pattern (Meketowa, Abbas-Hawks, Vorhees and Hadfield, 2003).

3.1.2 Lipid based methods

Lipid components and fatty acids can serve as target substances for animal species identification. The percentage of the composition between saturated, monounsaturated and polyunsaturated fatty acids is a possible animal species marker, which can be determined by means of gas chromatography (GC) or gas chromatography coupled with mass spectroscopy (GC-MS). However, analytical practice shows that this method is tainted with large variations leading to less reliable results in single species identification and furthermore in composed meat products consisting of mixtures of different animal species. (Honikel, Gempel and Schwägele, 2002).

3.1.3 DNA based methods

In recent years, DNA analytical techniques have been applied to food research and food control. The first DNA tests for species identification in foods were performed using specific DNA probes in hybridisation assays (Chikuni, Ozutsumi, Koishikawa and Kato, 1990; Wintero, Thomsen and Davies, 1990). Polymerase Chain Reaction (PCR) has been developed into a key technology for species

identification in foods and feeds (Saiki, Gelfand, Stoffel, Scharf, Higuchi, Horn, Mullis, and Erlich, 1988). PCR-RFLP (restriction fragment length polymorphism) has been used for the species identification of food relevant animals and plants (Meyer, Höfelein, Lüthy and Candrian (1995); Verkaar, Boutaga, Nijman and Lenstra, 2001).

Random amplified polymorphic DNA-PCR (RAPD-PCR) as well as assays based on single strand conformation pattern (SSCP) were developed for species and variety-specific identification of different animals and plants (Kaemmer, Afza, Weising, Kahl and Novak, 1992; Rehbein et al., 1999; Weder, 2002). Many species-specific PCR systems have been described for animal and plant species (Behrens, Unthan, Brinkmann, Buchholz and Latus, 1999; Kingombe, Lüthi, Schlosser, Howald, Kuhn and Jemmi, 2001; Altmann, Binke and Schwägele, 2004). Even in foods that have been produced under severe processing conditions (e.g. sterilisation) DNA techniques are effective. The limit of detection is usually $\leq 0.1\%$, but is dependent upon the PCR method (Schwägele, 2003). Recently a PCR method was developed for authentication of the most common poultry species in very complex samples with high sensitivity (Stirtzel, André, Seuß-Baum & Schwägele, 2007; Fig. 3).

Species identification and quantification can also be performed using real time PCR (Wurz, Bluth,

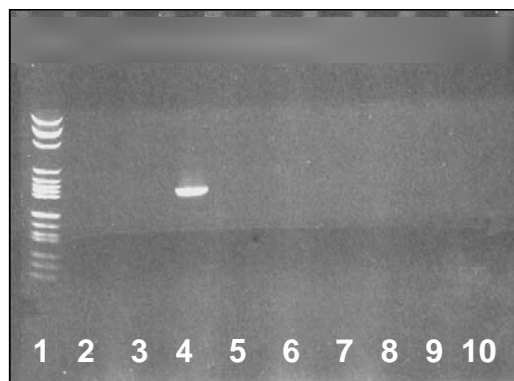


Figure 3. Detection of guinea fowl in commercially available meat-products using the primer system for guinea fowl; 1: marker pBR322, 2: quail terrine, 3: pheasant terrine, 4: guinea fowl terrine, 5: goose à l'Orange, 6: muscovy duck savoury, 7: turkey-Gelbwurst, 8: poultry-wiener, 9: pig, 10: NTC

Slika 3. Detekcija prisustva gvinejske peradi u komercijalnim proizvodima od mesa korišćenjem prajmer sistema za gvinejsku perad: 1: marker pBR322, 2: prepelica; 3. fazan; 4. gvinejska perad; 5. guske; 6. Moskovi patka; 7. ćurka Gelbwurst; 8. živinska kobasica; 9. svinja; 10. NTC

Zeltz, Pfeifer and Willmund R., 1999). In general, these techniques are more developed and reliable for the quantification of genetically modified organisms (Pöpping, 2001) than for natural animal or plant species (Binke, Altmann, Fischer, Müller and Schwägele, 2004; Binke, Altmann and Schwägele, 2003).

DNA sequence information can be used for species identification. The development of modern molecular biological techniques, including various sequencing techniques, has led to a large number of DNA sequences. Unfortunately, not all of them are available in the various DNA databases. For species identification, the mitochondrial DNA (mtDNA) is the most widely used target molecule. The main reason to use mtDNA for this kind of analysis is the availability of numerous sequences in databases and the high genetic variability of mtDNA, which allows sophisticated primer design for sequencing. DNA sequencing is theoretically the most informative and precise technique but requires samples consisting only of a single species. Sequencing allows species identification without reference material, if the generated sequence is available in a database. The technique also has been named FINS (Forensically Informative Nucleotide Sequencing; Bartlett and Davidson, 1992).

3.2 Authenticity, geographical origin and detection of fraud

To ensure authenticity as well as geographical origin and to detect fraud in the area of meat and meat products the above-mentioned electrophoretic, chromatographic, and molecular biological methods combined with other chemical and physical procedures can be very effectively applied to traceability as noted below.

- i) Protected Designation of Origin (PDO)
PDO covers the term used to describe foodstuffs, which are produced, processed, and prepared in a given geographical area using recognized methodology (Dinde de Bresse; Parma ham, Jamon de Terual).
- ii) Protected Geographical Indication (PGI)
This geographical link must cover at least one of the stages of production, processing or preparation. Furthermore the product can benefit from a good reputation (Canard à foie gras du Sud-Ouest; Pollo y Capón del Prat; Schwarzwälder Schinken; Nürnberger Bratwürste; Thüringer Rostbratwürste).
- iii) Certificate of Specific Character (CSC)
CSC means recognition of all member states of the EU that a foodstuff possesses specific characteristics, which distinguish it clearly

from similar products in the same category (Münchner Weißwürste; Salami Milanese).

3.2.1 NMR and MS based methods

Authentication strategies involving the use of multi-isotopic parameters (^2H , ^{13}C , ^{15}N , ^{18}O , ^{34}S and ^{87}Sr) facilitated by increasingly rapid measurement procedures present a complex analytical challenge because of many compounding factors, such as imported feed, origin of animal tissue, and metabolic turnover of tissue-specific substances.

Stable isotope analyses are considered an excellent tool for origin assessment. The ratio $^{13}\text{C}/^{12}\text{C}$ gives straightforward responses concerning the primary photosynthetic metabolism of feed plants (O'Leary, 1981), and the ratios of the stable isotopes of oxygen ($^{16}\text{O}/^{18}\text{O}$) and hydrogen ($^2\text{H}/^1\text{H}$) are good indicators of environmental conditions e.g. H_2O (Ziegler, Osmond, Stichler and Trimborn, 1976) and enables the tracing of the origin of animal material. The two main techniques used to determine the isotope ratios of natural products are isotope ratio mass spectrometry (IRMS) and site-specific natural isotope fractionation from nuclear magnetic resonance (SNIF-NMR). NMR has the advantage over IRMS in that the natural abundance of ^2H isotopomers may be precisely identified in compounds and accurately quantified by SNIF-NMR (Martin and Martin, 1991), whereas IRMS only gives a mean value of the deuterium content of a given chemical species.

Both, low and high resolution NMR can be used for the detection of plant species and genetically modified plant or animal material in food, but specific marker components must be isolated prior to analysis.

The geographic origin of a foodstuff can affect its composition and associated food-borne risks to the "food-to-farm" chain. Also, less expensive ingredients or components of dubious geographical origin may be fraudulently included for monetary gain. A need exists to develop a protocol enabling a foodstuff's geographic origin to be assessed. Techniques can be used to "fingerprint" the geographic origins of certain plant and animal materials and these methodologies can form part of a suite of traceability tests (Polychroniadou and Vafopoulou, 1985). Geographical effects arise due to differences in the geological origin of the soils, soil pH, anthropogenic contaminants, atmospheric and climatic differences and the interaction among certain trace elements. Zoonoses risks can vary considerably from one country to another (e.g. H5N1 risk in China >> Germany). Trace element analysis by inductively coupled plasma mass spectroscopy (ICP-MS) has

been used to determine the geographic origin of soils and plants (Anderson, Magnuson, Tschirgi and Smith, 1999). Trace element signatures can be used to identify the geographical provenance of a sample because organisms accumulate in their tissues, from the water, food and air, the elements available from the environment where they live. Differences in the isotope distributions of these trace elements among different geographical locations give different “signatures” of isotopes in the organic tissues.

GC-MS and liquid chromatography in combination with mass spectroscopy (LC-MS) have been successfully applied to the analysis of organic contaminants (PCBs, Dioxins, etc) in the origin of various feed and food materials.

3.2.2 Infrared spectroscopy

Both near infrared (NIR) and mid infrared (MIR) spectroscopy can be used for analysis of the main components of foods as well as animal feeds inclusive minerals and vitamins. Pires, Lemos and Kessler (2001) demonstrated the potential of NIR to measure the concentration of 11 vitamin levels in poultry feeds. Garnsworthy, Wiseman and Fegeros (2000) reported the application of NIR to the prediction of chemical, nutritive, and agronomic characteristics of wheat.

3.3 Traceability of production process and storage

To determine the “history of meat and meat products” with respect to the production processes and changes occurring during storage, a number of technologies (DNA based methods; electrophoresis including capillary electrophoresis CE; immunological methods; high pressure liquid chromatography HPLC including HPLC-MS; lipid based methods GC, GC-MS, and GCxGC-MS; IR and NMR spectroscopy; Electron Microscopy can be used.

One of the most important but widely unresolved issues in food traceability is to quantify the degree of batch mixing associated with a given blend of raw materials. There is a need for considerable research designed to address this issue.

The reliable use of “tracer substances” has to be investigated since they can be used to augment details concerning batch mixing (e.g. detection of enzyme activities and proteomics serving as indicators for the degree of sterilisation.). Tracers can be endogenous (i.e. compounds present in the food due to its make up or processing history) or purposely added to facilitate detection. However, adding tracers needs to be carefully considered as the tracer must not be harmful to the end users and must comp-

ly with all legislative requirements. For example, endogenous tracers can be used for fermented, Hungarian style salami, where possible tracer techniques include testing for lipid degradation, lactic acid or volatile components that occur during the ripening process. In addition, holistic (i.e. measuring nearly all compounds) analysis of all compounds in food (metabolites and proteins) and multivariate statistics can be used to characterise food. Characteristic metabolite profiles of foodstuffs can be obtained by holistic analytical methods (GC-MS, LC-MS, and NMR). Bioinformatics can be used to develop models and identify clusters of compounds correlating with certain production methods (organic processing, conservation, etc) and ingredients. This would allow the identification of new (endogenous) markers for the production methods, origin and others. If methods and tools developed especially for metabolite analysis are available, other natural tracers, such as specific isotopes, are not necessary for this purpose. The same strategy could be applied to proteins using techniques and tools developed for proteomics.

In many cases it is possible to infer the degree of sterilisation through certain indicators, such as the degree of protein degradation or the degradation of a marker added to the material prior to the sterilisation step. The addition of tracers is a very powerful adjunct to normal traceability techniques.

Isaksson, Ellekjaer and Hildrum (1989) and Ellekjaer and Isaksson (1992) concluded that NIR could be used for determination of heat treatments in the temperature range 50 to 85° C with an associated prediction error of 2.0 to 2.1 K. Thyholt, Enersen and Isaksson, (1998) described the use of NIR reflectance spectroscopy to determine endpoint temperature in previously heated meat.

Despite the high costs and consumer concerns, the number and quantity of foods being irradiated is increasing steadily. Currently about 250,000 tons of food are irradiated annually. In the USA and Europe it is a requirement that irradiated food products must be labelled. However, monitoring programs are in place in only a few European countries.

One of the significant challenges to identify irradiated food products is the different techniques necessary to cover the entire spectrum of products. Typical methods used include immunological methods, comet assay, photon-stimulated luminescence, thermoluminescence, and electron spin resonance. However, only a limited number of laboratories worldwide have the necessary capability for the reliable determination of food irradiation.

3.4 Cross contamination or carry over in food and feed

In several food production facilities, ingredients or raw materials are used that are known to have allergenic properties in human, e.g. egg and milk proteins. Subsequent processing of products using the machines or transport facilities previously used for allergen containing products, may lead to cross contamination of allergens to products not intended to contain these allergens. Manufacturers of food products should therefore have a high awareness of the risks of cross-contamination of allergenic proteins during the production process of their products. Knowledge of threshold levels for sensitive patients, the use of specific ingredients, cleaning strategies, etc. is helpful to reduce unwanted contamination with allergens. This information can be used to identify (within a given level of tolerance) the critical control points during processing and the aspects to be monitored for the most effective tracking information to be generated.

The same considerations apply to the manufacture of animal feeds formulated to contain antibiotics, coccidiostats and similar components. The use of veterinary drugs within the European Union is regulated by means of the Council Regulation (EEC) No. 2377/90. The prohibition of the use of growth promoting substances, such as hormones or β -agonists, is established with Council Directives No. 96/22/EC and 2003/74/EC. Since January, 1st 2006 according to Regulation (EC) No. 1831/2003 the use of antibiotic growth promoting substances as additives for use in animal nutrition is forbidden. However certain substances with coccidiostatic and histomonostatic effects are still considered as feed additives according to Regulation (EC) No. 1831/2003. If feed-mixing facilities are used to make feed with and without veterinary drug pre-mixes or feed additives, cross contamination is a distinct possibility and appropriate controls are essential.

3.5 Application of biosensors

Immunosensors, based on the antibody antigen recognition, are rapid, simple and sensitive methods that have been developed for the measurement of a wide range of target compounds such as bacteria (*Yersinia pestis*), alpha-toxin, ricin, brevetoxin, okadaic acid (Vaughan, Geary, Pravda and Guilbault, 2003), pesticides such as atrazine (Schipper, Rauchalles, Kooyman, Hock and Greve, 1998) and veterinary drug residues (Baxter, O'Connor, Haughey, Crooks and Elliott, 1999). These techniques offer considerable potential for traceability within the full poultry chain.

The aim of immunosensors is to develop a system capable of performing a single point determination without calibration between each measurement. Various transduction systems, based on potentiometry (Khomutov, Zherdev, Dzantiev and Reshetilov, 1994), electrochemiluminescence (Marquette, Coulet and Blum, 1999) and chemiluminescence (Samsonova, Baxter, Crooks, Small and Elliott, 2001) have been used successfully.

Biosensors basically have two components, biological or sensor molecules and a signal transducer. The biological component consists of an antigen or antibody. The transducer detects the change in one or more physicochemical properties of the biological molecule. Increasing attention is being paid to the development of immunobiosensors, especially to assay clinical samples. This technology uses novel biosensor techniques which can combine very specific antibody-antigen interaction with very sensitive signal transduction to enable faster, more sensitive and reliable techniques, which can also be applied to routine monitoring and quality control protocols in the food chain.

The most commonly used biosensors are the piezo electric (PZ) crystal, where the PZ crystal oscillator can be used as a microbalance to detect a change in mass of the crystal due to the formation of antigen-antibody complex, thus permitting it to be utilized as an immunobiosensor. Immuno-electrode and optic fibre biosensors have been used for the detection of Ivermectin in animal carcasses (Samsonova, Baxter, Crooks, Small and Elliott, 2001).

3.6. Tracking technology

Electronic data management (Automatic Identification and Data Capture [AIDC]) plays an important role in improving operational efficiency and accuracy of information handling in the "food to farm" chain. Since there are no industry standards for handling electronic data through out the complete food chain, the use of the European Article Numbering Association codes (EAN-UCC, 2002) is proposed to improve data tracking. For successful operation of this technology, the environment in which it operates must be relatively clean and this is not always achievable on the farm.

Technologies such as RFID (Radio Frequency Identification) overcome this problem by using radio signals instead of line of sight for identification, and can be integrated into a prototype recording system. However, product identifiers (tags) are not currently in widespread use, and are expensive in comparison to the barcode. Matrix codes are 2D, but information is stored by blanking out areas of a defined array, rather than in bars. These codes are generally only

used in specialist applications, including the marking of very small components. Scanners can operate with a 90% success rate where contamination levels are kept below 10% and barcodes are kept clean and undamaged. The performance of the laser scanner is such that any level of contamination will substantially reduce read success rate. Studies undertaken by *Watts, Miller and Godwin (2003)* indicate that the RFID achieve successful reads over 98% of the time, with unprotected and reused tags.

In electronic tracking and tracing systems, EAN-UCC (2002) is universally accepted as an identification and communication system that facilitates efficient global commerce and improves the effectiveness of recording and exchanging information between supply chain participants. The system uniquely identifies products, locations, services and assets and also includes a series of standard data structures known as Application Identifiers (AIs), which allow secondary information about a product such as batch, expiry and lot number to be encoded.

The EAN-UCC (2002) system consists of 3 components:

- i) Identification Numbers - used to identify a product, location, logistic unit, service or asset.
- ii) Data Carriers - the barcodes or radio frequency tags used to represent these numbers. The data carriers vary according to the level of information required or the space available. For space-constrained products, the use of reduced space symbology (RSS) barcode is ideal. For traceability purposes, an EAN 128 barcode is used to encode the identification and supplementary information relating to an item.
- iii) Electronic Messages - the means of connecting the physical flow of goods with the electronic flow of information. These technologies have been used in meat traceability, providing a robust tracking system for most elements of the meat chain (Harmonised Electronic Data Interchange, HEDI). Such electronic tracking systems play a key role in food labelling.

3.7 Computer modelling and risk assessment

Computer modelling can be a powerful tool to estimate the contamination and transmission pathways for pathogens and food contaminants. It can also help to assess the reliability and accuracy of a decision tree, composed of a suite of test pathways. Many epidemiological parameters have been estimated using models where direct measurement is

almost impossible. Risk assessment modelling can be used to help manage food chain risk and make policy decisions regarding the safety of the food chain from food-to-farm. Any food traceability system requires associated risk assessment models in order to evaluate the potential health risks to humans and animals (*Greiner, Mueller-Graf, Hiller, Schrader, Gervelmeyer, Ellerbroek and Appel, 2007; Serratos, Ribo, Correia and Pittman, 2007*). *Stark, Boyd and Mousing (2002)* illustrated how available information can be organised systematically within a risk model and a quantitative decision support can be provided quickly making optimal use of all available information. Risk assessment methodologies are being used increasingly to quantitatively assess risks to human health imposed by the food chain.

4. Conclusions

Regulation (EC) 178 (2002):

- i) stipulates that the delivery of safe food and animal feed belongs to specific food and feed producers,
- ii) specifies that foodstuffs, animal feed and feed ingredients must be traceable,
- iii) includes clear procedures for developing food law and dealing with emergencies,
- iv) gives the European Commission new powers to take emergency measures when national authorities are unable to contain an emerging food risk,
- v) establishes the “Standing Committee on the Food Chain and Animal Health, in the place of three Standing Committees”, bringing together Member States representatives with important roles in decision-making on food safety issues.

In the area of poultry and poultry products there is a need for fast and reliable systems to enable traceability along the full chain to provide safe and high quality food for the consumer with respect to origin and processing. Traceability cannot only be considered as a request of the legislation addressed to the food business operators (primary production, processing, distribution, retailing, and consumption); moreover it has to be their very own interest in terms of product liability to find practicable ways to implement the new regulation. Within the 5th and 6th framework program, the European Commission has funded various research and development projects such as [MOLSPEC-ID (2004); ENOSEFOODMICRODETECT (2003); QUALITYLOWINPUTFOOD (2005); ENTRANS-FOOD (2003); ΣChain (2006)] dealing with traceability along the food chain.

References

- Altmann Katrin, Binke, R., Schwägele, F., 2004.** Qualitativer Nachweis von Ziege in Fleisch- und Milcherzeugnissen – Nachweis auf Basis des nukleären single-copy Gens beta-casein. *Fleischwirtschaft*, 84, 115–116;
- Anderson, K. A., Magnuson, B. A., Tschirgi, M. L., Smith, B., 1999.** Determining the geographic origin of potatoes with trace element analysis using statistical and neural network classifiers. *Journal of Agricultural Food Chemistry*, 47, 1568–1574;
- Bartlett, S. E., Davidson, W. S., 1992.** FINS (Forensically Informative Nucleotide Sequencing): a procedure for identifying the animal origin of biological specimens. *Biotechniques*, 12 (3), 408–411;
- Behrens, M., Unthan, M., Brinkmann, Y., Buchholz, R., Latus, N., 1999.** Identification of animal species in heated and complex meat products using species specific PCR reactions. *Fleischwirtschaft International*, 6, 16–21;
- Baxter, G. A., O'Connor, M., Haughey, S. A., Crooks, S. R. H., Elliott, C. T., 1999.** Evaluation of an immunobiosensor for the on-site testing of veterinary drug residues at an abattoir. *Analyst*, 124 (9), 1315–1318;
- Binke, R., Altmann Katrin, Fischer Karin, Müller Edith, Schwägele, F., 2004.** Semiquantitative Bestimmung von Ziegengewebe in Fleischerzeugnissen mittels PCR: Bestimmung auf Basis der nucleären single-copy Gene *beta-Casein* and *Myostatin*. *Mitteilungsblatt BAFF*, 43 (164) 155–161;
- Binke, R., Altmann Katrin, Schwägele, F., 2003.** Influencing factors for the quantification of animal species in meat by means of PCR. *Innovations in Food Technology*, 21, 130.
- Chikuni, K., Ozutsumi, K., Koishikawa, T., Kato, S., 1990.** Species identification of cooked meats by DNA hybridisation assay. *Meat Science*, 27, 119–128.
- Directive 2003/74/EC, 2003:** of the European Parliament and of the Council amending Council Directive 96/22/EC concerning the prohibition on the use in stock farming of certain substances having a hormonal or thyrostatic action and of beta-agonists. *Official Journal of the European Union*, L 28, p. 45–50;
- Council Regulation (EEC) No 2377/90, 1990:** laying down a Community procedure for the establishment of maximum residue limits of veterinary medicinal products in foodstuffs of animal origin *Official Journal of the European Union*, L 224, p. 1–8;
- Council Directive 96/22/EC, 1996:** concerning the prohibition on the use in stock farming of certain substances having a hormonal or thyrostatic action and of β -agonists, and repealing Directives 81/602/EEC, 88/146/EEC and 88/299/EEC *Official Journal of the European Union*, L 125, p. 3–9;
- Cowie, W., 1968.** Identification of fish species by thin slab polyacrylamide gel electrophoresis. *Journal of the Science of Food and Agriculture*, 19, 226–229.
- Decision (EC) 468, 1999.** Laying down the procedures for the exercise of implementing powers conferred on the Commission. *Official Journal of the European Communities*, L 184/23 – L184/26;
- Directive (EC) 89, 2003.** Amending Directive (EC) 13 (2000) as regards indication of the ingredients present in foodstuffs. *Official Journal of the European Communities*, L 308/15 – L308/18.
- EAN-UCC, 2002.** European Article Numbering Association. EAN International and the Uniform Code Council. Available: <http://www.ean.ucc.org>;
- Ellekjaer, M. R., Isaksson, T., 1992.** Assessment of maximum cooking temperatures of previously heat treated beef. Part 1: Near Infrared Spectroscopy. *Journal of the Science of Food and Agriculture*, 59, 335 – 343;
- ENOSEFOODMICRODETECT, 2003.** 5th Framework Programme (EC) Project. Rapid detection of microbial contaminants in food products using electronic nose technology. Available: <http://www.e-nose.net>;
- ENTRANSFOOD, 2003.** 5th Framework Programme (EC) Project. European network safety assessment of genetically modified food crops. Available: <http://www.entransfood.com>.
- Gajendragad, R., Kamath, K. N. Y., Anil, P. Y., Prabhudas, K. Natarajan, C., 2001.** Development and standardization of a piezo electric immunobiosensor for foot and mouth disease virus typing. *Veterinary Microbiology*, 78, 319–330;
- Garnsworthy, P., Wiseman, J., Fegeros, K., 2000.** Predication of chemical, nutritive and agronomic characteristics of wheat by NIR spectroscopy. *Journal of Agricultural Science*, 135, 409–417;
- Greiner, M., Mueller-Graf, C., Hiller, P., Schrader, C., Gervelmeyer, A., Ellerbroek, L., Appel, B., 2007.** Expert opinion based modelling of the risk of human infection with H5N1 through the consumption of poultry meat in Germany. *Berliner und Münchener Tierärztliche Wochenschrift*, 120, 98 – 107;
- Hofmann, K., 1986.** Grundlegende Probleme bei der Identifizierung der Tierart von Muskelfleisch mit Hilfe elektrophoretischer Methoden. *Fleischwirtschaft*, 66, 91 – 98;
- Hofmann, K., 1997.** Nachweis der Tierart bei Fleisch und Fleischerzeugnissen. *Fleischwirtschaft*, 77, 151 – 154;
- Hofmann, K., Blüchel, E., 1986.** Bestimmung der Tierart von rohem Muskelfleisch anhand der Myoglobinemuster im pH-Gradienten-Gel. *Fleischwirtschaft*, 66, 916 – 921;
- Honikel, K. O., Gempel Gabriele, Schwägele, F., 2002.** Tierartidentifikation auf Protein-, DNA- und Fettsäure-Basis bei Fleisch, Fleischerzeugnissen und Tiermehl. *Mitteilungsblatt BAFF*, 41(156), 125–133;
- Isaksson, T., Ellekjaer, H. R., Hildrum, K. I., 1989.** Determination of the previous maximum temperature of heat treated minced meat by NIRS. *Journal of the Science of Food and Agriculture*, 69, 385–387;
- Jemmi, T., Schlosser H., 1993.** Tierartbestimmung aus mariniertem und erhitztem mariniertem Fleisch mittels isoelektrischer Fokussierung. *Fleischwirtschaft*, 73, 600–602;
- Khomutov, S. M., Zherdev, A.V., Dzantiev, B. B., Reshetilov, A. N., 1994.** Immunodetection of herbicide 2,4-dichlorophenoxyacetic acid by field-effect transistor-based biosensors. *Analytical Letters*, 27, 2983–2995;
- Kaemmer, D., Afza, R., Weising, K., Kahl, G., Novak, F. J., 1992.** Oligonucleotide and amplification fingerprinting of wild species and cultivars of banana (*Musa spp.*). *Biotechnology*, 10, 1030 – 1034;
- Kingombe C. I. B., Lüthi E., Schlosser H., Howald, D., Kuhn M., Jemmi T., 2001.** A PCR-based test for species-specific determination of heat treatment conditions of animal meals as an effective prophylactic method for bovine spongiform encephalopathy. *Meat Science*, 57, 35–41;
- Mackie, I. M., 1980.** A review of some recent applications of electrophoresis and iso-electricfocusing in the identification of fish species in fish and fish products. In J.J. Connell, *Advances in fish science and technology*, London: Fishing News Books, pp. 444–450;
- Marquette, C. A., Coulet, P. R., Blum, L. J., 1999.** Semi-automated membrane chemiluminiscent immunosensor for flow injection analysis of okadaic acid in mussels. *Analytica Chimica Acta*, 398, 173–182;

- Martin, G. J., Martin, M. L., 1991. Deuterium labelling at the natural abundance level as studied by high field quantitative $^2\text{H-NMR}$. *Tetrahedron Letters*, 22, 3525–3528;
- Meketowa, P., Abbas-Hawks, C., Vorhees K. J., Hadfield T. L., 2003. Microorganism gram type differentiation of whole cells based on pyrolysis high resolution mass spectrometry data. *Journal of Analytical and Applied Pyrolysis*, 211, 213–217.
- Meyer, R., Höfelein, Ch., Lüthy, J., Candrian, U., 1995. Polymerase Chain Reaction-Restriction Fragment Length Polymorphism analysis: a simple method for species identification on food. *Journal of AOAC International*, 78 (6), 1542–1551;
- MOLSPEC-ID, 2004. 5th Framework Programme (EC) Project. *Development of quantitative and qualitative molecular biological methods to identify plant and animal species in foods*. Available: <http://www.molspec.org>;
- O'Leary, M., 1981. Carbon isotope fraction in plants. *Phytochemistry*, 20, 553–567.
- Pires F., Lemos, M. C., Kessler, A. M., 2001. Use of NIR reflectance spectroscopy to analyse vitamin content. *Journal of Applied Poultry Research*, 14 (4), 412–418;
- Polychroniadou, A., Vafopoulou, A., 1985. *Journal of Dairy Science*, 68, 147–150;
- Pöpping, B., 2001. Are you ready for a roundup? - What chemistry has to do with genetic modification. *Journal of Chemical Education*, 78, 752–756;
- Regulation (EC) 178, 2002. Laying down the general principles and requirements of food law, establishing the European Food Safety Authority and laying down procedures in matters of food safety. *Official Journal of the European Communities*. L31/1 – L31/24;
- Regulation (EC) No 1831/2003, 2003: of the European Parliament and of the Council on additives for use in animal nutrition. *Official Journal of the European Union*, L 268, p. 29–43;
- Rehbein, H., Mackie, I. M., Pryde, S., González-Sotelo, C., Medina, I., Pérez-Martín, R. I., Quinteiro, J., Rey-Méndez, M., 1999. Fish species identification in canned tuna by PCR-SSCP: validation by a collaborative study and investigation of intra-species variability of the DNA patterns. *Food Chemistry*, 64, 263–268;
- QUALITYLOWINPUTFOOD, 2005. 6th Framework Programme (EC) Project. Improving quality and safety and reduction of cost in the European organic and “low input” food supply chains. Available: <http://www.qlif.org>;
- Saiki, R. K., Gelfand, D. H., Stoffel, S., Scharf, S. J., Higuchi, R., Horn, G.T., Mullis, K. B., Erlich, H. A., 1988. Primer-directed enzymatic amplification of DNA with a thermostable DNA polymerase. *Science*, 239, 487–490;
- Samsonova, J. V., Baxter, G. A., Crooks, S. R. H., Small, A. E., Elliott, C.T., 2001. Determination of ivermectin in bovine liver by optical immunobiosensor. *Biosensors and Bioelectronics*, 17, 523–529;
- Serratos, J., Ribo, O., Correia, S., Pittman, M., 2007. EFSA scientific risk assessment on animal health and welfare aspects of avian influenza (EFSA-Q-2004-075). *Avian Diseases*, 51 (1), 501–503;
- Schipper, E. F., Rauchalles, S., Kooyman, R. P. H., Hock, B., Greve, J., 1998. The waveguide mach-zender interferometer as atrazine sensor. *Analytical Chemistry*, 70, 1192–1197;
- Schwägele, F., 2001. Analytik bei Fleisch. Bewertung immunologischer und gentechnischer Methoden. *Fleischwirtschaft*, 81, 78–81;
- Schwägele, F., 2003. Noch Forschungsbedarf bei PCR. *Fleischwirtschaft*, 83, 78–79;
- Schwägele, F., 2005. Traceability from a European perspective. *Meat Science*, 71, p. 164–173;
- ΣChain, 2006: 6th Framework Programme (EC) Project. Developing a Stakeholders' Guide on the vulnerability of food and feed chains to dangerous agents and substances. (FP6 – 518451) Available: <http://www.sigmachain.eu>;
- Stark, K. D. C., Boyd, H. B., Mousing, J., 2002. Risk assessment following the hypothetical import of dioxin-contaminated feed for pigs - an example of quantitative decision-support under emergency conditions. *Food Control*, 13, 1–11;
- Stirtzel Sonja, André Sabine, Seuß-Baum Ingrid, Schwägele, F., 2007. Authentifizierung der gebräuchlichsten Geflügelarten mittels PCR. *Fleischwirtschaft*, 87, 86–89;
- Thyholt, K., Enersen, G., Isaksson, T., 1998. Determination of endpoint temperatures in previously heat treated beef using reflectance spectroscopy. *Meat Science*, 48, 1/2, 49–63;
- Vaughan, R. D., Geary, E., Pravda, M., Guilbault, G. G., 2003. Piezoelectric immunosensors for environmental monitoring. *International Journal of Environmental & Analytical Chemistry*, 83, 555–571;
- Von Bittner, G., Windhorst, H., 2005. Geflügelmast in Deutschland. *Weißer Reihe*; Band 24;
- Verkaar, E. L. C., Boutaga, K., Nijman, I. J., Lenstra, J. A., 2001. Differentiation of bovine species in beef by PCR-RFLP of mitochondrial and satellite DNA. *Meat Science*, 60, 365–369;
- Watts, A. J., Miller, P. C. H., Godwin R. J., 2003. Automatically recording sprayer inputs to improve traceability and control. In *Proceedings of the 2003 BCPC Crop Science and Technology Conference*. (pp. 323 - 328), Glasgow: BCPC publications UK;
- Weder, J. K., 2002. Identification of plant food raw material by RAPD-PCR: legumes. *Journal of Agricultural Food Chemistry*, 50, 4456–4463;
- Wintero, A. K., Thomsen P. D., Davies, W., 1990. A comparison of DNA-hybridization, immunodiffusion, counter current immunoelectrophoresis and isoelectric focusing for detecting the admixture of pork to beef. *Meat Science*, 27, 75 – 85;
- Wurz, A., Bluth, A., Zeltz, P., Pfeifer, C., Willmund R., 1999. Quantitative analysis of genetically modified organisms (GMO) in processed food by PCR-based methods. *Food Control*, 10, 385 – 389;
- Ziegler, H., Osmond, C. B., Stichler, W., Trimborn, P., 1976. Hydrogen isotope discrimination in higher plants: correlations with photosynthetic pathway and environment. *Planta*, 128, 85 –92.

STATE-OF-THE-ART OF THE INVESTIGATIONS IN THE FIELD OF QUALITY AND SAFETY CONTROL OF MEAT RAW MATERIALS AND MEAT PRODUCTS IN RUSSIA*

Lisitsyn A. B.

Abstract: Safety and quality of food products, including meat products, is an urgent problem now and will continue to be such in the near future.

Due to this fact, many countries developed novel systems for ensuring food safety and quality. Russia has developed "Complex system of safety and quality control of foods" that is based on the utilisation of: hurdle technologies, HACCP system, prediction microbiology, system of complex continuous monitoring of technological flow including the system of distribution of transport flows and the system of production management.

The All-Russian Meat research Institute (VNIIMP), in the last couple of years performs more complete and reliable safety and quality controls of meat within the complex system. It developed new standards for detection and identification of *L. monocytogenes*, investigated the possibilities of utilisation of natural spices mixtures that lowers the danger of occurrence of *L. monocytogenes* in meat products; defined new antioxidants; introduced new technological procedures for increasing the shelf-life of packaged meat and meat products; defined critical control points for HACCP application in slaughterlines and in meat processing facilities; defined and introduced into practice the system of voluntary HACCP-meat certification in meat industry; the Institute conducts Monitoring programme of toxic substances in meat and meat products; it developed and applied histological method of product components identification; applied electronic nose system (VOC meter) for determination of freshness and meat species; developed the production of kits and primers for PCR methods.

Research and development programmes ensure that meat production and control systems are maintained in accordance with contemporary achievements in science and needs for efficient consumers protection.

Key words: safety, quality, methods, investigations, freshness, meat species

Savremena ispitivanja na polju kvaliteta i kontrole bezbednosti mesnih sirovina i proizvoda od mesa u Rusiji

Sadržaj: Bezbednost i kvalitet hrane, uključujući i proizvode od mesa, je sada, veoma aktuelan problem, a očekuje se da će tako biti i u bliskoj budućnosti.

Zbog toga su mnoge zemlje, ili njihove grupacije, razradile nove sisteme za osiguranje bezbednosti i traženog kvaliteta hrane. U Rusiji je razrađen "Kompleksan sistem za kontrolu bezbednosti i traženog kvaliteta hrane", koji je baziran na korišćenju: tehnologije prepreka, HACCP-a, mikrobioloških predviđanja, kontinuiranog monitoringa tehnološkog procesa, uključujući transport, distribuciju i načine upravljanja proizvodnjom.

Sve-ruski naučno-istraživački institut industrije mesa (VNIIMP) poslednjih godina, radi potpunije kontrole i osiguranja pouzdanje bezbednosti i kvaliteta mesa, u okviru kompletnog sistema, izradio je nove standarde za detekciju i identifikaciju *L. monocytogenes*, izučio mogućnost korišćenja smeša prirodnih začina koje smanjuju opasnost od pojave ove vrste bakterija u proizvodima od mesa; definisao nove antioksidanse; uveo nove tehnološke postupke za produženje održivosti upakovanog mesa i pojedinih proizvoda od mesa; definisao kontrole kritične tačke za primenu HACCP-a na linijama klanja i prerade mesa; razradio i, u praksu, uveo sistem dobrovoljne „HACCP-meat sertification“ u pogonima industrije mesa; sprovodi monitoring program kontrole toksičnih supstanci u mesu i proizvodima od mesa; razradio, i kroz, monitoring primenio histološki metod identifikacije komponenata sastava proizvoda; za određivanje svežine i vrsta mesa primenio sistem (VOCmeter) elektronskog nosa i za korišćenje PCR-a metoda obezbedio proizvodnju potrebnih kitova i prajmera.

Istraživački i razvojni programi su i dalje usmereni da se sistemi proizvodnje i kontrole mesa i proizvoda od mesa, održavaju u skladu sa aktuelnim dostignućima nauke i potrebama efikasne zaštite potrošača.

Cljučne reči: bezbednost, kvalitet, metode, ispitivanje, svežina, vrste mesa

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTHOR: A.B. Lisitsyn, GNU the V.M. Gorbатов All-Russian Meat Research Institute, Talalihina 26, 109316, Moscow, Russia.

AUTOR: A.B. Lisitsin, Sveruski naučnoistraživački institut industrije mesa, VNIIMP, Gorbatova Roselholcakademi, Talalihina 26, 109316, Moskva, Rusija.

Introduction

Safety and quality of food products, including meat products, is an urgent problem now and will continue to be such in the near future (*Lisicin et al.* 1997).

The incidence of foodborne diseases has increased in the world. Integration between countries and globalization of food trade have led to changes in the existing systems of production and distribution of food products. This will create conditions, when both the known and new foodborne diseases can develop (*Lisicin et al.* 2008).

In the Russian Federation the quality and safety requirements for foods are stated in a number of laws: "On quality and safety of foods", "On protection of consumers' rights", "On technical regulation". Their main task is protection of consumers by ensuring high level of food products' safety (*Lisicin and Veselova* 2004).

The world practice shows that safety of foods can be ensured only through the control of production on the scheme "from field to table". It is already recognized that control should be provided on every stage of food chain – from the production of initial raw materials to final treatment, because there can always be situations when potentially dangerous substances for human health can enter to foods (*Lisicin et al.* 1997).

During last decades, the scientists from the V.M. Gorbatov All-Russian Meat research Institute have paid special attention to these problems, connected with harmonization of exothrophic chain - from production of meat products to their marketing (*Lisicin et al.* 1997). We base our work on extension of our knowledge about technological adequacy of meat raw materials, monitoring the production of safe and high quality products, optimization and ecologization of component composition of the product, using food nutrients, meeting the requirements of food quality, safety and dietetics. At the same time we develop new and improve the existing methods for raw materials treatment, ensuring safety and sanitary welfare of final product; we also work on the development of the methods for the determination of quality and safety indicators of meat raw materials and products.

A practical solution to this problem would be a COMPLEX SYSTEM OF SAFETY AND QUALITY CONTROL OF FOODS in Russia, based upon the use of: hurdle technologies, HACCP system, prediction microbiology, system of complex continuous monitoring of technological flows, including the system of distribution of transport flows and the system of production management (*Lisicin et al.* 2008).

Within the frame of this system, scientific investigations in VNIIMP are carried out in all five directions: scientific approaches, methodical and legal basis and the tools for introduction of this system at meat plants. Traceability system of the whole process of raising domestic animals and raw materials technological processing is studied and put into practical use.

No doubt, it is the prevention of different diseases of domestic animals that is the main factor of safety of food products, and the main challenge for sanitary microbiology is detection and monitoring of pathogens posing threat to safety of the product.

Most pathogens, veterinarians fight now with, have been known for a long time. Salmonella, pathogenic staphylococci, botulism agents, Coli group of bacteria have been for a long time associated with foodborne diseases. However, of special concern is the appearance of new pathogenic strains and, frequency, of cases when the known microorganisms can be found in new, non-typical products for them. One can not explain yet why pathogens are capable to spread all over the world very quickly (*Lisicin et al.* 2002).

For example, *Listeria*. Until recently, there was the opinion that mainly the animals catch listeriosis. And, if in the past *Listeria* were found only in some regions, now the cases of detection of pathogenic *Listeria* are registered all over the world, from New Zealand to the USA (*Lisicin et al.* 2008). Listeriosis is a disease, dangerous for humans, because mortality is 30-40% from the number of infected people, and the damage from this infection is much higher than from other infections.

Scientists from VNIIMP, together with workers from 8 research centers of the Academy of Medical Science, have created a national system of safety assurance and control of food products for the presence of the agent of Listeriosis; also the standard GOST P 51921-2002 has been developed - "Food products. Methods of detection and determination of bacteria *L. monocytogenes*" (*Lisicin and Veselova* 2004).

Sanitary stability of the product, as well as its safety is based upon combination of factors, or as they are also called – hurdles. One of such hurdles is biopreservation, whose position among the methods and techniques of quality preservation of products becomes stronger (*Lisicin et al.* 2008).

Spices can also be considered as one of the biopreservatives or non-traditional methods of products treatment to preserve their sanitary welfare. Thus, in the investigation of the curing process and influence of the main recipe mixtures of spices on viability of

Listeria, it was found that such spices, as cardamom, coriander and nutmeg, at 0.005%, reduce viability of *Listeria* 10-fold, and adding garlic emulsion in the same concentrations reduces viability of *Listeria* 100-fold (Fig.1).

sausages at high positive temperatures using the additional hurdles, preventing their spoilage (*Lisicin et al.* 2007).

The results of the investigations have shown that, in case of similar level of such hurdles, as initial

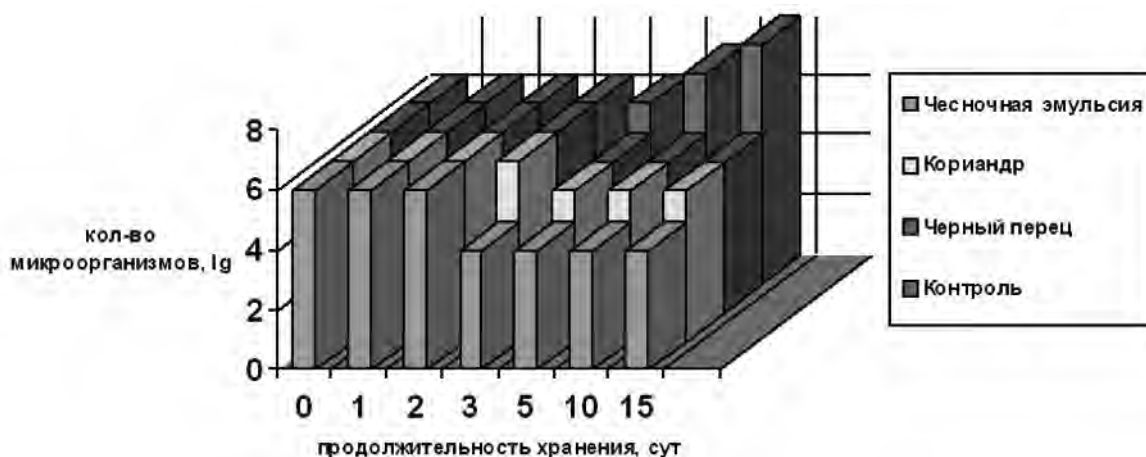


Figure 1. Dependence of viability of *Listeria monocytogenes* (serovar 1/2b) from added spices
Slika 1. Zavisnost viabilnosti *Listeria monocytogenes* (serovar 1/2b) od dodatih začina

Antioxidant activity of dihydroquercetin (DHQ) in thermally treated and non-treated meat products was also studied, and a possibility of increase of its activity as an antioxidant was proved. The efficient doses of dihydroquercetin were determined for mechanically separated poultry meat, which is subjected to oxidative spoilage to the most extent (*Lisicin et al.* 2008).

The investigations have shown that in the sample with dihydroquercetin, at 0.02% to the mass of the raw materials, hydrolytic and oxidative changes occurred 3-fold slower, as compared to the control group (without DHQ). Study of genotoxicity of dihydroquercetin by the "DNA-comet" method at its dosage of 1.5 and 150 mg/kg, demonstrated its safety.

Comparative evaluation of natural antioxidants, including monomer DHQ, has allowed ranking them by their antioxidant properties as follows: for fat products (on the example of raw fat) – DHQ > tocopherol > rosemary extract > tea catechins; for products with high moisture content (>70%) (on the example of MSPM) – DHQ > rosemary extract > tea catechins > tocopherols.

Based on the study of solubility and stability of DHQ in solutions, a possibility of creation of commercial form of DHQ for meat industry as a solution containing 2-5% DHQ and 2-5% of ascorbic acid was established.

To develop technologies of cooked and smoked sausages, not requiring cold storage, VNIIMP specialists were studying keepability of cooked-smoked

count of microorganisms, sodium nitrite content and pH value, changes in thermal treatment of sausages (reducing the time of smoking and elimination of secondary smoking) to increase the final product yield will lead to changes in the content of salt, moisture and water activity. This will result in reduction of their hurdle effect to the levels that will not ensure stable storage of sausages, even at low positive temperatures (2-6°C).

The studies have shown that the introduction of additional hurdles into technology – vacuum packaging and additional thermal treatment (72-76°C during 15 minutes) – will increase shelf life of cooked-smoked sausages, manufactured according to the proposed recipes, to 25 days at 18-20°C, instead of 3 days (Fig.2).

Further investigations will include the study of the influence of thermal treatment conditions, different doses of food additives with hurdle effect on quality and safety of cooked-smoked sausages in storage at high positive temperatures to increase their shelf life up to 45-50 days.

One of the approaches to the prevention of foodborne diseases and for safety of products is the use of HACCP system, which has been functioning at food plants of EC countries for many years (*Lisicin et al.* 2008).

The investigations carried out at the Institute will allow producers to reveal critical control points in production of different meat products, which should be controlled for removal of risk factors or elimination to minimum a possibility of their

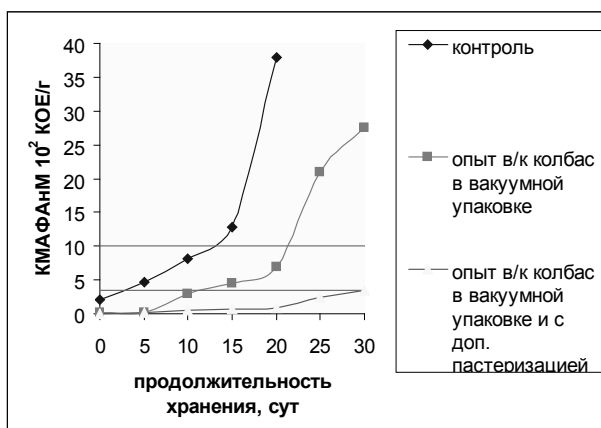


Figure 2. Change in the count of microorganisms in the experimental samples of cooked-smoked sausages during storage at 18-20°C

Slika 2. Promena broja mikroorganizama u eksperimentalnim uzorcima kuvano-dimljenih kobasica tokom skladištenja na 18-20°C

occurrence and also to compose a list of the most frequent non-conformities at meat plants.

Analysis of production chain for sausage products was carried out, beginning from raw materials supply and finishing with laboratory investigation of final products.

The obtained data suggest that frozen raw materials, supplied in sides, had larger microbial load

in cartons, which significantly reduced the possibility of additional contamination of the raw materials.

The obtained results show that the first link of the internal traceability - laboratory control of incoming raw materials - is an important component of safety management of foods, because knowledge of the extent of microbial contamination of raw materials allows managing logistics of the warehouse more efficiently and thus helps preventing non-conformities with regards to the biologically dangerous factor at the initial stage of technological process.

Besides, the amount of microbial contamination of the casings, both artificial (polyamide), and natural, were investigated, as well as spices, wash-outs of the hands of workers and equipment.

The results of the investigations have shown that adoption of a system of monitoring and traceability of hazards at the plant will allow a more efficient management of technological process and control safety of produced foods.

At the All-Russian Meat Research Institute the System of voluntary certification HACCP-MEAT has been developed and registered. It provides for the development of the system of safety and quality management at a meat plant as applicable to the specifics of meat industry plants of Russia.

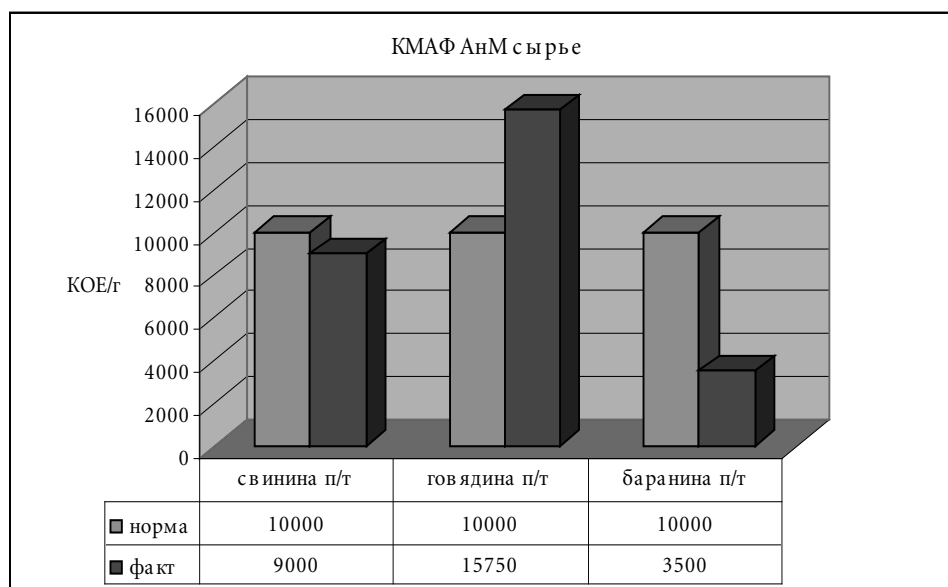


Figure 3. Average total plate count in sides

Slika 3. Prosečan ukupni broj mikroorganizama u mesu

(Fig. 3) than those in blocks. One can suppose that during transportation and unloading/loading of sides and quarters their surface was not protected from the contact with the environment, while the meat in blocks was first packed in film and then

A Methodical Center has been established and functioning at the Institute, which renders consultancy to meat industry plants with regards to the development, implementation and preparation for certification of quality management system and

products safety assurance on HACCP principles (Lisicin *et al.* 2008).

At present, there are more than 15 certified meat plants in this system: in Noginsk, Obninsk, Tcherepovets, Yoshkar-Ola, Borisov, etc.

Based on the principles of traceability, the scientists of the Institute are developing the system of complex monitoring and control of toxic substances content in meat products. In the North-Caucasus region of RF a data bank is being created with the analysis of toxic substances content in organs and tissues of slaughter animals; dynamics of their accumulation is determined, and critical control points of toxic substances in organs and tissues of farm animals and poultry are indicated. Comparison of data (Table 1) of 2008 with the results of the investigations, carried out in 1986-89 has shown, that content of toxic substances in farm animals during last 20 years has increased on average by 3.5-4 times (Lisicin *et al.* 2002).

Thus, monitoring of composition of cooked sausages “Doctorskaya”, “Molochnaya”, and “Rus-skaya” for 2008 supposedly manufactured according to GOST, was carried out by histological method of identification of meat products composition (Lisicin *et al.* 2008). It demonstrated that the share of plants, whose products contain large amounts of one or several not allowed additives, constituted more than 66.5% of the total number of the monitored plants. The percentage of plants which don't use the additives, not allowed by GOST at all, is only 3,5% (to compare: in 2006 - 24%, in 2007 – 11%).

Use of instrumental methods – multi-sensory system “Electronic nose” - allowed developing the methods for the evaluation of freshness of pork, showing also good prospects for the determination of species of meat on “VOCmeter (Germany), which is intended for conducting quality and quantity evaluation of gas mixtures (Černuha *et al.* 2008). The scientists of the Institute have determined the regi-

Table 1. Content of residues of harmful substances in pigs' organisms
Tabela 1. Sadržaj rezidua štetnih supstanci kod svinja

Name	Contents, mg/kg											
	Krasnodar region (the highest values) 1986-1989.				Rostov region Unfavorable zone (2007-2008)				Lipetsk region Unfavorable zone (2007-2008)			
	Pb	Cd	Cu	Zn	Pb	Cd	Cu	Zn	Pb	Cd	Cu	Zn
Muscle tissue	0.02	0.01	0.8	33.8	0.18	0.01	7.1	17.6	0.16	0.03	1.2	6.5
Liver	0.06	0.1	9.2	63.4	0.5	0.079	15.6	50.2	0.17	0.03	3.5	12.5
Heart	0.02	0.01	4.2	21.3	0.46	0.087	18.1	56.4	0.23	0.02	8	29.1
Kidneys	0.11	0.75	3.9	24.3	0.98	0.047	17.5	52.3	0.26	0.045	7.4	33.5

The situation relating to quality control and safety of foods has become more acute with sharply increased import of food products. According to the Federal Customs Service, in the period January-November 2008, the value of import of raw materials and food products to Russia constituted US\$31.9 blns., which is 30.5% more, than in the corresponding period of 2007 (US\$24.4 blns.).

Development of the method of identification and detection of adulteration of raw materials and foods, and also the control over observation of scientifically based recipes and determination of raw materials composition are great challenges in Russia now.

Specialists of the Institute have developed GOST R 51604 “Meat and meat products. Identification of the composition by histological method,” which makes possible identification of animal and plant components in the raw materials used in the manufacture of meat products (Hvilja and Paršenkova 2006).

ons of points in coordinate system of the instrument, characteristic of the samples “fresh”, “doubtfully fresh” and “not fresh”, and of the meat of different animals: beef, pork, chicken meat, turkey, ostrich, deer meat (Fig.4).

The other method, which has good prospects for the determination of species of the tissues of animal and plant origin in the meat raw materials and meat products, is the method of polymerase-chain reaction, which allows revealing the species of meat even in minor quantity, including thermally treated meat products.

Specialists of the Institute have conducted investigations on the determination of nucleotide sequence, based on which synthesis of species specific primers to the fragments of DNA of animal and plant origin (beef, pork, chicken meat, turkey meat and soya) has been accomplished. Study of species composition of meat raw materials and meat products of foreign and domestic origin has shown that

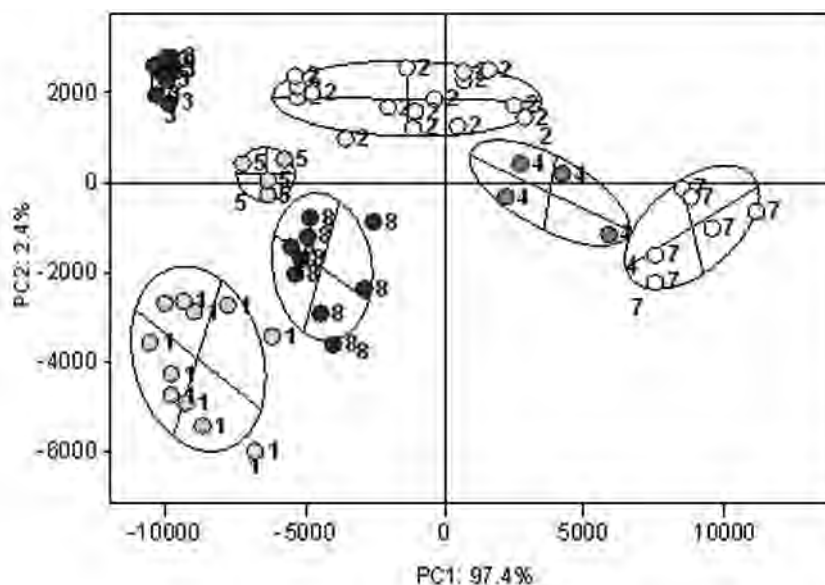


Figure 4. Use of multi-sensory system for the determination of meat species: 1 – pork; 2 – beef; 3 – chicken meat; 4- fish; 5 – mutton; 6 – deer meat; 7 – ostrich meat; 8 – turkey

Slika 4. Korišćenje multisenzornog sistema za određivanje vrste mesa: 1 – svinjsko meso; 2 – goveđe meso; 3 – pileće meso; 4 – riba; 5 – ovčije meso; 6 – meso jelena; 7 – meso noja; 8 – meso ćuraka.

the raw materials of 18% of the studied samples by their raw materials composition did not correspond to the information, indicated on the label.

Production of pig meat is on the increase in Russia at the present time, and production of chilled meat is of great interest due to its best quality traits. Chilled, aged meat with temperature from 0 to 4°C in the core has tender consistency, juiciness, pronounced flavor and aroma more intensive than of defrosted meat. Such meat is better for digestion, and it is more suitable for the manufacture of half-prepared products in pieces.

In Russia, chilled meat is delivered to meat-processing plants mainly in cuts, their shelf life at 0 ... -1°C is 10 days, and in sides – 12-16 days, while chilled meat delivered to Russia from abroad, for example from Argentina, can be stored during 90 days, and from Brazil – 120 days,

At the present time VNIIMP studies changes in sanitary-microbiological indices of chilled pork (boneless, bone-in) during long-term storage. The studied dynamics of changes of microorganisms count in deep layers of meat has shown that microflora penetrated into deep layers of meat from its surface, and this primarily related to motile forms of bacteria (Fig.5).

After the first day of chilling there was no microflora in deep layers of muscular tissue. Up to 12-15 days of storage deep layers of cuts turned to be sterile. During further storage changes in microbiological state were found in deep layers of muscle tissue: the number of lactic acid bacteria

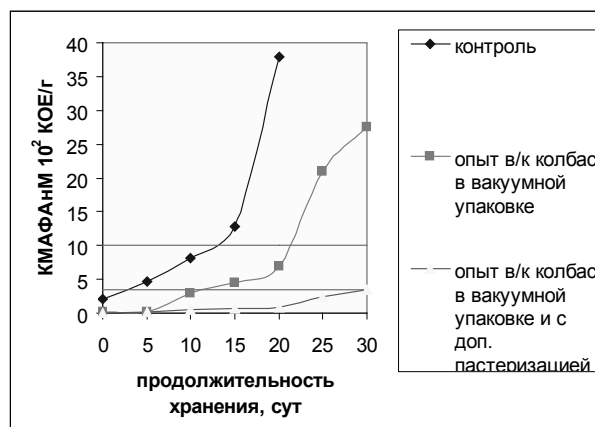


Figure 5. Changes in sanitary-microbiological indicators of chilled pork during long-term storage

Slika 5. Promene sanitarno-mikrobioloških indikatora u ohlađenom svinjskom mesu tokom dugotrajnog skladištenja

(LAB) and the index of total plate count increased. However, Coli group of bacteria, sulfite-reducing Clostridia, yeast, microorganisms of genera Salmonella, Listeria were not found in deep layers of cuts of chilled pork, stored under vacuum throughout all the period of investigations (up to 35 days). The obtained data will be used for the development of reference values in microbiological control of vacuum-packed meat cuts during long shelf life periods.

The future of meat science is the development of the methods of safety and quality improvement of

meat. It is necessary to combine knowledge about the processes taking place on molecular and cell levels, with our knowledge about the live organism on the whole, to understand more clearly the

mechanisms taking place in tissues of live animals and transformations in these tissues after slaughter. This will give us the opportunity to supply high quality, nutritive and safe products to consumers.

References

- Lisicin, A. B., 1997.** Tehnologičeskie aspekti povišenija ekzotrofičeskoj effektivnosti promišljennoj prerabotki mjasnogo sirja. Aftoreferat disertacii;
- Lisicin, A. B., Ivankin, A. N., Nekljudov, A. D., 2002.** Metodi praktičeskoj biotehnologiji, M.: VNIIMP.–408 s;
- Lisicin, A. B., Krilova, V. B., 2003.** Sostojanie i vazmožnosti primenenija polimernih materialov v konzervnoj otrasli. Svjo o mjase, No3, s. 46–47;
- Lisicin, A. B., Veselova, P. P., 2004.** O tehničeskom regulirovanii bezopasnosti mjasa i mjasnih roduktov. Mjasnaja industrija, No 11, s. 28–30;
- Lisicin, A. B., Smetanina, L. B., Kostenko, J. G., Gutnik, B. E., Černuha, I. M., Zaharov, A. N., 2007.** Savremenije aspekti toplovogo konzervirovanija mjasoproduktov. Pod obščej redakciej akademika RASHN Lisicina, A.B. – M.: VNIIMP.–576 s;
- Lisicin, A. B., Lipatov, N. N., Kudrjašov, L. S., Aleksahina, V. A., Černuha, I. M., 2008.** Teorija i praktika prerabotki mjasa. Vserossijskij naučno-isledovatel'skij institut mjasnoj promišlennosti im. V.M. Gorbatov. Moskva;
- Lisicin, A. B., Hvilja, S. I., Burlakova, S. S., Pčelkina, V. A., 2008.** Rezultati monitoringa mjasnih produktov za 2008. god. Sbornik dokladov XI Meždunarodnoj naučnoj konferenciji, Moskva, s. 97–102;
- Hvilja, S. I., Paršenkova, R. V., 2006.** Razrabotka novogo standarta dlja uskorennoj identifikacii sastava gistologičeskim metodom. Svjo o mjasu, No 2., s. 34–35;
- Černuha, I. M., Kuznjecova, T. G., Selivanova, E. B., Ivankin, A.N.: 2008.** Isledovanie vazmnožnostej ispoljovanija pribora „VOCmeter“ dlja ocenki svežosti mjasa. Mjasnaja industrija, No 3, s. 49-51.

Paper recieved: 13.04.2009.

MEAT AND MEAT PRODUCTS – HAZARDS AND RISK - NORWEGIAN STRATEGIES AND EXPERIENCES*

Alvseike, O.

A b s t r a c t: In general, Norway benefits from a low incidence rates of most zoonoses. This results mainly from systematic work and collaboration between authorities and private stakeholders for more than 100 years. The approaches have varied dependent of the hazards addressed and the risk they represent. Globalization and free trade challenge established systems. In Norway, the consequences have been delayed by the protection from import fees. Hopefully, we will be able to develop balanced risk based strategies that provide the consumers with thrust in their food supply.

Key words: zoonoses hazards, risk, meat, Norway

Meso i proizvodi od mesa – opasnosti i rizik – strategije i iskustva Norveške

S a d r ž a j: Uopšteno govoreći, Norveška je u prednosti s obzirom na nizak stepen pojavljivanja najčešće prisutnih zoonoza. Ovo je pretežno rezultat sistematskog rada i saradnje između vlasti i privatnih stočara koja se odvija više od sto godina. Pristupi ovom problemu se razlikuju u zavisnosti od vrste rizika sa kojima se suočavamo i mogućih posledica. Može se reći da su globalizacija i izazovi slobodne trgovine uticali na uspostavljanje sistema. U Norveškoj, posledice su odložene zaštitom od uvoznih dažbina. Nadamo se da ćemo uspeti da razvijemo balansiranu strategiju rizika koja će obezbediti poverenje potrošača u snabdevanju hranom.

Ključne reči: zoonoze, opasnosti, rizik, meso, Norveška

Introduction

Food security (enough food) is the most primary need for a human being. The ability to collect and store food was the keystone of the first civilizations on Earth. Grains in Mesopotamia and Egypt, rice in China and corn, squash and beans in Southern America (Diamond, 1997).

The first Norwegian animal health decree was proclaimed in 1732 to protect the country from rinderpest, an epidemic causing ravages to European livestock populations in those days: "Decree, that in Denmark and Norway no kind of Livestock, Meat, Hide, or Hair from Livestock from foreign Places shall be imported; due to Livestock: Illness in Poland".

A law giving powers to ban the import of live animals was passed in 1854 (Sandvik, 1992). The restrictions to import of animal products would have a reducing effect on several zoonoses too, but food safety was not seriously addressed in Norwegian laws until 1860 with the first "Health law". The concept of food safety is rather modern, and has gradually

been developed the latest centuries due to increased scientific knowledge, improved economies and an increased time to worry. Earlier, man was concerned for the food for tomorrow, today the vast majority in Europe is concerned by the foods wholesomeness, palatability, price, etc.

The Codex alimentarius definition of a hazard is "a biological, chemical or physical agent in, or condition of, food with the potential to cause an adverse health effect", and furthermore risk is defined as "a function of the probability of an adverse health effect and the severity of that effect, consequential to a hazard in food".

In 1892 meat inspection was introduced in Norway, according to the methods of the German scientist Ostertag. In practice, this hazard oriented approximation has been the core strategy up to day. Critics have argued for 30 years that the official meat inspection is targeted at diseases that no longer threaten the Norwegian public health, like tuberculosis and trikinosis, i.e the strategy is not risk based as important pathogens, like *Campylobacter*,

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mauntain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

Salmonella and *Toxoplasma* are ignored by the classical approach alone (Nesbakken *et al*, 1996).

The meat inspection legislation today, based on the EEA/EU-directives, is also based on an os-tertagian philosophy, even though the Hygiene package opens carefully for customization to national risk levels. However, demands to authorization, facilities, labeling, GHP, HACCP, surveillance and control have added important dimensions to improve meat safety. Still meat-borne disease occurs and question is: What is an appropriate response?

In general, Norway benefits from a low incidence rates of most zoonoses (Nesbakken *et al* 1996). The aim of this paper is to give a brief overview of Norwegian experiences and strategies against meat associated hazards and the risk they represent.

Discussion

To obtain safe meat one has to apply preventive measures in many dimensions. Some are given from Nature, some are cultural and others are biologically targeted.

Geography and climate

Geography has been important factor to protect the country from epidemics. The North Sea and Skagerrak have been efficient obstacles for many contagious diseases from the European continent. Also the Baltic Sea has protected both Sweden and Norway, and along the border to Sweden, Finland and Russia, it is mainly woods, mountains or arctic conditions with low density of both animals and humans.

Climate has also been important for some diseases like vector-borne infections and some parasitic infections. However, the cold climate is in general not regarded that important for most bacterial zoonoses. Febris undulans (*Brucella mellitensis*) is an exception, and yersinosis (*Yersinia enterocolitica*) may be an example that seems to occur more frequent in cold climates.

Infrastructure and organization

Infrastructure and organization are results of history and culture. The Scandinavian countries have benefited from relative stable political conditions. Norwegians thrust their authorities and the agricultural private sector is thoroughly organized and regulated. The basic idea is that food safety and contagious diseases are not a national competitive element and that the control measures should be made and financed to a large extent in common. However, a sound livestock is a very important competitive advantage for export of genes and live animals.

The Norwegian combats of diseases have benefited from collaboration between the authorities and private stakeholders. The trend of private responsibility for food safety and animal health standards may undermine the situation in the future, if the farmers' organizations and the industry are not able to coordinate or finance common actions and obtain confidence among the producers and companies.

Norwegian Food Authority has become considerably consumer oriented in few years. Then expensive Utopian demands, like zero-risk level, sometimes replace balanced risk management. The paradox is that unrealistic demands may out-compete national production that has achieved a very high level, for imports from countries in a less favourable epidemiological situation.

Norwegian herds have traditionally been small, but are now increasing significantly. Herd size has been regarded an important factor for prevalence of infectious diseases. Infections depend on infective and a critical number of susceptible individuals. If the group size is below a critical number, the infection will burn out (endemic fade-out) (Anderson *et al*, 1991). The effect of increased herd size on zoonotic incidences in Norway remains to be documented.

Feed control

The importance of animal feed has been terribly underlined the last decades by serious food scandals in Europe. Chemical contamination from e.g. dioxins and cadmium has raised great concern about the European food chain. The BSE and vCJD, caused by a transmissible protein, has not directly affected Norway. No cases of BSE or vCJD have been detected in spite of substantial testing according to the EU schemes. Status is due to decisions made by a former chief veterinary officer, Olav Sandvik, who banned the use of bone meal originating from the same species and the fat extraction method applied from the 80'ies, e.g. in Britain. The decisions were based on the precautionary principle that here could be simplified to "cannibalism is dangerous" and that it is important to apply measures that break cycles of transmissible agents. Norway is still the only country in the lowest risk group in Western Europe (Hogasen *et al*, 2007; Skjerve *et al*, 1996).

A pandemic like salmonellosis due to *S. Enteritidis* in layer hens has not established in Norway. This is probably due to strictly organised egg production systems and control regimes for concentrate feed. Also, infections due to *S. Typhimurium* and other serovars are seldom acquired from domestic animals and products thereof. The most likely preventive factor is again control regimes of concentrate feed. A significant proportion of protein feed is imported

and different salmonella serovars are isolated regularly from raw feed. The concentrate feed undergoes mandatory heat treatment and the positive effect on feed hygiene seems obvious.

Control measures on farm and in industry

Import restrictions of livestock and animal product have been the rule of thumb since 1732 to 1995. Norway has for centuries been dependant of import calories, i.e. grains. After the 2nd World War the policy has been 50 % self supply. However, animal food products, like milk and meat, have been protected by law and import duties. In 1995, The WTO Agreement on the Application of Sanitary and Phytosanitary Measures (SPS Agreement) ended the ban of import principle: "Reaffirming that no Member should be prevented from adopting or enforcing measures necessary to protect human, animal or plant life or health, subject to the requirement that these measures are not applied in a manner which would constitute a means of arbitrary or unjustifiable discrimination between Members where the same conditions prevail or a disguised restriction on international trade." Since then, the protection of Norwegian animal production is heavily dependent on import duties. Additionally, documentation that the prevailing conditions are favourable in Norway has been important for improved protection of the animal health and zoonosis situation. However, this is regarded to be a more labile situation.

Eradication programmes has been applied from late 1890'ies in Norway for antrax, bovine tuberculosis and brucellosis. Fowl typhoid (*S. Gallinarium*) in hens was actually eradicated twice, before and after the 2nd World War. This most radical strategy has been successful many times. The campaigns have not always been subject to cost-benefit analyses.

The pasteurization was introduced for milk in the 1920's in Norway. The original objective was

References

- Diamond, J. M., 1997.** Guns, germs and steel: a short history of everybody for the last 13,000 years. W.W. Norton & Co, New York;
- Sandvik, O., 1992.** Animal Health Standards in Norway. The Royal Ministry of Agriculture. Norway;
- Nesbakken, T., Skjerve, E., 1996.** Interruption of microbial cycles in farm animals from farm to table. Meat Science 43 (No. S), S47-S57;
- Anderson, R. M., May, R. M., 1991.** Infectious diseases of humans Dynamics and control. Oxford University Press, UK;
- Hogasen, H. R., de Koeijer A. A., 2007.** Quantitative risk assessment for bovine spongiform encephalopathy in low- or zero-prevalence countries: the example of Norway. Risk Anal., 27,5,1105-17;
- Skjerve, E., Gronstol, H., Rimstad, E., Sandvik, O., 1996.** Matvarekvalitet, dyrehelse, og internasjonal handel:

again to prevent animal disease like brucellosis, foot and mouth disease, etc. A by effect was a tremendous reduction on human diseases like scarlatina (*Strept. pyogenes*). Paradoxially, steam pasteurization of carcasses has not been allowed. The authorities argue that it may reduce the focus on general hygiene in the industry. A serious EHEC-outbreak (*Schimmer et al, 2008*) has challenged this policy, and it is likely that steam cabinets will be allowed in the future.

Surveillance and control have become the modern response to zoonoses like salmonellosis. Test positives on farm without clinical signs are challenging. Should the zoonotic agents or the zoonoses be targeted? Bacteriological samples have specificity close to 100 %, but their sensitivity may be low, which means false negatives is easily missed. A serological test may both have quite good sensitivity and specificity, but high numbers of screened individuals tend to cause a serious number of false positives as well. The surveillance systems introduced with the EEA agreement from 1994 have gained some knowledge of prevalence of many infectious agents, but it is not obvious that they have reduced efficiently the human burden of corresponding diseases (*Sandberg et al, 2002*). On the other hand, the documented reduction of human incidence rates for yersinosis is most likely a result of "bagging", a simple improvement of dressing procedures of pork carcasses applied in Norway (*Nesbakken et al, 1994*).

Conclusions

Future protection of meat safety will depend on several preventive measures along the value chain from farm to table. Hopefully, private and governmental bodies will be able to collaborate and coordinate balanced risk based strategies that provide the consumers with thrust in their food supply.

risikoforbundet med økt import av levende dyr, kjøtt og andre dyreprodukter fra EØS-området og andre land. Nor.Vet.Tidskr. 108(6B);

- Schimmer, B., Nygard, K., Eriksen, H. M., Lassen, J., Lindstedt, B. A., Brandal, L. T., 2008.** Outbreak of haemolytic uraemic syndrome in Norway caused by stx2-positive *Escherichia coli* O103:H25 traced to cured mutton sausages. BMC.Infect.Dis. 8, 41;
- Sandberg Marianne, Hopp, P., Jarp, J., Skjerve, E., 2002.** An evaluation of the Norwegian Salmonella surveillance and control program in live pig and pork. International Journal of Food Microbiology. 72, 1-2,1-11;
- Nesbakken T, Nerbrink E, Rotterud OJ, Borch E. 1994.** Reduction of *Yersinia enterocolitica* and *Listeria* spp. on pig carcasses by enclosure of the rectum during slaughter. Int.J.Food Microbiol.23, 2,197-208.

PROCEDURES IN IMPROVEMENT OF THE CONTROL OF THE QUALITY OF MEAT PRODUCTS – CONSUMER PROTECTION STRATEGY*

Matekalo-Sverak Vesna, Turubatović L., Petronijević R.

A b s t r a c t: Introduction of new parameters of control of the quality of meat products, as well as constant improvement of analytical methods used for examination of all major components of the meat products, would considerably contribute primarily to the improvement of the consumer health, as well as protection of their economical, ethical and religious interests. Identification of the main raw material in meat products, certain additives of which some can have detrimental effect on health safety of certain consumers, as well as control of type and quantity of certain additives would greatly contribute to the development of consumer protection strategy and strengthen the confidence of consumers in quality and safety of meat products on domestic market.

Key words: quality of meat products, consumer protection strategy, control of the quality of meat products

Postupci unapređenja kontrole kvaliteta proizvoda od mesa – strategija zaštite potrošača

S a d r ž a j: Uvođenjem novih parametara kontrole kvaliteta proizvoda od mesa, kao i stalnim unapređenjem analitičkih metoda kojima bi se svi važni sastojci koji čine proizvod od mesa, mogli ispitati, značajno bi se uticalo, pre svega na unapređenje zaštite zdravlja potrošača, kao i na zaštitu njihovih ekonomskih, etičkih i religioznih interesa. Identifikacija osnovne sirovine u proizvodima od mesa, određenih dodataka od kojih neki mogu imati negativan uticaj na zdravstvenu bezbednost pojedinih potrošača, kao i kontrola vrste i količine pojedinih aditiva znatno bi doprinela u razvoju strategije zaštite potrošača i pojačala poverenje potrošača u kvalitet i ispravnost proizvoda od mesa na domaćem tržištu.

Ključne reči: kvalitet proizvoda od mesa, strategija zaštite potrošača, kontrola kvaliteta proizvoda od mesa

Introduction

In the World, special attention is directed to protection and safety of consumers in all branches of production. Consumer must not be deceived, and product he is purchasing must be completely safe and cannot endanger human health in the lowest degree (Turubatović *et al.*, 2005).. In European Union countries, Canada and USA, consumer protection strategies are being developed mainly directed to production and marketing/ trade of food products, and in this way not only the health of consumers is protected but also their economical, ethical and religious interests. In developed countries, and in Serbia, consumers are protected by laws when the food is concerned. However, regardless of this fact, recently, food producers, and especially food producers in the meat industry –numerous incidents

(BSE, *E. coli*, utilization of not allowed meat species in meat products, genetically modified organisms, dyoxine, melamine, and in our country, utilization of prohibited additive potassium meta bisulphate in chopped meat for forming, so called čevapčići and pljeskavice/hamburgers, etc.) have contributed to lack of trust and confidence of consumers towards food producers – have increased the measures aimed at protection of consumers and developed strategy for improvement and implementation of these measures.

Since year 1998, it has become clear that consumers are concerned about the use of genetically modified food and they demanded that food products to which genetically modified ingredients have been added be labeled accordingly, (Joop de Boer *et al.*, 2007) i.e. on labels within the declaration of the product, it has to be declared that products has been

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTHORS: Vesna Matekalo-Sverak PhD, vesna@inmesbgd.com, Lazar Turubatović PhD, Radivoj Petronijević MSc, Institute of Meat Hygiene and Technology, Belgrade

AUTORI: dr Vesna Matekalo-Sverak, vesna@inmesbgd.com, dr Lazar Turubatović, mr Radivoj Petronijević, Institut za higijenu i tehnologiju mesa, Beograd

manufactured with addition of genetically modified food. In Great Britain, consumers even demanded that also restaurants serving food consisting of genetically modified food stuffs should be visibly marked. Consumer panic is completely understandable because of the suspicion that genetically modified organisms (GMO) are responsible for more frequent incidence of allergies occurring subsequent to consumption of certain types of food products and meat products (*Jamenez-Colmenero et al*, 2001). About the same time, a currently very modern term “safe food” was created, and in accordance to that term “safe meat products”. However, the difference between hygienically safe/correct meat product and safe meat product must be underlined. Meat product, which is correct, from the aspect of hygiene and health doesn't have to be safe at the same time. Certain ingredients used in manufacturing of different types of meat products, as well as certain meat species, can cause in specific group of consumers, due to allergic reactions or reactions due to intolerance to certain specific ingredient, more or less severe health problems, and consequences for consumer, when allergic reaction is in question, can sometimes be even lethal (*Giese*, 2003). Therefore, food producer is obligated to declare on the label of the product all information of significance to the consumer (*FDA*, 1999). Expanded content of declaration of food products and meat products compared to previous declarations/labels which only had some main parameters of the product composition presented, are result of this consumer protection strategy and have been adopted in the majority of world countries and also in Serbia. In any case, this is an excellent measure for maintaining of insight into the quality of meat product, but it is not sufficient and it is not the only measure. The most important issue is that the declaration reflects the true situation and that the food producer is manufacturing meat product from raw material, supplements and additives which have been declared, as well as that the name of the product is not misleading in any way. Following measure in implementation of the consumer protection strategy are inspection in production and trade, and education of consumers. One of unavoidable measures is also upgrading of analytical methods in the control of the quality of meat products and learning how to use modern methods.

Quality of meat product

In our society, the concept of quality of meat product mainly includes product composition and its sensory properties. Quality of meat product es-

entially, beside sensory properties, includes also its microbiological and health status, i.e. presence or absence of environment contaminants, heavy metals and pesticides, and residues of veterinary drugs. However, regardless of this fact, inspection examination of meat products, in our country, includes in inspection of the quality of meat products, beside sensory properties and declaration control, determination of main chemical parameters and, in regard to additives, determination of the residual nitrite, total phosphorus and nitrate in fermented sausages. Health correctness of meat product includes microbiological correctness, control of residues, examination of the radio activity and sensory examination, although, Law on health adequacy of food stuffs and objects of general use clearly defines that food stuff has to have issued quality – composition in order to be deemed adequate from the aspect of health.

Quality, composition of meat product in all organized countries is regulated by regulations adjusted to consumer habits, technological capability and development of the country, control possibilities, religious demands, etc (*Arihara Keizo*, 2006.) Some countries, for instance Australia and New Zealand, in their regulations and provisions for different meat products, issue different minimum quantities and species of meat, which have to be complied to. In certain countries, the content of water or lipids is limited, or the minimum content of protein for certain products is issued (which is case in our country). However, for almost all countries it is characteristic that there is a group of product of high quality or products of protected origin which are manufactured according to protected procedures and which often have better price than remaining meat products. Serbian Regulation on quality and other requirements regarding meat products issues for certain products possibility or impossibility for use of different additives of food stuffs. Regulation on quality and other requirements for additives used in food products, determines conditions for use of additives in meat products. So, it is evident that in our regulations the quality of each product is unambiguously determined and with compulsory declaration represents significant contribution to the consumer protection strategy.

In our country, control of the technological process is carried out in production facilities where meat products are manufactured, also production specifications and declarations are controlled, as well as main chemical investigations, such as protein content, relative content of protein of binding tissue, content of total phosphates, nitrites and nitrates, etc. Establishment of the presence of proteins ori-

ginating from different meat species is done only in meat products intended for export and specifically the presence of bovine protein is compulsory. Determination of the content of soy protein, gluten, supplements obtained from milk, carrageenens and colors in meat products, as far as we know, is not done on national level.

The most important issue relating to the part of the strategy determining that meat products manufactured according to precisely defined procedure and technology (defined production specification), are marketed with full declaration – information presented on the label where it is unambiguously stated which raw material was used for manufacturing of that product and which supplements and additives have been used. The accuracy of information on declaration is checked by the inspection at the production facility as well as by analysis of meat product, by applying acknowledged analytical methods. Such control of the quality of meat product is sufficient for large production facilities of the meat industry where an experienced team of experts are working and where inspection authorities are present on regular basis. In smaller facilities for meat processing, which are present in our country in great number, usually only one technologist is employed, and inspection authorities are not present there on daily basis, declaration/statement on composition of the meat product must be controlled on broader basis, and same relates to imported meat products which are also present on our market in significant quantities. Also, it is necessary to intensify the control of meat products and dishes containing meat sold in fast food restaurants, since these products are mainly consumed by children and young people. From the intensified control also the regular food and dishes containing meat prepared and served in conventional, traditional restaurants should not be excluded.

There are cases known in practice and described in literature of allergies on meat, most of hypersensitive persons are allergic to red meat, and in some cases there are allergic manifestations occurring after consumption of poultry meat or mutton (Aoyama, et al, 2000; Davidson, 2002; Givens, et al, 2006). Apart from that, there are many other health reasons or conditions such as gout, hypertension, diabetes, increased cholesterol in blood plasma, which limit the consumption of certain species of meat in nutrition. It is very important to point out that certain religions prohibit the consumption of pork, and some of beef, therefore, strict control of meat products in regard to its main ingredient, raw material is necessary. The latest, current Regulation on quality and other requirements for meat products,

which has been harmonized with similar regulations of EU countries and adjusted to our conditions for certain types of meat products issue even the type of raw material, which is additional reason confirming the necessity for development and application of methods for identification of species of meat in meat products.

Regardless of many health benefits contributed to soy bean preparations, (Aoyama et al, 2000; Hoffman & Wiklund, 2006; Hoogenkamp, 2007; Pszczola, 2003) which was also confirmed by scientific results, utilization of soy protein in food products, also in meat products, has to be properly declared on the product label, and soy bean preparations have to be declared also on cosmetic products. Similar problems to different species of meat can also occur in cases when soy bean preparations are used in manufacturing of meat products, however, more people are allergic to soy protein than to protein originating from certain species of meat. It is not widely known that soy bean as food stuff is considered as one of the greatest allergens and is on the list consisting of nine food stuffs “severe allergens”, with strict control over their use in food products. Main instigators of allergy in soy bean are proteins glycinine and three units of BETA conglycinine, i.e. soy proteins which are carriers of the functionality and are present in all soy bean products used as food supplements. Problems with allergies on to soy bean are especially present in developed world countries where the use of industrial food and semi-finished finished dishes is mostly present. Canadians state that they have recorded significant increase of number of allergic incidents in humans, especially children, during the nineties of the last century which coincides with increased utilization of different protein products in food processing. Although it is regulated by legislation provisions that the use of soy bean in food products has to be declared, in this country by inspection and control products are registered for which the use of soy bean has not been declared. Such occurrences must not immediately be regarded as intentional mistakes by the food producer. Many spice mixtures, especially spice extracts, contain as carrier soy proteins, so food producers in meat industry are sometime unaware of the fact that they are using soy bean in the production and therefore not declaring it. If for this reason, or intentional deceit of consumers, but in Spain hams were discovered which were manufactures with soy bean, but manufacturer has not declared its presence on the label. In regard to increase of number of allergic reactions to soy bean, according to official data from New Zealand, of all severe allergic incidences in humans caused by food, soy bean was

the cause of allergy in 25% of cases. Also, in Great Britain, it was established that consumption of soy bean milk in childhood, as substitute for cow milk, causes occurrences of allergies to peanuts later on. Soy bean in nutrition is not recommended in certain physiological conditions, such as pregnancy and nursing, also it is not recommended for consumers with heart problems since it causes blowing up for which oligo saccharides contained in soy bean are responsible. Based on all stated it can be concluded that content of soy bean in meat products must be declared, however, regardless of that, experiences from other countries tell us that we cannot even rely on the declarations with high certainty and that it is necessary to develop methods for verification of its presence in meat products, since consequences can be fatal for some people. In spite of the fact of the detrimental effect of soy bean on health, it is used as way to substitute parts of high valuable and expensive meat by cheap supplement (*Hoogenkam, 2007; Tsumara et. al, 2005*). If we mention that by only one kilogram of soy bean isolate it is possible, with hydration in meat products, to substitute five to six kilograms of meat, and at the same time maintain the content of total protein in meat product, it is clear that by conventional analytical methods, and such substitution cannot be precisely detected.

Gluten, wheat protein (*Pietrasik et. al, 2007*), is very often used in food products and meat products since it is very good emulgator and stabilizer and influences good consistency of the product, it has no significant effect on taste, odor and color of meat product, so it can't be easily identified organoleptically. Gluten is known allergen and declaration of gluten is compulsory. However, gluten is often used in mixtures which are intended for use in meat products, such as emulgators and stabilizers, and meat industry is buying them according to different trade names, so it can easily be used in manufacturing of meat products, without it being declared, most often due to lack of information or poor knowledge or expertise of the person applying it.

Karagenan is hydrocolloid with exceptional ability of hydration (*CyberColloids, 2007*). Its ability to provide for meat, through brine injected into it, up to 100% of hydration, i.e. to double the mass of meat, influenced the decision of producers in meat industry to use it more than technologically justified. Apart from this, karagenan, which is functioning as condenser, stabilizer and gelling agent and is often found in composition of different mixtures intended for use in meat industry, similar to gluten, can also unintentionally be left out of the product declaration. Considering that many consumers wish to consume products free of this additive since it can cause dige-

stion disturbances of different intensity, and we are familiar with cases of the use of this additive without declaring it on the label, the development of the method for determination of the presence of karagenan in meat products is of exceptional importance.

Included in the list of additives used in meat industry, and for which in our country there is still no developed determination method, and therefore are not controlled, beside karagenan, are also colors used in food industry.

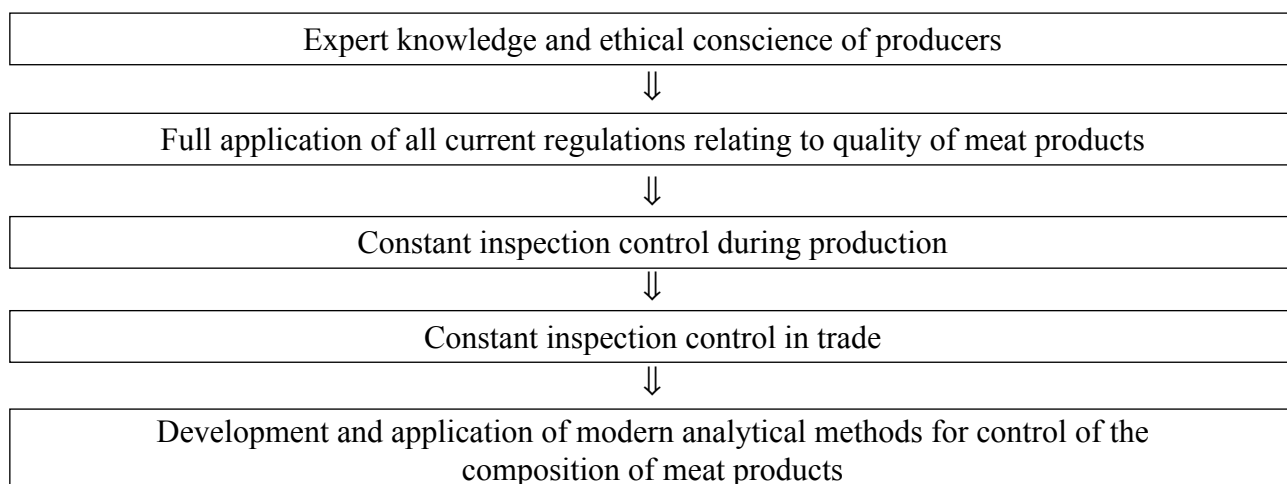
Certain colors have been allowed for use since December 2004 in meat products but only in certain products and in regulated quantities, so, it is understandable that in our country we still are not capable of using adequate analytical methodology for determination of their presence and quantity in meat products. Accordingly, of course, there are no requirements for their control. It has to be mentioned that Kosenilo is allowed for use in certain meat products, whereas Ponso 4P is not on the list of allowed additives for use in meat industry, but we suspect that because of its desired traits and effect on color quality of the product, in spite of ban, it is still used in processing, like some other additives which are not intended for use in meat products. Because of its property to affect positively the quality of color of certain meat products, food color Ponso is used also in manufacturing of some spice mixtures used all around the world in manufacturing of food products.

So, we think that beside conscience and expert knowledge of people working in the meat industry, obligation to comply with regulations relating to quality of meat products, use of additives and obligation to declare composition of products, inspection control during production and in trade, it is necessary to control the quality of meat product in more detailed manner and constantly develop and apply analytical modern analytical methods in assessment and control of meat products.

Selected analytical methods for more complete control of quality of meat products

In laboratories of the Institute of meat Hygiene and Technology, high quality methods for determination of species of meat have been developed and validated; also for determination of quantity and quality of soy bean and gluten proteins in meat products; for determination of quantity and quality of carrageenans and determination of quantity of food colors Kosenilo and Ponso.

Identification of proteins of muscle tissue originating from different animals in developed countries is done using method ELISA. Method is based on enzyme immune reaction (ELISA). Contrary to



Scheme 1. Elements of development of consumer protection strategy

Shema 1. Elementi razvoja strategije zaštite potrošača

other methods with same purpose, it takes very little time and it is very reliable and fast. Enzyme immune reaction is based on determination of presence of thermo-stable proteins which are characteristic/species specific. For detection and identification of different meat species in meat products also PCR technique is used which is also very fast and highly sensitive (Arslan Ali *et al.*, 2009; Ghovvati *et al.*, 2009; Gurdeep Rastogi *et al.*, 2007; Kesmen *et al.*, 2007; Weibin Bai *et al.*, 2009; Rea *et al.*, 2009). In this technique, gene targeted is cytochrome b coded by mitochondrial circular DNA molecule. This gene is highly preserved during evolution and can be found in numerous copies which enable its easy species specific identification.

Methods used for quantitative and qualitative determination of the content of soy protein in meat products are microscopy, SDS poly acryl amide gel electrophoresis and analysis of peptides, and all mentioned methods require lot of time and don't give sufficiently precise results (Tsumara *et al.*, 2005). Soy bean can be identified in meat products microscopically, but organoleptic/sensory evaluation is not neglected and represents the first step in further analytical procedure. ELISA method is also widely used all over the world and principles of enzyme immune reaction, as method sensitive and soy protein specific even in products where other proteins are present such as other proteins of plant or animal origin and other proteins. Presence of soy proteins in meat products, according to literature data, can also be established by reverse highly efficiency liquid chromatography. The latest literature data present fast/rapid, specific and sensitive method for determination of additional soy proteins in meat products which is also based on reverse-phase high efficiency liquid chromatography, but phytoestrogens are

detected, main isoflavones from soy bean - daidzein and genistein. Using this procedure it is possible to detect in meat products amounts of soy bean below 0.1%. However, we couldn't find data on if this procedure is used in regular inspection control.

Gluten, as well as dairy supplements, is easily and rapidly identified by analytical methods, primarily by ELISA technique, in meat products.

The most frequent methods for determination of karagenan stated in foreign literature and papers by different authors are chromatography methods (primarily methods of gas chromatography and high efficiency liquid chromatography) (Sebranek, & Bacus, 2007). Recently, there are methods presented which use infrared spectral-photometry with Fourier transformation. Advantage of spectral-photometry methods compared to chromatography is simpler preparation of samples for analysis and shorter time for carrying out of the analysis, whereas chromatography techniques have higher sensitivity and selectivity. All of these analytical methods and techniques are applied for determination of carrageenan and other hydrocolloid polysaccharides in products intended for human consumption.

Methods for determination of food colors in meat products, primarily those of interest to our market and products which can be found on our market, E 120 and E 124, are spectral-photometry methods, kinetic methods, and in more recent studies also liquid chromatography methods (high efficiency liquid chromatography) with different types of detectors (UV/VIS, PDA) are mentioned. Ponso 4R, as azo color, is frequently determined multi-residually with other azo colors, for instance together with Sunset yellow, Sudanese azo colors, etc (Straub, 2005).

Application of new methods and expanding of the list of parameters which are used within the

control of meat products, would greatly contribute to improvement of the efficiency of the consumer protection strategy, first of all of their health and of protection of their economical, ethical and religious interests. Identification of the main raw material in meat products is important because of the cases when during manufacturing process more expensive meat which is declared on the label, is partially substituted with cheaper meat species. This is not only economical and ethical violation, but it can seriously endanger health of those consumers who are intolerant or hypersensitive to certain types of meat proteins.

Conclusion

Introduction of new parameters of control in assessment of the quality of meat products offers

References

- Aoyama, T., Fukui, K., Takamatsu, K., Hashimoto, Y., Yamamoto, T., 2000. Soy protein isolate and its hydrolysate reduce body fat of dietary obese rats and genetically obese mice, *Nutrition*, 16, 349-354;
- Arihara Keizo, 2006. Strategies for designing novel functional meat products, *Meat Science*, 74, 219-229;
- Arslan Ali, O. Irfan Ilhak, Mehmet Calicioglu, 2006. Effect of method of cooking on identification of heat processed beef using polymerase chain reaction (PCR) technique, *Meat Science*, 72, 326-330;
- CyberColloids, 2007, www.CyberColloids.net/carrageenan;
- Davidson, A., 2002. Penguin Book, Rosebank, ISBN 0-14-051522-4;
- FDA, 1999. Food labeling health claims, Federal Register, 64, 57699-57733;
- Ghovvati, S., M. R. Nassiri, S. Z., Mirhoseini, A. Heravi Moussavi, A. Javadmanesh, 2009. Fraud identification in industrial meat products by multiplex PCR assay, *Food Control*, 20, 696-699;
- Giese, J., 2003. Food Allergen Testing, *Food Technology*, Vol. 57, No. 7, 98-100;
- Givens, D.I., Kliem, K. E., Gibbs, R. A., 2006. The role of meat as a source of n-3 polyunsaturated fatty acids in the human diet, *Meat Science*, 74, 209-218;
- Gurdeep Rastogi, Mahesh S. Dharne, Sandeep Walujkar, Ashutosh Kumar, Milind S. Patole, Yogesh S. Shouche, 2007. Species identification and authentication of tissues of animal origin using mitochondrial and nuclear markers, *Meat Science*, 76, 666-674;
- Hoffman, L. C., Wiklund, E., 2006. Game and venison -meat for the modern consumer, *Meat Science*, 74, 197-208;
- Hoogenkamp H., 2007. The Soy industry's love-hate relationship with meat, *Meat*, 17, 2-11;
- Jamenez-Colmenero, F., Carballo, J., Cofrades, S., 2001. Healthier meat and meat products: their role as functional foods, *Meat Science*, 59, 5-13;
- Joop de Boer, Hoogland, T., Carolien, Boersema, J. J., 2007. Towards more sustainable food choices: Value priorities and motivational orientations, *Food Quality and Preference*, 18, 985-996;
- Kesmen, Z., F. Sahin, H., Yetim, 2007. PCR assay for the identification of animal species in cooked sausages, *Meat Science*, 77, 649-653;
- Pietrasik, Z., Jarmoluk, A. Shand, P. J., 2007. Effect of non-meat proteins on hydration and textural properties of pork meat gels enhanced with microbial transglutaminase, *LWT*, 40, 915-920;
- Pszczola, D. E., 2003. Ten Ingredient Developments That May Impact the Future of Food, *Food Technology*, 57, 7, 76-89;
- Rea S, G. Storani, N. Mascaro, R. Stocchi, A.R. Loschi, 2009. Species identification in anchovy pastes from the market by PCR-RFLP technique, *Food Control*, 20, 515-520;
- Sebranek, J. G., Bacus, J. N., 2007. Cured meat products without addition of nitrite or nitrate: what are the issues, *Meat Science*, 77, 712-717;
- Straub, A. M., 2005. Colourful Demands, *Food Ingredients*, April - May, 16-19;
- Tsumara, K., Saito, T., Tsuge, K., Ashida, H., Kugimiya, W., Inouze, K., 2005. Functional properties of soy protein hydrolysates obtained by selective proteolysis, *LWT*, 38, 255-261;
- Turubatović, L. Matekalo-Sverak Vesna, Milanović-Stevanović Mirjana, 2005. Uticaj aditiva, začina i dodatnih sastojaka na bezbednost proizvoda od mesa, „Tehnologija Mesa“, Vol. 47, 2006-2010;
- Weibin Bai, Wentao Xu, Kunlun Huang, Yanfang Yuan, Si-shuo Cao, Yunbo Luo, 2009. A novel common primer multiplex PCR (CP-M-PCR) method for the simultaneous detection of meat species, *Food Control*, 20, 366-370.

Paper received: 25.05.2009.

Note: Study was financed by the Ministry of Science and Technological Development of the Republic of Serbia, within the project „Improvement of methods of control of safety and quality of meat products for the purpose of complete protection of consumer health and interests“. Project number 20145

NEUE TECHNOLOGIEN BEI DER SCHLACHTUNG, GROB - UND FEINZERLEGUNG – EINFLÜSSE AUF SICHERHEIT UND QUALITÄT DES FLEISCHES*

Troeger K.

Kurzerueberblick: Die Entwicklung neuer Technologien zielt häufig auf eine zunehmende Rationalisierung und Automatisierung von Prozessen ab. Dabei muss die Auswirkung der Innovation auf die Sicherheit und Qualität der Produkte ebenfalls in Betracht gezogen werden. In den letzten Jahren wurden seitens der Schlachthofausrüster vermehrt Anstrengungen unternommen, den Prozess der Fleischgewinnung möglichst weitgehend zu automatisieren. Nachdem sich ein Konzept, das auf Spezialmaschinen für jeden Arbeitsschritt basierte, am Markt nicht durchsetzen konnte, setzt man jetzt auf Standard-Industrieroboter, wie sie etwa in der Autoindustrie zahlreich im Einsatz sind. Erste Ergebnisse zeigten, dass die Roboter zuverlässiger und hygienischer arbeiten als der Mensch. Auch für den Bereich der Grobzerlegung stehen mittlerweile leistungsfähige Roboter zur Verfügung. Für die Feinzerlegung bzw. das Schneiden von Fleisch wurde am MRI Kulmbach eine weitere neue Technologie geprüft und bewertet. Es wurden Versuche mit Hochdruck-Wasserstrahl (3800 bar) zum Schneiden von Schweinelachsen (*M. longissimus dorsi*) durchgeführt. Die Ergebnisse zeigten, dass die frischen Fleischoberflächen weitgehend keimfrei waren und dass die Mindesthaltbarkeit dieses Fleisches unter SB-Handelsbedingungen deutlich länger war als die von konventionell mit einem Slicer geschnittenen Fleischscheiben.

Schlüsselwörter: Neue Technologien, Sicherheit, Qualität, Schlachtung, Roboter, Grobzerlegung, Feinzerlegung, Wasserstrahl-Schneiden

Nove tehnologije tokom klanja, grubog i finog rasecanja – uticaj na bezbednost i kvalitet mesa

S a d r ž a j: razvoj novih tehnologija usmeren je na racionalnije i automatizovanije procese. Zbog toga, uticaj inovacija na bezbednost i kvalitet proizvoda, takođe, mora da se uzme u obzir. Poslednjih nekoliko godina, snabdevači opremom za industriju mesa, ulažu velike napore u cilju automatizacije procesa klanja, kako iz ekonomskih razloga, tako i zbog higijene. Posle pokušaja da se implementira koncept korišćenja specijalnih mašina, koje su se pokazale neuspešnim na tržištu, pažnja je usmerena na standardne industrijske robote koji se koriste u mnogim industrijama, a naročito u automobilskoj. Početno iskustvo ukazuje da su roboti pouzdaniji i čistiji od čoveka. U međuvremenu, efikasni roboti su dostupni takođe, za primenu u primarnom rasecanju. U oblasti rasecanja mesa, druge nove tehnologije su ocenjivane u Max-Rubner Institutu u Kulmbachu. Eksperimenti su obavljani rasecanjem svinjskog mesa (*M. longissimus dorsi*) u nareške korišćenjem vodenog mlaza pod visokim pritiskom (3800 bar). Rezultati su pokazali da su površine svežeg mesa skoro sterilne i da je održivost ovog mesa (u uslovima prodaje) bila mnogo duža u odnosu na konvencionalno sečene odreske.

Ključne reči: nove tehnologije, sigurnost, kvalitet, klanje, roboti, grubo i fino rasecanje, vodeni mlaz visokog pritiska

New Technologies in Slaughtering, Pre-Cutting and Cutting – Influence on Safety and Quality of Meat

A b s t r a c t: The development of new technologies often directs to more rational and automatic processes. Thereby, the influence of the innovation on safety and quality of products must also be taken into consideration. Slaughterhouse equipment suppliers made increased efforts in the last few years to automate the process of slaughtering as far as

possible for economic reasons and for reasons of hygiene. After the attempt to implement a concept of using special machines failed in the market, one is now concentrating on standard industrial robots, as they are being used in large numbers for example in the automobile industry. Initial experience indicates that robots are more reliable and hygienic than human beings. In the meantime, efficient robots are available also for the purpose of primal cutting. In the field of meat cutting, another new technology was evaluated in MRI Kulmbach. Experiments were made cutting pork (*M. longissimus dorsi*) in slices using a high pressure waterjet (3800 bar). The results showed, that the fesh meat surfaces nearly were sterile and the shelf-life of this meat (under retail conditions) was much longer compared to conventionally slicer-cut steaks.

Key words: new technologies, safety, quality, slaughtering, robots, pre-cutting, cutting, waterjet-cutting

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTHOR: Klaus Troeger, klaus.troeger@mri.bund.de, Max Rubner-Institut (MRI) Standort Kulmbach, Bundesforschungsinstitut für Ernährung und Lebensmittel, E.-C.-Baumann-Str. 20, 95326, Deutschland

AUTOR: Klaus Troeger, klaus.troeger@mri.bund.de, Max Rubner Institut (MRI), Standort, Kulmbach, Bundesforschungsinstitut für Ernährung und Lebensmittel, E.-C.-Baumann-Str. 20, 95326, Nemačka

Einleitung

Die Forderung nach Produktsicherheit und damit der Stellenwert der Hygiene bei der Fleischgewinnung und –bearbeitung hat, auch aufgrund der zunehmenden Konzentration der Schlacht- und Zerlegebetriebe mit entsprechend längeren Distributionswegen für das Fleisch, in den letzten Jahren deutlich zugenommen. Zum einen ist die erforderliche Haltbarkeit des Frischfleisches nur bei konsequenter hygienischer Gewinnung und Behandlung erreichbar. Zum anderen dienen hygienische Maßnahmen dem Gesundheitsschutz des Verbrauchers: das Fleisch klinisch gesunder Tiere sollte beim Schlacht- und Zerlegungsprozess nicht mit pathogenen Mikroorganismen (Salmonellen, Listerien, Staphylococcus aureus, shigatoxinbildende *E. coli* u.a.) kontaminiert werden. Dies erfordert geeignete bauliche, technische und organisatorische Maßnahmen der Betriebe.

Neue Technologien der Fleischgewinnung und Zerlegung werden häufig mit dem Ziel einer zunehmenden Automatisierung der Prozesse entwickelt (z.B. Robotereinsatz in Schlachtung und Zerlegung). Die Erfüllung von grundlegenden Hygieneanforderungen, wie beispielsweise eine Vermeidung von Kreuzkontaminationen zwischen verschiedenen Schlachtkörpern in der Schlachtkette, muss dabei jedoch gewährleistet sein. Andererseits können neue technische Anwendungen auch primär auf eine bessere Prozesshygiene abzielen. Ein Beispiel hierfür ist das Schneiden von Fleisch mit Hilfe eines Hochdruck-Wasserstrahls (waterjet).

Einsatz von Industrie-Robotern

Bei der **industriellen Schweineschlachtung** ist heute bereits ein hoher Automatisierungsgrad möglich. Im reinen Bereich der Schlachtlinie verbleiben gegenwärtig als manuelle Tätigkeiten die Entnahme des Urogenitaltrakts, die Separierung der Innereien, die amtliche Fleischuntersuchung sowie die Herrichtung gemäß den Vermarktungsnormen der EU und das Trimmen.

Neben Spezialmaschinen werden zunehmend Standard-Industrieroboter mit eigens entwickelter Software eingesetzt. Die Schwierigkeit besteht darin, Standard-Industrieroboter an die spezifischen Bedingungen eines Schlachtbetriebs zu adaptieren. Im Gegensatz zur Automobilindustrie hat im Schlachtbetriebe jedes „Werkstück“ eine andere Größe und Form. Die Roboter können nicht immer das gleiche, sich wiederholende Bewegungsmuster ausführen – vielmehr müssen die Bewegungen bzw. die Schnittführung durch einen Hochgeschwindigkeits-PC für jeden Schlachtkörper individuell neu berechnet werden. Dazu passieren die Schlachtkörper einen oder mehrere Laserscanner (Abb. 1); eine speziell entwickelte Software liefert die Koordinaten für ein dreidimensionales Bild jedes Schlachtkörpers. Die für die Robotersteuerung erforderlichen Daten stehen innerhalb von wenigen hundert Millisekunden zur Verfügung. Die Bewegungen der Roboter laufen synchron mit der Bewegung des Schlachtförderers.

An Ausführung und Funktion der Roboter sind auch hygienische Anforderungen zu stellen.



Abb. 1. Laserscanner und Industrie-Roboter mit „Bauch- und Brustbeinöffner“ (Fa. Banss, Biedenkopf)
Slika 1. Laserski skener i industrijski robot za rasecanje grudno-trbušnog dela (Fa. Banss, Biedenkopf)

Die Konstruktion muß eine effektive Reinigung erlauben. Die Schutzhülle muß aus für Lebensmittel geeignetem Gewebe bestehen, welches mit Hochdruckwasserstrahl gereinigt werden kann und widerstandsfähig gegen Desinfektionsmittel ist. Die Roboterwerkzeuge müssen nach jedem Arbeitsgang einer effektiven Zwischenreinigung und –desinfektion unterzogen werden. Inwieweit für eine Desinfektion anstelle von 82-gradigem Wasser auch Heißdampf eingesetzt werden kann, ist Gegenstand laufender Untersuchungen.

Bisher sind – bei einer Stundenleistung von bis 650 Schweinen – Roboter für folgende Arbeitsschritte im Einsatz: „Vorderklauen kneifen“, „Rektum freischneiden“ (Abb. 2), „Schlossknochen öffnen“, „Bauch und Brustbein öffnen“ (Abb. 1) sowie „Nacken kneifen“ (Abb. 3). Seit kurzer Zeit ist auch ein Roboter geführter Schweinespalter verfügbar. Fleischhygienische Fragestellungen, insbesondere auch zur Effektivität der automatischen Zwischenreinigung und –sterilisation der Roboterwerkzeuge wurden und werden vom Institut für Sicherheit und Qualität bei Fleisch des MRI, Standort Kulmbach, untersucht. So wurden vergleichende Untersuchungen zur mikrobiellen Kontamination von Schlachtkörpern im Beckenbereich nach Einsatz eines manuellen bzw. Roboter-Bung Droppers (Rektum Freischneider, Abb. 2)

durchgeführt. Es wurden Oberflächen-Muskelpalten aus dem caudalen Beckenbereich destruktiv mit Hilfe einer Stanze (Durchmesser 25 mm) und eines Skalpells bei 101 Schlachtkörpern nach manuellem Bung Dropper-Einsatz und bei 100 Schlachtkörpern nach Roboter Bung Dropper-Einsatz entnommen und die aeroben Gesamtkeimzahlen sowie die Gehalte an Enterobacteriaceen bestimmt. Die Ergebnisse zeigten hygienische Vorteile für die Robotertechnik. Höhere Keimzahlen (10^4 bis $< 10^5$ Gesamtkeime pro cm^2) wurden bei 32 % der manuell bearbeiteten Schlachtkörper, aber nur bei 9 % der Roboter bearbeiteten Schlachtkörper ermittelt (Abb. 4; Troeger, 2008). Ein noch deutlicherer Unterschied zugunsten der Robotertechnik ergab sich bezüglich der Keimgehalte der Nackenmuskulatur nach Einsatz eines manuell bzw. Roboter geführten Nackenkneifers (Abb. 3, Moje, 2009). Der Grund für das bessere Abschneiden der Roboter dürfte in der effektiveren Zwischenreinigung und –sterilisation der Werkzeuge liegen.

Bei der **Grobzerlegung** von Schweinehälften sind ebenfalls bereits Industrie-Roboter im Einsatz. Die Arbeitsstation besteht aus einer vertikalen Fixationsvorrichtung für die Schweinehälften, einem dualen Kamerasystem mit PC zur Bildverarbeitung und einem Industrie-Roboter mit Hygiene-Design. Als Schneidwerkzeug dient eine kleine Krei-



Abb. 2. Industrie-Roboter mit „Rektum-Freischneider“ (Fa. Banss, Biedenkopf)
Slika 2. Industrijski robot za opsecanje rektuma (Fa. Banss, Biedenkopf)

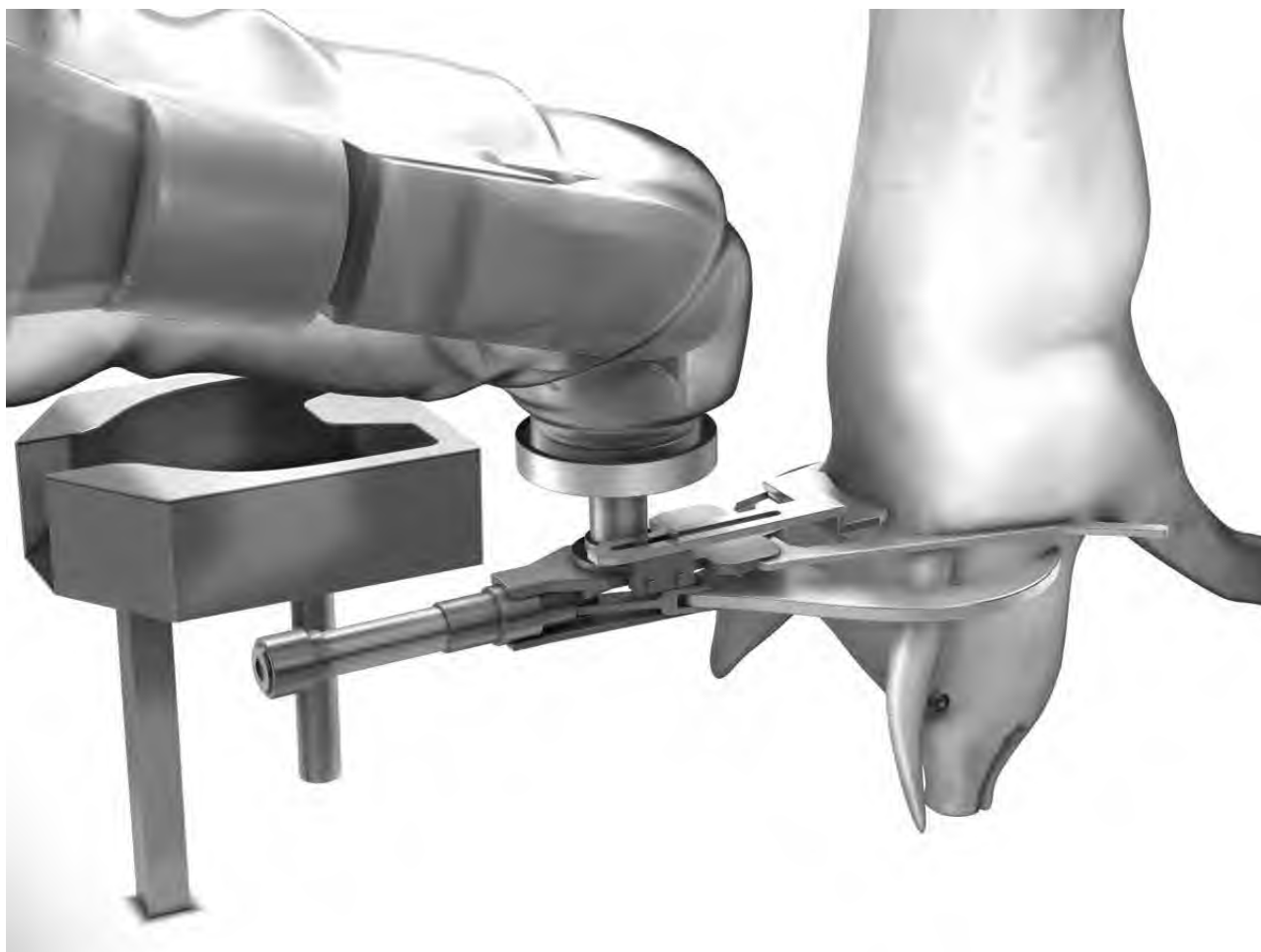


Abb. 3. Industrie-Roboter mit „Nackenkneifer“ (Fa. Banss, Biedenkopf)
Slika 3. Industrijski robot za odvajanje od vrata (Fa. Banss, Biedenkopf)

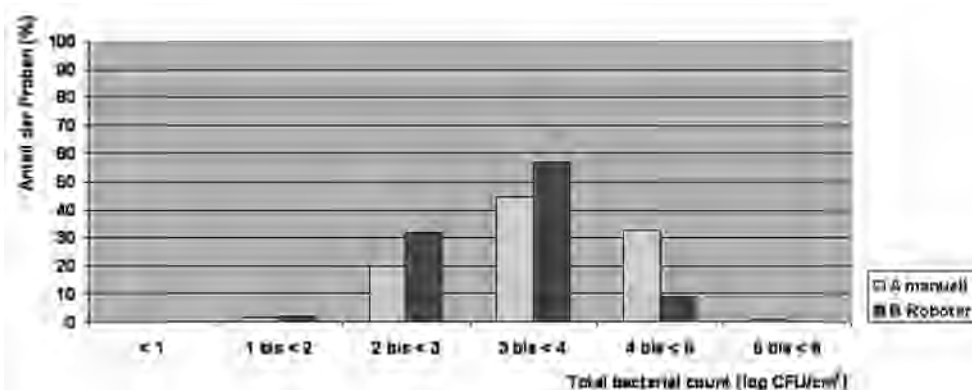


Abb. 4. Gesamtkeimzahlen der Beckenmuskulatur nach manuellem (n = 101) oder Roboter - Bung Dropper – Einsatz (n = 100); 600 Schweine pro Stunde

Slika 4. Ukupan broj bakterija u regiji karlice posle manualnog (n = 101) ili automatskog opsecanja rektuma (n = 101); 600 svinja na sat

ssäge, mit welcher sowohl lineare als auch Kurven-Schnitte ausgeführt werden können. Die Berechnung der Schnittführung orientiert sich an den anatomischen Gegebenheiten. Die Schnittlinien sind nach Kundenanforderungen frei wählbar, saisonale Variationen leicht zu programmieren. Ein Roboter

kann pro Stunde bis zu 1600 individuelle Schnitte ausführen. Die Variation der Schnitte (Abweichung von der Ideallinie) ist beim Roboter mit + - 5 mm deutlich geringer als beim manuellen Sägen (+ - 20 mm). Dies bedingt höhere Ausbeuten an höherwertigen Teilstücken.

Für ein weiteres Zerlegen und das **Entbeinen** von Teilstücken nach Grobzerlegung wurden bereits einige Spezialmaschinen entwickelt. Die sog. Mittelstück-Schneidemaschine trennt das Kotelett vom Bauch (Folkmann and Christensen, 2003). Die sog. Vorderviertel-Maschine entfernt die Rippen und die Halswirbelsäule aus einem Schweinevorderviertel (Hansen, 2004). An einer weiteren Automatisierung des Ausbeinungsprozesses wird weltweit gearbeitet.

Schneiden von Fleisch mit Hochdruck-Wasserstrahl (Waterjet)

Wasserstrahlschneiden wird in einer Reihe von Industriezweigen, wie der Luft- und Raumfahrtindustrie, dem Maschinenbau, der Glas-, Holzverarbeitungs-, Textil-, Papier-, Automobil- und Lebensmittelindustrie routinemäßig eingesetzt. Über Anwendungen in der Fleisch-, Geflügel- und Fischindustrie wurde berichtet (N.N., 2001; Wang and Shanmugam, 2009).

Im Rahmen eines Forschungsprojekts wurde der Einsatz eines Wasserstrahlschneidsystems (Hochdruckpumpe Typ Standard HP19/37-S, Fa. Uhde High Pressure Technologies, Hagen; Edelstahl-Schneidtisch mit Schneiddüse mit variabler Vorschubgeschwindigkeit, Fa. Banns Meat Technologies, Biedenkopf) zum Schneiden frischer Schweinerückenmuskulatur erprobt. An fünf Versuchstagen wurden je zwei ganze entbeinte Schweinerücken aus laufender Produktion eines Zerlegebetriebes entnommen und mittels Wasserstrahl (3800 bar, 0,15 mm Düsendurchmesser, 140 cm/min Vorschubgeschwindigkeit) in jeweils 12 ca. 2 cm dicke Scheiben (= 24 Scheiben pro Versuchstag) geschnitten. Die Rückenmuskeln wurden vor dem Schneiden zwei Stunden in einem Gefrierraum bei -18°C gelagert, so dass die Fleischtemperaturen zum Zeitpunkt des Wasserstrahlschneidens ca. -1,5°C in 2 cm Tiefe und im Kern ca. 0°C betragen. Die Rückenmuskel-Steaks wurden in Plastik-Trays unter Schutzgas ($O_2/CO_2 = 60/40$) verpackt und bei 5°C für 9 bzw. 16 Tage gelagert. Als Kontrollen dienten pro Versuchstag 24 Rückenmuskel-Scheiben („Minuten-Steaks“), die im Zerlegebetrieb aus gleicher Zerlegung mit einem konventionellen Slicer geschnitten und unter Schutzgas ($O_2/CO_2 = 60/40$) verpackt worden waren. Die Kühlung der mit Waterjet geschnittenen und der Kontrollscheiben erfolgte im selben Kühlraum. Es wurden physikalische (Farbe, Tropfsaft) und mikrobiologische Untersuchungen (Enterobakteriazeen-Zahl, aerobe Gesamtkeimzahl) durchgeführt.

Die mit Wasserstrahl geschnittenen Rückenmuskelscheiben waren nach Kühlung etwas heller als die mit dem Messer (Slicer) geschnittenen Kontrollen. Die Tropfsaftverluste (= Flüssigkeit in den Trays) nach 9 bzw. 16 Tagen Kühlung betragen bei den mit Wasserstrahl geschnittenen Scheiben im Mittel 8,9 bzw. 10,3 %, bei den Kontrollscheiben entsprechend 10,2 und 12,2 %. Die gravierendsten Unterschiede traten bei den Oberflächenkeimzahlen nach Lagerung auf. Bei der Mehrzahl der mit Waterjet geschnittenen Rückenmuskelscheiben lag die Gesamtkeimzahl nach 16 Tagen Kühlung unter der Nachweisgrenze von 10 Keimen pro cm^2 . Auch der Maximalwert von 10^4 Keimen/ cm^2 ist, verglichen mit den in der Praxis üblichen Keimbelastungen, noch sehr niedrig. Der Oberflächenkeimgehalt der Kontrollscheiben lag nach 16tägiger Kühlung im Mittel (Median) bei $2,5 \times 10^5$ Gesamtkeimen pro cm^2 (Abb. 5).

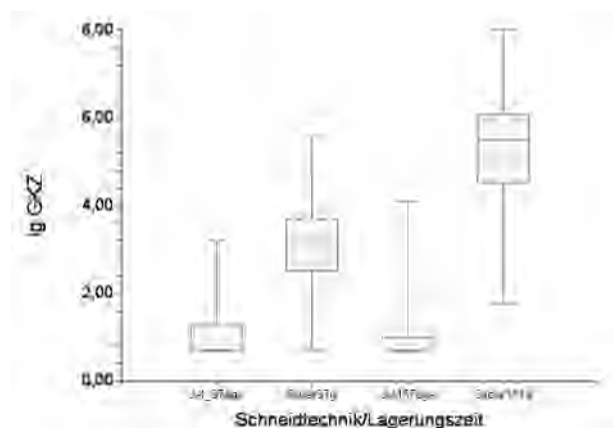


Abb. 5. Gesamtkeimzahlen (GKZ) auf Schweinerücken-Steaks in SB-Schutzgasverpackungen, mit Hochdruck-Wasserstrahl oder Slicer geschnitten, nach Kühlung bei 5°C für 9 bzw. 16 Tage
Slika 5. Ukupan broj bakterija u odrescima svinjskog mesa (lumbalni deo) u MAP, isečenog vodenim nožem ili uredajem za narezivanje pri temperaturi od 5°C u toku 9-16 dana

Die weitgehend keimfreien Oberflächen der mit Waterjet geschnittenen Steaks resultieren wahrscheinlich aus einer fehlenden Kontamination der Schnittflächen mit von der Fleischoberfläche verschleppten Keimen. Während es beim Schneiden mit dem Messer zwangsläufig zu einer gewissen Kontamination des Schneidwerkzeugs und damit der frischen Schnittflächen kommt, zerstört der Hochdruck-Wasserstrahl möglicherweise die Ober-

flächenkeime beim Aufprall. Außerdem wird Material entlang der Schnittebene in der Breite des Strahls (steril) abgetragen. Einer praktischen Anwendung der Technik in diesem Bereich sind jedoch aufgrund der, im Vergleich zu konventionellen Slicern, relativ geringen Schnittgeschwindigkeiten noch Grenzen gesetzt. Andererseits erscheint eine Anwendung

des Wasserstrahl-Schneidens bei Schlachtung und Zerlegung durch einen Industrie-Roboter mittelfristig realisierbar und aufgrund der zu erwartenden Hygienevorteile auch sinnvoll. Roboter, die mit Wasserstrahl schneiden, sind in anderen Industriebereichen bereits im Einsatz (N.N., 2008).

References

- Folkmann, P., Christensen, F. H., 2003.** Technology behind a new machine for automatic cutting of pork middles. *Fleischwirtschaft International* 3, 52–54;
- Hansen, F., 2004.** Robot ready for 47 million fore-ends. The world's first robot for removal of surface bones is installed at Danish Crown in Saeby. *Fleischwirtschaft International* 1, 31–32;
- Moje, M., 2009.** Robotereinsatz in der industriellen Schweineschlachtung – hygienische und wirtschaftliche Aspekte. *Mitteilungsblatt der Fleischforschung Kulmbach* Nr. 184, im Druck;
- N.N. 2001.** Waterjet cutting in the food industrie. A white paper. Flow Intern. Corp., Kent, WA, USA;

- N.N. 2008.** Roboter für komplexe Prozesse fit machen. *Robotworld* 03/08, 27;
- Troeger, K., 2008.** Aktuelle Forschungsschwerpunkte des Max Rubner-Instituts Kulmbach. Proc. 11. Internationale wissenschaftliche Konferenz gewidmet V. M. Gorbатов, 21-30 (kyrillisch). Gorbатов's All-Russisches Fleischforschungsinstitut, Moskau;
- Wang, J., Shanmugam, D. K., 2009.** Cutting meat with bone using an ultrahigh pressure abrasive waterjet. *Meat Sci.* 81, 671–677;

Paper recieved: 15.04.2009.

CAUSE AND POSSIBLE WAYS TO ELIMINATE BOAR TAINT IN PORK*

Zamaratskaia Galia

Abstract: Boar taint, an undesirable odour from meat from some entire male pigs, is caused by the naturally occurring compounds androstenone and skatole. The level of boar taint can be minimized by decreasing the concentrations of these compounds in adipose tissue. Immunocastration substantially reduces the levels of both. Skatole levels can be also reduced by dietary manipulations and improved rearing conditions; however, this approach has no or little effect on androstenone. Genetic selection against high androstenone and skatole levels is an attractive alternative if achieved without negatively affecting reproduction and economic efficiency. If entire male pigs are to be used in pork industry, methods to detect tainted carcasses are needed. Tainted carcasses can be used for processed meat products. Meat processing can probably reduce or mask boar taint; however, more studies are needed to investigate possible processing techniques and consumer attitudes towards final pork product. Thus, in future, surgical castration of male piglets can be avoided and replaced by practical and ethically acceptable alternatives. At the moment, castration using anaesthesia and analgesia, or immunocastration can be used as temporary solutions. This article reviews the development of some alternatives to surgical castration of entire male piglets to control boar taint.

Key words: boar taint; Surgical castration; Anaesthesia and analgesia; Immunocastration; Use of entire male pigs

Uzrok i mogućnosti eliminacije polnog mirisa u svinjskom mesu

Sadržaj: Polni miris nerastova, neprijatni miris koji potiče od mesa kod muških jedinki, uzrokovan je jedinjenjima androstenon i skatol, koji se prirodno nalaze kod ovih životinja. Intenzitet polnog mirisa može da se umanjí smanjenjem koncentracije navedenih jedinjenja u masnom tkivu. Imunokastracija značajno smanjuje nivoe i androstenona i skatola.

Nivo skatola može da se smanji i promenom ishrane kao i poboljšanjem uslova uzgoja. Međutim, ovakav pristup ne utiče na nivo androstenona. Genetska selekcija jedinki sa manjim nivoima androstenona i skatola je pogodna alternativa ukoliko ne utiče negativno na reprodukciju i ekonomsku isplativost uzgoja. Ukoliko se nerastovi koriste u industriji neophodno je razviti metode detekcije polnog mirisa trupova. Ovakvi trupovi mogu da se iskoriste u preradi. Prerada verovatno može da umanjí ili maskira polni miris. Međutim, potrebne su dalje studije koje bi istražile moguće tehnike kao i odnos potrošača prema gotovom proizvodu. Sioga je u budućnosti moguće izbeći hiruršku kastraciju prasadi muškog pola i zameniti je praktičnijim i etički prihvatljivijim alternativama. Kastracija pod anestezijom ili analgezijom, kao i imunokastracija su u ovom trenutku privremena rešenja. Ovaj rad pruža osvrt na razvoj nekih alternativa hirurškoj kastraciji prasadi muškog pola radi kontrolisanja polnog mirisa.

ključne reči: polni miris, hirurška kastracija, anestezija i analgezija, imunokastracija, korišćenje nerastova

Introduction

Surgical castration of entire male piglets not intended for breeding is routinely performed in many European countries to reduce the risk of boar taint, an off-flavour in heated pork products. Boar taint occurs in some entire male pigs at puberty and is primarily caused by high levels of androstenone and/or skatole in pig carcasses. Although surgical castration reduces the levels of both compounds and therefore decreases the risk of boar taint, this approach is not fully satisfactory. Entire male pigs compared to castrates have a superior feed efficiency and higher lean yield of the carcasses. Moreover,

surgical castration has been increasingly disparaged because of its negative effects on animal health and welfare. Therefore, to prevent boar taint, methods other than surgical castration are required. Ideally, such methods should be easy for use on farms and effectively reduce taint in entire male pigs. Various factors are known to regulate the levels of skatole and androstenone in pig carcasses and these factors have been regularly reviewed (Bonneau, 1982; Claus *et al.*, 1994; Bonneau, 1998; Lundström and Zamaratskaia, 2006; Zamaratskaia and Squires, 2009). The purpose of the present short review is to highlight selected aspects of the boar taint problem and to provide a summary of current knowledge on

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

skatole and androstenone. Special focus is given to the potential alternatives to surgical castration.

Cause of boar taint

Skatole (3-methylindole) and androstenone (5 α -androst-16-en-3-one) are two major compounds responsible for boar taint (Dijksterhuis *et al.*, 2000). Skatole is produced by the microbial degradation of tryptophan in the large intestine of pigs. Androstenone is a steroid produced in the Leydig cells of the testis in mature pigs. Skatole is perceived by most people as a faecal-like odour, whereas ability to detect androstenone is highly variable between people with different genetic background (Wysocki and Beauchamp, 1984). Descriptions of androstenone odour vary from sweat- or urine-like to perfume- or flower-like odour. There are indications that other compounds can contribute to boar taint, such as indole (Garcia-Requeiro and Diaz, 1989) and androstenol (Brooks and Pearson, 1989). Their contribution, however, seems to be of less importance because of relatively weak odour.

Effects of surgical castration

Surgical removal of testicles is an effective method to remove the source of testicular steroids, including the boar taint compound androstenone. Surgical castration also prevents the accumulation of another boar taint compound – skatole – due to enhanced skatole metabolic clearance in the absence of testicular steroids (Doran *et al.*, 2002; Zamaratskaia *et al.*, 2007). Furthermore, surgical castration removes the source of spermatozoa and prevents the male pigs from unplanned breeding. However, surgical castration of piglets negatively affects animal welfare which is a severe drawback of this method. Currently there are no widely accepted alternative to surgical castration.

“Humane” castration

Use of local anaesthesia

Effective local anaesthesia is one option to reduce pain in piglets during surgical castration. Surgical castration with anaesthesia using 10 mg of Procaine (Procasel 2%®, Selectavet, Germany) per testis reduces the intensity of pain during castration as assessed by changes in vocalisation and defence behaviour of piglets (Leidig *et al.*, 2009). Marx *et al.* (2003) demonstrated that piglets castrated without anaesthesia produced significantly more screams

than piglets castrated with local anaesthesia with lidocain (Ursocain 2%; 0.5 ml per testis). However, piglets during injection are subjected to an additional distress due to prolonged handling and pain due to injection. The use of local anaesthesia improves, although not completely, the welfare status of piglets but increases the costs of the procedure. Additionally, anaesthesia lasts for a short time, and the use of extra analgesic agents is recommended. Finally, the procedure should only be performed by veterinarians in most countries.

Immunocastration

Some progress has recently been made in development of a vaccine for immunization against gonadotrophin releasing factor (GnRF). Considerable experimental evidence supports the notion that this is a reliable non-surgical method to control both boar taint and aggressive behavior of entire male pigs. Blocking the action of GnRF by creating GnRF antibodies stops testicular function, thus producing a temporary castration effect and preventing accumulation of androstenone and skatole in boar's tissues. A potentially promising vaccine, Improvac™, has recently been tested in some countries (Dunshea *et al.*, 2001; Jaros *et al.*, 2005; Zamaratskaia *et al.*, 2008a,b; Font i Furnols *et al.*, 2008). Immunocastration with Improvac consistently reduced the production of testicular steroids and androstenone along with the size of reproductive organs, as well as skatole levels.

Possibility of use of entire male pigs

Reduction of slaughter weight

Slaughter at a younger age/lower live weight (before puberty) can reduce the risk of increased levels of androstenone and skatole. In some countries, e.g. Ireland and the United Kingdom, male pigs are produced intact. This approach does not negatively affect animal welfare; however, from an economic point of view it is not an attractive option. Additionally, slaughter at lower weight does not entirely eliminate boar taint (Aldal *et al.*, 2005; Zamaratskaia *et al.*, 2005a).

Management strategies (diet and hygienic conditions)

Given that skatole originates from tryptophan in porcine large intestine, it is not surprising that dietary composition is an important factor affecting skatole levels. Reduction of skatole levels by dietary means has been a subject of a considerable research

effort over the past decades (Lundström *et al.*, 1994; Jensen *et al.*, 1995; Claus *et al.*, 2003; Zamaratskaia *et al.*, 2005a). Non-digestible carbohydrates are known to reduce intestinal production of skatole. For instance, Jensen *et al.* (1995) and Whittington *et al.* (2004) found reduced skatole levels in fat in pigs fed sugar beet pulp. Dietary supplement of raw potato starch reliably reduced skatole levels in porcine tissues in castrated male pigs (Claus *et al.*, 2003), entire male pigs (Zamaratskaia *et al.*, 2005a) and female pigs (Zamaratskaia *et al.*, 2006). This reduction might be due to the inhibition of cell apoptosis in the colon and thus reduced tryptophan availability for skatole production (Claus *et al.*, 2003). Butyrate, which is formed in high quantities when the supply of resistant starch is high, can cause a reduction in apoptosis of epithelial cells and reduces availability of the skatole precursor tryptophan (Claus *et al.*, 2003). Additionally, changes in dietary composition may modify intestinal transit time and the microbial activity in the intestine (Jensen *et al.*, 1995).

Environment is also an important factor affecting skatole production in the intestine. Temperature and ventilation in the stable as well as stocking rate were shown to affect skatole levels (Hansen *et al.*, 1994).

Genetic selection for 'low taint' pigs

Some selection experiments have been performed to reduce androstenone levels (Sellier and Bonneau, 1988; Willeke and Pirchner, 1989; Sellier *et al.*, 2000). However, the selection against androstenone can lead to reduced levels of anabolic hormones as well, which in turn negatively affects growth performance of entire male pigs and onset of puberty in gilts and boars (Sellier and Bonneau, 1988; Willeke and Pirchner, 1989). To eliminate the undesirable side-effects of selection procedures it is essential to detect pigs that express low androstenone levels at sexual maturity. The development of genetic markers for pigs with low androstenone and skatole levels would allow the selection of taint-free pigs. The subject has been reviewed in more detail elsewhere (Zamaratskaia and Squires, 2009).

Detection of boar taint

The use of entire male pigs in pork industry requires identification of pigs with high level of boar taint to insure that no tainted meat reaches the consumers. Rapid, cheap and reliable methods to detect boar taint are needed. There are a number of analytical methods, e.g. HPLC, LC-MS, GC, GC-MS, RIA and ELISA, developed for the measurement of

concentrations of skatole, androstenone and both in adipose tissue. However, the application of these methods on the slaughter line is not realistic because of complicated sample preparation and purification steps. The colorimetric method to measure skatole equivalents in adipose tissue (Mortensen and Sørensen, 1984) has been used online in Danish slaughterhouses. This method is rapid and simple; however, it does not provide information about the levels of the other important boar taint compound, androstenone. A colorimetric method for total 16-androstenes was also developed (Squires, 1990) but never used at slaughterhouses. It was recently suggested that measurements of boar taint levels in carcasses can be performed using an electronic nose based on ion mobility spectrometry (Vestergaard *et al.*, 2006). However, an automatization of an on-line system based on electronic nose technology requires further development. As discussed by Vestergaard *et al.* (2006), "this would not necessarily imply the need for a skatole and androstenone specific sensor array, since also other possible compounds may be involved in the sensory perception of boar taint, but rather a broad-selectivity sensor array that matches the sensory perception of boar taint, which in turn should be calibrated against national consumer thresholds".

Besides, other simple methods to detect pigs with high boar taint levels have been proposed, e.g. measurement of reproductive organ sizes. Bonneau and Russeil (1985) suggested that the measurement of bulbourethral glands could be used as an indirect estimation of androstenone levels in fat from entire male pigs. Zamaratskaia *et al.* (2005b) showed that pigs of a crossbred (Swedish Yorkshire dams×Swedish Landrace sires) with testes weight below 565 g and a bulbourethral gland length below 90 mm had low skatole levels; low androstenone levels in this study could not be predicted by the size of reproductive organs. Pigs with reproductive organs above those levels should further be tested for skatole concentrations in fat. Thus, the use of such a method can reduce the number of carcasses for chemical analyses, but cannot be used as the basis on which to reject carcasses. Therefore, further investigations are required to develop a rapid and sensitive method for the systematic analysis of boar carcasses. The tainted meat could then be used for processed meat products.

Camouflage of boar taint

Except for potential presence of boar taint, meat from entire male pigs does not substantially differ from that from female or castrated pigs. The-

refore, tainted meat can be used after diluting with non-tainted meat. Processing of meat from entire male pigs can also neutralize the perception of boar taint. It was suggested that liquid smoke was able to mask the taint perception in sausages from entire male pigs (Stolzenbach *et al.*, 2009). Wood *et al.* (1993) demonstrated the importance of the cooking temperature on the acceptability of meat from entire male pigs. Finally, consumption of cold products from tainted meat does not induce such strong negative reactions among consumers as consumption of products immediately after heating (Pearson *et al.*, 1971). However, development of processing technology to camouflage boar taint needs more research.

Sorting sperm for sex pre-selection

Gender selection has lately been discussed as a promising tool for the pork industry (Johnson, 2000). Production of female-only herds through sex pre-selection is an alternative to surgical castration. However, the technique for gender selection is not commercially available at present. Large quantities of sperm are required for such a selection because of sperm losses and cell damage during selection. The other severe drawback of this method is an image of “manipulating nature”. However, the technique might become a promising strategy in pork production if it is effective and precise, and

costs of sperm separation are low. The current status of sexing technology in the pig and methodological developments is reviewed in Vazquez *et al.* (2009).

Conclusion

Boar taint, an undesirable odour from meat from some entire male pigs, is caused by the naturally occurring compounds androstenone and skatole. The level of boar taint can be minimized by decreasing the concentrations of those compounds in adipose tissue, e.g. via immunocastration, genetic selection, dietary manipulations and improved rearing conditions. Meat processing can probably reduce or mask boar taint; however, more studies are needed to investigate possible processing techniques and consumers attitudes towards final pork product. Genetic selection against high boar taint is probably the most attractive alternative, but is not realistic in the near future. At the moment, the best temporary solutions are “humane” castration using anaesthesia and analgesia, or immunocastration. The advantages and disadvantages of alternative methods should be carefully studied before the final decision is made about how to prevent boar taint without the need of stressful and painful surgical castration. It is generally believed that in future, surgical castration of male piglets can be avoided and replaced by practical and ethically acceptable alternatives.

References

- Aldal I., Andresen O., Egeli A. K., Haugen J.-E., Grodum A., Fjetland O., Eikaas, J. L. H., 2005. Levels of androstenone and skatole and the occurrence of boar taint in fat from young boars. *Livestock Production*, 95, 1–2, 121–129;
- Bonneau M., 1982. Compounds responsible for boar taint, with special emphasis on androstenone: a review. *Livestock Production Science*, 9, 6, 687–705;
- Bonneau M., Russeil P., 1985. The size of Cowper’s (bulbourethral) glands as an estimate of boar taint on the slaughter line. *Livestock Production Science*, 13, 2, 169–178;
- Bonneau M., 1998. Use of entire males for pig meat in the European Union. *Meat Science*, 49, Supplement 1, S257–S272;
- Brooks R. I., Pearson, A. M., 1989. Odor thresholds of the C19- δ 16-steroids responsible for boar odor in pork. *Meat Science* 25, 1, 11–19;
- Claus R., Weiler U., Herzog A., 1994. Physiological aspects of androstenone and skatole formation in the boar—a review with experimental data. *Meat Science*, 38, 2, 289–305;
- Claus R., Lösel D., Lacorn M., Mentschel J., Schenkel H., 2003. Effects of butyrate on apoptosis in the pig colon and its consequences for skatole formation and tissue accumulation. *Journal of Animal Science*, 81, 1, 239–248;
- Dijksterhuis G. B., Engel B., Walstra P., Font-i-Furnolls M., Agerhem H., Fischer K., Oliver M. A., Claudi-Magnussen C., Siret F., Beague M. P., Homer D. B., Bonneau M., 2000. An international study on the importance of androstenone and skatole for boar taint. II. Sensory evaluation by trained panels in seven European countries. *Meat Science*, 54, 3, 261–269;
- Doran E., Whittington F. M., Wood J. D., McGivan J. D., 2002. Cytochrome P450IIE1 (CYP2E1) is induced by skatole and this induction is blocked by androstenone in isolated pig hepatocytes. *Chemico-Biological Interactions*, 140, 1, 81–92;
- Dunshea F. R., Colantoni C., Howard K., McCauley I., Jackson P., Long K. A., Lopaticki S., Nugent E. A., Simons J. A., Walker J., Hennessy D. P., 2001. Vaccination of boars with a GnRH vaccine (Improvac) eliminates boar taint and increases growth performance. *Journal of Animal Science*, 79, 10, 2524–2535;
- Font i Furnols M., Gisbert M., Guerrero L., Velarde A., Tibau J., Soler J., Hortós M., García-Regueiro J. A., Pérez J., Suárez P., Oliver M. A., 2008. Consumers’ sensory acceptability of pork from immunocastrated male pigs. *Meat Science*, 80, 4, 1013–1018;
- Garcia-Regueiro J. A., Diaz I., 1989. Evaluation of the contribution of skatole, indole, androstenone and androstenols to boar-taint in back fat of pigs by HPLC and capillary gas chromatography (CGC). *Meat Science*, 25, 4, 307–316;
- Hansen L. L., Larsen A. E., Jensen B. B., Hansen-Møller J., Barton-Gade P., 1994. Influence of stocking rate and

- faeces deposition in the pen at different temperatures on skatole concentration (boar taint) in subcutaneous fat. *Animal Production*, 59, 1, 99–110;
- Jaros P., Bürgi E., Stärk K. D. C., Claus R., Hennessy D., Thun R., 2005.** Effect of active immunization against GnRH on androstenone concentration, growth performance and carcass quality in intact male pigs. *Livestock Production Science*, 92, 1, 31–38;
- Jensen M. T., Cox R. P., Jensen B. B., 1995.** Microbial production of skatole in the hind gut of pigs given different diets and its relation to skatole deposition in backfat. *Animal Science*, 61, 293–304;
- Johnson L. A., 2000.** Sexing mammalian sperm for production of offspring: the state-of-the-art. *Animal Reproduction Science*, 61–62, 93–107;
- Leidig M. S., Hertrampf B., Failing K., Schumann A., Reiner G., 2009.** Pain and discomfort in male piglets during surgical castration with and without local anaesthesia as determined by vocalisation and defence behaviour. *Applied Animal Behaviour Science*, 116, 2–4, 174–178;
- Lundström K., Malmfors B., Stern S., Rydhmer L., Eliasson-Selling L., Mortensen A. B., Mortensen H. P., 1994.** Skatole levels in pigs selected for high lean tissue growth rate on different dietary protein levels. *Livestock Production Science*, 38, 2, 125–132;
- Lundström K., Zamaratskaia G., 2006.** Moving towards taint-free pork – alternatives to surgical castration. *Acta Veterinaria Scandinavica* 48 Suppl 1. art. no. S1;
- Marx G., Horn T., Thielebein J., Knubel B., Von Borell E., 2003.** Analysis of pain-related vocalization in young pigs. *Journal of Sound and Vibration*, 266, 3, 687–698;
- Mortensen A. B., Sørensen S. E., 1984.** Relationship between boar taint and skatole determination with a new analysis method. In *Proceedings of 30th European Meeting Meat Research Workers*, Bristol, UK, 394–396;
- Pearson A. M., Ngoddy S., Price J. F., Larzelere H. E., 1971.** Panel acceptability of products containing boar meat. *Journal of Animal Science*, 33, 26–29;
- Sellier P., Bonneau M., 1988.** Genetic relationships between fat androstenone levels in males and development of male and female genital tract in pigs. *Journal of Animal Breeding and Genetics* 105, 11–20;
- Sellier P., Le Roy P., Fouilloux M. N., Gruand J., Bonneau M., 2000.** Responses to restricted index selection and genetic parameters for fat androstenone level and sexual maturity status of young boars. *Livestock Production Science*, 63, 3, 265–274;
- Stolzenbach S., Lindahl G., Lundström K., Chen G., Byrne D. V., 2009.** Perceptual masking of boar taint in Swedish fermented sausages. *Meat Science*, 81, 4, 580–588;
- Squires E. J., 1990.** Studies on the suitability of a colorimetric test for androst-16-ene steroids in the submaxillary gland and fat of pigs as a simple chemical test for boar taint. *Canadian Journal of Animal Science*, 70, 1029–1040;
- Vazquez J. M., Parrilla I., Roca J., Gil M. A., Cuello C., Vazquez J. L., Martínez E. A., 2009.** Sex-sorting sperm by flow cytometry in pigs: Issues and perspectives. *Theriogenology*, 71, 1, 80–88;
- Vestergaard J. S., Haugen J.-E., Byrne D. V., 2006.** Application of an electronic nose for measurements of boar taint in entire male pigs. *Meat Science*, 74, 3, 564–577;
- Whittington F. M., Nute G. R., Hughes S. I., McGivan J. D., Lean I. J., Wood J. D., Doran E., 2004.** Relationships between skatole and androstenone accumulation, and cytochrome P4502E1 expression in Meishan × Large White pigs. *Meat Science*, 67, 4, 569–576;
- Willeke H., Pirchner F., 1989.** Selection for high and low level of 5 α -androst-16-en-3-one in boars. II. Correlations between growth traits and 5-androstenone. *Journal of Animal Breeding and Genetics*, 106, 312–317;
- Wood J. D., Nute G. R., Cuthbertson A., 1993.** Optimum cooking conditions for pork. *Meat Focus International*, 453–455;
- Wysocki C. J., Beauchamp G. K., 1984.** Ability to smell androstenone is genetically determined. *Proceedings of the National Academy of Sciences of the United States of America*, 81, 15, 4899–4902;
- Zamaratskaia G., Babol J., Andersson H. K., Andersson K., Lundström K., 2005a.** Effect of live weight and dietary supplement of raw potato starch on the levels of skatole, androstenone, testosterone and oestrone sulphate in entire male pigs. *Livestock Production Science* 93, 3, 235–243;
- Zamaratskaia G., Rydhmer L., Chen G., Madej A., Andersson H. K., Lundström K., 2005b.** Boar taint is related to endocrine and anatomical changes at puberty but not to aggressive behaviour in entire male pigs. *Reproduction in Domestic Animals*, 40, 6, 500–506;
- Zamaratskaia G., Chen G., Lundström K., 2006.** Effects of sex, weight, diet and hCG administration on levels of skatole and indole in the liver and hepatic activities of cytochromes P4502E1 and P4502A6 in pigs. *Meat Science*, 72, 2, 331–338;
- Zamaratskaia G., Gilmore W. J., Lundström K., Squires E. J., 2007.** Effect of testicular steroids on catalytic activities of cytochrome P450 enzymes in porcine liver microsomes. *Food and Chemical Toxicology* 45, 4, 676–681;
- Zamaratskaia G., Andersson H. K., Chen G., Andersson K., Madej A., Lundström K., 2008a.** Effect of a gonadotropin-releasing hormone vaccine (Improvac®) on steroid hormones, boar taint and performance in entire male pigs. *Reproduction in Domestic Animals*, 43, 3, 351–359;
- Zamaratskaia G., Rydhmer L., Andersson H. K., Chen G., Lowagie S., Andersson K., Lundström K., 2008b.** Long-term effect of vaccination against gonadotropin-releasing hormone, using Improvac®, on hormonal profile and behaviour of male pigs. *Animal Reproduction Science*, 108, 1–2, 37–48;
- Zamaratskaia G., Squires E. J., 2009.** Biochemical, nutritional and genetic effects on boar taint in entire male pigs. *Animal* (doi:10.1017/S1751731108003674).

Paper received: 15.04.2009.

HOCHDRUCKBEHANDLUNG BEI FLEISCHERZEUGNISSEN*

Dederer Irina

Zusammenfassung: Es wurden die durch die Hochdruckbehandlung (HDB) von Fleischerzeugnissen hervorgerufenen chemisch-physikalischen, mikrobiologischen und sensorischen Veränderungen diskutiert. Die Wirkung vom hohen hydrostatischen Druck wurde auf die Inaktivierung der produktspezifischen Kontaminationsflora in vakuumverpacktem Brühwurstaufschnitt und der Bakteriensporen in Brühwurstkonserven untersucht. Durch die HDB mit 600 MPa bei 20 °C konnte kein Wachstum von nicht sporenbildenden Bakterien festgestellt werden und somit wurde die Haltbarkeit des untersuchten Brühwurstaufschnittes wesentlich verbessert. Bei der kombinierten Anwendung von Hochdruck- und Wärmebehandlung war es möglich durch die druckinduzierte Auskeimung bei einem moderaten Druck von 300 MPa und nachfolgender Pasteurisation alle Sporenbildner zu inaktivieren. Die so hergestellten tropenlagerfähigen Brühwurstkonserven waren von sehr guter sensorischer Qualität.

Schlüsselwörter: hochdruckbehandlung, fleischerzeugnisse, brühwürstaufschnitt

High-pressure treatment of meat products

Abstract: Chemical-physical, microbiological and sensory changes caused by the high-pressure treatment (HPT) of meat products were reported. The effect of the high hydrostatic pressure was examined for the inactivation of the contamination flora specific for vacuum-packed sliced cooked sausages and the bacterial spores in canned cooked sausages. By the HPT of 600 MPa and 20 °C it was ascertained no growth of the not sporeformers and therefore the stability of the examined sliced cooked sausages was substantially improved. With the combined application of high-pressure and heat treatment it was possible to inactivate all sporeformers by the pressure-induced germination with a moderate pressure of 300 MPa and the following pasteurization. Tropical-storable canned cooked sausages made in this way were of very good sensory quality.

Key words: high-pressure treatment, meat products, cooked sausages

Tretiranje proizvoda od mesa visokim pritiskom

Sadržaj: U radu su prikazane fizičko-hemijske, mikrobiološke i senzorne promene proizvoda od mesa izazvane tretmanom sa visokim pritiskom (TVP). Ispitivan je efekat visokog hidrostatskog pritiska na inaktivaciju mikroflora specifične za vakuum pakovanje slajsovane kuvane kobasice i bakterijskih spora kod kuvanih kobasica u konzervi. Primenom tretmana sa visokim pritiskom (TVP) od 600 Mpa, na temperaturi od 20 stepeni, postignuta je potpuna inhibicija sporogenih mikroorganizama, i na taj način, značajno povećana stabilnost ispitivanih kuvanih kobasica. Kombinovanom primenom tretmana sa visokim pritiskom (TVP) i termičkog tretmana inaktivisani su svi sporogeni mikroorganizmi, germinacijom koja je izazvana pritiskom, primenom umerenog pritiska od 300 Mpa i pratećom pasterizacijom. Ovakvo izrađene tropske kuvane kobasice bile su veoma dobrog senzornog kvaliteta.

Ključne reči: tretman visokim pritiska, proizvodi od mesa, kuvane kobasice

Einleitung

Die Anwendung hoher hydrostatischer Drücke als neues und zukunftsweisendes Verfahren der Lebensmittelbehandlung hat sich innerhalb des letzten Jahrzehnts in einigen Lebensmittelbereichen praktisch durchgesetzt. Die Lebensmittel, wie auch Fleischerzeugnisse, können auf relativ schonende Weise, ohne Anwendung hoher Temperaturen haltbar gemacht werden. Es gibt aber bei Fleisch und

Fleischerzeugnissen wenige Forschungsergebnisse, die die Vorteile dieser Technologie gegenüber konventionellen Konservierungsverfahren belegen.

Auf dem internationalen Markt gibt es bereits einige Produkte, die mit großem Erfolg vermarktet werden. Die folgenden Einsatzgebiete geben einen Eindruck von den vielfältigen Möglichkeiten dieses neuen Konservierungsverfahrens: Japan: Säfte, Konfitüren, Desserts, Fruchtkonzentrate; USA: Avocadopürees, Direktsäfte; England:

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

Milchprodukte; Frankreich: Gänseleberpastete, Direktsäfte; Spanien: Roh- und Kochschinkenprodukte; Kanada: Geflügelfleischprodukte; Deutschland: Rohschinken, Fruchtzubereitungen. Bei den genannten Produkten brachte die Hochdrucktechnologie deutliche Qualitätsvorteile gegenüber der konventionellen Hitzekonservierung.

Zweck der HDB von Lebensmitteln: Konservierung (Abtötung von Mikroorganismen), Veränderung von Reaktionskinetiken, Proteindenaturierung, Enzyminaktivierung oder –aktivierung, Änderung der Eigenschaften von Polymeren (Kohlenhydraten und Fetten). Die hydrostatischen Drücke, die im Lebensmittelbereich angewandt werden, bewegen sich im Bereich zwischen 100 und 1000 MPa. Die Druckgefäßgrößen kommerzieller Anlagen liegen heute zwischen 100 und 500 Litern. Die Behandlung erfolgt meist diskontinuierlich. Wegen der augenblicklich noch hohen Gerätekosten beschränkt sich die Anwendung auf qualitativ hochwertige Produkte. Es ist allerdings abzusehen, dass sich mit fortschreitender technischer Entwicklung und größerer praktischer Erfahrung die Anzahl vermarktungsfähiger Produkte erhöhen wird und die Kosten dieser umweltfreundlichen Technologie zurückgehen werden. Die weitaus meisten Anwendungen und Patente befassen sich bisher mit Obst und Gemüse, während Lebensmittel wie Milch, Fleisch und Fisch, vor allem hinsichtlich chemisch-physikalischer Wirkungen einer HDB, eine untergeordnete Rolle spielen. Dies mag damit zusammenhängen, dass die durch die HDB ausgelösten Umsetzungen in den protein- und fetthaltigen Lebensmitteln tierischer Herkunft vielgestaltiger und damit unübersichtlicher sind als in der Matrix eines Gemüses oder eines Obstsaftes.

Untersuchungen an Fleisch und Fleischerzeugnissen zeigten, dass die sensorische Qualität nur bedingt erhalten werden kann. Die Vorbehandlung – Denaturierung, Abtrocknungsgrad, Oberflächen-Volumen-Verhältnis – sowie Rezeptur und Umgebungsbedingungen haben Einfluss auf durch die HDB bewirkte Effekte in Farbe, Konsistenz, Geschmack und Mikrobiologie. Um für die Praxis relevante Resultate zu erhalten, müssen konkrete Untersuchungen an einzelnen Lebensmitteln durchgeführt werden.

Einfluss der HDB auf die stoffliche Zusammensetzung

Wasserlösliche **Vitamine** wie Vitamin C, Vitamine B1, B2 und B6 und Folsäure scheinen durch die Druckbehandlung unter realistischen Produktionsbedingungen nicht oder nur wenig (Serfert, 2002) beeinflusst zu werden.

Über die **Oxidation der Fette** im Lebensmittel durch Hochdruckbehandlung finden sich widersprüchliche Aussagen, die oftmals nicht deutlich gegen die Veränderungen während der Lagerung abgegrenzt sind. Enzymatische Restaktivitäten, Fettsäurespektrum, Wassergehalte, pH-Wert, Oxidationsgrad vor der Druckbehandlung, Pro- und Antioxidantien haben einen entscheidenden Einfluss auf die druckinduzierte Veränderung der Lipide und den Oxidationsverlauf während der Lagerung. Strukturveränderungen der Zellmembran bis hin zur Zerstörung des Zellverbundes beeinflussen ebenfalls die Oxidation der Lipide. Ergebnisse unserer Untersuchung der Fettoxidation bei Brühwurstkonserven zeigten, dass die HDB nur einen minimalen Einfluss auf die Fettoxidation hat. Bei der Rohwurst kam es durch HDB zu einem leichten Anstieg der Fettoxidationsparameter. Während der nachfolgenden Lagerung traten geringfügige oxidative hochdruckinduzierte Fettveränderungen auf.

Kohlenhydrate zeigen sich weitgehend unempfindlich gegen Druck. Jedoch können Polysaccharide hinsichtlich Wasserbindungs- und Gelbildungseigenschaften beeinflusst werden. Die Veränderungen betreffen jedoch die funktionalen Eigenschaften und beinhalten nicht strukturelle Änderungen. (Pfister, 2000).

Die Primärstruktur der **Proteine** wird durch Druck nicht beeinflusst. Der Druck beeinflusst hydrophobe Wechselwirkungen und damit die Quartärstruktur, die Tertiärstruktur durch reversibles Entfalten und die Sekundärstruktur durch irreversibles Entfalten des Proteins. Druckinduzierte Gele haben andere rheologische Eigenschaften als hitzeinduzierte. Die Protease-Abbaubarkeit druckmodifizierter Proteine ist erhöht, was möglicherweise auf eine höhere Wasserbindungskapazität hindeutet. Von besonderem Interesse ist das Verhalten von **Prion-Proteinen**. So führte Hochdruckbehandlung von (Hamster und Rinder) Prion-Proteinen zu einer Verringerung der Proteolyseresistenz der Prionen (Heinz, Kortschack, 2002).

Bei **Enzymen** kann durch Druckbehandlung sowohl die Aktivität als auch die Substratspezifität beeinflusst werden. Auch eine partielle Inaktivierung ist möglich, Reaktivierung der Enzymaktivität, z.B. während der Lagerung, kann u.a. zur Bildung unerwünschter Stoffe führen. In einigen Fällen ist auch eine Aktivitätssteigerung von Enzymen unter Druck zu beobachten, was während der Druckaufbauphase zu Fehlparfömen führen könnte. Bildung toxischer Verbindungen aufgrund veränderter Substratspezifität unter Druck wurde bisher nicht beobachtet (Fernandez Garcia, 2002)

Einfluss der HDB auf die vegetativen Mikroorganismen

Da bei HDB um ein Konservierungsverfahren geht, wird in der ersten Linie über die Inaktivierung der Bakterien gesprochen. Grundsätzlich sind zwei antimikrobielle Wirkungen zu unterscheiden: Wachstumsverzögerung und Abtötung der Keime. Eine vollständige Abtötung aller vorhandenen vegetativen Keime gelingt oft nur bei sehr hohem Druck. Vegetative Zellen der Bakterien werden durch hydrostatischen Druck im Bereich von 150–800 MPa abgetötet. Mit dem steigenden Druck erhöht sich auch die Inaktivierungsrate. Elektronenmikroskopische Aufnahmen von druckbehandelten Bakterienzellen zeigten, dass es nur selten zu einer sichtbaren Zerstörung der Zellen kommt. Meist bleiben die Zellen in ihrer Struktur erhalten; weisen dann nur einige Veränderungen der Membranen auf. Direkt nach der Behandlung sind die Zellen nur eingeschränkt wachstumsfähig. Sie können in geeigneten Medien sich erholen und wieder vermehren. Wichtig ist, dass auf die Empfindlichkeit von Bakterien gegenüber dem Hochdruck eine ganze Reihe von Faktoren beeinflussen. Hierzu gibt es eine Vielzahl von Untersuchungen auch mit pathogenen Mikroorganismen. Das Überleben vegetativer Zellen während und nach einer HDB hängt stark von der Lebensmittelmatrix.

Neuere Literaturergebnisse deuten darauf hin, dass die Hochdruckbehandlung (300 MPa, 17 °C, 10 min) eine zusätzliche Hürde für die mikrobiologische Stabilität von schwachsaurer Rohwurst hin-

sichtlich der Salmonellen darstellt. Für die Inaktivierung von Listerien war Druck von 600 MPa für 10 Minuten notwendig.

Die folgenden mikrobiologischen Daten unserer Untersuchung beziehen sich auf die Entwicklung der Keimflora, die im Wesentlichen durch Rekontamination auf die thermisch behandelte Brühwurst (Mortadella) und damit in die Packung gelangt ist. Verglichen werden jeweils die nach unterschiedlichen Verfahren hochdruckbehandelte Ware und eine nicht hochdruckbehandelte Kontrollcharge. Erfasst wurden die aerobe, mesophile Gesamtkeimzahl, die Gruppe der Laktobazillen sowie die Vertreter der *Enterobacteriaceae*.

Die Hochdruckbehandlung wurde in einer Hochdruckanlage der Fa. EPSI (Belgien) bei Raumtemperatur (20°C) mit 600 MPa als Intervallbehandlung mit zweimal 4 Minuten Druckhaltezeit und 2 Minuten Pause durchgeführt. Unmittelbar nach der Hochdruckbehandlung und nach 7, 14, 21, 28, 35 und 38 Tagen bei 7 °C Kühlung wurden die Brühwurstproben mikrobiologisch untersucht.

Durch die Druckbehandlung wurde die Gesamtkeimzahl um ca. 1 Zehnerpotenz reduziert (Abb.1). Die Zahl der Milchsäurebakterien in behandelten Proben lag unter der Nachweisgrenze. Die *Enterobacteriaceae* waren in keinem Fall nachweisbar. Während der Lagerung stieg die Gesamtkeimzahl der Kontrollen bis zum 35. Lagerungstag bis auf 10^8 KBE/g an. Die Mikroflora der Kontrollen bestand überwiegend aus den Milchsäurebakterien. Während der gesamten Lage-

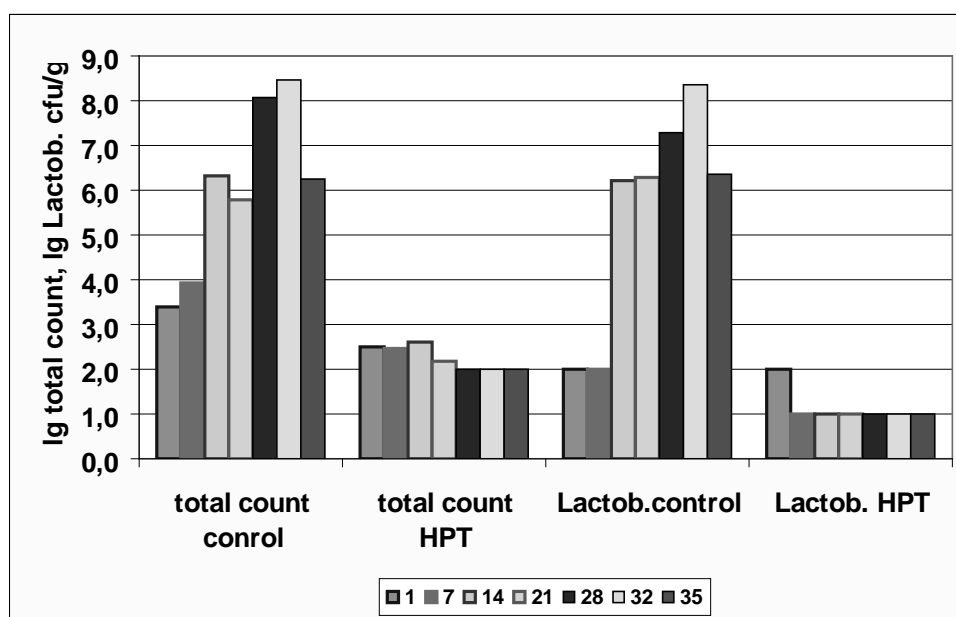


Abb. 1. Entwicklung der Keimzahlen bei HDB von Brühwurstaufschnitt

Figure 1. Praćenje broja mikroorganizama pomoću HPT kod narezanih kuvanih kobasica

zung der druckbehandelten Proben kam es nicht zum Anstieg der Gesamtkeimzahl sowie der Milchsäurebakterien. Bis zum 38. Lagerungstag blieben die Milchsäurebakterien unter der Nachweisgrenze. Die im Rahmen der Gesamtkeimzahl nachgewiesenen Bakterien der HDB-Proben waren ausschließlich Bazillen.

Ergebnisse der Inaktivierung der Sporen in Fleischerzeugnissen durch die HDB

Bei den Bakteriensporen verläuft das ganz anders. Der nicht einfacher Mechanismus der Inaktivierung beruht darauf, dass sich zwei wirkende Wege überlagern: die druckinduzierte Auskeimung und die subletale Schädigung der Sporen. Der Druck allein ist für die Inaktivierung der Sporen nicht ausreichend, dazu werden noch zusätzliche Faktoren benötigt. Eine kombinierte Anwendung der Hochdruck- und der Wärmebehandlung waren in unseren Versuchen für die vollständige Inaktivierung der Bakteriensporen notwendig. Dabei sollen zwei prinzipiell mögliche Vorgehensweisen geprüft werden. Die Druckbehandlung kann direkt im Aufschluss an die Pasteurisation erfolgen, in der vortemperierten Kammer. Zweite Möglichkeit ist die zeitversetzte Anwendung der Wärme- und Hochdruckbehandlung. Hierbei wird die Tatsache benutzt, dass die Sporen nach einer Hochdruckbehandlung – druckinduziert - auskeimen. Die ausgekeimten Sporen weisen dann eine ge-

ringere Resistenz auf als die Sporen. Die wichtigsten Parameter einer HDB sind die Höhe des Drucks, die Behandlungstemperatur, die Art der Druckbehandlung und die Druckhaltezeit. Diese Parameter wurden in den Inaktivierungsversuchen variiert.

Für die Inaktivierungsexperimente wurde Brühwurstbrät aus 64 % Rindfleisch, 18 % Sonnenblumenöl und 18 % Eis hergestellt. Als Zutaten wurden 16 g/kg Nitritpökelsalz, 3,0 g/kg Phosphat, 7 g/kg Gewürzmischung und 0,3 g/kg Ascorbat verarbeitet. Um eine ausreichende Sicherheit zu gewährleisten, erfolgte die Beimpfung des Brühwurstbrätes mit einem Pool aus aeroben und anaeroben, mesophilen und thermophilen Sporen (*Bacillus subtilis* ATCC 6633, *Clostridium sporogenes* PA 3679, *Bacillus stearothermophilus* DSM B171 und *Clostridium thermosaccharolyticum* DSM) mit jeweils 10^5 Sporen/g. Das beimpfte Brät wurde in 50 g Alu-Dosen abgefüllt, verschlossen und anschließend erhitzt und hochdruckbehandelt.

Die Ergebnisse der Untersuchungen zeigten, dass die Sporen von *Clostridium thermosaccharolyticum* DSM und *Bacillus stearothermophilus* DSM B171 nach der Pasteurisation und anschließender Hochdruckbehandlung mit 500 MPa und *Bacillus subtilis*-Sporen ab 600 MPa bei der Temperatur von 75 °C nicht mehr nachweisbar waren. *Clostridium sporogenes* erwies sich in den bisherigen Untersuchungen als der mit Abstand druckresistenteste Sporenbildner.

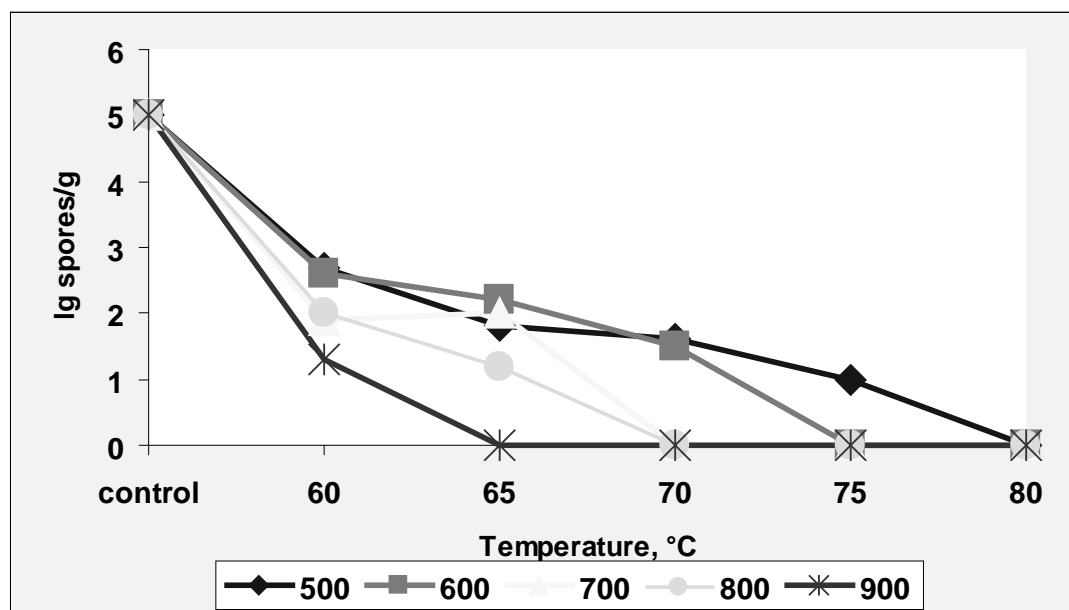


Abb. 2. Einfluss verschiedener Druckhöhen und Temperaturen auf die Inaktivierung der Sporen von *Clostridium sporogenes*
Slika 2. Uticaj različitih vrednosti pritiska i temperatura na inaktivaciju spora *Clostridium sporogenes*

Für die vollständige Inaktivierung der Clostridien-Sporen in den untersuchten Brühwurstbräten waren ein hoher Druck von 900 MPa bei einer niedrigeren Temperatur von 65 °C oder eine höhere Temperatur von 80 °C bei einem niedrigeren Druck von 600 MPa notwendig (Abb. 2). Jedoch war diese Behandlung für die Struktur des Produktes schädlich, deshalb wurde die zweite Möglichkeit – druckinduzierte Auskeimung der Sporen untersucht. Dafür wurde das mit dem obengenannten Pool der Sporenbildner (mit jeweils 10^5 Sporen/g) beimpfte Brühwurstbrät sofort nach dem Abfüllen in Dosen bei einem moderaten Druck von 300 MPa in zwei Zyklen von 2 Mal 4 Minuten im auf 50 °C vortemperierten HD-Behälter, hochdruckbehandelt.

Danach erfolgte die druckinduzierte Auskeimung der Sporen bei unterschiedlichen Bebrütungszeiten von 20 bis 100 Minuten mit dem 10-minütigen Abstand bei 37°C für die mesophilen und bei 60 °C

Wie die Ergebnisse der Bebrütung zeigten, waren während der Inkubationszeiten zwischen 20 und 100 Minuten ca. 10^2 als Sporen, mit zunehmender Tendenz zwischen 10^4 und nahe 10^5 als vegetative Keime nachweisbar. Nach der Bebrütung wurden die Proben bei 95°C unterschiedlich lang erhitzt. Nach 20-minütiger Erhitzung überlebten nur 10 vegetative *Clostridium sporogenes* in Proben, ab 30-minütiger Bebrütungszeit konnte in den erhitzten Proben kein Wachstum aller untersuchten Sporenbildner mehr festgestellt werden.

Durch eine zweistufige zeitversetzte druckinduzierte Hochdruckbehandlung gelang es mit einem moderaten Druck von 300 MPa die Sporen zum Auskeimen anzuregen. Bei geeigneter Bebrütungszeit ab 30 min verlieren die Sporen so viel von ihrer Hitzeresistenz, dass sie bei einer Kerntemperatur von 95°C nach 20 min vollständig inaktiviert werden konnten.



Abb. 3: Druckinduzierte Auskeimung mit nachfolgender Hitzeinaktivierung von *Clostridium sporogenes* bei 95°C und der Erhitzungsdauer von 20 Minuten

Figure 3. Germinacija spora klostridija inukovana pritiskom sa pratećom pasterizacijom na 95°C tokom 20 minuta

für die thermophilen Sporenbildner. Dabei sollte bei der Sporenauskeimung die Zeit festgestellt werden, in der die Sporen noch nicht vollständig ausgekeimt sind, aber ihre sporenspezifische Hitzeresistenz bereits verloren haben. Zu Beginn der Auskeimung der Sporen zur Entwicklung einer vegetativen Bakterienzelle wird die für die Hitzeresistenz verantwortliche Dipicolinsäure in der Zellwand der Spore abgebaut. Daher war es nicht notwendig so lange zu inkubieren bis alle Sporen vollständig ausgekeimt sind.

Die so hergestellten Brühwurstkonserven hatten nach sensorischen Bewertung Frischwarencharakter. Nach 24-monatiger Bebrütung der mit den 4 vorgenannten Sporenbildnern beimpften Brühwurstkonserven bei 37°C und 55°C gab es keine Bombagen. Bei mikrobiologischen Untersuchung konnten keine Bakterien oder Sporen nachgewiesen werden. Diese Ergebnisse sind ausschließlich repräsentativ für die untersuchte Rezeptur und die verwendeten Sporenbildner. Bei anderen Rezepturen

(z. B. pH-Wert, aW- Wert, Kochsalzgehalt, Pökelfstoffe) bzw. anderen Sporenbildnern sind abweichende Ergebnisse nicht auszuschließen.

Schlussfolgerungen

Hohe hydrostatische Drücke können in Abhängigkeit von den gewählten Temperatur/Zeit-Bedingungen zur teilweisen oder vollständigen In-

aktivierung von vegetativen Keimen sowie Sporen eingesetzt werden. HDB stellt eine zusätzliche Hürde hinsichtlich der mikrobiologischen Stabilität dar. Die sensorische Qualität der Fleischerzeugnisse kann durch diese neue Technologie nur bedingt erhalten werden.

Als alternative oder ergänzende Maßnahme zur schonenden Haltbarmachung mikrobiologisch kritischer Fleischerzeugnisse könnte die HDB deshalb vom großen technologischen Interesse sein.

Literatur

Pfister, M. K.-H., Butz, P., Heinz, V., Dehne, L. I., Knorr, D., Tauscher, B. 2000. Der Einfluss der Hochdruckbehandlung auf chemische Veränderungen in Lebensmitteln. - Bundesinstitut für gesundheitlichen Verbraucherschutz und Veterinärmedizin. Berlin (BgVV-Hefte), Nr. 3;
Serfert, Y. 2002. Diplomarbeit, FH Bernburg;

Heinz, V., Kortschack, F. 2002. Method for modifying the protein structure of PrP(sc) in a targeted manner, Patent WO 02/49460;

Fernandez Garcia, A., Butz, P., Tauscher, B. 2002. Mechanism-based irreversible inactivation of horseradish peroxidase at 500 MPa. Biotechnol. Prog. 18, 1076-1081.

Paper recieved: 15.04.2009.

FAST DRYING OF DRY-CURED MEAT PRODUCTS: QUICK-DRY-SLICE (QDS) PROCESS TECHNOLOGY*

Comaposada J., Arnau J., Garriga Margarita, Ferrini G., Xargayó Marta, Bernardo J., Corominas Montserrat, Gou P., Freixanet, L., Lagares J., Monfort J. M.

Abstract: The traditional process for dry-cured meat products is time consuming. A Quick-Dry-Slice process based on a continuous system that combines both convective and vacuum drying could accelerate the drying of slices after the desired pH is reached in fermented sausages.

Key words: sausage, rapid, dehydration, fermentation, vacuum

Brzo sušenje suvih i salamurenih proizvoda od mesa: tehnologija brzog sušenja odrezaka

Sadržaj: Tradicionalni proces sušenja i salamurenja proizvoda od mesa zahteva mnogo vremena. Proces brzog sušenja odrezaka zasnovan je na kontinualnom sistemu koji kombinuje sušenje konvekcijom i vakuum sušenje, a može da ubrza sušenje odrezaka nakon postizanja željenog pH u fermentisanim kobasicama.

Ključne reči: kobasica, brzo, dehidracija, fermentacija, vakuum

Introduction

In the manufacture of dry-cured meat products by traditional methods, the drying stage is the most time consuming. In traditionally used drying methods this stage takes 1-2 weeks for small caliber fermented sausages, three to six weeks in the case of fermented sausages with higher diameter, and 1.5-3 years in Iberian dry-cured hams. During the fermentation process the pH drops, the pieces of meat bind and so facilitate the slicing process. During the drying phase, the product undergoes a dehydration process that is accompanied by a series of biochemical reactions produced by endogenous and microbial enzymes, which break down part of the lipids and proteins which gives the product its characteristic texture and flavor. In conventional dryers, dry air is injected by nozzles located in a series of perimeter conduits and the moist air is returned through a series of centrally mounted conduits located on the ceiling of the drying chamber. The design of these dryers causes the air passing over the meat products located next the nozzle exits to have different properties than the air passing over the products in other parts of the dryer.

The drying process is affected by the resistance of the meat to the flow of water and the distance that the water must travel until it reaches the surface of the product in order to be extracted (Crank, 1975). The objective of this study is the evaluation of a drying process for slices of meat products after fermentation, where the drying process consists of a convection phase followed by a vacuum drying phase (Quick-Dry-Slice process).

Drying technology based on the “Quick-Dry-Slice process”

Quick-Dry-Slice (QDS) drying technology is based on the drying and maturing method for sliced products proposed by Comaposada *et al.*, 2004. In this technology, the sausages are fermented until they attain the desired pH, then frozen, sliced and dried following a convective drying stage and a subsequent vacuum drying stage. With this drying process it is possible to obtain the desired water content and texture in only 30 minutes (Figure 1).

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mauntain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTHORS: Josep Comaposada, josep.comaposada@irta.es, Jacint Arnau, Margarita Garriga, Gabriele Ferrini, Pere Gou, Josep M^o Monfort. IRTA, Finca Camps i Armet, E-17121 Monells, Girona, Spain; Marta Xargayó, Llorenç Freixanet, Josep Lagares. METALQUIMIA S.A., St. Ponç de la Barca, s/n, 17003 Girona, Spain; Jordi Bernardo, Montserrat Corominas. CASADEMONT S.A., Paratge Constantins, s/n, 17164 Bonmatí, Girona, Spain.

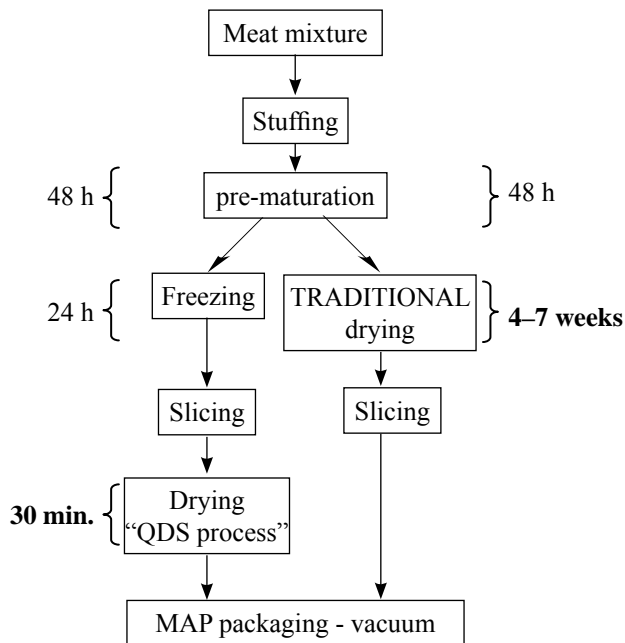


Figure 1. Time comparison between the traditional drying process and the process using QDS technology for drying dry-cured meat products with a diameter of approximately 80 mm.

Slika 1. Poređenje između tradicionalnog postupka sušenja i QDS tehnologije kod sušenja salamurenih proizvoda od mesa prečnika 80 mm

The QDS system

The QDS system developed by Metalquimia S.A. (Figure 2) was designed according to a continuous production system. There is a charging zone for frozen slices, a tempering and pre-drying zone with air circulation, and a vacuum drying zone in which the required moisture is extracted from the slices. Finally, and depending on the exit temperature of the slices, the product is tempered again prior to being packaged in order to prevent condensation or adherence of fat to the packaging. The slices are placed on a stainless steel belt designed to facilitate the extraction of moisture from the slices, both during drying by convection and during the vacuum drying phase. The air used for drying and tempering during the forced convection stage is purified by means of a high efficiency particulate air (HEPA) filter in order to minimize contamination of the air coming into contact with the product. In addition, the speed of the tempering and drying processes can be adjusted by controlling the temperature, relative humidity and velocity of the air passing over the product. The vacuum drying stage is controlled mainly via the operating pressure and the heating temperature. The different stages of the processes are linked with one another by means of conveyor

belts and slice loading/unloading mechanisms. The complete process is controlled by a PLC which additionally enables monitoring and recording of the control parameters.

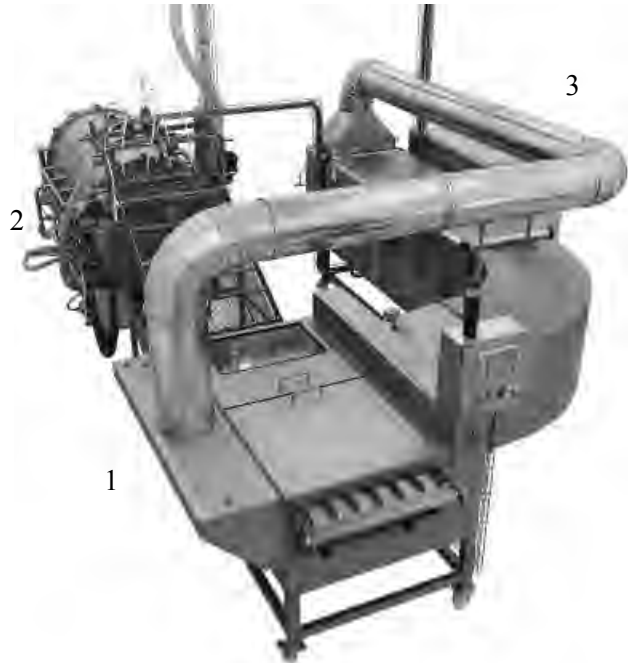


Figure 2. The QDS system: 1. Tempering and pre-drying section; 2. Vacuum drying section; 3. Air purification circuit with HEPA filter

Slika 2. QDS sistem: 1. temperiranje i faza predušenja; 2. faza vakuum sušenja; 3. sklop za prečišćavanje vazduha sa HEPA filterom

Microbiological and sensory evaluation

A number of studies were carried out to compare the safety and the sensory properties of the dry-cured meat products produced by the traditional method with those produced by the QDS process. In these studies the microbiological quality of „salchichón” sausages was evaluated after determining the following parameters: bacterial counts of *Staphylococcus aureus*, sulfite-reducing clostridia, *Escherichia coli* and *Listeria monocytogenes*. In addition, the presence / absence of *Salmonella* was also investigated in 25 g samples. The study also included the pH measurement of the products at different sampling times, as well as the water activity of the final product.

The pH of the fermented sausages dried by the traditional method was lower than that of those dried by means of the QDS process (Table 1). Moreover, an additional reduction of pH was observed in fermented sausages stored at 13°C, an effect that was not found at 1°C.

Table 1. Average losses obtained for the different drying processes and pH of fermented sausages at different sampling times**Tabela 1.** Prosečan kalo dobijen za različite procese sušenja i pH za fermentisane kobasice pri različitim vremenima uzorkovanja

Drying process	Batch	Diam.	Drying time / days	Drying losses %	a _w End of drying	pH			
						Before drying	End of drying	3 month storage	
								1°C	13°C
Traditional	1	80	38	28.6	0.907	5.32	4.89	5.00	4.70
	2	80	38	26.5	0.917	5.21	4.85	4.99	4.63
QDS	1	80	<1	30.7	0.902	5.32	5.25	5.14	4.99
	2	80	<1	32.8	0.887	5.21	5.15	5.20	5.15

The results of the microbiological analyses and the *Staphylococcus aureus*, sulfite-reducing clostridia and *Escherichia coli* counts for each sampling time are shown in Tables 2, 3 and 4, respectively. The results show that both drying processes (tra-

ditional and QDS), as well as the subsequent storage of the vacuum-packed slices of „salchichón“ sausage, achieve similar results in terms of reducing the number of microorganisms below the detection limit.

Table 2. *Staphylococcus aureus* (log cfu/g) counts in fermented sausages depending on the drying process**Tabela 2.** Broj *Staphylococcus aureus* (log cfu/g) u fermentisanim kobasicama u zavisnosti od procesa sušenja

Drying process	Batch	Before drying	End of drying	Storage		
				15 days 4°C	3 months 1°C	3 months 13°C
Traditional	1	1.94	<1.00	<1.00	1.10	<1.00
	2	2.26	1.03	<1.00	<1.00	<1.00
QDS	1	1.94	1.77	<1.00	<1.00	<1.00
	2	2.26	1.91	1.27	<1.00	1.10

Table 3. Sulfite-reducing clostridia (log cfu/g) counts in fermented sausages depending on the drying process.**Tabela 3.** Broj sulfitoredukujućih klostridija (log cfu/g) u fermentisanim kobasicama u zavisnosti od procesa sušenja

Drying process	Batch	Before drying	End of drying	Storage		
				15 days 4°C	3 months 1°C	3 months 13°C
Traditional	1	1.22	<1.00	<1.00	<1.00	<1.00
	2	1.46	<1.00	<1.00	<1.00	<1.00
QDS	1	1.22	<1.00	<1.00	<1.00	<1.00
	2	1.46	1.09	<1.00	<1.00	<1.00

Table 4. *Escherichia coli* (log cfu/g) counts in fermented sausages depending on the drying process**Tabela 4.** Broj *Escherichia coli* (log cfu/g) u fermentisanim kobasicama u zavisnosti od procesa sušenja

Drying process	Batch	Before drying	End of drying	Storage		
				15 days 4°C	3 months 1°C	3 months 13°C
Traditional	1	3.45	1.76	1.43	<1.00	<1.00
	2	3.45	1.86	1.62	<1.00	<1.00
QDS	1	3.45	2.97	1.47	<1.00	<1.00
	2	3.45	2.89	1.22	<1.00	<1.00

As to the prevalence of *Salmonella* in the fermented sausages, it was observed that in the case of a raw material contaminated with this pathogen (presence in 25 g) prior to drying, the presence of *Salmonella* could still be detected in the 25 g sample of the final product, regardless of the process followed (traditional or QDS). The studies carried out by Smith *et al.*, (1975a, 1975b) report the incidence of dry-cured meat products showing the presence of *Salmonella* in those cases where the traditional drying method was used. The study concludes that in the cases where the pathogen is present after the pre-maturation stage, it is difficult to guarantee its absence in the final product by the reduction of the water activity which takes place during the drying process. In view of these problems and in compliance with the Commission Regulation (EC) No. 2073/2005 on microbiological criteria for foodstuffs, which requires the absence of *Salmonella* in a 25 g sample for these types of products, the QDS process facilitates the integration of elements that inactivate this microorganism and could therefore improve the safety of the dry-cured meat product. To evaluate this possibility, a very low dose (<3 NMP/g) of *Salmonella* was inoculated and 2 g/kg of sodium acetate were added to the mixture to be processed by the QDS method. The QDS process showed better results (greater number of 25 g samples showing an absence of the pathogen) than the conventional process (Garriga *et al.*, unpublished results). These preliminary results will be validated in future investigations.

The *Listeria monocytogenes* counts, carried out in all the fermented sausages analyzed, were all below the detection limit (<20 cfu/g) for all the sampling times (end of pre-maturation, end of dry-

ing and storage). It can hence be concluded that, starting from raw materials having low counts of the pathogen in question, it is possible to produce safe dry-cured meat products in compliance with the Commission Regulation (EC) No. 2073/2005 on microbiological criteria for foodstuffs, which limits the *L. monocytogenes* counts to <100 cfu/g for this type of products.

In order to further investigate the effects of QDS drying on raw materials contaminated with *L. monocytogenes*, an experiment was conducted in which the pathogen was inoculated under controlled conditions. In this experiment, the initial meat mass was first inoculated with a mixture of 5 different cultures of the pathogen, with counts in the order of 3×10^3 cfu/g. It was then subjected to the fermentation and maturation/drying processes by both the traditional and the QDS methods. In both cases, similar reductions in the pathogen counts were achieved, which shows that the QDS process is an efficient process with respect to food safety.

It is important to emphasize that there are complementary technologies, such as those based on high pressures, which have provided satisfactory results in minimizing the risk when applied to sliced products (Garriga *et al.*, 2003). High pressure affects appearance texture and flavour (Fulladosa *et al.*, 2009). However, the effect is small at the water content at which fermented sausages are commercialized. Color parameter L* increases when the water content increases, while a* parameter decreases (Comaposada *et al.*, 2009). The effect of high pressure on colour parameters was more important at a higher water content and hardly apparent at a lower water content (Fig. 3).

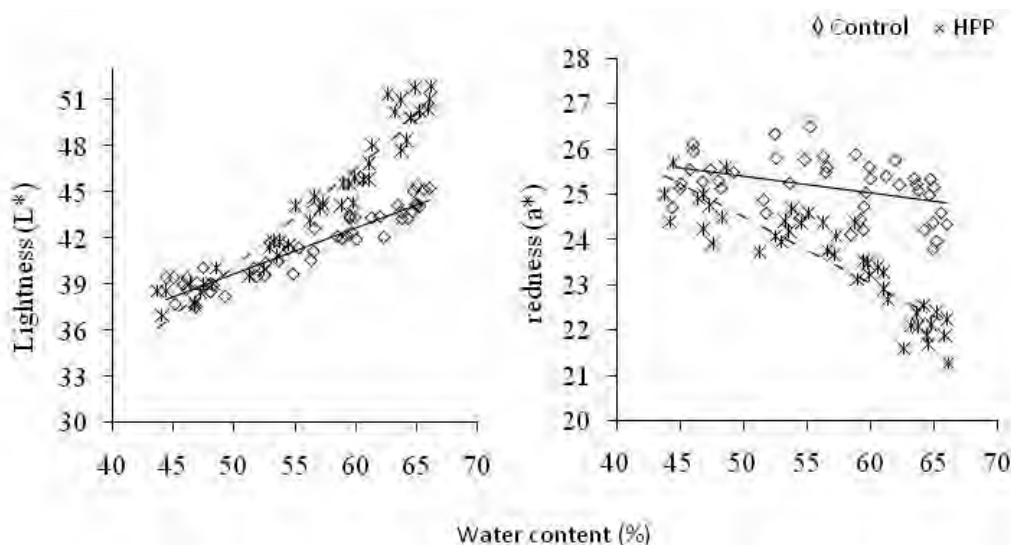


Figure 3. L* and a* color parameter depending on water content in dry-cured meat product with high pressure processing (7 min. at 500 MPa) and without (control)

Slika 3. L* (svetlina) i a* (stepen intenziteta crvene boje) u zavisnosti od sadržaja vode u suvim i salamurenim proizvodima od mesa proizvedenih pod visokim pritiskom (7 min. na 500 MPa) i bez visokog pritiska (kontrola)

With regard to the sensory evaluation, the slices of products made by the QDS process presented a less acidic aroma and taste than those made in the traditional manner (Table 5). This could be attributed to the lack of acidification during drying and the absence of an acidity gradient between the external and the internal parts of the slice. In addition, the volatile acids may have been partially eliminated during the drying stage. For this reason, in the QDS process, the pH may decrease to values below those of the conventional process during pre-maturation. The colour was also found to be more intense in the case of the QDS process because the intensity of the coloring agent Ponceau 4R was not reduced during the process. The flavor of the product produced by the traditional method was more balanced and it was therefore necessary to modify the initial mixture of spices and flavoring agents in the case of the QDS process in order to obtain equivalent products in both cases. Similarly, slight differences in appearance were found depending on the product. Figure 4 shows various products obtained by both methods after vacuum-packing.

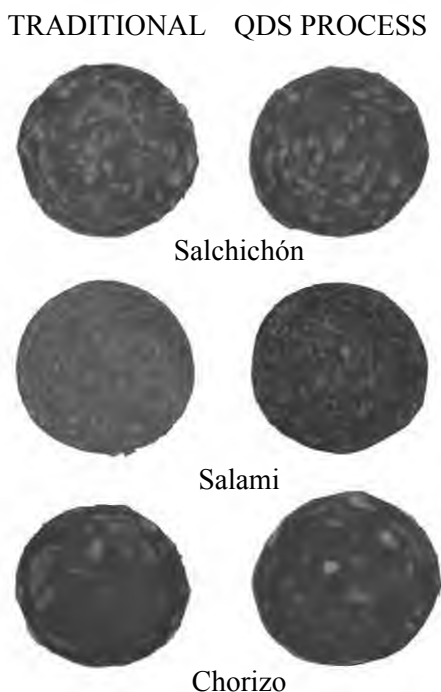


Figure 4. Visual comparison between “salchichón”, “salami” and “chorizo” sausages obtained by means of the traditional method and the QDS method after 7 days of storage in vacuum packs

Slika 4. Vizuelno poređenje između “salchichón”, “salami” i “chorizo” kobasica tradicionalnom metodom i QDS metodom nakon sedam dana skladištenja u vakuum pakovanju

Table 5. Evaluation of sensory parameters of “salchichón” sausage produced by the traditional method and by the QDS method

Tabela 5. Ocena senzornih parametara “salchichón” kobasice proizvedene tradicionalnim metodom i QDS metodom

	Drying process	
	Traditional	QDS
Roughness	0.21a	2.33b
Color	5.96a	6.67b
Flavor cured	6.00a	4.50b
Acidity	5.00a	1.00b

Advantages of the QDS method and technological challenges

For the commercialization of dry-cured meat products in slices, QDS technology offers numerous advantages relative to conventional drying methods. There are advantages of a technological nature and others related to the operation and management of the production process. Among the technological advantages of the QDS process it is worth mentioning the ability to obtain more homogeneous products showing a less acidic flavour. Furthermore, the products are free from fungi and product safety control is enhanced thanks to a more precise monitoring of the process and of the product itself. In addition, the application of the QDS process results in increased productivity and decreased residues.

With regard to the production process, the QDS method offers enhanced production flexibility, an increase in speed, the possibility to implement just-in-time systems as well as requiring less space than conventional methods.

The QDS process may contribute to the development of new formats and products in line with the trends and lifestyles of today’s consumers, who demand ready-to-use products in a small format. It is also important to develop products aimed at especially sensitive consumer groups (people with high blood pressure, elderly people, immune-depressed patients, diabetics, obese people, etc.), as well as other types of products that will help to achieve the objectives set forth by the NAOS strategy, which was agreed between the Public Administration (represented by the Ministry of Health and Consumer Affairs, Ministry of Industry, Ministry of Education and Science), the Spanish Food Safety Authority (AESAs), the Spanish Food and Drinks Federation, (FIAB) together with large food producers and the majority of the Health Departments of the

Autonomous Communities of Spain, as an attempt to communicate the need to reduce the daily intake of fats and salt, among other things.

Acknowledgements

This work was supported by the CDTI (Cenit Futural project) and the integrated projects within

the Sixth RTD Framework programme: Truefood (Traditional United Europe Food- Integrated Project FOOD-CT-2006-016264) and Q-Porkchains (Integrated Project FOOD-CT-2007-036245). The views expressed in this publication are the sole responsibility of the authors and do not necessarily reflect the views of the European Commission.

References

- Comaposada, J., Arnau, J., Gou, P., Monfort, J. M., 2004.** Accelerated method for drying and maturing sliced food products, Patent WO/2005/092109;
- Comaposada, J., Ferrini, G., Arnau, J., Gou, P., 2009.** The effect of high pressure process on colour of dry-cured meat at different water content, The SAFE consortium 2nd International Congress on Food Safety, Girona, Spain;
- Crank, J., 1975.** The mathematics of diffusion, Oxford University Press, London;
- Fulladosa Elena, Serra, X., Gou, P., Arnau, J., 2009.** Effects of potassium lactate and high pressure on transglutaminase restructured dry-cured hams with reduced salt content, Meat Science, Volume 82, Issue 2 213-218;
- Garriga Margarita, Marcos Begoña, Aymerich Teresa, Hugas Marta, 2003.** Prospectiva de aplicación de altas presiones para la minimización de riesgos asociados a Salmonella y Listeria monocytogenes en embutidos madurados en frío, Eurocarne, 121, 93-99;
- Smith, J. L., Huhtanen, C. N., Kissinger, J. C., Palumbo, S. A., 1975a.** Survival of salmonellae during pepperoni manufacture, Appl. Environ. Microbiol. 30(5), 759-763;
- Smith, J. L., Palumbo, S. A., Kissinger, J. C., Huhtanen C. N., 1975 b.** Survival of Salmonellae dublin and Salmonella typhimurium in lebanon bologna, J. Milk Food Technol. 38(3), 150-154

Paper received: 10.04.2009.

THE MICROBIOLOGICAL ECOSYSTEM OF TRADITIONAL FERMENTED SAUSAGES IN SERBIA – POSSIBILITY TO CREATE OUR OWN STARTER CULTURES*

Vesković-Moračanin Slavica, Obradović D.

A b s t r a c t: Today in Serbia, according to the existing world trends, a growing number of meat industries are implementing in the production active starter cultures. Bearing in mind that in Serbia there is no commercial production of such cultures, the domestic industry is obliged to purchase such cultures from foreign manufacturers. Such cultures, are as a rule adapted for the needs of other markets and usually do not result in products which have traditional sensory characteristics which are acceptable to our customers.

The Project "Technological and protective features of autochthonous bacterial strains isolated from traditional fermented sausages and possibilities for their application in the meat industry" is financed by the Serbian Ministry of Science and Technology and has the aim to study the diversity of a number of bacteria such as: *Lactobacillus*, *Micrococcus*, *Staphylococcus* and *Streptococcus* which carry out the fermentation in narrow diameter sausages "Levačka", "Sremska" and "Užicka" from three different regions in Serbia, as well as to determine the possibility of their use within industrial conditions of production. Adequate selection and choice of bacterial strains, after their detailed morphological, biochemical, molecular and genetical, as well as potential technological, protective and probiotic features characterization would make the presumptions needed for a qualitative step forward in the production of our own starter cultures. With their regular use specific national products with distinctive sensory features to which our population is accustomed and with improved quality parameters would be obtained.

By realizing the scope the rationale for the use of autochthonous strains of LAB in the production of fermented sausages their authenticity will be preserved, uniform quality can be obtained, production mistakes avoided, the fermentation and maturation time shortened and at the same time the typical sensory features preserved and/or improved.

Key words: traditional fermented sausages, isolates, LAB, starter cultures, meat industry

Mikrobiološki ekosistem tradicionalnih fermentisanih kobasica u Srbiji – mogućnosti stvaranja sopstvenih starter kultura*

S a d r ž a j: Danas u Srbiji, a u skladu sa savremenim svetskim trendom, sve veći broj industrija mesa u svojoj proizvodnji primenjuje aktivne starter kulture. Obzirom da kod nas ne postoji njihova komercijalna proizvodnja, domaće industrije mesa su primorane da ih nabavljaju od stranih proizvođača. Takve starter kulture, po pravilu, prilagođene su potrebama drugih tržišta pa najčešće ne daju proizvode sa tradicionalnim senzorskim svojstvima koja su najprihvatljivija za naše potrošače.

Projekat "Tehnološke i protektivne osobine autohtonih sojeva bakterija mlečne kiseline izolovanih iz tradicionalnih fermentisanih kobasica i mogućnosti njihove primene u industriji mesa", koji finansira Ministarstva za nauku i tehnološki razvoj Republike Srbije, ima za cilj sagledavanje diverziteta različitih bakterijskih vrsta iz roda *Lactobacillus*, *Micrococcus*, *Staphylococcus* i *Streptococcus*, koji su nosioci fermentacije u kobasicama uskog dijametra („levačka“, „sremska“ i „užicka“) sa tri područja Srbije, kao i utvrđivanje mogućnosti njihove primene u industrijskim uslovima. Adekvatnom selekcijom i odabirom određenih sojeva bakterija, nakon njihovih detaljnih morfoloških, biohemijskih, molekularno-genetskih ispitivanja, kao i utvrđivanja potencijalno tehnoloških, protetektivnih i probiotskih svojstava, stvorile bi se pretpostavke za drugi, kvalitetan iskorak u pravcu sopstvene proizvodnje starter kultura. Njihovom primenom dobili bi se specifični nacionalni proizvodi sa karakterističnim i prepoznatljivim senzorskim svojstvima na koje je naše stanovništvo naviklo, sa, istovremeno, unapređenim parametrima kvaliteta.

Realizacijom postavljenih zadataka dokazaće se svrsishodnost korišćenja autohtonih sojeva BMK u proizvodnji fermentisanih kobasica, sačuvaće se autentičnost proizvoda, dobiće se ujednačen kvalitet, izbeći će se manje proizvodne greške, skraćuje se proces zrenja i sušenja, a pri tome će biti očuvana i/ili unapređena karakteristična senzorska svojstva proizvoda.

Cljučne reči: tradicionalne fermentisane kobasice, izolati, BMK, starter kulture, industrija mesa

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

Introduction

The increasing manufacture of fermented products, after the Second World War, has conditioned the need for standardized and economical production on one side, and a safe product on the other. Nowadays, in order to fulfill these requirements the modern industry uses specially selected and chosen microorganisms, the so called starter cultures (Caplice, Fitzgerald, 1999). As during the traditional production of fermented meat products, the lactic fermentation is a spontaneous process, often uncontrolled and based on the activity of the "wild" epiphytic microflora, the quality of the products present on the market is variable and often lacking the specific sensory characteristics. The direction taken by these processes is guided by the accidentally present microflora which can give to the fermentation in an unwanted direction resulting in spoilage. The manufacture of good products with a standard quality is possible only if in the meat are the dominant useful homofermentative strains of LAB are present. If not, mistakes are not rare (Coretti, 1971, 1975).

The production of a safe product with standard uniform quality characteristics is an imperative for every serious producer. By respecting this principle on one side continuous production can be obtained and on the other customer confidence can be achieved. Starter cultures, which today are used in the meat industry, have the purpose not only to achieve desirable sensory characteristics, but by ensuring optimal microbiological processes to ensure a safe production.

The idea to inoculate *Lactobacillus* cultures for the production of fermented sausages was presented for the first time in 1940 by Jensen and Paddock (US Patent 2, 225, 783) as a way to shorten the maturation time and to obtain the desired quality and aroma. The first used starter cultures in the USA meat industry in 1955 were LAB, such as *Pediococcus cerevisiae* (Erkkila S., 2001). At the same time in Europe Niinivaara is (1955) used the *Micrococcus* M53 M53 (Slavica Vesković, 2009).

Lactic Acid Bacteria (LAB) as the carriers of lactic fermentation

The ability of LAB to ferment sugars down to lactic acid is the main principle which determines their use in the production of fermented meat products (Figure 1). Thus, the stability to ferment carbohydrates, which occurs at the level of phosphorylated substrates, is the key feature of LAB (Slavica Vesković Moračanin, 2007). They produce lactic acid

as the result of glucose breakdown during glycolysis or 6-phosphogluconate/ phosphoketolase reaction, depending if homo or heterofermentative bacteria are employed (Kandler, 1983; Axelsson, 1998). Homofermentative LAB genus *Lactococcus*, *Pediococcus*, *Streptococcus* and certain strains of *Lactobacilli* convert during anaerobic glycolysis 1 mol of glucose into 2 moles of lactate, while the heterofermentative group of LAB (*Leuconostoc* and some *Lactobacillus*) during anaerobic glucose catabolism produce lactic acid, carbon dioxide, ethanol and half the energy (Ros et al., 2002; Caplice & Fitzgerald, 1999).

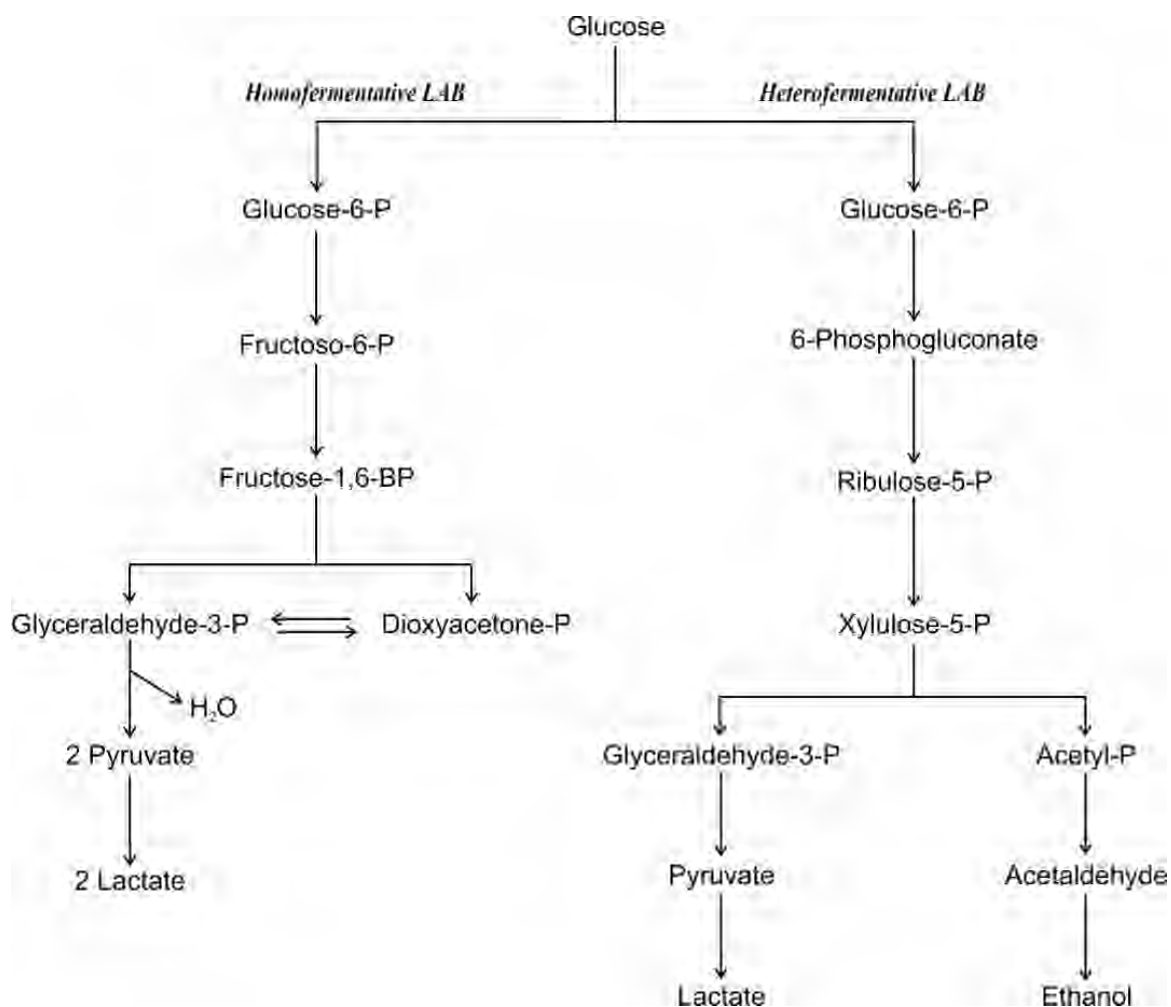
LAB besides having effects on the acidity by producing absolute and relative quantities of lactic and acetic acid, influence the taste of the final product by producing some substances which are under the sensitivity detection limit. By lowering the pH of the sausage filling during the fermentation process enzymes which regulate lipolysis (Garcia et al., 1992; Molly et al., 1996) and proteolysis (Demeyer, 1992) become activated.

Microorganisms, specially the catalase positive cocci, influence the aroma and taste of fermented sausages through direct breakdown of lipids and proteins into compounds which contribute to the desired sausage aroma. At the same time their nitrate – reducing activity results into the formation of a stable color (Lücke, 2000). The most important quality parameters affected by the starter cultures are shown in Table 1.

Nowadays a few important companies which produce starter cultures (needed for the meat industry) provide pure *Lactobacillus* spp., *Pediococcus acidilactici*, *P. pentosaceus*, *Staphylococcus xylosum* or *S. carnosus* (Daly & Davis, 1998, Hammes et al., 1985) cultures.

LAB in biological food protection

The growing need for natural and safe food have lead to an increased interest for the use of bacteriocin-producing LAB bacteria which are used as protective cultures in the meat industry for the making of fermented products. The principle on which biological protection of these cultures (Lindgren & Dobrogosr, 1990) is achieved is based on lowering the number of unwanted spoilage bacteria, but without influencing the quality of the final product. The biological protection of LAB through the presence of bacteria and/or their metabolic products are achieved by: production of lactic acid or other volatile organic



Šema 1. Metabolizam glukoza kod homo- i heterofermentativnih BMK

Table 1. Effect of starter cultures on raw sausages
Tabela 1. Efekat starter kultura u sirovim kobasicama

Quality parameters	Mode of action	LAB	Catalase positive cocci
Color	- reduction of nitrates - lowering of pH - decreased O ₂ content in the sausages (Eh) - H ₂ O ₂ degradation	- +++ - -	+++ - ++ ++
Aroma	- acid production - proteolysis - lipless - rancidity (antioxidative)	+++ - - -	- + ++ ++
Consistency	- lowering of pH	+++	-
Shelf time	- lowering of pH - reduction of nitrates - suppression of unwanted microflora	+++ - ++	- ++ -
Low content of residues	- reduction of nitrates	+	++

+++ very important role
 ++ important role
 + no importance

acids resulting in lowered pH; production of other primary metabolites such as hydrogen peroxide, carbon dioxide, diacetyl, reuterin and bacteriocine production which is a specific antibacterial compounds (*De Vuyst and Vandamme, 1994*) (Table 2).

tainty of the growth rate and metabolic intensities of the protective and unwanted bacteria.

However, even if many producers of starter cultures suggest that they can solve or even eliminate problems relative to hygienic standards of the basic components of meat products we have to be very cautious not to expect an immaculate product if we

Table 2. Metabolic products of LAB and their antimicrobial effect

Tabela 2. Metabolički produkti BMK sa antimikrobnim efektom

LAB products	Target microorganisms
Organic acids	
Lactic acid	Rotting and GR-ve bacteria, some fungi
Acetic acid	Rotting bacteria, clostridia, some yeasts and molds
Hydrogen peroxide (H₂O₂)	Pathogenic bacteria and bacterial contamination especially in high protein food
Enzymes	
Lactoperoxidase system with H ₂ O ₂	Pathogenic bacteria and bacterial contamination (milk and milk products)
Lysosimes (tech recombinant RNA DNK)	Unwanted GR +ve bacteria
Low molecular weight metabolites	
Reuterin (3-OH-propionaldehyde)	Wide spectrum bacteria, moulds and yeasts
Diacetyl	GR -ve bacteria
Fatty acids	Various bacteria
Bacteriocines	
Nisine	Some LA and GR+ve bacteria, specially those which are spore producing
Other bacteriocines	GR+ve bacteria, inhibitory spectrum

In order for the use of starter cultures to be justified it is needed that the used cultures fulfill the following conditions (*Holzappel et alr., 1995, Slavica Vesković, 2005, 2007, 2009*): *they should not be harmful for the consumer* (i.e. do not produce toxins, biogenic amines or other metabolites which can harm human's health and are not part of pathogenic bacteria); *they must contribute towards desired effects in the product* (they have to be adapted for the product, must have a consistent protective effect, reliable metabolic activity and must suppress any undesirable microflora) and *must not have negative effects upon good manufacturing practice* (do not produce unwanted acids, gas, slime...).

At the same time from the ideal bacterial culture it is expected to be able to produce bacteriocines during the fermentation process. The formed bacteriocines must be stable in the meat matrix and inactivation by ingredients from the stuffing should not be allowed. The starter culture has to be preserved relative to other bacteriocine producing bacteria (*Slavica Vesković, 2005, 2009*).

The use of protective cultures bears some unknown elements, relative especially to the uncer-

have second rate raw materials. Bearing all of this in mind, the use of protective cultures has to be seen in the light of only an added security measure in the production processes in the meat industry.

Microflora of traditional fermented sausages

Following the modern trends a growing number of meat producers in Serbia are using active growing cultures. As in Serbia there is no commercial production of starter cultures, the meat industries are compelled to purchase from foreign producers. Such starter cultures are often adapted to the needs of other markets and do not result in products which have such quality to which our customers are used.

The use of LAB as starter cultures isolated from autochthonous fermented meat products would be the solution to this problem. Strains isolated from autochthonous fermented products would be the basis for a potential production of domestic starter cultures. This could be achieved independently or joined to existing established international companies.

The need of every modern state is to study the natural resources and have an overview on the possessed potentials. In Serbia, up to now there were no extensive studies on the diversity and characteristics of autochthonous microflora of traditionally fermented sausages. This resulted in insufficient data and knowledge needed for a direct application in the meat industry.

The justification of the project "Technological and protective features of autochthonous bacterial strains isolated from traditional fermented sausages and possibilities for their application in the meat industry", financed by the Serbian Ministry of Science and Technology, is based on the need to form a data bank of autochthonous LAB strains. Such a data bank would make the foundation for the production of starter cultures in Serbia.

In order to reach this goal in this research we have studied three different traditionally fermented sausages ("Uzicka", "Levacka" and "Sremska").

In the meat industry AD "Juhor" - Jagodina out of the raw material obtained by slaughtering the animals raised in the Levca region the "Levacka" and "Sremska" sausage were produced. The sausages were produced according to the standard (producer's

specification) which is used in the regular production by AD "Juhor". Fermentation and smoking were based on traditional principles.

The "Sremska" sausage was made of pork meat, bacon and spices, while "Levacka" was made of equal quantities of pork and beef and firm fat tissue. The prepared stuffing was filled into pork's small intestine. The production process lasted for 21 days.

The traditional "Uzicka" sausage was manufactured in the household of Nikola Brkovic, on the Zlatibor slopes in the village of Kacer. "Uzicka" was made of beef and pork meat, minced beef, firm fat tissue, nitrites, salt and S/ (Alimenta) in a three month period (November 2008 – January 2009). The sausage filling was stuffed into beef's small intestines and the fermentation process lasted for 21 days. The characteristics, ingredients and procedures studied within the Project are shown in Table 3.

Besides the basic studies which encompassed procedures for the isolation and characterization of LAB at different stages of maturation extensive work on physical, chemical, microbiological and sensory characteristics of raw materials, spices salt and additives used in sausage production have been carried out. By doing so, in an indirect way

Table 3. Characteristics, ingredients and procedures for traditional fermented sausages
Tabela 3. Karakteristike, sastojci i procedura zrenja njih ispitivtradicionalnih fermentisanih kobasica

Type of fermented sausage	Dimensions and weight	Wrapping	Ingredients	Quantity (100 kg)	Duration of maturation
„Sremska“ sausage	34-36 mm ø 24 cm length 300 g weight	<i>natural</i> (pork's small intestine)	Pork meat Pork shoulder Firm fat tissue Nitrite salt 1% Sugar – saccharose Ground hot and sweet pepper Black popper extract Garlic extract	45 kg 25 kg 30 kg 2.47 kg 330 g 140 g 70 g 35 g	Smoking - 3 days at 20°C, 66% RVV. Maturation - 21 days on 12 to 29°C, 58% - 80% RVV
„Levacka“ sausage	34-36 mm ø 19 cm length 250 g weight	<i>natural</i> (pork's small intestine)	Pork meat Beef meat Firm fat tissue Nitrite salt 1% Sugar – saccharose Ground hot and sweet pepper Black popper extract Garlic extract	47 kg 20 kg 33 kg 2.5 kg 330 g 140 g 70 g 35 g	Smoking - 3 days at 20°C, 66% RVV. Maturation - 21 days on 12 to 29°C, 58% - 80% RVV
„Uzička“ sausage	40 mm ø 41 cm length 700 g weight	<i>natural</i> (beef small intestine)	Pork meat Beef meat Firm fat tissue Nitrite salt Sodium chloride Spice S77 Alimenta	70 kg 20 kg 10 kg 2.5 kg 300 g 850 g	Maturation - 21 days at 2 - 13°C, 64% - 88% RVV

it has been tried out to determine the presence and diversity of the characteristic achromous microflora in three different regions in Serbia (Zlatibor, Levac, and Pomoravlje). At the same time the physical and chemical sensory changes which occurred during the process of fermentation and maturation of traditional sausages. Not only, but all data relative to the manufacturing of pork meat, microclimatic changes during the production process (temperature, relative humidity and air currents) were collected and recorded.

The aim of the physical, chemical and microbiological studies was to register the changes which occurred during the fermentation and maturation process. The aim of the sensory studies was to establish when do start and with what intensity the desired sensory changes which are reflected in the final product.

In order to obtain reliable indicators and results during the Project all studies were repeated as triplicates in all three types of traditional sausages. Within each fermentation 50 LAB isolates were collected and 50 catalase positive cocci (staphylococci and micrococci). Resulting in a collection of 450 LAB isolates and 450 micrococci isolates which have been morphologically and biochemically studied and by API tests were closely identified.

The results of a number of papers related to studies of the microflora of traditional fermented sausages have shown that out of the total number of LAB the most predominant are the *Lb. sakei* (Amor *et al.*, 2005) and *Lb. curvatus*. *Lb. sakei* making more than 55% of the total number of isolates (Hugas & Monfort, 1997). Both of these LABS have an interesting metabolic potential upon which the possibility of their application in the meat industry is based. A smaller part of the microflora of fermented sausages which mature spontaneously, are *Lb. plantarum*, *Lb. brevis*, *Lb. paracasei* i *Lb. buchneri*. Nowadays, some of them, specially *Lb. plantarum* and *Lb. sakei* (commonly referred to as the "good" technological bacterium due to their production of antimicrobial substances – bacteriocines) and *Lb. curvatus* and *Lb. pentosus* are used as starter cultures in the production of fermented sausages. Their use is the result of detailed studies, taxonomic determination, identification of morphologic, physiologic and functional biotechnologic properties.

For further projects it is planned that isolated LAB strains should undergo detailed morphological and biochemical studies, as well as molecular identification. Within all identified strains the most important technological and protective features will be studied. Thus, all isolates will be studied in order

to enhance secondary metabolic compounds i. e. bactericines

LAB bactericines are natural antimicrobial peptides or proteins with a very interesting potential application in the food industry, as bioprotectors (Cleveland *et al.*, 2001), with the aim to protect health (Turcotte *et al.*, 2004) with a simultaneous increase in shelf life (Slavica Vesković, 2007, 2007-1). Bacteriocines are polypeptides synthesized on ribosomes, have a potent bactericidal activity and are quickly digested by the proteases of the human alimentary tract (Joerger *et al.*, 2000).

In the published papers very often they are compared with antibiotics (Hansen, 1993; Hurst, 1981). However, being not equal to therapeutic antibiotics their use is seldom associated to allergic reactions in man (Cleveland *et al.*, 2001). What meant to the society the discovery by Alexander Fleming (1929) of penicillin regard human health, from the aspect of food safety and natural protection is represented by the bacteriocines.

Bearing in mind the expressed bacteriocidal and bacteriostatic effects of bacteriocines on some pathogenic strains, in the last years their application within the meat industry has been reviewed. On the other side, direct consumers have a consistently negative approach on the question of the use of chemical additives in food production. As a result consumers are not sure on the use of treated foods, with the exception of fresh food. Such a trend on one side (so called green technology - Ross *et al.*, 2002) and the continuous development of modern protective technologies in the XX and XXI century have included the exploitation of biological protectors such as bacteriocines. However, for bacteriocines to be used in the food industry they must be approved as legal additives („GRAS“ Generally Regarded As Safe). Up to now only nisine has this status.

Bacteriocines as bioprotectors can be used in food production in one of the following ways (Schillinger *et al.*, 1996):

- by adding them to the food LAB which produce bacteriocines within the product ("in situ" production);
- direct use and/or semi purified bacteriocines as additives
- use of previously fermented products containing bacteriocine producing bacterial strains.

Chosen strains of LAB with clear technological and/or protective features will be used for experimental production of fermented sausages in industrial conditions.

Instead of a conclusion

The predicted studies within the Project "Technological and protective features of autochthonous bacterial strains isolated from traditional fermented sausages and possibilities for their application in the meat industry" are aimed to improve the safety of food production, decrease of production costs, preservation and even improvement of sensory characteristics of traditionally fermented meat products, development of a national collection of LAB and a positive influence on the population's health. The determination of critical factors on which depends the standard quality of traditionally fermented meat products is of great importance not only to Project participants, but to the national public, as well.

Basic HACCP principles can not be implemented without adequate data on the physical and chemical, microbiological and sensory characteristics of

all ingredients, standard operating procedures (SOP) for the technological process and modern methods for the control of individual production phases as well as for the control of the final products.

By isolation and adequate selection of domestic, ephythic microflora and by forming a collection of LAB the foundations for the determination and their possible use in the production of autochthonous starter and/or protective cultures. In such a way can be maintained the preservation of the sensory characteristics of traditionally fermented sausages

It is known that due to their sensory characteristics traditional fermented sausages are highly rated not only on the domestic market, but there is also a great interest for them on foreign markets. In order to export products on foreign markets it is important to ensure a consistently good quality product with characteristic sensory features, all at a good price.

References:

- Caplice, E., Fitzgerald G., 1999. Food fermentations: Role of microorganisms in food production and preservation. *Int. J. Food Microbiol.* 50: 131-149;
- Coretti, K., 1971. Rohwurstreifung und Fehlerzeugnisse bei der Rohwurstherstellung. *Fleischforschung und Praxis, Schriftenreihe Heft 5*, Verlag der Rhein Hessischen Druckwerkstate Alzey;
- Coretti, K., 1975. Rohwurst und Fohlfleischwaren I Teil: Rohwurst. *Fleischwirtschaft*, 55, 174-181;
- Erkkila S., 2001. Bioprotective and probiotic meat starter cultures for the fermentation of dry sausages. Academic dissertation. University of Helsinki. Department of Food Technology;
- Niinivaara, F., 1955. Über den Einfluss von Bakterienreinkulturen auf die Reifung und Umrotung der Rohwurst. *Avta Agr. Fenn.*, Helsinki, 84:128;
- Kandler, O., 1983. Carbohydrate metabolism in lactic acid bacteria. *Ant. van Leeuwenhoek*, 49: 209-224;
- Axelsson, L., 1998. Lactic Acid Bacteria: Classification and Physiology. In: Salminen, S., von Wright, A. (Eds.), *Lactic acid bacteria: Microbiology and Functional Aspects*, 2nd Edition, Marcel Dekker Inc., New York, pp. 1-72;
- Ross, R. P., Morgan, S., Hill, C., 2002. Preservation and fermentation: past, present and future. *Int. J. Food Microbiol.*, 79: 3-16;
- Garcia, M. L., Selgas, M. D., Fernandez, M., Ordóñez, J.A., 1992. Microorganisms and lipolysis in the ripening of dry fermented sausages. *Int. J. Food Sci. Technol.*, 27: 675-682;
- Molly, K., Demeyer, D., Civera, T., Verplaetse, A., 1996. Lipolysis in a Belgian sausage: relative importance of endogenous and bacterial enzymes. *Meat Sci.*, 43: 235-244;
- Demeyer, D. I., 1992. Meat fermentation as an integrated process. In *New technologies for meat and meat products*, eds. F.J.M. Smulders, F. Toldrá, J. Flores and M. Prieto, *Audet Tijdschriften*, Nijmegen, pp. 21-36;
- Lücke, F. K., 2000. Utilization of microbes to process and to preserve meat. *Meat Sci.*, 56: 105-115;
- Daly, C., Davis, R., 1998. The biotechnology of lactic acid bacteria with emphasis on applications in food safety and human health. *Agr. food Sci.*, Finland, 7: 251-265;
- Hammes, W., Rolz, I., Banteon, A., 1985. *Microbiologische Untersuchung der auf dem deutschen Markt vorhandenen Starterkulturpräparate für die Rohwurstbereitung*;
- Lindgren, S. E., Dobrogosz, W. J., 1990. Antagonistic activities of lactic acid bacteria in food and feed fermentations. *FEMS Microbiol. Rev.*, 7: 149-163;
- De Vuyst, L., Vandamme, E., 1994. Nisin, a lantibiotic by *Lactococcus lactis* subsp. *lactis*: properties, biosynthesis, fermentation and applications. In: De Vuyst, L., Vandamme, E (Eds.). *Bacteriocins of lactic acid bacteria*. Blackie, London, Glasgow, New York, Tokyo, Melbourne, Madras, pp. 151-221;
- Holzapfel, W., Geisen, R., Schillinger, U., 1995. Biological preservation of foods with reference to protective cultures, bacteriocins and food-grade enzymes. *Int. J. Food Microbiol.*, 24: 343-362;
- Vesković Slavica, 2005. „Uticaj bakteriocina *Leuconostoc mesenteroides* E 131 i *Lactobacillus sakei* I 154 na *Listeria monocytogenes* u toku proizvodnje Sremske kobasice“, Magistarska teza, Poljoprivredni fakultet, Zemun – Beograd.
- Vesković Slavica, 2007. „Uticaj *Lactobacillus sakei* I 151, bakteriocina *Leuconostoc mesenteroides* E 131 i MAP na održivost Sremske kobasice“, Doktorska disertacija, Poljoprivredni fakultet, Zemun – Beograd.
- Vesković Slavica, 2009. Bakteriocini BMK – Mogućnosti primene u proizvodnji fermentisanih kobasica, Monografija, 1-89. Izdavač: Zadužbina Andrejević, Beograd.
- Leistner, L., 1985. *Mikrobiologie und Qualität von Rohwurst und Rohschinken*. Herausgeber: Institut für Mikrobiologie, Toxikologie und Histologie der BAFF, Kulmbach, pp. 219-244.
- Ammor, S., Dufour, E., Zagorec, M., Chaillou, S., Chevallier, I., 2005. Characterization and selection of *Lactobacillus sakei* strains isolated from traditional dry sausage for their potential use as starter cultures. *Food Microbiol.*, 22: 529-538;
- Hugas, M., Monfort, J. M., 1997. Bacterial starter cultures for meat fermentation. *Food Chem.*, 59: 547-554.
- Cleveland, J., Montville, T. J., Nes, I. F., Chikindas, M. L., 2001. Bacteriocins: Safe, natural antimicrobials for food preservation. *Int. J. Food Microbiol.*, 71: 1-20.

- Turcotte, C., Lacroix, C., Kheadr, E., Grignon, L., Fliss, I., 2004.** A rapid turbidometric microplate bioassay for accurate quantification of lactic acid bacteria bacteriocins. *Int. J. Food Microbiol.*, 90: 283-293;
- Slavica Vesković Moračanin, Turubatović, L., Stjepanović, A., 2007-1.** Application of protective cultures and bacteriocines in the production of traditionally fermented sausages, Uvodno predavanje (predavanje po pozivu) na 10-oj Međunarodnoj načnoj konferenciji „Aktuelni problemi u industriji: inovacije, kvalitet i rukovodjenje“, Moskva, Rusija 4-6 decembar 2007 god. Zbornik radova, pp.7-16;
- Joerger, R. D., Hoover, D. G., Barefoot, S. F., Harmon, K. M., Grinstead, D. A., Nettles-Cutter, C. G., 2000.** Bacteriocins. In: Lederberg, editor. *Encyclopeda o microbiology*, Vol. 1, 2nd edition. San Diego: Academic Press, Inc. pp. 383-97;
- Hansen, J. N., 1993.** Antibiotics synthesized by post translational modification. *Annu. Rev. Microbiol.*, 47: 535-564;
- Hurst, A., 1981.** Nisin. *Adv. Appl. Microbiol.*, 27: 85-123;
- Cleveland, J., Montville, T. J., Nes, I. F., Chikindas, M. L., 2001.** Bacteriocins: Safe, natural antimicrobials for food preservation. *Int. J. Food Microbiol.*, 71: 1-20;
- Ross, R. P., Morgan, S., Hill, C., 2002.** Preservation and fermentation: past, present and future. *Int. J. Food Microbiol.*, 79: 3-16;
- Schillinger, U., Geisen R., Holzapfel, W. H., 1996.** Potential of antagonistic microorganisms and bacteriocins for the biological preservation of foods. *Trends Food Sci Technol.*, 7: 158-64.

Paper received: 23.04.2009.

Note: Research Project: “Technological and protective features of autochthonous bacterial strains isolated from traditional fermented sausages and possibilities for their application in the meat industry” is financed by the Ministry of Science and Technological Development of the Republic of Serbia

NEKI PARAMETRI KVALITETA I NUTRITIVNA VREDNOST FUNKCIONALNIH FERMENTISANIH KOBASICA*

Vuković, I., Saičić Snežana, Vasilev D., Tubić M., Vasiljević Nađa, Milanović-Stevanović Mirjana

S a d r ž a j: U radu su prikazani rezultati ispitivanja nekih parametara kvaliteta i nutritivne vrednosti funkcionalnih fermentisanih kobasica. Funkcionalne fermentisane kobasice, dobijene od svinjskog i goveđeg mesa prve kategorije (75–80 posto), masnog tkiva svinja i biljne masti, uz dodatak inulina, vlakana, omega-3 masnih kiselina i probiotske starter kulture, sadrže 24 posto proteina i do 30 posto masti, a energetska vrednost manja je za oko 400 kJ (95 kcal/100 g) nego konvencionalnih. Zamenom masnog tkiva svinja palminom masti postiže se povoljniji odnos između sadržaja nezasićenih i zasićenih masnih kiselina. Palmina mast u količini do 15 posto u izvesnom stepenu menja, ali ne utiče negativno na boju fermentisanih kobasica i daje proizvodima čvrstoću i konzistenciju. U funkcionalnim fermentisanim kobasicama probiotska bakterija *Lactobacillus casei* LC 01 dostiže broj veći od 8,0 log cfu/g, fermetiše šećere i stvara povoljne uslove za zrenje. Povoljan sastav, kao i prisustvo probiotika, inulina, vlakana i omega-3 masnih kiselina, čini funkcionalne fermentisane kobasice kvalitetnom namirnicom visoke biološke vrednosti, koja poseduje značajan potencijal da pozitivno utiče na zdravlje ljudi.

Ključne reči: fermentisane kobasice, funkcionalna hrana, kvalitet, nutritivna vrednost

SOME QUALITY PARAMETERS AND NUTRITIONAL VALUE OF FUNCTIONAL FERMENTED SAUSAGES

A b s t r a c t: in this paper results of studies on some quality parameters and nutritional value of functional fermented sausages are shown. Functional fermented sausages, made of first class pork and beef meat (75 - 80%), lard and vegetable fats, with the addition of inulin, fibers, omega - 3 fatty acids and probiotic starter cultures contain 24% protein and 30% fat. Their energetic value is smaller by 400kJ (95 kcal/100 g) compared to traditional ones. By replacing the pork fat with palm fat a favorable relationship between saturated and unsaturated fatty acids content is obtained. Palm fat, supplied in a quantity of up to 15% changes to a certain degree the color of fermented sausages and gives them a degree of firmness and consistency. In functional fermented sausages the probiotic bacteria *Lactobacillus casei* LC 01 reaches values above 8.0 log cfu/g and it ferments sugars thus creating optimal conditions for ripening. A superior composition, as well as the presence of probiotics, inulin, fibers and omega - 3 fatty acids makes this product a foodstuff of high nutritional value with a high potential to have positive effects on human health status.

Key words: fermented sausages, functional foodstuff, quality, nutritional value

Uvod

Pojam funkcionalna hrana odnosi se na namirnice koje, pored osnovnih nutritijenata, sadrže i sastojke koji pozitivno utiču na zdravlje ljudi. Dodaci koji neku namirnicu mogu da čine funkcionalnom hranom su različiti i u njih se ubrajaju probiotici, prebiotici, antioksidansi, omega-3 masne kiseline, biljne masti i ulja, bioaktivni peptidi, vlakna, mine-

ralne materije, mikroelementi, vitamini, i drugo (Jimenez-Colmenero, 2001; Arihira, 2006). Dosađajna ispitivanja pokazuju da fermentisane kobasice mogu da se proizvode i kao funkcionalna hrana (Mendosa i sar., 2001; Muguersa i sar., 2004; Müller, 2006; Vuković i sar., 2007; Vasilev i sar., 2007). Fermentisane kobasice se dobijaju od usitnjenog mesa i masnog tkiva, zatim začina, šećera, aditiva, starter kultura i drugih dodataka, koji se

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa održanom 15–17. juna 2009. na Tari

AUTORI: Ilija Vuković, vukovic@eunet.yu, Dragan Vasilev, Fakultet veterinarske medicine, Beograd; Snežana Saičić, Mirjana Milanović-Stevanović, Institut za higijenu i tehnologiju mesa, Beograd; Nađa Vasiljević, Medicinski fakultet u Beogradu, Miodrag Tubić, Kompanija Big-Bull, Klanica i prerada mesa, Bačinci.

AUTHORS: Ilija Vukovic, vukovic@eunet.yu, Dragan Vasilev, Faculty of Veterinary Medicine, Belgrade; Snežana Saicic, Mirjana Milanovic-Stevanovic, Institute of Meat Hygiene and Technology, Belgrade; Nadja Vasiljevic, Faculty of Medicine, Miodrag Tubic, Company Big-Bull, Meat Industry, Bacinci

posle nadevanja u omotače konzervisu sušenjem, sa ili bez dimljenja, pri čemu kobasice sazrevaju i dobijaju karakteristične osobine kvaliteta i postaju mikrobiološki i hemijski stabilniji, odnosno održivi proizvodi (Vuković, 2006). Činjenica da se ove kobasice u toku proizvodnje ne obrađuju toplotom omogućava, s jedne strane da nutritivno vredni sastojci mesa ostaju bitnije nepromenjeni u smislu smanjenja biološke vrednosti, a s druge strane otvara se mogućnost upotrebe probiotika kao starter kultura, koji u funkcionalnim fermentisanim kobasicama imaju poseban značaj (Vuković i sar., 2007).

Meso je na osnovu sadržaja proteina, vitamina, mikroelemenata i drugih sastojaka, bez sumnje, najvažnija „funkcionalna“ komponenta fermentisanih kobasica. U proteinima mišića oko 43 posto su esencijalne aminokiseline. U intramuskularnoj masti mesa sadržana je značajna količina polinezasićenih masnih kiselina, a masti preživara sadrže i konjugovanu linolnu kiselinu kojoj se pripisuje antioksidativno, antiteratogeno i antikancerogeno dejstvo. I neki drugi sastojci mesa, kao što su anserin, karnizin i glutation, imaju važnu ulogu kao antioksidansi. Meso je jedan od najbogatijih izvora vitamina B grupe za čoveka, kao što su tiamin (B₁), riboflavin (B₂), niacin, folna kiselina, piridoksin (B₆) i kobaltamin (B₁₂). Meso je, takođe, vrlo važan izvor gvožđa; najviše gvožđa sadrže konjsko i goveđe meso, a u manjoj meri svinjsko i živinsko meso. Gvožđe iz mesa oko pet puta se bolje iskorišćava od gvožđa iz biljaka i ujedno pomaže resorpciju gvožđa biljnog porekla. Gvožđe se bolje iskorišćava zajedno sa bakrom koji se, takođe, nalazi u mesu. Crveno meso je vrlo dobar izvor cinka koji se, isto tako, bolje resorbuje iz mesa nego iz biljaka. Cink je naročito potreban starijim osobama, jer utiče na smanjenje koncentracije lipidnih peroksida u krvnoj plazmi. Meso je značajan izvor selena, koji je kao antioksidans, poput vitamina E i C, važan sastojak enzima koji štite ćelije od oksidacije (Prändl, i sar., 1988; Gašparin i sar., 2002).

Od svih proizvoda od mesa jedino fermentisane kobasice, kada se proizvode kao funkcionalna hrana, sadrže probiotske bakterije. Kao probiotici se koriste, pre svega, bakterije vrste *Bifidobacterium* spp. i vrste roda *Lactobacillus* (*Lb. acidophilus*, *Lb. casei* i *Lb. rhamnosus*). Inače, prirodnu mikrofloru fermentisanih kobasica čine pretežno bakterije roda *Lactobacillus*, prvenstveno *Lb. sakei* i *Lb. curvatus*, a manjim delom *Lb. plantarum*, *Lb. brevis*, *Lb. paracasei* i *Lb. buchneri*. Opšte prihvaćeni zahtevi za probiotske mikroorganizme su da predstavljaju značajan deo crevne flore zdravog čoveka, da preživljavaju pasažu kroz želudac i creva (otporne na kiseline i žuč) i da mogu da se adhezuju na ćelije

crevnog epitela. Mikroflora koja učestvuje u zrenju kobasica, sa izuzetkom *Lb. plantarum*, nema probiotski značaj. Probiotske bakterije koje se danas nalaze na tržištu razvijene su, pretežno, za potrebe industrije mleka i samo neke od njih mogu da rastu u fermentisanim kobasicama. Da bi se ostvarilo merljivo probiotsko dejstvo smatra se da sa jednim gramom fermentisane kobasice treba uneti u organizam najmanje jedan milion probiotskih bakterija. Uticaj probiotske hrane na zdravlje čoveka nije, do danas, potpuno izučen, ali neke studije potvrđuju da konzumiranje 50 g/dan fermentisane kobasice proizvedene sa jednim sojem *Lb. paracasei* utiče pozitivno na imuni sistem čoveka (Kröckel, 2006).

Prebiotici su nesvarljivi sastojci hrane koji stimulišu rast i/ili aktivnost jedne ili manjeg broja vrsta bakterija u debelom crevu i povoljno deluju na zdravlje domaćina. U fermentisane kobasice dodaju se najčešće inulin i dijetalna vlakna. Inulin je oligosaharid koji nije svarljiv u tankom crevu. Međutim, bakterije prisutne u debelom crevu razlažu inulin do laktata i acetata kao krajnjih produkata fermentacije, što ima kao posledicu pozitivne promene u crevnoj flori čoveka. Inulin, takođe, doprinosi boljem iskorišćavanju kalcijuma, smanjenju rizika od stvaranja pretkanceroznih lezija i opadanju nivoa triglicerida u krvi. Inulin ne utiče nepovoljno na teksturu, sočnost i elastičnost fermentisanih kobasica i može da zameni jedan deo masnog tkiva u nadevu. Dijetalna vlakna (ovas, šećerna repa, soja, jabuka, i grašak), biljni proteini (soja, suncokret, pšenica, kukuruz, ovas i seme pamuka) i njihovi proizvodi, pored toga što povoljno deluju na zdravlje ljudi, mogu da se koriste i kao zamena za masno tkivo u proizvodima od mesa. Takođe se koriste i sinbiotici koji predstavljaju mešavinu probiotika i prebiotika (Jackson i sar., 1996; Causey i sar., 2000; Rao, 2001; Roberford, 2002; Mendosa i sar., 2006).

Osim prebiotika, masno tkivo u fermentisanim kobasicama može da se delimično ili čak potpuno zameni biljnim uljima (repičino, laneno, maslinovo i kukuruzno), mastima dobijenim od ovih ulja i emulzija ulja sa proteinima. Upotrebom ulja povećava se sadržaj nezasićenih i polinezasićenih masnih kiselina, a posebno omega-3 masnih kiselina. Iako najviše omega-3 masnih kiselina sadrži riblje ulje, ono se retko koristi, jer nepovoljno utiče na aromu proizvoda. U proizvodnji funkcionalne hrane značajno mesto imaju biljni steroli, koji mogu da budu nezasićeni – fitosteroli i zasićeni – fitostanoli, i po građi i funkciji su slični holesterolu. Fitosteroli snižavaju nivo LDL holesterola u krvi i štite organizam od kardiovaskularnih oboljenja. Fitosteroli se dodaju u različite namaze, jogurt i mleko, koji su obogaćeni slobodnim fitosterolima ili estrima fitosteril ili fito-

stanil masnih kiselina. Najvažniji izvor biljnih stero-
la su biljna ulja i margarina (*Simopoulos i sar.*, 2000;
Lagarda i sar., 2006; *Jimenez-Colmenero*, 2007).

Polazeći od podataka iz literature i sve strože
zahteve u pogledu sastava i nutritivne vrednosti
hrane, postavljen je cilj da se razvije nova generacija
fermentisanih kobasica kao funkcionalne hrane, koje
bi imale visoku biološku vrednost, i po kvalitetu, bi-
le prihvatljive na domaćem tržištu, odnosno odgova-
rale navikama naših potrošača. U koncipiranju ovog
rada pošlo se i od Istraživanja zdravlja stanovništva
Republike Srbije (2007), čiji rezultati pokazuju da
je u Srbiji svaka peta osoba gojazna ($BMI \geq 30$), a
svaka treća osoba sa predgojaznošću ($BMI \geq 25$),
što je bez sumnje posledica ishrane. U ovom radu
prikazan je deo rezultata dobijen realizacijom pro-
jekta TR-20073, koji finansira Ministarstvo nauke i
tehnološkog razvoja Republike Srbije.

Materijal i metode

U eksperimentima je izrađeno i ispitano više
različitih formulacija funkcionalnih fermentisanih
kobasica. Kao rezultat toga, razvijena su tri nova
proizvoda: (1) funkcionalna fermentisana kobasica
sa masnim tkivom svinja (Probio), (2) funkcionalna
fermentisana kobasica sa masnim tkivom svinja i
biljnom masti (Probiomiks) i (3) funkcionalna fer-
mentisana kobasica sa palminom masti (Probiofit).
Fermentisane kobasice su spravljane na uobičajeni
način od svinjskog i govedeg mesa prve kategori-
je (75–80 posto), čvrstog masnog tkiva, mešavine
čvrstog masnog tkiva i biljne masti ili samo biljne
masti, koji su bili u takvom odnosu da sadržaj masti
u nadevu na početku zrenja bude ujednačen i iznosi
18–19 posto. U nadev je dodato 2,0 posto inulina,
1,0 posto vlakana graška, 50 mg/100 g omega-3
masnih kiselina (Den omega Gat) i probiotska kul-
tura *Lactobacillus casei* 01 (LC 01). Na 1,0 kilo-
gram nadeva dodato je 28 grama nitritne soli za
salamurenje, 5,0 grama šećera i mešavina začina.
Nadev je usitnjavan finije do veličine komadića tkiva,
od oko 2 mm, i posle punjenja u kolagene omotače
prečnika 65 mm, kobasice su podvrgnute sušenju,
odnosno zrenju na temperaturama koje su opadale od
24 do 16°C. Sušenje i zrenje trajalo je 20 dana.

Funkcionalne fermentisane (polusuve) kobasice
su ispitivane standardnim fizičkim, fizičko-hemijskim,
hemijskim, bakteriološkim i senzornim metodama.

Fizičke metode: a) Određivanje gubitka mase
(kalo sušenja) gravimetrijski; b) Instrumentalno
merenje boje (CIE L*, a*, b*) uređajem Minolta
Co. Ltd. Chromameter CR-400); c) Instrumentalno
merenje čvrstoće aparatom Instron 4301 (sila pre-
secanja i sila penetracije u N).

Fizičko-hemijske metode: Određivanje pH-
vrednosti (pH-meter WTW, 340i) i aktivnosti vode
(a_w -Wert-Messer, Luft Durotherm, Stuttgart).

Hemijske metode: a) Određivanje sadržaja vlage
(SRPS ISO 1442/1998); b) Određivanje sadržaja
ukupne masti (SRPS ISO 1443/1992); c) Određivanje
sadržaja proteina (SRPS ISO 937/1992); d) Određivanje
sadržaja hidroksiprolina (JUS ISO 3496/2002); e)
Određivanje indeksa proteolize (*Careri i sar.*, 1993);
f) Određivanje sadržaja natrijum–hlorida (SRPS ISO
1841-2 /1999); g) Određivanje sadržaja pepela (SRPS
ISO 936/1999); h) Određivanje sadržaja nitrita (SRPS
ISO 2918/1999); i) Određivanje kiselinskog broja
(SRPS ISO 660/1996), peroksidnog broja (SRPS ISO
3960/2001) i TBARS-broja (*Tarladgis i sar.*, 1964
i *Holland*, 1971); j) Određivanje sadržaja masnih
kiselina (ekstrakcija lipida metodom po *Garces i
Manuelu*, 1993, a potom određivanje masnih kiselina
gasnom hromatografijom (FAME MIX 37, kolona
100 m, signal 28,5, split 30).

Bakteriološke metode: Određivanje broja pro-
biotske bakterije *Lactobacillus casei* 01 na MRS-
agaru, Merck, sa dodatkom moksalaktama u količi-
ni od 112 mg/L, Sigma M-8158, pri 37 °C/72 časa u
anaerobnoj sredini (*Kröckel*, 2006).

Senzorne metode: Ukupan senzorni kvalitet
fermentisanih kobasica po metodi korigovanog pe-
tobalnog bod sistema (*Radovanović i Popov-Raljić*,
2001).

Rezultati ispitivanja i diskusija

Na osnovu ispitivanja hemijskog sastava eks-
perimentalnih funkcionalnih fermentisanih kobasica
(tabela 1) može da se zaključi da se ove kobasice
odlikuju visokim sadržajem proteina (24 posto) i
relativno malim sadržajem ukupne masti (<30 posto),
pri čemu odnos između sadržaja proteina i masti
nije veći od 1:1,25. S druge strane, konvencionalne
fermentisane kobasice, dobijene pretežno od svinj-
skog i govedeg mesa druge kategorije (oko 70 posto)
i čvrstog masnog tkiva (oko 30 posto), zavisno od
stepena sušenja, sadrže 18–20 posto proteina i
42–44 posto masti, pa je kod njih sadržaj masti više
od dva puta veći od sadržaja proteina. Upotrebom
mesa prve kategorije dobijen je oko četiri puta
manji relativan sadržaj proteina vezivnog tkiva u
proteinima mesa koji, prema Pravilniku o kvalitetu
i drugim zahtevima za proizvode od mesa (2004),
za fermentisane kobasice ne sme da bude veći od 15
posto. Ovo pokazuje da je sadržaj proteina mišićnog
tkiva, i apsolutno i relativno veći kod funkcionalnih
nego konvencionalnih fermentisanih kobasica. Sa
dodatkom 3 posto prebiotika, njihov sadržaj u goto-
vom proizvodu dostiže vrednost oko 5 posto.

Tabela 1. Važniji pokazatelji hemijskog sastava funkcionalnih fermentisanih kobasica**Table 1.** Relevant indicators of chemical composition of fermented sausages

Sastojaci (%)	Probio	Probiomiks	Probiofit
Voda	36,26	36,70	34,25
Masti	28,99	29,41	30,55
Proteini	24,13	23,98	24,93
Relativan sadržaj proteina vezivnog tkiva	3,38	3,38	3,61
Natrijum hlorid	4,00	3,76	4,03
Natrijum-nitrit (mg/kg)	3,2	2,4	2,1
Inulin i vlakna*	4,87	4,95	5,16

* Određeno iz razlike do 100 posto

Funkcionalna fermentisana kobasica sa palmynom masti (Probiofit) sadrži više nezasićenih masnih kiselina od funkcionalnih kobasica sa masnim tkivom svinja ili mešavinom masnog tkiva i biljne masti (tabela 2), i na osnovu toga, ima povoljnije odnose između sadržaja zasićenih i nezasićenih masnih kiselina u mastima proizvoda. Odnos između sadržaja zasićenih i nezasićenih masnih kiselina i kod funkcionalnih fermentisanih kobasica sa masnim tkivom je, takođe, povoljan sa gledišta dijetetike. Vrednostima za sadržaj nezasićenih masnih kiselina treba dodati i omega-3 masne kiseline, čiji sadržaj zbog nedovoljne specifičnosti primenjene metode nije mogao da bude određen, a koje su ovim proizvodima dodate u količini od 50 mg/100 g.

Tabela 2. Sadržaj masnih kiselina funkcionalnih fermentisanih kobasica i odnosi između zasićenih i nezasićenih masnih kiselina**Table 2.** Fatty acids content of functional fermented sausages and relationship between saturated and unsaturated fats

Masne kiseline (g/100g)	Probio	Probiomiks	Probiofit
Zasićene	12,83	14,23	12,95
Nezasićene	14,74	14,61	16,06
Polinezasićene	3,48	2,92	3,69
Polinezasićene/zasićene	0,27	0,20	0,28
Nezasićene/zasićene	1,15	1,03	1,24
Zasićene/nezasićene	0,87	0,97	0,81

U poređenju sa konvencionalnom fermentisanom kobasicom, funkcionalne fermentisane koba-

sice sadrže više proteina mesa za 4 g/100 g i manje masti za 12 g/100 g (tabela 3). Funkcionalne fermentisane kobasice sadrže, takođe, do 5 g/100 g prebiotika (inulin i biljna vlakna) i najmanje 50 mg/100 g omega-3 masnih kiselina. Energetska vrednost funkcionalnih fermentisanih kobasica je za 400 kJ/100 g (95 kcal/100 g) manja nego konvencionalne kobasice. Kako je relativan sadržaj proteina vezivnog tkiva u proteinima mesa funkcionalnih fermentisanih kobasica oko četiri puta manji, tako je i sadržaj proteina mišićnog tkiva, kao osnovnog izvora esencijalnih amino kiselina u hrani, i apsolutno i relativno veći kod funkcionalnih nego konvencionalnih fermentisanih kobasica. Proteini mišićnog tkiva sadrže 43 posto esencijalnih amino kiselina, a protein vezivnog tkiva 23 posto kolagena. Veća količina mesa u funkcionalnim fermentisanim kobasicama ima kao posledicu ne samo veći sadržaj proteina, već i više vitamina B grupe, zatim gvožđa, cinka i drugih nutritivno važnih sastojaka. Sa dodatkom 3 posto prebiotika, njihov sadržaj u gotovom proizvodu dostiže vrednost oko 5 posto. Kada se, pored ovoga, uzme u obzir da ove kobasice sadrže i probiotske bakterije koje, kao i omega-3 masne kiseline, pozitivno utiču na zdravlje ljudi, može da se zaključi da funkcionalne fermentisane kobasice predstavljaju hranu vrlo visoke biološke vrednosti sa značajnim potencijalom da pozitivno utiču na zdravlje ljudi.

Tabela 3. Usporedni prikaz važnijih parametara nutritivne vrednosti funkcionalne konvencionalne fermentisane kobasice**Table 3.** Comparative display of important parameters of nutritive values of functional and conventional sausages

Parametar	Funkcionalna	Konvencionalna
Energetska vrednost, kJ/100 g (kcal/100 g)	1578 (377)	1978 (472)
Proteini, g/100 g	24,0	20
Masti, g/100 g	30,0	42
Inulin i biljna vlakna, g/100g	5,0	–
Omega-3 masne kiseline, mg/100 g	50	–

Probiotska bakterija *Lactobacillus casei* LC 01, koja je razvijena za primenu u mlecarskoj industriji, pokazalo se da može dobro da se razmnožava i u funkcionalnim fermentisanim kobasicama i da dostiže broj veći od 8,0 log cfu/g (tabela 4). Prema usvojenim standardima, broj probiotskih bakterija u funkcionalnim fermentisanim proizvodima mora da

bude veći od 6,0 log cfu/g. Vrednost pH funkcionalnih fermentisanih kobasica koje sadrže palminu mast niža je od pH vrednosti fermentisane kobasice koja sadrži masno tkivo svinja. Funkcionalne fermentisane kobasice sa biljnom masti, takođe, imaju manju aktivnost vode. Na osnovu ovih vrednosti može se zaključiti da funkcionalne fermentisane kobasice imaju vrlo dobru mikrobiološku stabilnost i da u njima ne postoje uslovi za razmnožavanje patogenih bakterija.

Tabela 4. Broj probiotske bakterije *Lactobacillus casei* 01 (LC 01) i pH i a_w vrednosti funkcionalnih fermentisanih kobasica

Table 4. The number of probiotic bacteria *Lactobacillus casei* (LC01) and pH and a_w values of functional fermented sausages

Fermentisana kobasica	Broj LC 01 (log cfu/g)	pH	a_w
Probio	8,43	4,94	0,90
Probiomiks	8,38	4,89	0,89
Probiofit	8,26	4,86	0,88

Rezultati instrumentalnog merenja boje preseka i čvrstoće funkcionalnih fermentisanih kobasica prikazani su u tabeli 5. Funkcionalna fermentisana kobasica sa palminom masti (Probiofit) ima veći udeo crvene (a^*) i žute (b^*) boje i manju L^* -vrednost, odnosno boja ove kobasice na preseku je nešto tamnija od boje kobasica u čijem sastavu dominira masno tkivo svinja. Instrumentalnim merenjem čvrstoće funkcionalnih fermentisanih kobasica, utvrđena je veća sila presecanja i veća sila penetracije

Tabela 5. Rezultati instrumentalnog merenja boje preseka i čvrstoće funkcionalnih fermentisanih kobasica

Table 5. Results of measurements of the color and firmness of functional fermented sausages

Fermentisana kobasica	L^*	a^*	b^*	Sila presecanja (N)	Sila penetracije (N)
Probio	43,02	18,16	6,56	25,19	19,18
Probiomiks	43,24	18,73	7,53	28,49	22,51
Probiofit	41,96	19,53	8,49	30,86	23,20

Tabela 6. Ukupan senzorni kvalitet funkcionalnih fermentisanih kobasica

Table 6. Total sensory quality of functional fermented sausages

Fermentisana kobasica	Spoljašnji izled	Izgled i sastav preseka	Boja i održivst boje	Miris i ukus	Tekstura	Ukupna ocena
Probio	9,43	20,00	19,43	27,86	18,86	95,57
Probiomiks	10,00	19,14	19,14	28,29	19,43	96,00
Probiofit	9,86	18,57	18,86	28,29	19,43	95,00

kod fermentisanih kobasica kod kojih je masno tkivo potupno ili delimično zamenjeno palminom masti. Iskustva dobijena u proizvodnji ovih kobasica su u punom skladu sa ovim rezultatima, jer se pokazalo da funkcionalne fermentisane kobasice sa biljnom masti brže postižu čvrstoću, odnosno njihova konzistencija je uvek čvršća.

Ukupan senzorni kvalitet funkcionalnih fermentisanih kobasica prikazan je u tabeli 6. Kao što ukazuju rezultati, funkcionalne fermentisane kobasice su prilikom senzornog ispitivanja dobile ocenu za ukupan senzorni kvalitet od 95,00 do 96,00, na osnovu koje bi ovim kobasicama na javnim ocenama kvaliteta (na primer, na Novosadskom sajmu) pripala zlana medalja za kvalitet.

Uporedni prikaz cene (u dinarima) sirovina i dodataka po 1,0 kilograma nadeva funkcionalnih fermentisanih kobasica i jedne konvencionalne kobasice, obračunate po cenama od 31. marta 2009. godine, dat je u tabeli 7. U poređenju sa konvencionalnom fermentisanom kobasicom, koja se dobija od mesa druge kategorije (70 posto) i čvrstog masnog tkiva (30 posto), cena sirovina i dodataka funkcionalnih fermentisanih kobasica je znatno veća. Na to utiču veća količina mesa u nadevu (75–80 posto), veća cena mesa prve kategorije (oko 40 posto), upotreba probiotske starter kulture, čija cena je nekoliko puta veća od cene drugih kultura, zatim inulina, vlakana i preparata omega-3 masnih kiselina. Prilikom određivanja proizvođačke cene funkcionalnih fermentisanih kobasica treba uzeti u obzir i troškove rada i utrošene energije (oko 100 din./kg) i gubitak mase (kalo) kobasica prilikom sušenja i zrenja (30–35 posto).

Tabela 7. Usporedni prikaz cene (u dinarima) sirovina i važnijih dodataka po 1,0 kg nadeva konvencionalne i funkcionalnih fermentisanih kobasica**Table 7.** Comparative results of the cost (in dinars) of raw materials and spices per 1.0kg of filling for conventional and functional sausages

Cene* u dinarima	Konvencionalna	Probio	Probiomiks	Probiofit
Meso prve kategorije	–	354	354	367
Meso druge kategorije	224	–	–	–
Masno tkivo	36	26	20	–
Biljna mast	–	-	6	21
Inulin	–	6	6	6
Vlakna	–	2	2	2
Starter kultura	6	–	–	–
Probiotska starter kultura	–	57	57	57
Omega-3 masne kiseline	–	13	13	13
Aditivi i začini	3	6	6	6
Ukupno	269	464	464	472

* Na dan 31. marta 2009. godine

Zaključak

Rezultati ispitivanja pokazuju da funkcionalne fermentisane kobasice dobijene od svinjskog i goveđeg mesa prve kategorije (75–80 posto), masnog tkiva svinja, palmine masti i prebiotika, sadrže 24 posto proteina i do 30 posto masti, a energetska vrednost ovih proizvoda manja je za 400 kJ/100 g (95 kcal/100 g) od konvencionalnih. Zamenom masnog tkiva svinja palminom masti postižu se povoljniji odnosi između sadržaja nezasićenih i zasićenih masnih kiselina. Palmina mast, u količini do

15 posto, u izvesnom stepenu menja, ali ne utiče negativno na boju, i povećava čvrstoću proizvoda. U funkcionalnim fermentisanim kobasicama probiotska bakterija *Lactobacillus casei* LC 01 dostiže broj veći od 8,0 log cfu/g, pri čemu fermentiše šećere i stvara povoljne uslove za zrenje kobasica. Povoljan sastav, kao i prisustvo probiotika, inulina, vlakana i omega-3 masnih kiselina, čini funkcionalne fermentisane kobasice vrlo kvalitetnom namirnicom visoke biološke vrednosti, sa značajnim potencijalom da pozitivno utiču na zdravlje ljudi.

Literatura

- Anon. 2007.** Istraživanje zdravlja stanovnika Republike Srbije, 2006. godina, Finalni izveštaj, Ministarstvo zdravlja Republike Srbije;
- Arihara, K. 2006.** Strategies for Designing Novel Functional Meat Products, *Meat Sci.*, 74, 219–229;
- Careri, M., Mangia, A., Barbieri, G., Bolzoni, L., Virgili, R., Parolari, G. 1993.** Sensory property relationships to chemical data of italian dry-cured ham, *Jour. Food Scie.*, 58, 968–972;
- Causey, J. L., Feirtag, J. M., Gallaher, D. D., Tunland, B. C., Slavin, J. L. 2000.** Effects of Dietary Inulin on Serum Lipids, Blood Glucose and the Gastrointestinal Environment in Hypercholesteoleic Men, *Nutrition Res.*, 20, 2, 191–201;
- Gašparin, L., Čepin, S., Žlender, B. 2002.** The Role of Meat and Meat Products as Functional Food, *Tehn. mesa*, 43, 3–6, 186–199;
- Jackson, K. G., Taylor G. R. J., Clohessy A. M., Williams C. M. 1999.** The effect of the daily Intake of Inulin on Fasting Lipid, Insulin and Glucose Concentracions in Middle-aged Men and Women, *Br. J. Nutri.*, 82, 23–30;
- Jimenez-Colmenero, F., Carballo, J., Cofrades, S. 2001.** Healthier Meat and Meat Products: their Role as Functional Foods, *Meat Sci.*, 59, 5–13;
- Jimenez-Colmenero, F. 2007.** Healthier lipid formulation approaches in meat-based functional foods. Technological options for replacement of meat fats by non-meat fats. *Trends in Food Sci. and Technol.*, 18, 567–578;
- Kröckel, L. 2006.** Einsatz probiotischer Bakterien bei Fleischerzeugnissen, *Mitteilungsblatt der Fleischforschung Kulmbach*, 45, 173, 163–172;
- Lagarda, M. J., Garcia-Liats, G., Farre, R. 2006.** Analysis of Phytosterols in Food, *Pharmaceuticals and Biomedical analysis*, 41, 1486–1496;
- Mendoza, E., Garcia M. L., Casa, C., Slgas, M. D. 2001.** Inulin as Fat Substitute in Low Fat Dry Fermented Sausages, *Meat Sci.*, 57, 387–393;
- Muguerza, E., Gimeno, O., Ansorena, D., Astiasaran, I. 2004.** New Formulations for Healthier Dry Fermented Sausage: a review, *Trends in Food Sci. and Technol.*, 15, 452–457;
- Müller, W.-D. 2006.** Funktionelle Fleischerzeugnisse - Rohwürste, *Mitteilungsblatt der Fleischforschung Kulmbach*, 45, 173, 185–191;
- Prändl, O., Fischer, A., Schmidhofer, T., Sinell, H.-J. 1988.** *Fleisch – Technologie und Hygiene der Gewinnung und Verarbeitung*, Verlag Eugen Ulmer, Stuttgart;

- Rao, V. A., 2001.** The Prebiotic Properties of Oligofructose at Low Intake Levels, *Nutrition Res.*, 21, 843–848;
- Radovanović, R., Popov-Raljić Jovanka, 2001.** Senzorna analiza prehrambenih proizvoda, Poljoprivredni fakultet, Beograd, i Tehnološki fakultet, Novi Sad;
- Roberford, M., 2002.** Functional Food Concept and its Application to Prebiotics, *Digest Liver Dis.*, 34,105–110;
- Simopoulos A. P., Leaf A., Salem Jr. N., 2000.** Workshop Statement on the Essentiality of and Recommended Dietary Intakes for Omega-6 and Omega-3 Fatty Acids, Prostaglandins, Leukotrienes and Essential Fatty Acids, 63, 3, 119–121;
- Vasilev, D., Tubić, M., Saičić Snežana, Nonković Svetlana, Milanović-Stevanović Mirjana, Vuković, I. 2007.** Važniji parametri kvaliteta fermentisanih polusuvih kobasica proizvedenih sa masnim tkivom i biljnim mastima, Međunarodno 54. savetovanje industrije mesa, Vrnjačka Banja, 18–20. 06. 2007, Zbornik kratkih sadržaja, 60–61;
- Vuković, I. 2006.** Osnove tehnologije mesa, treće izdanje, Veterinarska komora Srbije, Beograd;
- Vuković, I., Vasilev, D., Vasiljević Nadja, 2007.** Fermentisane kobasice kao funkcionalna hrana, I Međunarodni kongres: Tehnologija, kvalitet i bezbednost hrane, Novi Sad, 13.–15. novembar, 2007, Zbornik radova, 119–123.

Rad primljen: 8.05.2009.

NAPOMENA

Rad iz realizovan u okviru istraživačkog projekta broj TR-20073, finasiran sredstvima Ministarstva za nauku i tehnološki razvoj Republike Srbije

IZJAVA ZAHVALNOSTI:

Autori izražavaju zahvalnost Kompaniji Big-Bull, Klanici i preradi mesa, Bačinci, na pomoći u realizaciji eksperimentalne proizvodnje funkcionalnih fermentisanih kobasica, i mr Vladimiru Tomoviću, asistentu Tehnološkog fakulteta Univerziteta u Novom Sadu, na pomoći prilikom instrumentalnog ispitivanja boje i konzistencije fermentisanih kobasica.

INTEGRATED MONITORING OF ZOOBOTIC FOODBORNE PATHOGENS IN THE MEAT CHAIN*

Nastasijević I.

A b s t r a c t: Zoonoses are diseases or infections, which are transmissible from animals to humans. These diseases can be acquired directly from animals but are most often acquired through ingestion of contaminated foods. The severity of these diseases in humans can vary from mild symptoms to life-threatening conditions. Although various foods can serve as sources of foodborne illness, meat and meat products are important sources of human infections with zoonotic foodborne pathogens: *Salmonella* spp., *Campylobacter jejuni/coli*, *Yersinia enterocolitica*, VTEC *E. coli* (including *E. coli* O157:H7) and, to some extent, *Listeria monocytogenes*. The most frequent chain of events leading to meat borne illness involves food animals as healthy carriers of the pathogens; these organisms are faecally excreted and subsequently transferred to humans through production, handling and consumption of meat and meat products. In order to prevent zoonoses from occurring, it is important to identify which animals and foodstuffs are the main sources of infections. Zoonoses Directive (2003/99/EC) covers the collection, evaluation and reporting data on: zoonoses, zoonotic agents, antimicrobial resistance, food-borne outbreaks and epidemiological investigation in the Member States of the EU. Zoonotic pathogens in foods, including meats, have to be controlled through a complete, continuous farm-to-fork system (i.e. Longitudinal and Integrated Safety Assurance – LISA) and should take into account not only the risk assessment, but also technical possibilities, consumers' attitude/behaviors, and cost-benefit analysis. This means that integrated concept for monitoring in all major phases along the meat chain should be implemented through modular approach: 1. Pre-harvest (on the farm), 2. Harvest (in abattoir), and 3. Post-harvest (meat processing-distribution-retail-consumer). This approach includes sampling, testing and reporting on pathogens' occurrences in those three main production modules. It is of utmost importance to control direct and indirect faecal contamination of carcasses, in abattoir, through efficient GHP/GMP and HACCP based process hygiene management systems.

Key words: zoonoses, contaminated foods, zoonotic foodborne pathogens, antimicrobial resistance, modular approach

Integrirani monitoring zoonotskih alimentarnih patogena u lancu mesa*

S a d r ŷ a j: Zoonoze su oboljenja ili infekcije koje su prenosive sa životinja na ljude. Ove bolesti mogu da nastanu direktno preko životinja, ali su najčešće stečene ingestijom kontaminirane hrane. Težina ovih oboljenja kod ljudi može da varira od blagih simptoma do stanja koja ugrožavaju život. Iako različita hrana može da bude izvor alimentarnih oboljenja, meso i proizvodi od mesa predstavljaju važne izvore infekcija ljudi, sa zoonotskim alimentarnim patogenima: *Salmonella* spp., *Campylobacter jejuni/coli*, *Yersinia enterocolitica*, VTEC *E. coli* (uključujući *E. coli* O157:H7) i, do određenog stepena, *Listeria monocytogenes*. Najčešći sled događaja koji dovodi do alimentarnih oboljenja preko mesa, uključuje zdrave životinje koje se koriste za proizvodnju hrane, kao nosioce patogena; ovi mikroorganizmi se fekalno izlučuju i posledično doprevaju do ljudi u toku proizvodnje, rukovanja i konzumiranja mesa i proizvoda od mesa. Radi sprečavanja nastajanja zoonotskih oboljenja, važno je da se identifikuju životinje i hrana koji predstavljaju glavne izvore infekcije. Direktiva o zoonozama (2003/99/EC) pokriva prikupljanje, ocenjivanje i izveštavanje o: zoonozama, zoonotskim agensima, antimikrobnoj rezistenciji, alimentarnim oboljenjima i epidemiološkim istragama u zemljama članicama EU. Zoonotski patogeni u hrani, uključujući meso, treba da budu kontrolisani preko kompletnog, kontinuiranog sistema od farme do trpeze (tj. Longitudinalno i integrirano osiguranje bezbednosti – LISA), pri čemu treba da se uzme u obzir ne samo ocena rizika, već takođe tehničke mogućnosti, stav/ponašanje potrošača i ekonomska opravdanost. To znači da koncept integriranog monitoringa u svim glavnim fazama duž lanca mesa treba da bude primenjen kroz modularni pristup: 1. farma (pre-harvest), 2. klanica (harvest), i 3. prerada mesa – distribucija – maloprodaja – potrošač (post-harvest). Ovakav pristup uključuje uzorkovanje, testiranje i izveštavanje o učestalosti patogena u ova, tri glavna proizvodna modula. Od najvećeg je značaja kontrolisanje direktne i indirektno fekalne kontaminacije trupova, u klanici, kroz efikasnu primenu GHP/GMP i HACCP – baziranih menadžment sistema za procesnu higijenu.

Ključne reči: zoonoze, kontaminirana hrana, zoonotski alimentarni patogeni, antimikrobna rezistencija, modularni pristup.

Introduction

Zoonoses are diseases or infections, which are transmissible from animals to humans. These diseases can be acquired directly from animals but are most

often acquired through ingestion of contaminated foods. The severity of these diseases in humans can vary from mild symptoms to life-threatening conditions. Zoonotic agents reportedly affected over 368, 000 persons in the EU in 2007 (Figure 1).

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTHOR: Ivan Nastasijević, ivann@inmesbgd.com, Institut of Meat Hygiene and Technology, Kacanskog 13, Belgrade Serbia.

AUTOR: Ivan Nastasijević, ivann@inmesbgd.com, Institut za higijenu i tehnologiju mesa, Kačanskog 13, Beograd, Srbija.

Although various foods can serve as sources of foodborne illness, meat and meat products are important sources of human infections with *Salmonella* spp., *Campylobacter jejuni/coli*, *Yersinia enterocolitica*, VTEC *E. coli* (including *E. coli* O157:H7) and, to some extent, *Listeria monocytogenes*. All these foodborne pathogens can be harbored in the gastrointestinal tract of food-producing animals. The most frequent chain of events leading to meat borne illness involves food animals as healthy carriers of the pathogens; these organisms are faecally excreted and subsequently transferred to humans through production, handling and consumption of meat and meat products. Occurrences of *Salmonella* spp., *C. jejuni/coli*, *Y. enterocolitica*, VTEC *E. coli* and *L. monocytogenes* in fresh red meat are variable, although most often are between 1% and 10%, depending on a range of factors including the organism, geographical factors, farming and/or meat production practices (Norrung and Buncic, 2008).

Zoonotic pathogens in foods, including meats, have to be controlled through a complete, continuous farm-to-fork system (i.e. Longitudinal and Integrated Safety Assurance – LISA) and should take into account not only the risk assessment, but also technical possibilities, consumers' attitude/behaviors, and cost-benefit analysis.

However, some aspects of the control system are pathogen-specific. Thus some pathogens in meats (e.g. *Salmonella* spp., *Campylobacter* spp., *Y. enterocolitica* and VTEC *E. coli*) are most efficiently controlled by the main interventions applied in the primary production, combined with optimization of the slaughter hygiene. For some others, such as more

environmentally ubiquitous *L. monocytogenes*, the main control measures are focused on later stages of the meat chain (Norrung and Buncic, 2008).

In order to prevent zoonoses from occurring, it is important to identify which animals and foodstuffs are the main sources of infections. For this purpose and to follow the developments on food safety in the European Union, information aimed at protecting human health is collected and analysed from all European Union Member States. Directive 2003/99/EC on the monitoring of zoonoses and zoonotic agents (Zoonoses Directive) covers the epidemiological investigation and reporting of food-borne outbreaks in the Member States (MSs) of the European Union (EU). Each MS has the obligation to collect relevant and, where applicable, comparable data of zoonoses, zoonotic agents, antimicrobial resistance and food-borne outbreaks. Thorough investigation of foodborne outbreaks aims to identify: 1. the pathogen, 2. the food vehicle involved, and 3. the factors in the food preparation and handling, contributing to the outbreak.

The data collection may allow the identification of emerging trends in the causative agents, and vehicles. Data regarding food-borne outbreaks provides important information on the number of humans affected annually and complements the picture of the burden of food-borne disease given by the total number of cases of disease in the Community. The added value concerns especially the information on the causative agent-food vehicle combinations responsible for the food-borne outbreaks. This information is necessary when targeting actions to improve food safety (EFSA, 2009a).

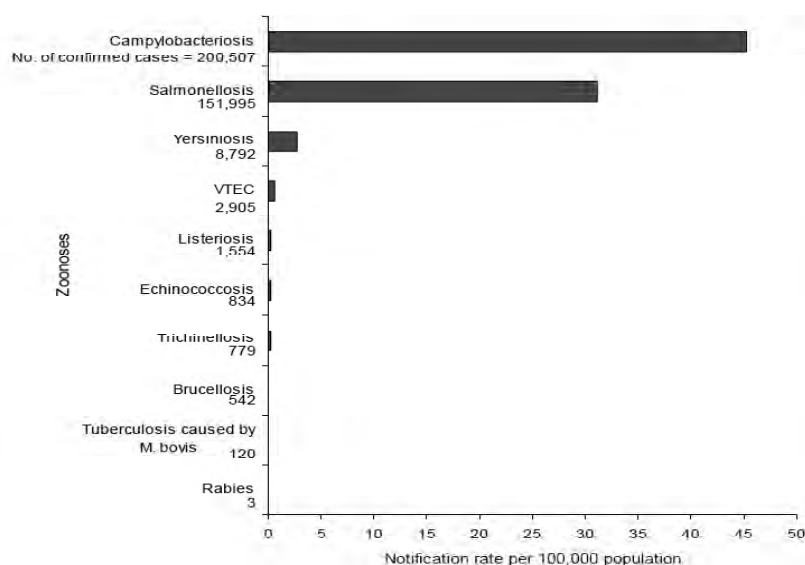


Figure 1. The reported notification zoonoses rates in confirmed human cases in the EU, 2007 (Adapted from EFSA, 2009b)

Slika 1. Incidenca prijavljenih zoonoza kod potvrđenih slučajeva u ljudi, na nivou EU, 2007 (preuzeto iz EFSA, 2009b)

2. Materials and methods

The present paper is not an detailed review of microbial zoonotic foodborne pathogens along the meat chain, but rather gives an overview of the main microbial meatborne risks, aspects of their control, and system of integrated monitoring (*Campylobacter* spp., *Salmonella* spp., *Yersinia enterocolitica*, VTEC *E. coli* and *Listeria monocytogenes*). Therefore, for the purposes of better understanding and explanation of the monitoring/surveillance and reporting system of microbial zoonotic foodborne pathogens, as well as, their control, reporting system and subsequent epidemiological investigation, the related documents issued by EFSA (European Food Safety Authority) and DG SANCO (EU Commission, Directorate General Health and Consumer Protection) have been used (Manual for Reporting of Food-borne Outbreaks in the framework of Directive 2003/99/EC; EFSA, 2009a, The Community Summary Report on Trends and Sources of Zoonoses and Zoonotic Agents in the European Union in 2007; EFSA, 2009b); as well as, the other relevant documents.

3. Main microbial meatborne infections in Europe

In 2007, campylobacteriosis was again the most frequently reported zoonotic disease in humans in the European Union, with 200,507 reported confirmed cases; most Member States (MSs) reporting an increased number of cases. Salmonellosis was still the second most commonly recorded zoonosis accounting for 151,995 confirmed human cases. However, the incidence of salmonellosis continues to decrease in the European Union with a statistically significant trend over the last four years.

3.1. *Campylobacter* spp.

Humans. In total, 200,507 confirmed cases of campylobacteriosis were reported by 24 MSs, which was a 14.2% increase compared to 2006. Children under the age of five had the highest notification rate (120 cases per population of 100,000). Other age groups varied between circa 32 to 53 cases per population of 100,000 (Figure 2, Figure 3).

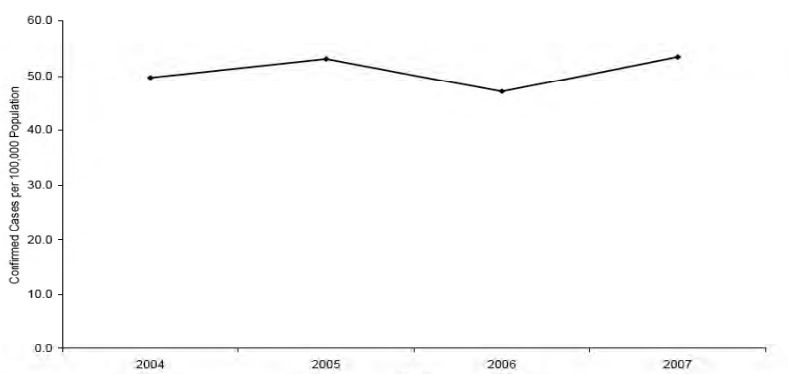


Figure 2. Notification rates of reported confirmed cases of human campylobacteriosis in the EU, 2004-2007 (Adapted from EFSA, 2009b)

Slika 2. Nivoi prijavljenih i potvrđenih slučajeva humanih kampilobakterioza u EU, 2004-2007 (preuzeto iz EFSA, 2009b)

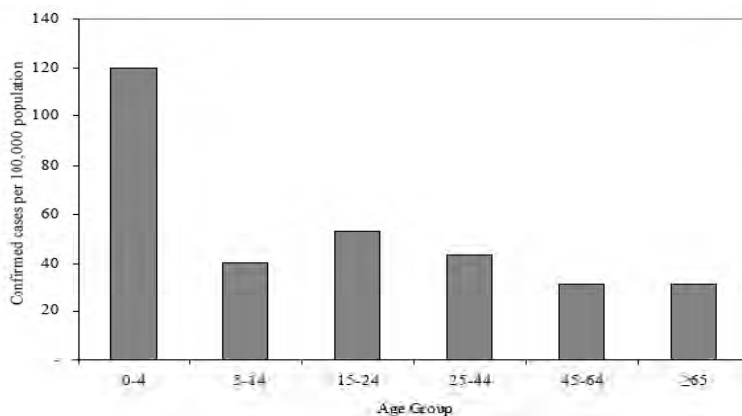


Figure 3. Age-specific distribution of reported confirmed cases of human campylobacteriosis (TESSy, 2007)

Slika 3. Distribucija prijavljenih i potvrđenih slučajeva humanih kampilobakterioza prema starosnoj kategoriji (TESSy, 2007)

Foodstuffs. Broiler meat was the most frequently sampled food category in 2007 and the reported occurrence of *Campylobacter* was generally at the same high level as in previous years. On average, 26.0% of fresh broiler meat samples tested *Campylobacter* positive at EU level and findings ranged from 0% to 86.5%. In samples of pig meat and bovine meat, *Campylobacter* was detected less frequently: 0.9% and 1.2% of the samples, respectively. Poultry meat appears still to be the most important food-borne source of *Campylobacter* as the occurrence of the bacteria remained at high levels throughout the food chain, from live animals to meat retail level (Figure 4).

Animals. In 2007, as in previous years, the majority of data on *Campylobacter* in animals was from investigations of broilers, but data from pigs and cattle was also reported. The recorded prevalence of *Campylobacter* positive broiler flocks was generally high: 25.2% at EU level ranging from 0% to 82.8% in MSs. High prevalence was also observed from the monitoring of pigs, 56.1% at EU level (ranging from 0.9% to 78.5%). In cattle, reported occurrences were somewhat lower, 5.9% on average in the EU, but prevalence up to 70.5% was reported by some MSs. However, *Campylobacter* contamination rates in pig and bovine meat typically decrease sharply following slaughter and remain low at retail (Figure 5).

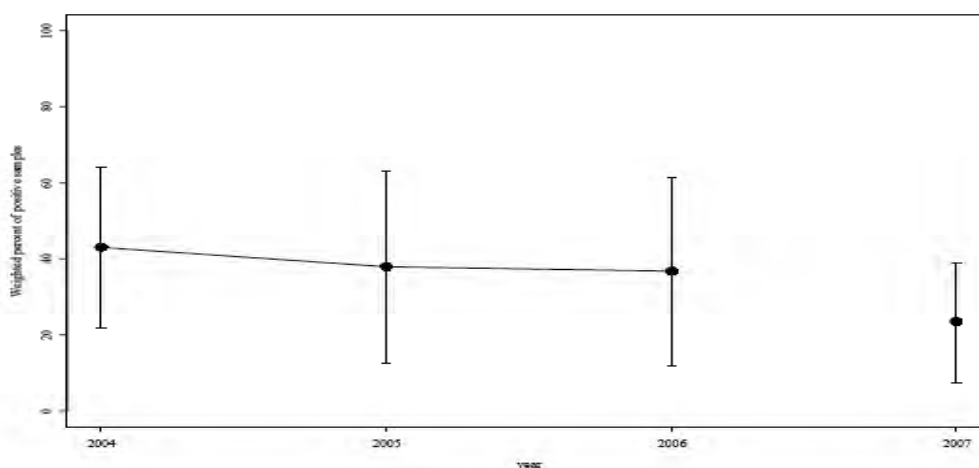


Figure 4. *Campylobacter* in fresh broiler meat* (Adapted from EFSA, 2009b)

*Combined data (samples taken at slaughter, at processing/cutting plant or at retail)

Slika 4. *Campylobacter* u svežem živinskom mesu (preuzeto iz EFSA, 2009b)

*Kombinovani podaci (uzorci uzeti na klanju, u pogonu za rasecanje ili u maloprodaji)

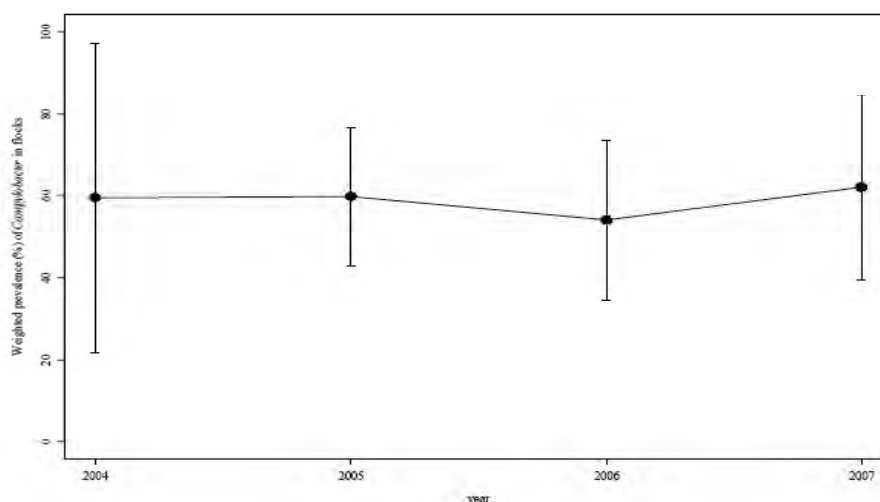


Figure 5. *Campylobacter* in broiler flocks (Adapted from EFSA, 2009b)

Slika 5. *Campylobacter* u jatima živine (preuzeto iz EFSA, 2009b)

3.2. *Salmonella* spp.

Humans. In 2007, a total of 151,995 confirmed cases of human salmonellosis (TESSy) were reported in the EU. The EU incidence rate was 31.1 cases per population of 100,000, ranging from 2.9 to 171.6 confirmed cases. In 2007, there was a 7.3% decrease comparing with 2006 and this was part of a significant, decreasing trend over the past four years. As in previous years, *S. Enteritidis* and *S. Typhimurium* were the most frequently reported serovars (81% of all known serovars in human cases) (Figure 6). The highest notification rate for human cases was for age groups 0 to 4 years and 5 to 14 years. A seasonal peak in the number of cases during the late summer and autumn was generally observed in all MSs and *S. Enteritidis* demonstrates a much more prominent peak than the other serovars.

Foodstuffs. Reported *Salmonella* findings were most frequently from investigations of poultry meat, followed by those of pig meat. The highest proportions of positive samples were also observed in investigations of these food categories. The overall proportion of positive samples in fresh broiler

meat was 5.5%, at EU level, varying between 0% and 55.6%. 1.1% of fresh pig meat samples were on average found *Salmonella* positive in the EU, ranging from 0% to 19.4% (this data is strongly influenced by the high numbers of samples reported by the Nordic MSs that have low prevalence). In bovine meat, most MSs reported very low (<1.0%) proportions of positive samples (Figure 7, Figure 8). Overall, 0.8% (range 0% to 5.8%) of tested egg units were found positive, which is the same level as in 2006 (0.8%). However, in general, the level of samples in noncompliance with the *Salmonella* criteria in 2007 was comparable to the findings in 2006.

Animals. 2007 was the first year when the new *Salmonella* control programmes in breeding flocks of *Gallus gallus* were implemented on a mandatory basis (Regulation (EC) No 2160/2003). The aim of the programmes is to meet the *Salmonella* reduction target set down by the Regulation (EC) No 1003/2005. The target states that the occurrence of *S. Enteritidis*, *S. Hadar*, *S. Infantis*, *S. Typhimurium* and *S. Virchow* should be reduced to 1% or less in adult breeding flocks comprising at least 250 birds by 31 December 2009. 15 MSs reported in 2007 a prevalence of

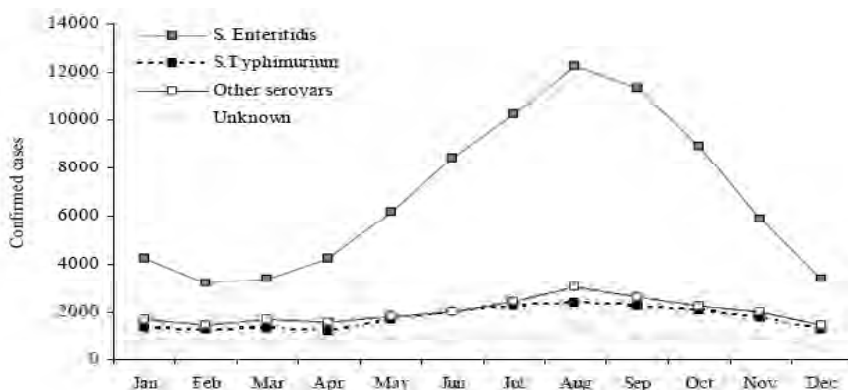


Figure 6. Number of reported confirmed salmonellosis cases in humans by month and serovar (Adapted from EFSA 2009b; TESSy, 2007)

Slika 6. Broj prijavljenih i potvrđenih slučajeva salmoneloza kod ljudi, prema mesecu i serovaru (preuzeto iz EFSA 2009b; TESSy, 2007)

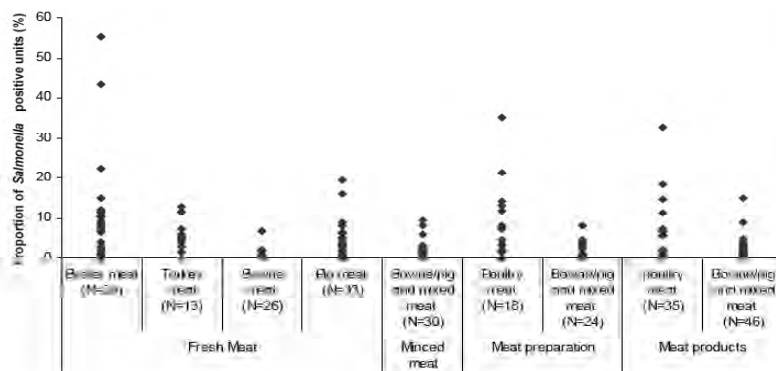


Figure 7. Proportions of *Salmonella* positive units, by meat category (Adapted from EFSA, 2009b)

Slika 7. Proporcija *Salmonella* pozitivnih proizvodnih jedinica, prema kategoriji mesa (preuzeto iz EFSA, 2009b)

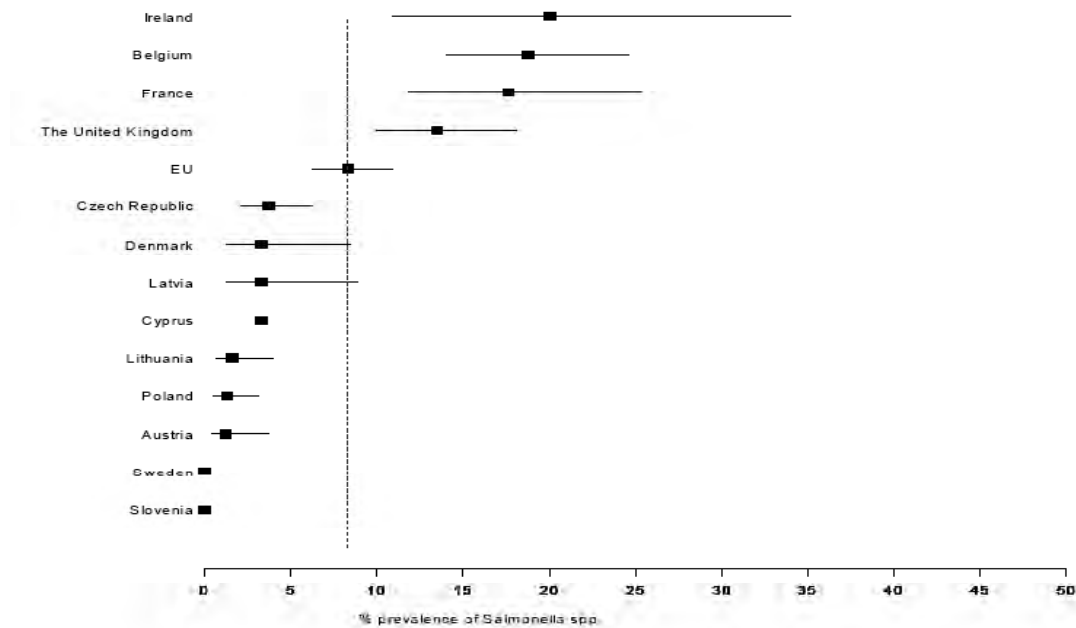


Figure 8. Observed prevalence of carcasses contaminated with *Salmonella* spp., baseline survey 2006-2007 (Adapted from EFSA, 2009b)

Slika 8. Prevalenca kontaminiranih trupova sa *Salmonella* spp., osnovno istraživanje 2006-2007 (preuzeto iz EFSA, 2009b)

these five target serovars that was lower than the target, whereas eight MSs reported prevalence of the five serovars ranging from 1.1% to 15.4%. A total of 4.3% (ranging between 0% and 27.1%) of the tested laying hen flocks were found infected during 2007, an overall occurrence slightly higher than in the two previous years. An EU-wide *Salmonella* baseline survey was carried out in slaughter pigs in 2006 to

2007 (*S. Typhimurium*). In total, 19,071 ileo-caecal lymph node samples were collected from slaughtered pigs and the EU weighted mean prevalence in pigs was 10.3% ranging between 0% and 29.0% in MSs. Few MSs have active monitoring of *Salmonella* in cattle, but two MSs both reported slaughter prevalence of 0.1% in cattle (*S. Typhimurium*, *Salmonella Dublin*) (Figure 9).

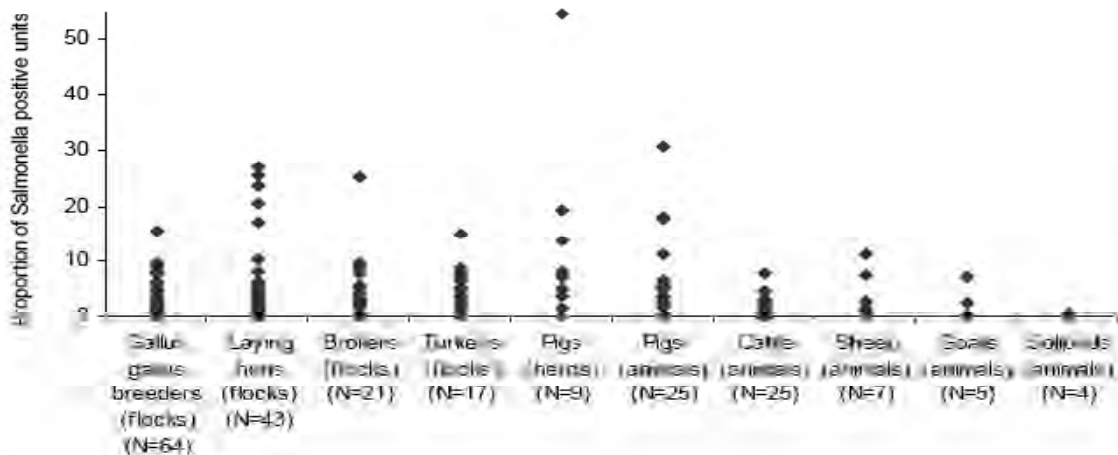


Figure 9. Reported *Salmonella* prevalence by animal species within the EU, in 2007* (Adapted from EFSA, 2009b)

*Data are only presented from sample size ≥ 25 . Results from HACCP and baseline surveys are excluded, as well as data based on suspicion or trace-back sampling.

Slika 9. Prevalenca *Salmonella*, prema vrstama životinja na nivou EU, u 2007* (preuzeto iz EFSA, 2009b)

*Podaci su predstavljeni samo za veličinu uzorka ≥ 25 . Rezultati iz HACCP-a i osnovnih studija nisu prikazani, kao i podaci o suspektom ili retroaktivnom uzorkovanju.

3.3. *Yersinia enterocolitica*

Humans. In 2007, 8,792 confirmed human cases of yersiniosis were reported in the EU. **Foodstuffs.** Findings of *Y. enterocolitica* were reported on average in 2.0% of pig meat samples. **Animals.** Findings of *Y. enterocolitica* were reported in 0% to 52% of pigs.

3.4. VTEC *E. coli*

Humans. In 2007, a total of 2,905 confirmed human VTEC cases were reported from 23 MSs. This is a slight decrease compared to 2006. The EU incidence rate was 0.6 per population of 100,000. The most

commonly identified VTEC serogroup was O157 (54%), although other serogroups were detected (i.e. O26, O103, O91, O145, O111, O128, O113, O146) (Figure 10). The notification rate was highest in 0 to 4 year old children and this group also accounted for almost 60% of the 103 HUS cases reported, mainly associated with VTEC O157 infections (Table 1).

Foodstuffs. The reported occurrence of VTEC bacteria in food was generally low, and has been relatively constant during the 2005 to 2007 period. In fresh bovine meat the proportion of samples positive for VTEC was 0.3% at EU level and 0.1% for the serogroup VTEC O157. Some MSs also reported, from bovine meat, the O26, O103, O111, and O113

Table 1. VTEC serogroups by country (TESSy, 2007)

Tabela 1. VTEC serogrupe po zemljama (TESSy, 2007)

Country	Serogroup										
	O157	NT	O26	O103	O91	O145	O111	O128	O113	O146	Other
Austria	17	41	1	3	2	7	2			2	7
Belgium	25	3	5	2	1	2	2		1	2	4
Denmark	25	1	28	16	9	5	4	8	5	8	47
Estonia	2							1			
Finland	9	3									
France	14	29	10		1		1	1			1
Germany	66	577	61	46	26	13	12	9	8	1	51
Hungary	1										
Ireland	94	5	13			1	1	1			
Italy	5	20	1				1				
Luxembourg	1										
Malta	4			3							
Netherlands	80	1	3	1	1						
Poland	2			6							
Slovakia	3	3									
Slovenia											4
Spain	18										
Sweden	85	138	13		3	1		1	2	1	12
United Kingdom	1,120	91	1			3					3
Total (19 MSs)	1,571	842	136	77	43	31	22	21	16	14	136
Iceland	13										
Norway	5	7	3	1		4		2		1	3

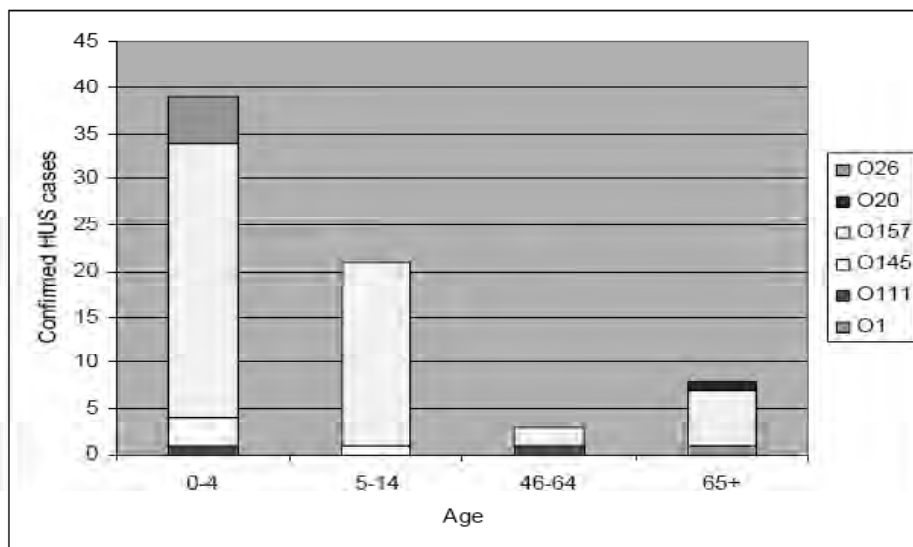


Figure 10. Haemolytic Uremic Syndrome (HUS) by age and serogroup (Adapted from EFSA, 2009b)
Slika 10. Hemolitički Uremički Sindrom (HUS) po godinama i serogrupama (preuzeto iz EFSA, 2009b)

serogroups that are all frequently isolated from human VTEC cases. (Figure 11).

Animals. In bovine animals the average VTEC prevalence was 3.6% and the proportion of VTEC O157 positive animals was 2.9%. The reported occurrence of VTEC ranged from 0% to 22.1%.

3.5. *Listeria monocytogenes*

Humans. A total of 1,554 confirmed cases of listeriosis were reported in 2007. The EU incidence

rate was 0.3 per population of 100,000. The highest notification rates were observed in Scandinavian countries. The number of confirmed cases of listeriosis almost reached the same level as in 2006. Listeriosis mainly occurred among elderly people, with 53.1% of cases (notification rate was 1.0 per population of 100,000) occurring in individuals over the age of 65. The notification rate among children under the age of five was 0.5 cases per population of 100,000. The case fatality rate for human listeriosis was 20% (mainly in elderly people) (Figure 12).

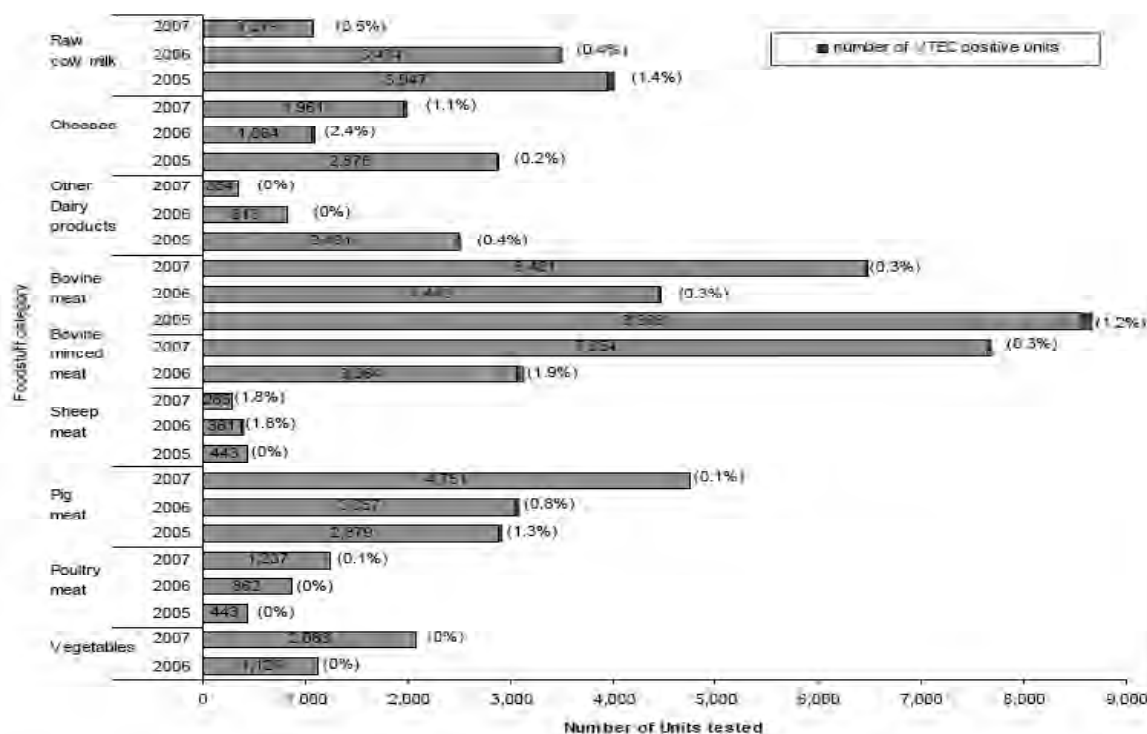


Figure 11. Number of food samples tested for VTEC by food category and number of VTEC positive units, 2005-2007 (Adapted from EFSA, 2009b)

Slika 11. Broj uzoraka hrane testiranih na VTEC prema kategoriji hrane i broju VTEC pozitivnih proizvodnih jedinica (preuzeto iz EFSA, 2009b)

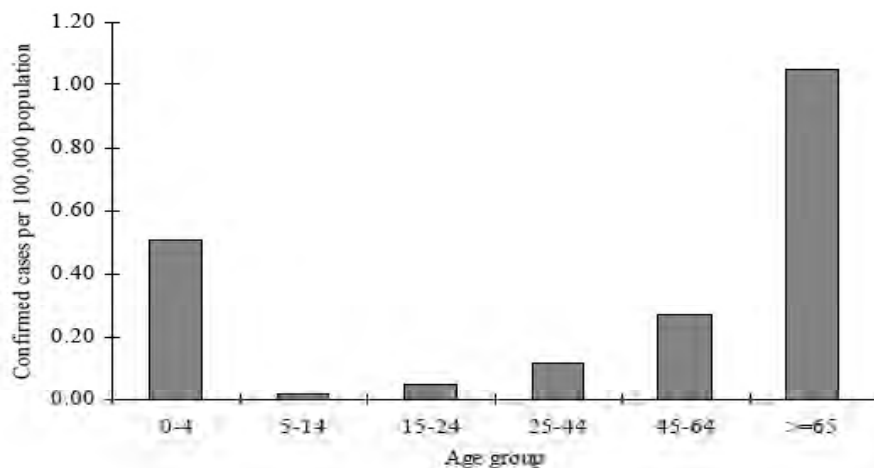


Figure 12. Age-specific distribution of reported confirmed cases of human listeriosis (TESSy, 2007)

Slika 12. Distribucija prijavljenih i potvrđenih slučajeva listerioza kod ljudi, prema starosnoj kategoriji (TESSy, 2007)

Foodstuffs. In 2007, a large number of investigations concerning ready-to-eat (RTE) foodstuffs were reported by MSs. The food categories most often covered were RTE meat products, dairy products, cheeses and fishery products (Figure 13). In general, *L. monocytogenes* was rarely detected in quantities exceeding the legal safety limit of 100 cfu/g (Regulation 2073/2005/EC). The proportion of the samples in non-compliance with the criterion was most often observed at retail in fishery products (1.7% and 2.2% for single products and batches, respectively), particularly in smoked fish, followed by meat products (0.3% and 0.7%).

on the EFSA zoonoses reporting homepage: (www.efsa.europa.eu/zoonoses).

For each reporting year, a national report is created in the web-based reporting system. For each zoonoses or other subject, text forms and reporting tables are provided. The text forms are used to enter the narrative part of the report, e.g. description of the monitoring system and the analyses of the results. The reporting tables are used to enter the results, e.g. number of samples and number of positive results.

The national report on zoonoses, antimicrobial resistance and foodborne outbreaks is divided into three sections:

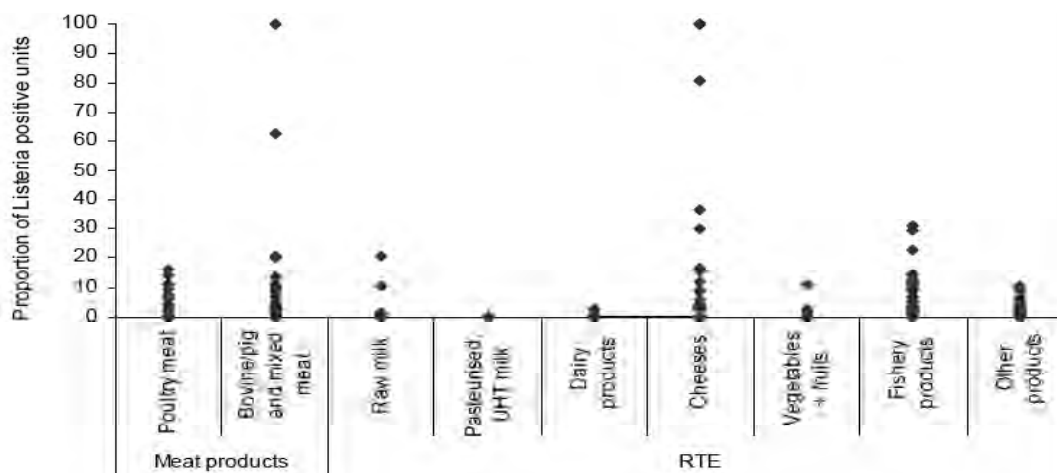


Figure 13. Proportions of *Listeria* positive samples by ready-to-eat food category (Adapted from EFSA, 2009b)

Slika 13. Proporcija *Listeria* pozitivnih uzoraka kod proizvoda spremnih za konzumiranje (preuzeto iz EFSA, 2009b)

Animals. In 2007, data on *L. monocytogenes* in animals and the bacterium was reported from various animal species. In some MSs the detected proportion of positive samples reached a moderate level in cattle and in small ruminants.

4. Structure of the web-based integrated monitoring system

The European Community (EC) system for monitoring and collection of information on zoonoses is established by Directive 2003/99/EC on the monitoring of zoonoses and zoonotic agents. This Directive requires Member States (MS) to collect, evaluate and report data on: zoonoses, zoonotic agents, antimicrobial resistance and food-borne outbreaks to the European Commission each year. Monitoring on zoonoses, antimicrobial resistance and foodborne outbreaks is web-based and accessible

1. Description of the national reporting system and national evaluation of the reported food-borne outbreaks;
2. Total number of food-borne outbreaks;
3. Data to be reported for verified food-borne outbreaks.

4.1. Relevant outbreaks and causative agents to be reported

The annual reporting system covers the results of the investigations of all food-borne outbreaks carried out in MSs. "Food-borne outbreak" is defined in the Zoonoses Directive "as an incidence, observed under given circumstances, of two or more human cases of the same disease and/or infection, or a situation in which the observed number of human cases exceeds the expected number and where the cases are linked, or are probably linked, to the same food source" (Directive 2003/99/EC).

For the purpose of the reporting system, this is understood to include food-borne outbreaks caused by any virus, bacterium, alga, fungus, parasite, and its products, such as toxins and biological amines (e.g. histamine). Reporting should not be limited to foodborne outbreaks caused by zoonotic agents only, but should include food-borne outbreaks caused by any of the agents above. Outbreaks caused by ingestion of drinking water are also considered food-borne (Regulation 178/2002), while food-borne outbreaks caused by chemical agents are not covered at this stage.

4.2. Mandatory reporting

In accordance with the Zoonoses Directive 2003/99/EC, all MS have to report on the following zoonoses, zoonotic agents (list A) and other subjects:

- Brucellosis and agents thereof;
- Campylobacteriosis and agents thereof;
- Echinococcosis and agents thereof;
- Listeriosis and agents thereof;
- Salmonellosis and agents thereof;
- Trichinellosis and agents thereof;
- Tuberculosis due to *Mycobacterium bovis*;
- Verotoxigenic *Escherichia coli*;
- Antimicrobial resistance in *Salmonella* and *Campylobacter* isolates from poultry, pigs and cattle and foodstuffs derived from these species;
- Food-borne outbreaks;
- Susceptible animal populations.

4.3. Reporting based on epidemiological situation

Other zoonoses are to be included in the monitoring and reporting according to the epidemiological situation in each MS. This means that if a certain zoonosis is of public health importance in a MS, this MS should report on that zoonosis, but the other MSs do not have the same obligation to report on it.

The zoonoses to be reported based on the epidemiological situation are listed in Directive 2003/99/EC (list B):

Viral zoonoses:

- Calicivirus;
- Hepatitis A virus;
- Influenza virus;
- Rabies;
- Viruses transmitted by arthropods.

Bacterial zoonoses:

- Borreliosis and agents thereof;
- Botulism and agents thereof;

- Leptospirosis and agents thereof;
- Psittacosis and agents thereof;
- Tuberculosis other than in point A;
- Vibriosis and agents thereof;
- Yersiniosis and agents thereof.

Parasitic zoonoses:

- Anisakiasis and agents thereof;
- Cryptosporidiosis and agents thereof;
- Cysticercosis and agents thereof;
- Toxoplasmosis and agents thereof.

Other zoonoses and zoonotic agents

Other non-zoonotic pathogenic microbiological and toxicological agents in foodstuffs (e.g. *Enterobacter sakazakii*, staphylococcal enterotoxins and histamine).

4.4. Monitoring system for zoonotic foodborne pathogens in the meat chain

Sampling strategy. The framework of the sampling is an important part of the strategy, and it should be stated if the sampling is part of a permanent or temporary monitoring programme, linked to surveillance or control programmes or if it is a question of a single survey, e.g. the sampling strategy chosen and the purpose of the sampling: whole country covered or only part of it; target population (entire or subset of animal population, categories of foodstuffs and feedingstuffs); geographical regions; size of the holdings; sampling protocol (objective, selective, suspected, convenient or census sampling); who is performing the sampling (competent authority – official sampling, by owners of animals, food or feed businesses in the context of HACCP / own-checks); where the samples are taken (at farm, at slaughterhouse, at hatchery, at food processing plant or at retail); stage of sampling (animal rearing period, production period, before or after a chilling of carcass in the slaughterhouses, before or after expiration of the shelf-life of foodstuffs).

Frequency of the sampling. This part is intended to explain how often samples are taken. The standard terms (e.g. every week, once a month, x times a year)

Type of specimen taken. The specimen taken from the units sampled is described (Table 2):

Animal species – cattle, pigs, broilers (specimens: faeces, blood, organs or milk)

Foodstuffs – beef, pork, poultry meat

Stage in the meat chain – preharvest (on the farm), harvest (slaughter), postharvest (processing/distribution/retail)

Methods of sampling. This should include information on the site of sampling (e.g. part of a car-

Table 2. Description of the sampling strategy for monitoring of *Salmonella* spp. in the meat chain (EFSA, 2009a; adapted by *Nastasijevic I.*)

Tabela 2. Opis strategije uzorkovanja za monitoring *Salmonella* spp. u lancu mesa (EFSA, 2009a; preuzeo i modificovao *Nastasijević I.*)

<i>Salmonella</i> spp.
The sampling strategy
<i>The control, surveillance and monitoring programmes</i>
<i>Who performs the sampling: competent authority (official sampling) or industry (own checks)</i>
<i>The type of sampling i.e. objective, selective or suspect</i>
<i>The place or stage at which the sample was taken (e.g. farm, slaughterhouse, processing plants, retail, border inspection posts)</i>
Type of specimen taken
<i>Meat and meat products</i>
<i>Animal species: broiler, bovine and pig meat, duck meat</i>
<i>Pre-harvest phase: faeces, environmental surfaces, animal waste, animal` hides, etc.</i>
<i>Harvest phase: carcass, fresh meat, trimming</i>
<i>Post-harvest phase: minced meat, meat preparations, meat products, retail</i>
<i>Status of meat: fresh/frozen/cooked</i>
<i>Intended to be consumed: raw or cooked</i>

case, part of the facilities for environmental sample), size of sample taken (e.g. in g, cm², ml), use of swabs or other instruments in the sampling, when relevant, the number of (sub)samples / sample units taken, pooling of samples when conducted (always refer the number of samples combined by pooling), the possible storage of samples, and the length of this storage.

Case definition / definition of a positive finding. This covers the description of when the sample is considered to be positive for the zoonotic agent or when the animal, herd or flock is considered to be infected with the zoonotic agent. Regarding food and feed, it should describe when the foodstuff, feedingstuff or the batch sampled is considered to be positive or contaminated with the zoonotic agent.

Diagnostic / analytical methods used. Under this title, the diagnostic or analytical methods used in the laboratory to test the specimens are described. Whenever possible, a reference to standard methods used is made (such as national, ISO or EN standard methods), or to the methods prescribed by the legislation.

Vaccination policy. This policy can cover different kinds of situations: vaccination of animal populations against the zoonotic agent may be prohibited or it may be mandatory or voluntary. There can be recommendations in place to vaccinate certain animal populations or to use a certain type of vaccination scheme. It may also be that there is no official policy regarding vaccination. If a vaccinati-

on policy exists, it should be described and if no policy exists, the established way of using the vaccines in the MS can be explained. The description should include, at least, a description of the vaccine, characteristics of the animals to be vaccinated (age, sex), area where vaccination is to be implemented, special measures for marking the vaccinated animals, etc.

Other preventive measures than vaccination in place. Other preventive measures may include actions taken at different levels of the food chain. Regarding animals, it may cover bio-security measures at the farms. For the foodstuffs, it may include recommendations on meat consumption for susceptible consumer groups.

4.5. Reporting on antimicrobial resistance

Trends on antimicrobial resistance. The information to be reported each year or at regular intervals (e.g. every 2. or 3. year).

4.5.1. Mandatory

Antimicrobial resistance on Salmonella spp.

Relevant animal species / food categories to be reported: Laying hens and broilers (*Gallus gallus*), turkeys, pigs and cattle, broiler meat, pig meat, bovine meat.

Relevant agent species / serovars to be reported:

In the qualitative antimicrobial susceptibility tables: *S. Enteritidis* and *S. Typhimurium* and the

next 5 most prevalent serovars in the country and the other serovars group together. In the quantitative antimicrobial susceptibility tables: *S. Enteritidis* and *S. Typhimurium* for poultry species and meat thereof; *S. Typhimurium* and *S. Derby* for pigs and pig meat, *S. Typhimurium* and *S. Dublin* for cattle and bovine meat, and other *Salmonella* serovars grouped together for all species.

Recommended antimicrobials to be reported: Ampicillin; Cefotaxime; Chloramphenicol; Ciprofloxacin; Gentamicin; Nalidixic acid; Streptomycin; Sulphonamides; Tetracycline; Trimethoprim.

Antimicrobial resistance on Campylobacter spp.

Relevant animal species / food categories to be reported: Broilers (*Gallus gallus*), turkeys, pigs, cattle, broiler meat, other poultry meat

Relevant agent species / serovars to be reported: *C. jejuni* and *C. coli* separately. Reporting of susceptibility data for *Campylobacter spp.* overall is discouraged because resistance patterns vary for different species.

Recommended antimicrobials to be reported: Erythromycin; Ciprofloxacin; Tetracycline; Streptomycin; Gentamicin.

4.5.2. Optionally

Antimicrobial resistance on E. coli (non-pathogenic).

Relevant animal species / food categories to be reported: Laying hen, broilers (*Gallus gallus*), turkeys, pigs, cattle, broiler, pig and bovine meat.

Recommended antimicrobials to be reported: Ampicillin; Cefotaxime; Chloramphenicol; Ciprofloxacin; Gentamicin; Nalidixic acid; Streptomycin; Sulphonamides; Tetracycline; Trimethoprim.

Antimicrobial resistance on Enterococcus spp.

Relevant animal species / food categories to be reported: Broilers (*Gallus gallus*), pigs, cattle, broiler meat, pig meat, bovine meat

Relevant agent species to be reported: *E. faecium* and *E. faecalis*, separately

Recommended antimicrobials to be reported: Aminoglycosides: streptomycin, gentamicin; Aminopenicillins: chloramphenicol; Beta-lactams or β -lactam inhibitors: ampicillin or amoxicillin; Glycopeptides: vancomycin; Macrolides: erythromycin; Streptogramins: preferably quinopristin/dalfopristin; Tetracyclines: tetracycline.

Diagnostic/analytical methods typically used. Three types of methods are used in antimicrobial

resistance testing for *Salmonella* and indicator bacteria: disk diffusion, agar dilution and broth dilution. For *Campylobacter*, only dilution methods are considered reproducible.

4.6. Control programmes/mechanisms

The control programmes / strategies in place.

Under this title, the control programmes in place are described. The control programmes may be national or regional, and they may be approved nationally or by the Commission and co-financed by the Community (Council Decision 90/424/EEC). The nature of the control programmes, e.g. voluntary, mandatory, national, regional, Community or national approval and co financing should be indicated.

Measures in case of the positive findings or single cases. Actions required by the legislation or control programmes as a consequence of findings of positive animals, foodstuffs or feedingstuffs are explained (e.g. withdrawal of the products from the market, destruction of animals and others).

Notification system in place. The notification system is described, including its legal basis and since when the disease or infection has been notifiable.

Recent actions taken to control the zoonoses. Specific measures undertaken during the recent years to control zoonoses, are described (Table 3).

4.7. Results of the investigation

National evaluation of the recent situation, the trends and sources of infection. The results are interpreted in relation to their importance to public health. It is essential to evaluate the trend when compared to the previous year, e.g. is there a decreasing or increasing trend or is the situation stabilized. The important sources of infections are also discussed.

Relevance of the findings in feedingstuffs / animals / foodstuffs and to human cases (as a source of infection). The importance of the feedingstuffs / animals / foodstuffs as sources of the human infections is evaluated. The role of feedingstuffs as a source of infection for animals, and similarly the role of animals as a source of contamination for foodstuffs are considered, as well.

History of the disease and / or infection. The history of the zoonoses cases in humans and animals in the past is reflected. For example, issues such as the number of cases in the past and the impact of control and eradication programmes can be addressed.

Additional information...

Table 3. Example of integrated monitoring and control programmes for VTEC *E. coli* in the meat chain (EFSA, 2009a; adapted by *Nastasijević I.*)**Tabela 3.** Primer integriranog monitoringa i programa za kontrolu VTEC *E. Coli* u lancu mesa (EFSA, 2009a; preuzeo i modifikovao *Nastasijević I.*)

Verotoxigenic <i>Escherichia coli</i> (VTEC) in foodstuffs
The sampling strategy
<i>The control, surveillance and monitoring programmes in place</i>
<i>Who performs the sampling: competent authority (official sampling) or industry (own checks)</i>
<i>The type of sampling i.e. objective, selective or suspect</i>
<i>The place or stage at which the sample was taken (e.g. farm, slaughterhouse, processing plants, retail, border inspection posts)</i>
Type of specimen taken
Meat and meat products
<i>Animal species: broiler, bovine, sheep, goat, game (ruminants)</i>
<i>Harvest phase: carcass, fresh meat, trimming</i>
<i>Post-harvest phase: minced meat, meat preparations, ready-to-eat fermented meat products, retail</i>
<i>Status of meat: fresh/frozen/cooked</i>
<i>Intended to be consumed: raw or cooked</i>
Relevant agent species / serovars / phage types to be reported:
<i>Strains of <i>E. coli</i> that are capable of producing vero- (shiga-) cytotoxin (i.e. VT+) and/or possess the genes coding for VT production.</i>
<i>Information on the serotype or the serogroup (O antigen) should be reported.</i>
<i>Serotypes of particular interest: O157 and non-O157, (e.g. O111, O103, O26, O145, O91).</i>
Case definition / definition of a positive sample
VTEC positive sample / batch – a sample / batch from which verotoxigenic <i>E. coli</i> has been isolated using a method specified below.
VTEC O157 or other serotype positive sample / batch - a sample / batch from which verotoxigenic <i>E. coli</i> O157 or other serotype has been isolated using a method specified below.
Diagnostic/analytical methods typically used
<i>The recommended method: EN/ISO ISO 16654 - molecular subtyping (PCR)</i>
<i>Currently, there is no internationally recognised standard method for detection of VTEC non-O157</i>
<i>Details should be provided on the diagnostic method used, including how verification of VTEC is carried out and the serotypes for which screening is carried out.</i>
<i>Other methods (the performance characteristics of the methods should be given in comparison to the EN/ISO or ISO standard reference methods or other reference methods- evidence of validation: ISO 16140:2003)</i>
Reporting the results in the tables
For reporting of data, use tables named:
<ul style="list-style-type: none"> • “VT <i>E. coli</i> in food”; Specific guidelines for reporting data in the prevalence table; • Sampling unit – “Single” or “Batch” should be used as the terms to be reported; • Total units positive for VTEC - the total number of units positive for Verotoxigenic <i>E. coli</i> (VTEC); • VTEC O157 and other serotypes – the number of units positive for the specific VTEC serotype; • VTEC, unspecified - the number of units positive for VTEC where the serotype is unknown.
Preventive and control measures in place
<i>National microbiological criteria or guidelines for foodstuffs</i>
<i>Provisions or recommendations concerning use of certain foodstuffs containing potentially hazardous agents</i>
<i>Special recommendations for susceptible populations of consumers</i>

5. Conclusions

There are many routes by which the zoonotic pathogens can reach consumers via meats including consumption of contaminated, uncooked or improperly cooked ready-to-eat (RTE) product and cross-contamination from raw to RTE foods. Better knowledge on the relative importance of these different routes is needed. For that, both epidemiological and microbiological approaches as well as risk assessments of specific pathogens in specific foods need to be applied. Such knowledge is important to tailor and optimise the risk management strategies and activities.

Therefore, zoonotic pathogens in meat have to be monitored and controlled through a complete, continuous farm-to-fork system. This means that integrated concept for monitoring in all major phases along the meat chain should be implemented through “modular approach”. Risk mitigation options were identified according to three lines of defence formulated by the World Health Organization (WHO): the first line focuses on the control of foodborne pathogens in the food producing animal (Pre-harvest control / on the farm), the second line deals with improvement of hygiene during slaughter and further processing of meat (Harvest control / in abattoir) and the third line concentrates on measures during the final preparation of the food and the education of the industry and the consumer concerning the application of effective hygienic measures (Post-harvest control / meat processing-distribution-retail-consumer) (WHO, 1980; EFSA,

2006). This approach includes sampling, testing and reporting on pathogens' occurrences in those three main production modules. In addition, it is of utmost importance to control direct and indirect faecal contamination of carcasses, in abattoir, through efficient GHP/GMP and HACCP based process hygiene management systems.

In Serbia, the integrated system for monitoring of zoonotic foodborne pathogens in the food (meat) chain should be developed and implemented in the foreseeable future, in the scope of necessary harmonization with Zoonoses Directive 2003/99/EC. The implementation of harmonized survey methods is needed. This will include targeted research to obtain top quality baseline data on occurrences of different pathogens along the meat chain and the characteristics on their antimicrobial resistance. Further on, proper science-based risk assessment of consumer exposure to related pathogens can be only achieved by effective intersectoral cooperation between veterinary and health authorities. This includes: 1. assessment of the monitoring systems for foodborne hazards in the food (meat) chain (veterinary authorities); 2. assessment of surveillance systems for foodborne diseases (health authorities); and, 3. assessment the interface, between the monitoring systems for foodborne hazards in the food (meat) chain and the surveillance systems for foodborne diseases (Figure 14). Finally, the ultimate objective regarding adequate level of public health protection, can be only achieved by effective professional integration of veterinary and health authorities, setting up all related activities to the integrated health concept.

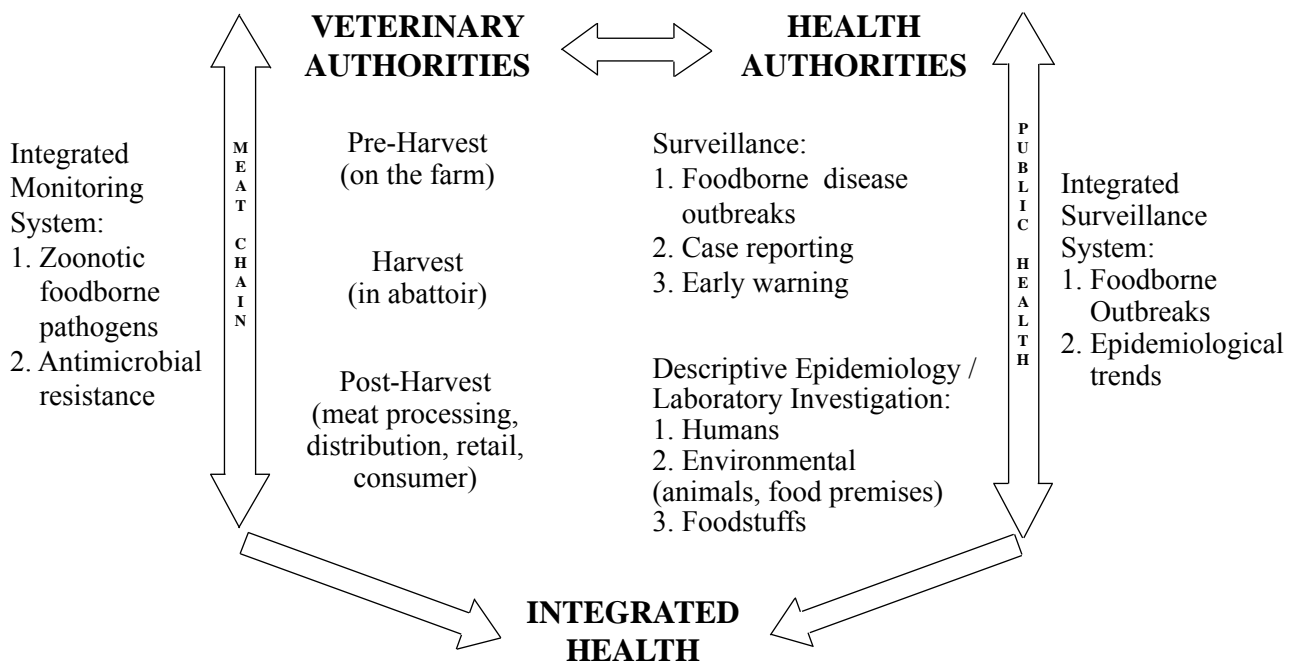


Figure 14. Integrated Health Concept (schematic overview, by Nastasijević, I.)
Slika 14. Koncept integrisanog zdravlja (šematski prikaz, prema Nastasijević, I.)

References

- Outbreaks in the Framework of Directive 2003/99/EC. The EFSA Journal, 257, 1–46;
- Anonymous, 2009 b.** The Community Summary Report on Trends and Sources of Zoonoses and Zoonotic Agents in the European Union in 2007. The EFSA Journal, 223, 1–217;
- Anonymous, 2006.** Risk Assessment and Risk Mitigation Options of *Salmonella* in pig production. The EFSA Journal, 341, 1–131;
- Commission Regulation (EC) No 1003/2005** of 30 June 2005 implementing Regulation (EC) No 2160/2003 as regards a Community target for the reduction of the prevalence of certain salmonella serotypes in breeding flocks of *Gallus gallus* and amending Regulation (EC) No 2160/2003;
- Commission Regulation (EC) No 2073/2005** of 15 November 2005 on microbiological criteria for foodstuffs;
- Commission Regulation (EC) No 2160/2003** of the European Parliament and of the Council of 17 November 2003, on the control of salmonella and other specified food-borne zoonotic agents;
- Commission Regulation (EC) No 178/2002** of the European Parliament and of the Council of 28 January 2002 laying down the general principles and requirements of food law, establishing the European Food Safety Authority and laying down procedures in matters of food safety;
- Council Decision 90/424/EEC** of 26 June 1990 on expenditure in the veterinary field;
- Directive 2003/99/EC** of the European Parliament and of the Council of 17 November 2003 on the monitoring of zoonoses and zoonotic agents, amending Council Decision 90/424/EEC and repealing Council Directive 92/117/EEC;
- Norrung, B., Buncic, S., 2008.** Microbial Safety of meat in the European Union. Meat Science, 78, 14–24;
- TESSy (The European Surveillance System). 2007.** European Centre for Disease Prevention and Control (ECDC). http://ecdc.europa.eu/documents/pdf/TESSy_workgroup.pdf (access on 5th May, 2009.);
- WHO 1980.** Report of the WHO/WAVFH Round Table Conference on the Present Status of the Salmonella Problem (Prevention and Control), Bilthoven, the Netherlands, WHO/VPH/81.27;

Paper received 11.05.2009.

COCCIDIOSIS IN POULTRY INDUSTRY*

Lilić S., Ilić Tamara, Dimitrijević Sanda

A b s t r a c t: Coccidiosis is a permanent health problem in poultry industry especially in intensive production systems. It is the most important poultry disease as far as economy is concerned since yearly costs of prophylaxis, as well as of therapy exceed 2 billion Euros, at the global level. In Serbia the disease has the highest prevalence in chicken, less in turkeys, gees, ducks and pheasants. The causes of the infection are protozoa belonging to the Eimeridae family, spore oocysts being the infective form. The source of the infection are already infected birds, whereas the disease can spread in the susceptible bird population by direct and indirect contact such as dust, objects on the farm, people, rodents, wild birds, as well as insects. The incidence of the disease depends on the lack of space on the farm, high temperature and high relative humidity, improper feeding, other diseases and all factors that can compromise bird immunity and general resistance to infectious diseases. Coccidiosis is the disease of the spring and fall, i.e. humid seasons with plenty of rain. The parasite development takes place in epithelial cells of the intestine of all bird species. The parasite can develop also in epithelial cells of the kidney glomerully in gees whereas merozoites and shizonts (as a developing form of the parasite) cause severe lesions and desquamation of the mucus. Local symptoms are accompanied with general health disturbance and typical diarrhea which is the characteristic symptom. Diagnosis is on the basis of the general symptoms, gross and microscopic findings as well as feces sample testing. To control coccidiosis in poultry, there is a vaccine or the disease is controlled by anticoccidials in the feed. Coccidiosis is possible to treat with anticoccidials (coccidiostatics and coccidiocides). Economical consequences of the coccidiosis in poultry are decreased feed conversion, smaller weight gain, inadequate feed conversion, smaller body weight at the end of the fattening period, prolonged fattening period, as well as therapy costs.

Body weight gain is reduced, as well as accumulation of abdominal fat. The disease has a negative impact on chemical and sensory meat appearance. One of the problems as far as coccidiosis is concerned is drug resistance. Today, coccidiosis control strategies are the „shuttle” and „switch” program of the prophylactic medication, good manufacturing praxis and proper sanitation.

Key words: coccidiosis, poultry, economical impact.

Kokcidioza u proizvodnji živine

S a d r ž a j: Kokcidioza je oboljenje koje predstavlja stalan zdravstveni problem, naročito u uslovima intenzivnog uzgoja živine. Najznačajnija je bolest živine u ekonomskom pogledu, jer godišnji troškovi za profilaksu i terapiju kokcidioze prevazilaze dve milijarde eura na globalnom nivou. U našoj zemlji najzastupljenija je kod kokošaka, a ređe se javlja kod čuraka, gusaka, pataka i fazana. Uročnici oboljenja su protozoe iz familije Eimeridae, a infektivni oblik predstavljaju sporulisane oociste. Izvor infekcije su inficirane jedinke, a bolest se prenosi direktnim i indirektnim kontaktom – pribor, oprema, prašina, ljudi, glodari, divlje ptice i insekti. Na rasprostranjenost bolesti utiču: nedovoljan prostor, visoka temperatura i relativna vlažnost vazduha, neadekvatna ishrana, pojava drugih oboljenja i svi faktori koji smanjuju otpornost organizma. Najčešće se javlja u proleće i jesen, odnosno u kišnim periodima. Razvoj parazita odigrava se u epitelnim ćelijama creva svih ptica, odnosno epitelu bubrenih kanalića gusaka, a razvojni oblici (merozoiti i šizonti) dovode do teških oštećenja i deskvamacije sluznice, praćenih promenom opšteg stanja i karakterističnim prolivom. Dijagnoza oboljenja postavlja se na osnovu kliničke slike, koprološkog, patomorfološkog i patohistološkog nalaza. Profilaksa oboljenja sprovodi se vakcinacijom ili primenom antikokcidijala u smešama za ishranu, dok se terapija sprovodi antikokcidijalima, koji mogu biti kokcidiostatici i kokcidiocidi. Ekonomski gubici ogledaju se u povećanom utrošku hrane, smanjenom prirastu, nižoj konverziji hrane, manjoj prosečnoj telesnoj masi na kraju tova, produženom trajanju tova i troškovima lečenja.

Prinos trupova i deponovanje abdominalnog masnog tkiva su manji, a oboljenje negativno utiče i na hemijske i senzorne parametre kvaliteta mesa. Problem u suzbijanju oboljenja je brz razvoj rezistencije na lekove, a kontrolne strategije u suzbijanju kokcidioze su „shuttle” i „switch” program profilaktičke medikacije, dobra proizvođačka praksa i sanitacija.

Ključne reči: kokcidioza, živina, ekonomski aspekti

Poultry coccidiosis

Coccidiosis is a parasitic disease that is a constant health problem, especially in intensive poultry industry. It is the most important infectious poultry

disease, as far as economy is concerned. Coccidiosis is a global disease and costs on yearly basis, for prophylaxis, as well as therapy exceed two billion Euros (Dallouil and Lillehoj, 2006).

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTHORS: Slobodan Lilić, slobo@inmesbgd.com, Institute of Meat Hygiene and Technology, Belgrade, Serbia; Tamara Ilić, Sanda Dimitrijević, Faculty of Veterinary Medicine, Belgrade, Serbia

AUTORI: Slobodan Lilić, slobo@inmesbgd.com, Institut za higijenu i tehnologiju mesa, Beograd, Srbija; Tamara Ilić, Sanda Dimitrijević, Fakultet veterinarske medicine, Beograd, Srbija.

Several domestic species are susceptible, however concerning the incidence, as well as economic consequences, coccidiosis is most important in poultry, rabbits, ruminants, carnivores and less in swine. In Serbia, coccidiosis is most important in the chicken industry, less in turkey, gees, ducks and pheasants.

Causative agent of the disease belong to phylum *Apicomplexa*, class *Sporozoa*, subclass *Coccidia*, ordo *Eucoccidia*, suborder *Eimerinae*, and family *Eimeridae* that has two ordo: *Eimeria* and *Tyzzeria*. Depending on the localization, disease in poultry has two forms: coccidiosis of the caecum that is caused by *Eimeria tenella* and intestinal coccidiosis that is caused by a number of parasites: *E. necatrix*, *E. acervulina*, *E. maxima*, *E. brunetti*, *E. mitis*, *E. mivati*, *E. praecox* and *E. hagani*. Coccidiosis of turkeys is caused by: *E. adenoides*, *E. meleagritidis*, *E. gallopavonis*, *E. dispersa*, *E. inocua*, *E. meleagridis* and *E. subrotunda*. In gees the disease can be in the form of renal infection *E. truncata* and intestinal coccidiosis: *E. anseris*, *E. nocens*, *E. parvula* and *E. stigmosa*. Duck coccidiosis is caused by *Tyzzeria perniciosus* however, *E. anatis* and *E. danailovi* can also cause the disease. In pheasants, coccidiosis is caused by *E. dispersa*, *E. phasiani*, *E. langeroni*, *E. pacifica*, *E. megalostomata*, *E. gennaeuscus*, *E. duodenalis*, *E. colchici*, *E. picta* and *E. tetartooimia*.

Epidemiology

The source of the infection varies and depends on the technology in the poultry industry. In the case of extensive poultry farming the source of infection is one bird. In case of intensive production the source of infection is the old bird population (Hammond and Long, 1973). In flock, disease is spreading by direct, as well as indirect contact (Williams, 2002). Oocysts that are infectious could be distributed by equipment, dust, people, rodents, wild birds as well as insects (Dimitrijević and Ilić, 2003). *Coleoptera* spp, which are usually present in the broiler population, can serve as mechanical vectors (Calnek, 1997).

Distribution and prevalence is influenced by several factors: high animal density cramped on a small space, high air temperature, high relative humidity, different (especially different age) categories of birds at same place, feed change, quality of feed, as well as all other factors that compromise resistance to the disease and general health status of the birds (Calnek, 1997). Onset of the disease depends on the age of the bird at the time of the first infection and number of passages of the infect (for one passage to be completed it is required 10 days), as well as on

ability of the bird to develop proper specific immune response (Hofstad, 1984; Ilić et al., 2003).

The highest incidence of coccidiosis is during spring and fall, especially when weather is cold and humid (rain). The incidence is significantly smaller during hot and dry weather conditions (Maungyai et al, 1990; Calnek, 1997; Razmi and Kalideri, 2000). The intensity of the infection depends on the number of oocysts that are ingested and the immune status of the bird (Hofstad, 1984). In case of release of the chicken on to the floor that was used for the previous flock. Šibalić and Cvetković (1996), reported the acute form of the coecal coccidiosis and mortality as soon as in eight days old chicken.

Infection of young chicken can not be avoided in intensive production systems, whatever prophylactic measures have been taken. So, infection takes place in the first weeks of life. Intensive poultry production systems, high density of totally susceptible birds and many passages of the causative agent in the new bird generation, pose almost ideal circumstances for infection to persist and spread within the flock (Jordan, 1990). To heavy load of infectious oocysts on the floor is one of the most important prerequisite conditions for infection to persist in the flock (Hofstad, 1984).

Clinical disease can be prevented by continuous adding of the anticoccidials in feed. However, persistence of the sub clinical disease is always a possibility. According to some authors (Braunis, 1980; Razmi and Kalideri, 2000), sub clinical forms of the disease depend on the size of the flock. Prevalence of the sub acute form of the disease is significantly higher in flocks with more than 40,000 birds in comparison to flocks with less of 10,000 birds. The subclinical form of the disease is most frequent in six weeks old chicken and infection occurs in nearly all flocks (Jordan and Pattison, 1996). Voeten (1987) showed that sub clinical coccidiosis is most prominent from four to six week old chicken in the case if anticoccidials are not added to the feed.

Pathogenesis

The infectious form of the causative agent are oocysts in the form of spores. Infection is by oral route, with contaminated feed and/or water. After ingestion, infectious oocysts exist, liberating the infective form: the sporozoit. Sporozoit infect epithelial cells of the intestine and kidney epithelial cells. Transfer of the sporozoits up to the locus of the primary lesion is with the help of intraepithelial lymphocytes (Lawn and Rose, 1982; Daszak, 1999). The pathogenic process starts during shizogonic phase of the parasite development. The pathogenic

process during the first generation of shizonts is negligible. However, the most pathological stadium is during the second generation of shizonts. Their development, deep in the cells of Lüberkinii glands, results in inflammation, mucus desquamation, capillary rupture and haemorrhagiae. This stadium of the disease is accompanied with severe clinical symptoms. In this stadium, possible outcome could be death of the bird. Death is a consequence of haemorrhagiae (bird can loose 60 to 80 percent of the blood volume), toxemia or as a consequence of gangrene or rupture of the intestinal wall.

During coccidiosis, there can be other infections such as reovirus infection, Marek disease, New Castle virus infection and infectious bronchitis virus infection. In such a case, symptoms are mixed depending on causative agents (Ruff, 1991). Especially in Nordic countries, there are mixed infections with *Eimeria* spp, *Cl. perfringens* or *E. coli*. This is because the use of antibiotics is banned (Van Der Stroom and Van der Sluis, 1999).

Endogenous development of renal coccidiosis in gees takes place in tubules of the kidney. As a result, there is desquamation of the epithelia, obstruction and dilatation of the tubuli by mature gamonts. Kidneys are enlarged, there are urate salts deposits in the urinary tract, as well as kidney failure.

Developmental cycle of the parasite

All coccidia develop in same way. There are two phases: endogenous and exogenous. *Vide infra* is the infectious cycle of the *E. tenella* which is highly pathogenic and the most prevalent in our region.

The endogenous phase is in the animal (bird) and there are two sub-phases: shizogonia (nonsexual sub phase) and gametogonia. Shizogonia is characterized by producing one after another generations of shizonts that carry merozoites as the infectious form of the parasite (Soulsby and Rose, 1972). During the sexual sub-phase (gametogonia), oocysts form that are responsible for further infection spreading. Exogenic phase take place out of the bird. During this phase, oocysts sporulate (sporogonia).

One to two hours (Lawn and Rose, 1982), after ingestion of the oocysts they excyst as the wall of the oocysts ruptures and releases sporocysts. From oocysts, by further degradation, release of the sporozoites occurs. Sporozoites attack the surface of the caecal epithelium (Patillo, 1959; Davies et al., 1963), penetrate the basal membrane and enter the lamina propria mucosae whether free or inside the macrophages. Finally, they attack epithelial cells that cover the bottom of the Lüberkinii cripts (Lillehoj and Trout, 1993).

In most cases, from the second generation of merozoites microgametocyte develop, as well as makrogametocytes. Sexual phase of the parasite development, takes place in the cells of the mucus and sub mucus. That phase starts from 6th day of the infection (Pellerdi, 1974). Microgametocytes (12,4 x 8,7 µm) (Tyzzer, 1929), enlarge and undergo through a number of divisions resulting in microgamete development (Davies et al, 1963). Microgametes are mobile, fusiform in the shape approximately 5 µm long with three active flagella evenly distributed on one end of the cell (Joyner and Kendall, 1963).

Macrogametes are as big as oocysts. During growth they transform into the macrogamete. Macrogamete have granular cytoplasm and centrally placed nucleus (Pellerdy, 1974). When micro and macrogametes join they form zygote. After the fertilization phase, the macrogamete mucoproteinaceous granule that is placed on the periphery of the cell, form the outer membrane of the zygote. From that form, nonporous oocyst develops.

Once the cyst wall is formed completely the oocyst leaves the host through feces. Prepatent period is the time from the start of the infection up to the moment when first oocyst could be found in feces. This period of time is unique for the species and in case of *E. tenella*, it is up to 6 to 7 days (Pellerdy, 1974). The maximal number of oocysts in feces is at 10th day after infection. After that time, number of the oocysts in feces sharply decline (Hammond and Long, 1973).

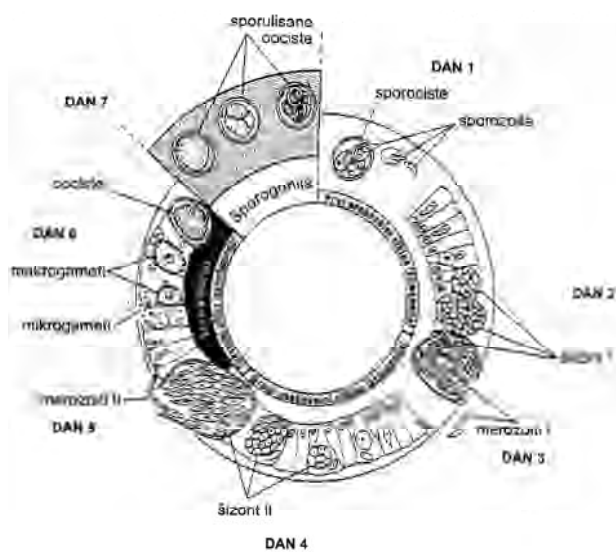


Figure 1. Scheme of the developing cycle of *Eimeria tenella*

Slika 1. Shematski prikaz razvojnog ciklusa *Eimeria tenella*

Diagnosis

Diagnosis of the coecal coccidiosis is made on the basis of clinical signs, coprology, patomorphological and patohistological analysis. Clinical diagnosis is not reliable. One of the basic symptoms that could lead to diagnosis is bloody diarrhea, as well as changes in feces appearance (Dimitrijević, 1999).

Disease signs (clinical)

The first and most frequent symptom is at the beginning yellow diarrhea. As the disease progresses, because of the blood in feces, feces are red or resemble the color of chocolate (Jordan, 1990). The feathers around the cloacae are covered with bloody deposits. Feces are stained with blood. Birds that survive first few days of the infection, can survive the next 10 to 15 days. During that time, birds are thirsty and rapidly loose weight (Calnek, 1997). Symptoms of the disease start to appear at the time when the second generation of shizonts starts rapidly to replicate, grow, mature and release the second generation of merozoites. Second generation of merozoites cause inflammation of the sub epithelial mucus, desquamation of the epithelia and capillary rupture in the caecum wall. As a consequence, bloody diarrhea occurs (Jordan, 1990).

Other signs of the disease are anorexia, thirst (Pellerdy, 1974), somnolentia, goose pimples, dropped wings, closed eyes, leg paralysis, anemia of the crest and outer mucous membranes, enterorrhagiae, skin depigmentation and in spite of the loss of appetite the gizzard is filled with feed (Ruff, 1991). Death usually occurs 5. and 6. day after infection (Hammond and Long, 1973).

It is postulated that death is the result of blood loss, as well as infection. However, the precise cause of death is not jet clear (Calnek, 1997). Death of the bird can be the result of gangrene or rupture of the caecal sac (Hofstad, 1984).

In gees with renal coccidiosis somnolence, leg weakness, birds are reluctant to move, eyes are closed, inapetencia, thirst, whitish diarrhea, dropped wings, nervous signs, neck twisting, weight loose, and death are present.

Coprology

Coprology is performed on native samples by flotation, using concentrated solutions of NaCl. The most reliable method is to find oocysts and count them by using the McMaster method. However, it is not enough to confirm the causative agent or cause of death since death can occur before onset of oocysts in the feces (Dimitrijević, 1999; Dimitrijević and Ilić, 2003). Positive results only show that there

is infection that is at least seven days old (Hofstad, 1984). In the case of renal coccidiosis of gees, oocysts can be found in feces however this finding is not enough for diagnosis, since there are difficulties to differentiate them and oocysts of the intestinal coccidias.

Patomorphological leasons

In cases of intestinal coccidiosis, the first and second day after infection, on the microscopic level (patohistology) there are focal lesions of the intestinal epithelium and small necrotic foci in the subepithelial connective tissue. Those changes are the result of first generation shizont maturation. On the third day, caecums are enlarged in diameter and there are regions with petechiae in the mucosa. The most prominent macroscopic lesions are from the fourth and fifth day after infection. It is obvious since in that period the second generation of the shizonts completely matures and on the fifth day after infection there is transformation into the second generation merozoites. Entrance of the second generation of merozoites into the healthy epithelial cells, mark the moment when haemorrhagiae of the caecum start. Such findings accompanied with heterofil infiltration of the *lamina propriae* and submucosis, as well (Calnek, 1997).

The intestine is shortened and the intestinal wall is thickened. The lumen is enlarged two to three times. The color is dark blue with sub serous petechiae. Mucosa is thickened; surface of the epithelium, as well as the epithelium of the Lüberkini crypts is desquamated with haemorrhagic patches. The intestinal content is watery, bright red in colour with desquamated cells, erythrocytes and plenty of coccidia in different stages of development. Later on, the content becomes thick and the colour is changed to dark red. Gradually, fibrinous tissue encirculates the content of the intestinum, resembling gray-yellow hard cork (Nešić, 1999).

During the sixth and seventh day of infection the content of the intestinum hardens and becomes dry. Epithel regeneration is fast and can be accomplished in 10 days after infection. However, as a consequence of intensive local lesions, it is possible that the epithel never returns to the previous condition (Calnek, 1997). Recovery starts with the appearance of fibroblasts and angioblasts (Pellerdy, 1952).

Microscopic examination of the intestinal wall, reveals plenty of parasites in different stages of maturation and development. Native sample-slide is especially useful since it shows oocysts and macrogamets (Jordan, 1990). The pathognomonic finding is the presence of shizonts in the material (Calnek, 1997).

Diagnosis is made on the basis of gross lesions in the intestine, as well as microscopically by using the content of the intestine as a sample (Calnek, 1997). Intensity of the infection can also be estimated especially if there is a doubt whether coccidias are the only cause of the fatal outcome of the disease. Intensity of infection is in proportion with the number of oocysts that were ingested and is in positive correlation to other parameters such as loss of body weight and changes in feces appearance (Hofstad, 1984).

Postmortem examination of geese that succumbed to renal coccidiosis are cahectic and gross lesions are localized only in kidney. Kidneys are enlarged, circular in shape, smooth and bright at the surface, grey-white or grey-yellow in color. Sometimes the color changes to gray-red and red-brown. The surface of the kidneys have plenty of softened foci that are white or yellow, circular and 0.5 to 1 mm in diameter. These foci are not clearly separated from rest of the kidney tissue. It is possible to find whitish stripes and petechiae (Dimitrijević and Ilić, 2003).

Histology

Using standard pathohistology staining procedures (hematoxylin-eosin) different stages of parasite development can be seen (Hofstad, 1984). In order to differentiate and identify them, it is better to use Shiff's reagent. Polysaccharides accompanied with refractory granula, as well as aggregates that form the macrogamete wall, stain bright-red (Calnek, 1997; Nešić, 1999).

Appart of the abovementioned standard technique, there are other more specialized diagnostic methods that use monoclonal antibodies conjugated with fluorescent markers (Calnek, 1997).

Microscopical examination of the sample reveals that second shizont generation migrate deep into the lamina propria. Around them, there is a strong inflammatory cell reaction with eosinophils, plasma cells and in some cases giant cells (Hofstad, 1984). Oocysts can be found in tissue sections, and the finding depends on the stage of the infection when the sample was taken. Oocysts can be seen in giant cells next to the muscular lamina of the intestinal wall (Pellerdy, 1974). The first shizont generation, that matures two to three days after infection, can be seen microscopically scattered as a wide belt. Small focal hemorrhagiae and necrosis can be seen in the vicinity of blood vessels in the stratum circulare internum of the intestinal wall muscular lamina (Jordan, 1990).

Kidney tubuli from infected geese are dilated and filled with epithelial cells and oocysts. Ureters

are dilated and filled with mucous yellow-brown mass. At some places, epithel of the renal tubuli totally disappeared and as a consequence, there are cists filled with parasites in different stages of development and cell detritus. Around most of the tubuli, there is fibrinous tissue proliferation with a number of inflammatory cells (Dimitrijević and Ilić, 2003).

Prophylaxis and therapy of coccidiosis

Coccidiosis can be treated with anticoccidials. They can act either as coccidiostatics, that inhibit growth and development of the intracellular parasite form or coccidiocides. Coccidiocides destroy the parasites during their developmental stages. Most of the anticoccidials are coccidiocides or they are at the beginning of the action coccidiostatics and in later stage, coccidiocides (Long and Jeffers, 1986). In order to prevent coccidiosis, it is possible to add some of the above mentioned substances in the feed for birds. In case therapy is needed, the drug is given diluted in drinking water.

Basically, anticoccidials are divided in 12 groups: benzenacethonitril derivatives (clazuril and diclazuril), benzyl-purin (arprinocid) derivatives, xarbanilid derivatives (nicarbazine), guanidine derivatives (robenidin), dinitrobenzamide derivatives (dinitolmid), ionofors-polyether antibiotics (monensin, lasalocid, narasin, salinomycin, maduramicin, alboriksin), piridins (klopidol), quinazolines (halofuginon), hinolons (dekokvinat, metilbenzalkvat), sulphonamides (sulphakvinoxalin), symmetric triazinons (toltrazuril) and tiamine antagonists (amprolium).

With the exception of ionophors, there is a possibility that coccidias develop resistance (Jordan, 1990; Dimitrijević et al., 1992; 1998). It is required only that several sporozoites survive and start the asexual cycle. That leads to production of several thousands of parasites that are resistant to a particular drug. In order to avoid resistance, it is better to use coccidiocides that act on the late stages of shizogony (Jezdimirović, 1997).

In order to minimize the possibility for resistance to develop, it is possible to use "shuttle" and "dual" program. The basis of such program is to change drugs during flock raising. Another program is the "switch" program i.e. changing the drug for the next flock. Whatever drug is in use, it is essential to change drugs according to the mode of action of the active substance. Only in that case there is a real chance to avoid development of resistance within the parasite population (Calnek, 1997; Dimitrijević and Ilić, 2003).

After treatment, whether prophylactic or in therapy, there is need to take care of drug withdrawal period. Nowadays, in order to prevent the disease, most often iodophors are in use. Drugs are omitted in the feed for the final fattening period. Nevertheless, even with ionophores there is a possibility for the parasite to develop resistance (Chapman, 1997).

Immunity

Broilers that survived coecal coccidiosis, immunity is life lasting and that is normal in natural infection (Pellerdy, 1974). Chicken, acquire immunity from their mothers only if hens are actively immunized against coccidiosis (Hammond and Long, 1973).

Immunity against parasitic diseases develops in the same way as protection against all other infectious diseases. It is dependent on the age (Ruff, 1991) and genetic background (Jeffers and Shirley, 1982). At the same time, it depends on the number of oocysts that are inoculated. Immunity against coccidiosis is highly specific and cross protection has not been documented. That means that different species of the parasite can cause disease in susceptible birds (Hofstad, 1984; Ilić et al., 2003a).

Early information on immunity against coccidiosis show that in order to stimulate the immune reaction, it is required to have, as immunogen, shizonts of the second generation. However, it has been shown that the immune reaction develops as early as 72 (Kendall and McCulloch, 1952) hours after ingestion or after intracutaneous injection (Pellerdy, 1974), of the infective oocysts at the time when there are not second generation of the shizonts developed yet.

Good protection in the case of coccidiosis means that there is no development of the parasites and onset of oocysts during reinfection. That is achieved after several natural infections. Better protection is achieved with every day infection of chickens with a small number of infective oocysts in comparison with one single dose (Joyner and Notrhon, 1973). In practice, simulation of multiple dose immunization is during floor husbandry when continuous reinfection keep the immune system in contact with the immunogen (Šibalić and Cvetković, 1996; Jordan, 1990).

The immune response to coccidia is complex. Animals infected with *Eimeria* spp. develop parasite-specific immunoglobulins that are present in the circulation, as well as on the mucous membranes, in secretions. However, it has been shown that specific antibodies play a minor role in the protection against coccidiosis. Nowadays there is evidence that cell

immunity plays a major role in the protection against infection (Challey and Burns, 1959; Pattillo, 1959; Davies and Sandar, 1963; Soulsby, 1972; Lillehoj and Trout, 1996; Ilić et al., 2003a, 2003b).

Early investigations show that the basis for protection against coccidiosis are of the humoral type (McDermot and Stauber, 1954; Itagaki and Tsubokura, 1955). However, today it has been shown that the protection is of the cellular type (Long and Pierce, 1963). Details of the protective mechanisms that are activated during infection are not clarified yet however, it is clear that cellular immunity plays the most important role in bird protection (Lillehoj and Bacon, 1991).

As a result of infection, T lymphocytes produce cytokines. At the same time, T lymphocytes are cytotoxic to infected cells (Lillehoj and Trout, 1996). However, detailed mechanisms of that protection are still obscure. One of the theory is that the major mechanism of protection is the presence of intestinal immune system of chickens, that means that the intestinal lymphoid tissue poses as the first specialized line of defence of the mucous surfaces. That system encirculates not only immunoregulatory, but effector cells, as well.

Vaccination

Because of the resistance against anticoccidials that often develops, vaccination is the most appropriate method for disease control (Augustine et al., 2001). Vaccination is the simplest and cheapest way to achieve immunoprophylaxis. In that way, the immune system is activated so natural infection causes a secondary immune reaction which is faster and better in comparison to the primary immune reaction (Naglić and Hajsig, 1993; Dimitrijević and Ilić, 2003a).

An ideal vaccine will stimulate long lasting immunity against all epitopes in the coccidia structure. That immunity has to be not only specific for the basic pathogenic coccidia species, but also against strains that develop during epizootia (Dimitrijević, 1993). The vaccine also has to be harmless for birds that are vaccinated. At the same time the vaccine must not contaminate the natural habitat with potentially pathogenic coccidia. Vaccines that are in use, can have attenuated (alive), recombinant or anti-idiotypic immunogens. As immunogens, attenuated vaccine can have non-virulent coccidia strains or can be produced on the basis of virulent coccidia strains (Lillehoj and Trout, 1993; Liand et al., 2005).

Live vaccines that possess virulent coccidia strains comprise of a mixture of all species of virulent coccidia. Such a vaccine can be used in drinking

water (Jordan, 1990). They elicit the most potent immune reaction since immunogenic characteristics match with the ability of the parasite to replicate and with the level of pathogenicity (Naglić and Hajsig, 1993). They are the best vaccines however, such vaccines have to be used in small doses in order pathogenic changes not to occur (Orlić *et al.*, 1996; Dimitrijević, 1997). For maximal effect, birds have to be revaccinated several times (Orlić *et al.*, 1996; Dimitrijević, 1997).

Recently, as the immunogen in vaccines, there are alive *Eimeria* species that are tolerant to iodophores. Advantage of such vaccines is that in vaccinated flock iodophores can be used in the first 3-4 weeks of bird life, at the time when immunity is not yet fully developed (Danforth, 2000). Vaccinated birds, for not yet clear reasons, have a smaller mortality in comparison untreated ones (Williams, 2002). Live, virulent immunogens (vaccines) are not quite appropriate for broilers since there is a possibility of accumulation of parasites in the floor (Lillehoj and Trout, 1993).

Live attenuated vaccines can be divided into two groups. The first group comprises of vaccines that are made of natural strains that are of low virulence. The second group of such vaccines, have laboratory produced low virulence strains as immunogens (Shirley, 1989).

Special advantages of live vaccines is that vaccine strains compete with natural, highly virulent strains that are resistant to drugs (Hofstad, 1984). In that case, vaccine strains overgrow natural strains in the vaccinated bird population. By attenuation of infectious oocysts, live cycle of coccidia can be shortened in order to enable required number of immunizing stages and still not possessing an infectious potential (Dimitrijević, 1993).

Advantages of attenuated vaccines, in comparison to virulent vaccines are that in the production of a great number of oocysts, there is minimal danger of infection to occur. The disadvantage is that there is only a partial protection against natural „field“ coccidia strains (Shirley, 1989; Augustine *et al.*, 1993).

Vaccines based on recombinant techniques consist of immunogens that were produced in bacterial vectors. In that way, large quantities of immunogen can be produced (Dimitrijević, 1997). They are a kind of cocktail consisting of different antigens originated from several coccidia species. At the same time, such vaccines consist of different antigens from the same coccidia species. To produce them, it is required to use complex technology and their production is still a matter of future in vaccinology. Disadvantages of such vaccines are

low immunogenicity and possible selection of mutant coccidias that do not possess the cloned gene. So, such mutant parasite can freely replicate in the vaccinated bird population. At the beginning, in few parasite generations, mutants represent a small population however, during epizootia, they became dominant. That means that in such a case, there is a need to produce new recombinant immunogens frequently (Lillehoj and Trout, 1993; Dimitrijević and Ilić, 2005).

Anti-idiotypic vaccines are a special variety of vaccines that use anti-idiotypic immunoglobulins (Lillehoj and Trout, 1993). The mode of action of such antibodies is based on idiotypic-antiidiotypic network. Anti-idiotypic vaccines open new possibilities in coccidiosis immunoprophylaxis however, they are very expensive. At the same time they lack immunogenicity (Naglić and Hajsig, 1993). In future, such immunization could be used for overcoming certain genetical limitations that are still causing problems in vaccination against some other diseases (Lillehoj and Trout, 1993).

Coccidiosis – economic impact

In the last few years the poultry industry and as a consequence chicken meat represents 80 percent of the whole production of meat originating from birds. Still, production is the fastest growing in the meat industry. According to analysis, production, as well as consumption of chicken meat, will rise because of: good feed conversion in comparison to other animal species, there is not religious aspect of poultry meat consumption, poultry meat is healthy (low fat and high protein content), has good sensory qualities, low price and fast production which mean a short generative time. Poultry, during coccidiosis and after therapy, have poor productive results. Daily feed quantity and feed conversion rise. Chicken daily growth weight is reduced, as well as body mass at the end of the fattening period (Jordan, 1990; Vermeulen *et al.*, 2001). As a result the fattening period should be prolonged. At the same time, care should be taken for the withdrawal period for the drug which further rises costs of production (Jordan, 1990; Williams, 2002).

Because of coccidiosis, carcass yield is smaller, as well as the proportion of more valuable parts of the body. Also, fat deposits are smaller in the abdominal fat tissue. In broilers' meat, there is higher water content and less proteins. Relative proportion of proteins of the fibrinous tissue in the total protein mass is higher. Sensory characteristics of the broilers' meat are bad in comparison to the population where coccidiosis was absent (Lilić, 2007). In liver of infected broilers, content of iron

and copper is smaller. Meat of infected broilers have a decreased iron manganese and phosphorous content (Koinarski et al., 1998).

A great economic problem is resistency to anticoccidial drugs. Such drugs are not easy to use. Also, development of new drug generations, that are for prophylaxis and therapy, is expensive. As an alternative, there are investigations whose target is to use immunological, biotechnical and genetical methods for prevention and control of coccidiosis (Grag et al, 1999). Of all coccidias that cause the disease, *Eimeria tenella* is widely distributed and serves as a gold standard in order to sequence the genetical material of the causative agent. At the same

time, *E. tenella* is the first candidate for eradication (Augustine et al., 2001).

Poultry meat consumption, at a global level is constantly rising. So, there is a need to intensify broiler production. In such a production system, the possibility for coccidiosis is higher inspite of using anticoccidials in feed. At contrary, world trends in food production are to produce organic meat, with no drugs added to the feed. This means that the risk of coccidiosis is higher. Nevertheless, strategies to control coccidiosis are still based on prophylactic medication through feed and vaccination (Vermeulen et al, 2001), not to exclude good production praxis and good hygiene and sanitation.

References

- Augustine, P. C., Danforth, H. D., McAndrew, S. J. 1993.** Monoclonal antibodies detecting antigenic differences in refractile bodies of avian *Eimeria* sporozoites, *Journal of parasitology*, 74:653-659;
- Augustine, C. P., Bartha, R. J., Innes, L., Müller, N. 2001.** Chasing coccidia - new tools enter the race, *Trends in Parasitology*, Volume 17, Issue 11, 1 November, 509-511;
- Braunis, W. W. 1980.** Monitoring the biological performance in broiler with special regard to subclinical coccidiosis, *Archiv für Geflügelkunde*, 44, 183-187;
- Calnek, M. 1997.** *Diseases Of Poultry*, Iowa State University Press, Ames;
- Challey, J. R., Burns, W. M. C., 1959.** The invasion of the caecal mucosa by *Eimeria tenella* sporozoites and their transport by macrophages, *Journal of protozoology*, 52:964-967;
- Chapman, H. D. 1999.** The development of immunity to *Eimeria* species in broilers given anticoccidial drugs, *Avian Pathology*, 28, 155-162;
- Dalloul, R. A., Lillehoj, H. S. 2006.** Poultry coccidiosis: recent advancements in control measures and vaccine development. *Exp. Rev. Vaccines* 5, 143-163;
- Danforth, H. D. 2000.** Increase in anticoccidial sensitivity seen after field trial studies with five oocysts vaccination of partially drug-resistant strains of avian *Eimeria* species, *Proceedings of the 75th Annual Meeting of the American Society of Parasitologists and the 53rd Annual Meeting of Protozoologists*, 90;
- Daszak, P. 1999.** Zoite migration during *Eimeria tenella* infection: parasite adaption to host defences, *Parasitology Today*, 2:67-72;
- Davies, S. F. M., Yoyner, L. P., Kendall, S. B. 1963.** Coccidiosis, Oliver and Boyd LTD. Edinburgh;
- Dimitrijević, S. 1993.** Citogenetske i imunološke promene pod uticajem kokcidiostatika, *Doktorska disertacija*, Univerzitet u Beogradu, Beograd;
- Dimitrijević, S. 1997.** Kokcidioza živine i načini preveniranja, *Živinarstvo*, 4-5:99-101;
- Dimitrijević, S. 1999.** Dijagnostika parazitskih bolesti, *Fakultet veterinarske medicine*, Beograd;
- Dimitrijević, S., Ilić, T. 2003.** Kokcidioza živine, monografija, *Fakultet veterinarske medicine*, Univerzitet u Beogradu;
- Dimitrijević, S., Ilić, T. 2005.** Biohemijske i imunološke karakteristike kokcidijalnih antigena, 17. Savetovanje veterinarara Srbije, *Zbornik radota i kratkih sadržaja*, zlatibor, 7-10. septembar, 223-224;
- Dimitrijević, S., Pujić, N., Čupić, V., Savovski, K., Dimitrijević, B. 1992.** Supression of thymocyte proliferation by the coccidiostatic salinomycin and derivates of penicillin, *Acta Veterinaria*, Vol.42, No.5-6:291-298;
- Dimitrijević, S., Savovski, K., Dimitrijević, B. 1998.** Genotoxicity of the anticoccidial agent salinomycin, *Acta Veterinaria*, 48(4).245-254;
- Dimitrijević, S., Ilić, T. 2003.** Najvažniji aspekti imunogenosti *Eimeria* spp., *Veterinarski glasnik*, 57(7-8):505-508;
- Grag, R., Banerjee, D. P., Gupta, S. K. 1999.** Immune responses in chickens against *Eimeria tenella* sporozoite antigen, *Veterinary Parasitology*, 81, 1-10;
- Hammond, D. M., Long, P. L. 1973.** *The Coccidia*, University Park Press, Baltimore, Buterworths, London;
- Hofstad, M. S. 1984.** *Diseases Of Poultry*, Iowa State University Press. Ames;
- Ilić, T., Knežević, M., Aleksić-Kovačević, S., Dimitrijević, S. 2003.** Neke karakteristike imunološkog odgovora na infekciju prouzrokovanu kokcidijama, 15. Savetovanja veterinarara Srbije, *Zlatibor*, 9-13. septembar, *Zbornik radova i kratkih sadržaja*, 174;
- Ilić, T., Knežević, M., Aleksić-Kovačević, S., Dimitrijević, S. 2003a.** Neke karakteristike imunološkog odgovora na infekciju prouzrokovanu kokcidijama, 15. Savetovanja veterinarara Srbije, *Zlatibor*, 9-13. septembar, *Zbornik radova i kratkih sadržaja*, 174;
- Ilić, T., Knežević, M., Dimitrijević, S., Nešić, V., Aleksić-Kovačević S. 2003b.** Study of the distribution of CD3-T lymphocytes in caeca of chickens experimentally infected with *Eimeria tenella*, *Acta Veterinaria*, 53 (5-6):385-391;
- Itagaki, K., Tsubokura, M. 1955.** Studies on coccidiosis in fowls, IV. On the agglutination by merozoites. *Jap. J. Vet. Sci.*, 17:139;
- Lilić, S. 2007.** Ispitivanje uticaja infekcije brojlera protozomom *Eimeria tenella* na proizvodne rezultate pilića u tovu i neke parametre kvaliteta mesa, doktorska, disertacija, *Fakultet veterinarske medicine*, Beograd
- Jezdimirović, B. M., 1997.** *Veterinarska farmakologija*, *Elit Medica*, Beograd;
- Jordan, F. W. T., Pattison, M. 1996.** *Poultry Disease*. Saundr, London, 497;
- Jordan, F. W. T. 1990.** *Poultry Diseases*. Eenglish Language Book Society, London;
- Joyner, L. P., Kendall, S. B. 1963.** Coccidiosis, Oliver and Boyd LTD. Edinburgh, Great Britain;
- Joyner, L. P., Northon, C. C. 1973.** The immunity arising from continuous low-level infection with *Eimeria tenella*, *Parasitology*, 67:333-338;

- Kendall, S. B., McCullough, F. S. 1952.** Relationships between sulphamethazine therapy and acquisition of immunity to *Eimeria tenella*, *Journal of Comparative Pathology*, 62:116;
- Koinarski, V., Georgieva D., Pavlov, A. 1998.** Effect of coccidiosis upon the chemical composition of broiler meat and liver, Poster session, *Parasitology International* 47 (Suppl.);
- Lawn, A. M., Rose, M. E., 1982.** Mucosal transport of *Eimeria tenella* in the cecum of the chicken. *Journal of Parasitology*, 68:1117—1123;
- Li, G. Q., Kanu, S., Xiao, S. M. and Xiang, F. Y. 2005.** Responses of chickens vaccinated with a live attenuated multi-valent ionophore-tolerant *Eimeria* vaccine, *Veterinary Parasitology*, Volume 129, Issues 3-4, 15 May, 179-186;
- Lillehoj, H. S., Bacon, L. D. 1991.** Increase of intestinal intraepithelial lymphocytes expressing CD8 antigen following challenge infection with *Eimeria acervulina*, *Avian Diseases*, 35:294-301;
- Lillehoj, H. S., Trout, J. M. 1993.** Coccidia: a review of recent advances on immunity and vaccine development, *Avian Pathology*, 22:3-31;
- Lillehoj, H. S., Trout, J. M. 1996.** Avian gut-associated lymphoid tissues and intestinal immune responses to *Eimeria* parasites, *Clin. Microbiol. Rev.*, 9:349-360;
- Long, P. L., Jeffers, T. K. 1986.** Control of chicken coccidiosis, *Parasitology today*, 2:236-240;
- Long, P. L., Pierce, A. E. 1963.** Role of cellular factors in the mediation of immunity to avian coccidiosis (*Eimeria tenella*), *Nature*, 2:426-427;
- Maungyai, M., Sirichokchatchawan, S., Juranukul, U. 1990.** Efficacy of Toltrazuril and Maduramicin in the control of coccidiosis in broilers, *Thailandian J.Vet. Med*, 20, 247-253;
- McDermot, J. J., Stauber, L. A. 1954.** Preparation and agglutination of merozoite suspensions of the chicken coccidian *Eimeria tenella*, *Journal of parasitology*, 40 (suppl.), 23;
- Naglić, T., Hajsig, D. 1993.** Veterinarska imunologija, Školska knjiga, Zagreb;
- Nešić, V. 1999.** Forenzička procena eksperimentalno izazvane cecalne kokcidioze brojlera u uslovima ishrane smešama sa zeolitom, Magistarska teza, Fakultet veterinarske medicine, Univerzitet u Beogradu, Beograd;
- Orlić, D., Kapetanov, M., Lalić, M., Mrđen, M., Gagić, M. 1996.** Suzbijanje i mere kontrole kokcidioze živine, *Veterinarski glasnik*, 50:585-589;
- Patillo, W. H., Becker, E. R. 1955.** Cytochemistry of *Eimeria brunetti* and *Eimeria acervulina* of the chicken, *Journal of Morphology*, 69:372-377;
- Patillo, W. H. 1959.** Invasion of the cecal mucosa of the chicken by sporozoites of the *Eimeria tenella*, *Journal of Parasitology*, 45:253-258;
- Pellerdy, L. 1952.** A vakbelcoccidiosis sulphonamid-therapi a janak hazai tapaszalathi, *MTA Agrartud. Oszt. Kozl.*, 3:133-143;
- Pellerdy, P. L. 1974.** Coccidia And Coccidiosis, Akademia Kiado, Budapest;
- Razmi, G. R., Ali Kalideri, G. 2000.** Prevalence of subclinical coccidiosis in broiler-chicken farms in the municipality of Mashhad, Khorasan, Iran, *Preventive Veterinary Medicine*, Volume 44, Issues 3-4, 28 April, 247-253;
- Ruff, M. D. 1991.** An overview of control measures for coccidiosis - present and future, *Proceedings of the Seventh International Poultry Breeders' Conference*, Auchincruive, UK, 29-38;
- Shirley, M. W. 1989.** Development of a live attenuated vaccine against coccidiosis of poultry, *Parasite Immunology*, 11:117-124;
- Soulsby, E. J. L. 1972.** *Immunity To Animal Parasites*, Academic Press, New York and London;
- Soulsby, E. J. L., Rose, M. E. 1972.** Immune response to intracellular parasites, *Journal of Parasitology*, 10:365-372;
- Šibalić, S., Cvetković, Lj. 1996.** Parazitske bolesti domaćih životinja. Veterinarski fakultet, Univerzitet u Beogradu, Beograd;
- Tyzer, E. E. 1929.** Coccidiosis in galinaceous bird, *Am. J. Hyg.*, 10:269;
- Van der Stroom, J. H., Van der Sluis, W. 1999.** The effect of intercurrent diseases on coccidiosis (Van der Sluis W. (Ed.), *World Poultry*. Elsevier, Amsterdam, 13-14;
- Vermeulen, A. N., Schaapk, D. C., Schetters, T. P. M. 2001.** Control of coccidiosis in chickens by vaccination, *Veterinary Parasitology*, 100, 13-20;
- Voeten, A. C. 1987.** Coccidiosis: a problem in broilers (Verstegen M.W.A., Henken A.M., *Energy Metabolism in Farm Animals: Effect of Housing, Stress and Disease*, Martinus Nijhoff, Dordrecht, 410-422);
- Williams, R. B. 2002.** Anticoccidial vaccines for broiler: pathways to success, *Avian Pathology*, 31, 317-353.

Paper recieved 3.04.2009.

MYCOTOXINS IN THE FOOD CHAIN – OLD PROBLEMS AND NEW SOLUTIONS*

Milicevic D.

A b s t r a c t: Mycotoxins are toxic compounds, produced by the secondary metabolism of toxigenic molds in the *Aspergillus*, *Penicillium*, *Fusarium*, *Alternaria* and *Claviceps* genera occurring in food and feed commodities both pre- and post-harvest. Adverse human health effects from the consumption of mycotoxins have occurred for many centuries. When ingested, mycotoxins may cause a mycotoxicosis which can result in an acute or chronic disease episode. Chronic conditions have a much greater impact, numerically, on human health in general, and induce diverse and powerful toxic effects in test systems: some are carcinogenic, mutagenic, teratogenic, estrogenic, hemorrhagic, immunotoxic, nephrotoxic, hepatotoxic, dermatotoxic and neurotoxic.

Although mycotoxin contamination of agricultural products still occurs in the developed world, the application of modern agricultural practices and the presence of a legislatively regulated food processing and marketing system have greatly reduced mycotoxin exposure in these populations. However, in the developing countries, where climatic and crop storage conditions are frequently conducive to fungal growth and mycotoxin production, much of the population relies on subsistence farming or on unregulated local markets. Therefore both producers and governmental control authorities are directing their efforts toward the implementation of a correct and reliable evaluation of the real status of contamination of a lot or food commodity and, consequently, of the impact of mycotoxins on human and animal health.

Key words: mycotoxins, human and animal health, risk analysis

Mikotoksini u lancu ishrane—stari problemi i nova rešenja

S a d r ž a j: Mikotoksini su toksična jedinjenja, proizvod sekundarnog metabolizma plesni iz roda *Aspergillus*, *Penicillium*, *Fusarium*, *Alternaria* i *Claviceps*, koja mogu da kontaminiraju hranu za ljude i životinje, kako u poljima tako i u skladištima. Štetni efekti upotrebe plesnive hrane zabeleženi su još od davnina. Alimentarnim unošenjem toksina plesni nastaju intoksikacije tzv. mikotoksikoze koje, s obzirom da su vezane za hranu, mogu da poprime akutne i hronične razmere. Hronični efekti nastali upotrebom hrane kontaminirane mikotoksinima imaju veoma veliki uticaj na zdravlje ljudi i rezultuju kancerogenim, mutagenim, teratogenim, estrogenim, hemoragičnim, imunotoksičnim, nefrotoksičnim, hepatotoksičnim, dermatoksičnim i neurotoksičnim efektima.

Iako je problem kontaminacije hrane za ljude i životinje mikotoksinima još uvek prisutan u razvijenim zemljama, primenom novih dostignuća u poljoprivrednoj proizvodnji i odgovarajućom zakonskom regulativom, značajno je smanjena izloženost ljudi i životinja mikotoksinima. U zemljama u razvoju u kojima su klimatski faktori i uslovi skladištenja hrane, često povoljni za kolonizaciju plesni i sintezu mikotoksina veliki deo stanovništva orijentisan jena poljoprivrednu proizvodnju ili na snabdevanje iz neuslovnih objekata prodaje. Iz tog razloga proizvođači hrane i organi državne uprave su svoje aktivnosti usmerili ka implementaciji tačne i pouzdane procene stvarnog stanja kontaminacije hrane mikotoksinima, a u cilju dobijanja relevantnih podataka o uticaju mikotoksina na zdravlje ljudi i životinja.

Ključne reči: mikotoksini, zdravlje ljudi i životinja, analiza rizika

Introduction

Mycotoxins are a structurally diverse group of mostly small molecular weight compounds, produced by the secondary metabolism of some filamentous fungi or molds of the *Aspergillus*, *Penicillium*, *Fusarium*, *Alternaria* and *Claviceps* genera, which, under suitable temperature and humidity conditions, may develop on various foods and fe-

eds, causing serious risks for human and animal health. In structural complexity, mycotoxins vary from simple C₁ compounds, e.g. moniliformin, to complex substances such as the phomopsins (*Culvenor*, 1989) and the tremorgenic mycotoxins (*Steyn*, 1985). Although currently more than 300 mycotoxins are known, scientific attention is focused mainly on those that have proven to be carcinogenic and/or toxic. Human exposure to mycotoxins may

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTHOR: Dragan Milicevic, dragan@inmesbgd.com, Institute of Meat Hygiene and Technology, Kacanskog 13, 11000 Belgrade, Republic of Serbia

AUTOR: Dragan Milićević, dragan@inmesbgd.com, Institut za higijenu i tehnologiju mesa, Kaćanskog 13, 11 000 Beograd, Srbija

result from consumption of plant derived foods that are contaminated with toxins, the carryover of mycotoxins and their metabolites into animal products such as milk, meat and eggs or exposure to air and dust containing toxins (Jarvis, 2002; CAST, 2003). Human food can be contaminated with mycotoxins at various stages in the food chain and the three most important genera of mycotoxigenic fungi are *Aspergillus*, *Fusarium* and *Penicillium*. The principal classes of mycotoxins include a metabolite of *Aspergillus flavus* and *Aspergillus parasiticus*, aflatoxin B₁, the most potent hepatocarcinogenic substance known, which has been recently proven to be genotoxic; ochratoxin A, produced by *Penicillium verrucosum* and *Aspergillus ochraceus*, which is known to be carcinogenic in rodents and nephrotoxic in humans. Although its genotoxic power has so far not been definitively established; zearalenone, produced by various species of *Fusarium*, in particular *F. graminearum* and *F. culmorum*, which has an estrogenous action and is significantly toxic to the reproductive system of animals. The tricothecens, a group of numerous metabolites produced by *Fusarium*, *Stachybotris*, and *Cephalosporium* species, cause mainly dermatotoxicity, immunotoxicity, and gastrointestinal disturbances; and the fumonisins, produced mainly by *Fusarium moniliforme*, may induce leukoencephalopathy in equines as well as hepatotoxicity in rats (Pohland, 1987).

The impact of mycotoxins on health depends on the amount of the mycotoxin consumed, the toxicity of the compound, e.g. acute or chronic (e.g. carcinogenic) effects, the body weight of the individual, the presence of other mycotoxins (synergistic effects) and other dietary effects (Kuiper-Goodman, 1991). The incidence and extent of mycotoxin contamination are strictly related to geographic and seasonal factors as well as cultivation, harvesting, stocking, and transport conditions (WHO, 1979). The evaluation of the incidence and extent of contamination of foodstuffs is crucial and has, in fact, been taken into account for many years by the various disciplines that concur in the definition and management of the risk associated with these toxins and its management (Gleadle, *in press*).

Chemistry of mycotoxins

Aflatoxins

The aflatoxins, a group of closely related hepatocarcinogenic bisdihydrofurano metabolites, produced by certain strains of *Aspergillus flavus* and *Aspergillus parasiticus*, led to the resurgence of interest in all aspects of mycotoxicology. Aflatoxin B₁

(AFB₁) is the most carcinogenic of the aflatoxins, also, it is the most commonly occurring aflatoxin and has been said to be the most potent hepatocarcinogen to rats and mice. AFB₁ excreted in the milk of lactating cows has toxic properties similar to AFB₁; it is therefore of great public concern, particularly with regards to young children. The potent hepatocarcinogenicity of the aflatoxins led to extensive studies of their carcinogenic properties; detailed information was obtained on their world-wide occurrence in foods and feeds, and their putative role as causal factors for human PLC (primary liver cancer). IARC declared the aflatoxins in 1987 as human carcinogens; the classification was confirmed by re-evaluation in 1992. The need to control aflatoxin exposure is based on 2 major concerns: the adverse short and long-term effects of aflatoxin-contaminated commodities on human and animal health and the presence of aflatoxin residues or metabolites in animal tissues and milk used as human food.

Ochratoxins

Ochratoxin A (OTA) is a pentaketide-derived dihydroisocoumarin moiety linked via the 12-carboxy group by a peptide bond to L-phenylalanine. There are several OTA analogues, ochratoxins B, C, and alkyl esters of ochratoxins that have similar structure but are less toxic. OTA was the first mycotoxic compound isolated from *Aspergillus ochraceus*, and later it was found in other *Aspergillus* and *Penicillium* species such as *Penicillium verucosum*. OTA is a main contaminant of cereals (corn, barley, wheat) and to some extent beans (coffee, soy, and cocoa). *Aspergillus* species are associated with OTA production in tropical areas, whereas OTA producing *Penicillium* species thrive and can produce OTA in a colder climate with temperatures as low as 5°C. The toxicity of OTA involves several mechanisms. OTA inhibits protein synthesis by competing with the phenylalanine aminoacylation reaction catalyzed by Phe-tRNA synthase (Creppy, 1984). This results in inhibition of protein as well as DNA and RNA synthesis. OTA also disrupts hepatic microsomal calcium homeostasis by impairing the endoplasmic reticulum membrane via lipid peroxidation (Omar, 1991). OTA became regarded as a very important mycotoxin since it plays a major role in the nephropathy occurring both in human and animal, particularly in swine (Danish porcine nephropathy) and poultry.

Tricothecenes

The *Fusarium* fungi are probably the most prevalent toxin-producing fungi of the northern

temperate regions and are commonly found on cereals grown in the temperate regions of America, Europe and Asia. A variety of *Fusarium* fungi, which are common soil fungi, produce a number of different mycotoxins of the class of trichothecenes: T-2 toxin, HT-2 toxin, deoxynivalenol (DON) and nivalenol and some other toxins zearalenone and fumonisins. The trichothecenes are a family of related cyclic sesquiterpenoids, which are divided into four groups (types A–D) according to their characteristic functional groups. Type-A and –B trichothecenes are the most common. Type A is represented by HT-2 toxin and T-2 toxin and type B is most frequently represented by DON, 3-acetyl-DON (3-Ac-DON), 15-acetyl-DON (15-Ac-DON), nivalenol (NIV), and fusarenon X (FUS-X). Whereas type-B trichothecenes possess a carbonyl functionality at C-8, type-A trichothecenes lack the keto group at that position and have other oxygen functions at C-8 instead. This chemical characteristic and the fact that type-A trichothecenes generally have fewer hydroxyl groups makes the type-A trichothecenes less polar, which affects analytical procedures from extraction and clean-up up to separation and detection. Toxinogenic *fusaria* have been implicated in human health diseases such as ATA (Yagen, 1977), Kashin-Beck disease, akakabiyu (scabby grain intoxication) and esophageal cancer, as well as in a number of animal diseases such as skin toxicity, bone marrow damage, haemorrhagic and estrogenic syndrome (zearalenone), and equine leukoencephalomalacia (ELEM, fumonisins).

Tremorgenic mycotoxins

A brief survey of the structural properties of the fungal tremorgens, namely penitremes, janthitrems, lolitrems, aflatrems, paxilline, paspaline, paspalicine, paspalinine and paspalitrems A and B, reveals their close biogenetic relationship. In the case of aflatrems and paspalitrems A and B, a unit is attached to the paspaline-type structure (Steyn, 1985). Tryptophan (Trp) is a common constituent of many secondary metabolites, several affecting the central nervous system, such as the ergot alkaloids. Trp is the biogenetic precursor of the cyclopiazonic acids (Steyn, 1975), tremorgenic substances such as fumitremorgens A and B (Yamazaki, 1971) and verruculogen (Fayos, 1974). In the structurally related metabolites, the brevianamides and austamides (Steyn, 1973), Trp and proline contribute the dioxopiperazine part of the molecules. Trp is again a building block of the tetrapeptide metabolites, the tryptoquivalines, which contain in addition anthranilic acid, valine and methylalanine. L-Trp and L-histidine are the

precursors of the dioxopiperazines, oxaline (Nagel, 1976) and roquefortine (Gorst-Allman, 1982), metabolites of *Penicillium oxalicum* and *Penicillium roqueforti*, respectively. Roquefortine, a compound which affects the central nervous system, is also produced by *Penicillium camemberti* and is as such a frequent contaminant of some cheeses.

Fumonisins

Fumonisins are water-soluble mycotoxins that are produced by several species of *Fusarium*, but primarily *F. verticillioides* and *F. proliferatum*. At least 28 different FBs have been reported (Rheeder *et al.*, 2002). Three groups of FBs (A–C) have been identified based on structural similarities. Groups A and B are characterized by the presence of an amide and amine group, respectively. Group C is similar to the B-group, except for the absence of the methyl group at the C1-terminal (Cole *et al.*, 2003). Of all the FBs identified to date, the fumonisin B₁ (FB₁), fumonisin B₂ (FB₂) and fumonisin B₃ (FB₃) are the most important. FB₁ usually constitute about 70% of the total FBs content found in naturally contaminated foods and feeds. These molecules differ by lacking one of the free hydroxyl groups at either C-10 position (FB₂) or C-5 (FB₃). In addition, FBs analogues have been identified in some processed foods, following hydrolysis (e.g. nixtamalization) or reaction with food components (sugar, starch and proteins).

Zearalenone

Zearalenone (ZEA), 6-(10-hydroxy-6-oxo-*trans*-1-undecenyl)- β -resorcylic acid lactone; CAS 17924-92-4), is produced as a secondary metabolite by a number of *Fusarium* species including *F. culmorum*, *F. graminearum* (Hestbjerg *et al.*, 2002; Glenn, 2007), as well as *F. equiseti* and *F. crookwellense* (Bennett and Klich, 2003). These species are known to infest wheat, barley, rice, maize, and some other crops (Yamashita *et al.*, 1995; Jimenez and Mateo, 1997). Despite its non-steroidal structure, ZEA activates estrogen receptors resulting in functional and morphological alteration in reproductive organs. ZEA interacts not only with both types of estrogen receptors but is also a substrate for hydroxysteroid dehydrogenases, which convert it into two stereoisomeric metabolites, α -zearalenol and β -zearalenol. A second reduction step yields the two minor metabolites α -zearalanol and β -zearalanol. Alpha-hydroxylation results in an increase in estrogenic potency as compared to the parent compound, and the species-specific rate of alpha-hydroxylation may account for the

susceptibility of certain animal species, including pigs, towards ZEA exposure.

The topic of conjugated or masked mycotoxins first caught attention in the mid-1980s because in some cases of mycotoxicoses, clinical observations in animals did not correlate with the low mycotoxin content determined in the corresponding feed. The unexpected high toxicity could, for instance be attributed to the occurrence of undetected, conjugated forms of mycotoxins that hydrolyze to the precursor toxins in the digestive tracts of animals (Gareis, 1994). It was shown that plants can reduce the toxicity of mycotoxins either by chemical modification and/or by inclusion into the plant matrix (Wallnöfer *et al.*, 1996). This detoxification process includes the conjugation of mycotoxins to polar substances such as sugars, amino acids, or sulfate (Schneweis *et al.*, 2002) and subsequent storage of the conjugates in vacuoles. So far, the natural occurrence of a zearalenone glucoside in wheat has been reported (Langseth *et al.*, 1998). High-performance liquid chromatography (HPLC) combined with tandem mass spectrometry (MS/MS) offers a powerful tool for identification and characterization of mycotoxin conjugates (Berthiller *et al.*, 2005).

Mycotoxin exposure and effect on human and animal health

A wide range of commodities can be contaminated with mycotoxins (Table 1) both pre- and post-harvest. (CAST, 2003). Aflatoxins are found in maize and peanuts as well as in tree nuts and dried fruits. Ochratoxin A is found mainly in cereals, but significant levels of contamination may also occur in wine, coffee, spices and dried fruits. Fumonisin are found mainly in maize and maize based products. Tricothecenes are chiefly associated with grain, as is zearalenone. Available evidence suggests that tissue accumulation of mycotoxins, or their metabolites, is very low and that residues are excreted in a few days. The hydroxylated metabolite of aflatoxin B₁, aflatoxin M₁, is excreted into milk from 1 to 6% of dietary intake. (Van Egmond, 1989, Veldman, 1992) Ochratoxin A has been detected in blood, kidneys, liver and muscle tissue from pigs in several European countries. (Leistner, 1984, Van Egmond, 1994, Milićević, 2008). Residues of cyclopiazonic acid (CPA), a co-contaminant with aflatoxin, have been found in meat, milk and eggs. (Bryden, 2001). After an extensive review of the literature, Pestka (1995) concluded that trace levels of mycotoxins and their metabolites may carry over into the edible tissue (meat) of food producing animals. However, he concluded that to date there is no evidence to

suggest that the levels of transmitted mycotoxins pose a threat of acute toxicity.

AFB₁ has been extensively linked to human primary liver cancer (PLC) in which it acts synergistically with HBV infection and was classified by the International Agency for Research on Cancer (IARC) as a human carcinogen (group 1 carcinogen), (IARC, 1993a). This combination represents a heavy cancer burden in developing countries. A recent comparison of the estimated population risk between Kenya and France highlighted the greater burden that can be placed on developing countries (Shephard, 2006). Based on respective estimates for aflatoxin exposure of 133 and 0.12 ng kg⁻¹ body weight day⁻¹ and respective HBV prevalence of 25 and 1%, the liver cancer risk would be 11 vs. 0.0015 cancers per year per 100.000 population, respectively. Given recently published liver cancer incidence rates in the European Union of 10.0 per 100.000 for males and 3.3 per 100.000 for females (Bray *et al.* 2002), it is clear that aflatoxin plays a significant role in liver cancer in developing countries, but not in the developed world where other risk factors such as cirrhosis are more important. Fumonisin have been implicated in one incident of acute food-borne disease in India in which the occurrence of borborygmy, abdominal pain, and diarrhea was associated with the consumption of maize and sorghum contaminated with high levels of fumonisins (Bhat *et al.* 1997). Fumonisin B₁, the most abundant of the numerous fumonisin analogues, was classified by the IARC as a group 2B carcinogen (possibly carcinogenic in humans), (IARC, 2002). Studies in the former Transkei region of South Africa and in Linxian and Cixian counties, China, have demonstrated an association between fumonisin exposure in rural subsistence farming areas and a high incidence of oesophageal cancer as well as with field outbreaks of ELEM in many countries such as Egypt, South Africa and the United States of America (Marasas, 1988) and pulmonary oedema in swine (Ross, 1990). ELEM is a fatal neurological disease of horses, characterized by liquefactive necrosis of the white matter of the brain. ELEM has been experimentally induced in horses by, either supplementing their diets with *F. moniliforme*-contaminated corn, or by the oral administration of fumonisin B, (FB₁), a toxin produced by *F. moniliforme* (Kellerman, 1990). Fumonisin, which inhibit the uptake of folic acid via the folate receptor (Stevens and Tang, 1997), have also been implicated in the high incidence of neural tube defects in rural populations known to consume contaminated maize, such as the former Transkei region of South Africa and areas of Northern China (Marasas *et al.* 2004). The other three agriculturally

important mycotoxins have also been associated with various outbreaks of human disease, mostly in developing countries. A number of occurrences of acute food-borne illness in India and China involving gastrointestinal symptoms have been attributed to the consumption of DON-contaminated cereals (Luo, 1988; Bhat *et al.* 1989). OTA has long been associated with Balkan endemic nephropathy (BEN), a fatal renal disease with histopathological similarities to OTA-induced nephropathy in swine and has been associated with the incidence of epithelial tumours of the upper urinary tract (Benford *et al.* 2001; Castegnaro *et al.* 2006). OTA was classified by the IARC as possibly carcinogenic to humans (group 2B carcinogen), (IARC, 1993b). ZON is a naturally occurring endocrine-disrupting chemical and has been associated with clinical manifestations of hyper-oestrogenism in humans and animals, including an outbreak of precocious pubertal changes in young children in Puerto Rico in the Caribbean (Saenz de Rodrigues *et al.* 1985) and gynecomastia with testicular atrophy in rural males in southern Africa (Campbell, 1991).

tained by monitoring food data since the latter may, as stated, be affected by sampling, subsampling, and analysis errors.

The role of sampling and analysis in mycotoxin contamination

The correct evaluation of mycotoxin contamination in foodstuffs depends principally on the degree of accuracy associated with the single steps by which this information is obtained. Because of the highly heterogeneous distribution of mycotoxins in a lot, taking a representative sample is the most critical stage. In Fig. 1 are presented the errors associated with sampling for Aflatoxins analysis (expressed as a coefficient of variation). From this it can be seen that the error associated with sampling procedures is notably higher than that associated with subsampling or analysis.

The most prominent reason for collecting food samples for the investigation of contaminants, such as mycotoxins, is to protect consumer health,

Table 1. Some human diseases in which mycotoxins have been implicated

Tabela 1. Neka oboljenja ljudi povezana sa mikotoksinima

Disease	Mycotoxin source	Fungus
Akakabio-byo	Wheat, barley, oats, rice	<i>Fusarium spp.</i>
Alimentary toxic aleukia	Cereal grains (toxic bread)	<i>Fusarium spp.</i>
Balkan nephropathy	Cereal grains	<i>Penicillium spp.</i>
Cardiac beriberi	Rice	<i>Aspergillus spp., Penicillium spp.</i>
Celery harvester's disease	Celery (Pink rot)	<i>Sclerotinia</i>
Ergotism	Rye, cereal grains	<i>Claviceps purpurea</i>
Hepatocarcinoma	Cereal grains, peanuts	<i>Aspergillus flavus, A. parasiticus</i>
Kwashiorkor	Cereal grains	<i>Aspergillus flavus, A. parasiticus</i>
Neural tube defects	Maize	<i>Fusarium verticillioides, F.proliferatum</i>
Oesophageal tumors	Corn	<i>Fusarium verticillioides, F.proliferatum</i>
Onyalai	Millet	<i>Phoma sorghina</i>
Reye's syndrome	Cereal grains (grain dust)	<i>Aspergillus</i>
Stachybotryotoxicosis	Cereal grains, (grain dust)	<i>Stachybotrys atra</i>

The evaluation of mycotoxins in biological fluids can provide useful indications of the dietary intake of mycotoxins. This approach can also constitute a valid, although indirect, evaluation of mycotoxin contamination in foodstuffs. This methodology, in fact, can somehow give a better estimate of the exposure of humans to mycotoxins than that ob-

mainly verifying the compliance of food and feed with acceptable safety standards. Sampling is one of the most crucial, but underestimated parts of the multifaceted and complex bulk of activities aimed at addressing and managing food issues. In practice, the overall objective of good sampling is to provide reliable samples to be analyzed that can represent the

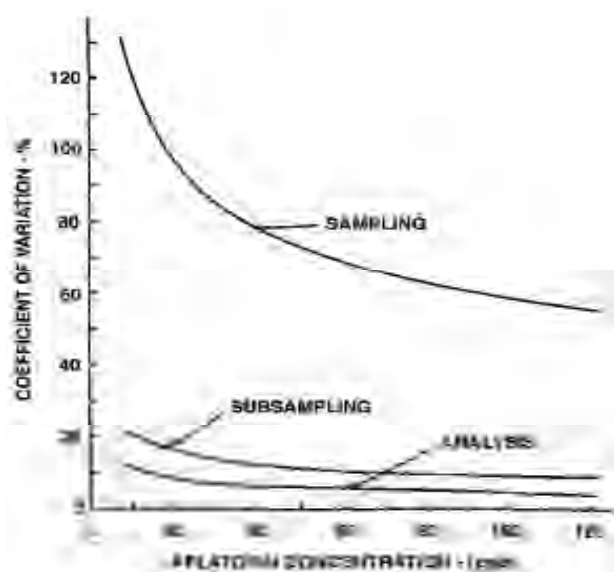


Figure 1. Coefficient of variation characterizing sampling, subsampling and analysis as a function of Aflatoxins concentration

Slika 1. Koeficijent varijacije koji karakteriše uzorkovanje, poduzorkovanje i analizu u funkciji koncentracije alfatoksina

basis for “fit for purpose” investigations. In most cases, meaningful sampling is a process comprising two very dissimilar steps:

(1) The first step (hereafter referred to as “primary sampling”) consists in taking the decision on “why, where and when” to collect the samples. In other words, the process of “statistically” locating the sites (populations) from which food samples should be taken;

(2) The second step (hereafter referred to as “secondary sampling”) consists of establishing how samples should be collected in order to be representative of the lot under investigation. For both steps the quality and the consequent reliability of the data are strongly dependent on the available resources and on the skill of the people involved.

For this class of contaminants, the need for statistically-based planning is particularly relevant for: (i) The multifaceted implications of mycotoxin contamination (health, trade, ethical issues related to developing countries’ difficulties), and (ii) the largely inhomogeneous distribution of the toxins within food commodities, with the consequent need for careful secondary sampling. Appropriate sampling plans are essential to ensure that the analytically-derived mean concentration of a sample is representative of the true mean concentration of a lot. Sampling plans are particularly relevant in the area of mycotoxins where it is known that the contamination of a commodity can be heterogeneously distri-

buted. Good primary sampling schemes have so far been developed for several classes of contaminants, such as dioxins and pesticides (*South et al.* 2004), in contrast to the very few valid ones so far proposed for mycotoxins. In contrast, a large number of papers have appeared, related to secondary sampling schemes for aflatoxin B₁ (particularly on its distribution in a lot and on related sampling plans), (*Whitaker et al.* 1979, 1994), but only a few studies deal with some *Fusarium* toxins (*Hart and Schabenberger* 1998; *Whitaker et al.* 1998; *Whitaker et al.* 2000). Conversely, specific studies focused on the distribution of OTA-contaminated units are not yet available, apart from the vague assumption that “representative sampling” for aflatoxins is more difficult than sampling for other known mycotoxins in food products. Sampling procedures recommended for aflatoxins should thus be adequate for other mycotoxins (*Dickens and Whitaker* 1982). Nevertheless, the European legislation dealing with sampling and methods of analysis of mycotoxins for official control was recently adopted (*EC*, 2006).

In conclusion, the analysis of sources of errors in evaluating the impact of mycotoxins on human health should be carefully performed taking into account many aspects such as planning and accomplishment of monitoring programs, consumer’s health protection, economic, political and commercial considerations.

Analysis

Legislation calls for monitoring methods. Reliable analytical methods must be available to enable enforcement of the regulations in daily practice. In addition to reliability, simplicity is desired, as it will affect the amount of data generated and the practicality of the ultimate measures taken. The reliability of mycotoxin analysis data can be improved by use of interlaboratory-validated methods of analysis (e.g. the methods of AOAC International and methods standardized by CEN). These methods have been largely developed in response to planned regulations for mycotoxins or regulations that came into force. The requirements for these methods were dictated by the needs, i.e. they had to be suitable for the (planned) regulated mycotoxin–matrix combination(s). The limits of determination of the methods had to be demonstrated to be low enough for precise and accurate determination of the mycotoxins of interest at regulatory levels. Methods were also developed and validated for toxin–matrix combinations for which there were no regulations (yet), but for which the scientific community saw a need, e.g. for surveillance purposes. These developments eased

the establishment of specific mycotoxin regulations. AOAC currently has approximately 45 analytical methods for determination of mycotoxins (AOAC, 2005). All have undergone extensive testing in interlaboratory validation studies, and subsequent review by the AOAC's rigorous approval process. AOAC methods are referred to as official methods in mycotoxin legislation in a few dozen countries (FAO, 2004). In Europe, CEN methods are becoming increasingly important. Ten mycotoxin methods have been standardized by the CEN, and this number will grow substantially in the years to come. Although CEN mycotoxin methods are not mandatory for official food control in the EU, all CEN mycotoxin methods can be used in the EU for official food-control purposes because their performance characteristics fulfill the criteria laid down in the EU regulation for sampling and analysis (EC, 2006). One of them, high performance liquid chromatography (HPLC) with different detectors, is frequently used both for routine analyses and as a confirmatory method for novel or screening techniques. For some mycotoxins, e.g. trichothecenes, gas chromatography (GC) is the method more often used (Krska, 2001). Except for direct mass spectrometric methods, all the other analytical methods used for mycotoxin determination are, either immunoassay based, or otherwise fall into the category of direct or indirect screening methods. The use of good, validated methods of analysis is no guarantee that reliable analytical results will be obtained in mycotoxin determination. Analytical quality assurance (AQA) is another prerequisite for adequate food-law enforcement. AQA includes, where possible, the use of (certified) reference materials (e.g. CRMs supplied by the European Commission's Joint Research Centre/Institute for Reference Materials and Measurements; JRC/IRMM, see <http://www.irmm.jrc.be>).

Factors affecting the mycotoxin regulations

Regulations relating to mycotoxins have been established in many countries to protect the consumer from the harmful effects of these compounds. Different factors play a role in the decision-making process of setting limits for mycotoxins. These include:

- the availability of toxicological data on mycotoxins,
- the availability of exposure data on mycotoxins,
- knowledge of the distribution of mycotoxins concentrations within commodity or product lots,
- the availability of analytical methods,
- legislation in other countries with which trade contacts exist,
- the need for sufficient food supply.

The first two factors provide the information necessary for hazard assessment and exposure assessment, respectively, the main bases of risk assessment. Risk assessment is the scientific evaluation of the probability of occurrence of known or potential adverse health effects resulting from human exposure to food-borne hazards. It is the primary scientific basis for promulgation of regulations. The third and fourth factors are important factors enabling practical enforcement of mycotoxin regulations through adequate sampling and analysis procedures. The last two factors are merely socio-economic in nature but are equally important in the decision-making process to establish meaningful regulations and limits for mycotoxins in food and feed. Risk assessment regulations are primarily based on known toxic effects. For the mycotoxins currently considered most significant (aflatoxins B₁, B₂, G₁ and G₂; aflatoxin M₁; ochratoxin A; patulin; fumonisins B₁, B₂ and B₃; zearalenone; T-2 and HT-2 toxins; and deoxynivalenol), the Joint Expert Committee on Food Additives (JECFA—a scientific advisory body of the World Health Organization WHO and the Food and Agriculture Organization FAO has evaluated their hazard in several sessions (WHO, 1999, 2000, 2002). In February 2001 a special JECFA session was devoted to entirely mycotoxins. Two reports have appeared on this session, a longer version (FAO, 2001) and a shorter version (WHO, 2002). These reports provide good and detailed insight into the process of risk assessment of mycotoxins. The reports addressed several concerns about the mycotoxins considered—their properties and metabolism, toxicological studies, and final risk evaluation. With the mycotoxin evaluations the Committee discussed general considerations on sampling, analytical methods, associated intake issues and control. Risks associated with mycotoxins depend on both hazard and exposure. The hazard of mycotoxins to individuals is probably, more or less, the same all over the world (although other factors are, sometimes, also important, e.g. hepatitis B virus infection in relation to the hazard of aflatoxins). Exposure is not the same, because of different levels of contamination and dietary habits in various parts of the world. Risk analysis framework for food safety is illustrated in Fig. 2.



Figure 2. Risk analysis framework for food safety
Slika 2. Okvir analize rizika za bezbednost hrane

The international mycotoxin regulatory situation

Since the discovery of the aflatoxins in 1960 and subsequent recognition that mycotoxins are of significant health concern to both humans and animals, regulations gradually developed for mycotoxins in food and feed. In the early days of mycotoxin regulations these measures focused mainly on the aflatoxins. They were established by industrialized countries and limits often had an advisory or guideline character. Over the years, the number of countries with known specific mycotoxin regulations has increased from 33 in 1981 (*Schuller, 1983*) to 56 in 1987 (*Van Egmond, 1989*), 77 in 1995 (*FAO, 1997*), and 100 in 2003 (*FAO, 2004*). Current regulations encompass 13 different mycotoxins or groups of mycotoxins and specific limits have been established for many food and feed commodities and products. Until the late 1990s setting of mycotoxin regulations was mostly a national affair. Gradually, several economic communities e.g. EU, European Union; MERCOSUR Mercado Común del Sur; Australia and New Zealand harmonized their mycotoxin regulations, thereby overruling existing national regulations. Current regulations are increasingly based on scientific opinions of authoritative bodies, for example the FAO/WHO Joint Expert Committee on Food Additives of the United Nations (JECFA) and the European Food Safety Authority (EFSA). At

the same time, requirements for adequate sampling and analytical methods put high demands on other professional organizations, for example AOAC International and the European Standardization Committee (CEN).

Economic impact

Mycotoxin contamination of the food chain has a major economic impact. However, the insidious nature of many mycotoxicoses makes it difficult to estimate incidence and cost (*CAST, 1989*). In addition to crop losses and reduced animal productivity, costs are derived from the efforts made by producers and distributors to counteract their initial loss, the cost of improved technologies for production, storage and transport, the cost of analytical testing, especially as detection or regulations become more stringent, and the development of sampling plans (*Whitaker, 1995*). There is also a considerable cost to society as a whole, in terms of monitoring; extra handling and distribution costs, increased processing costs and loss of consumer confidence in the safety of food products. It is estimated that in developing countries, the greatest economic impact is associated with human health. (*Miller, 1998*). Delineating economic impact reflects the complexity of a mycotoxin contamination within the food chain. There is a clear need to protect consumers through regulations but

at what cost? A comprehensive risk and economic analysis of lowering the acceptable levels for fumonisins and aflatoxin in world trade demonstrated that the United States would experience significant economic losses from tighter controls (Wu, 2004). The developing countries, China and Argentina, were more likely to experience greater economic losses than sub-Saharan Africa. The disturbing outcome of this detailed analysis was that tighter controls were unlikely to decrease health risks and may have the opposite effect (Wu, 2004). In other words, very stringent international trade regulations could lead to the situation where exporting countries, especially developing countries, would retain higher risk commodities which would subsequently be available for their own populations; communities which are already exposed to higher levels of mycotoxins than consumers in developed countries.

Strategies to Prevent Mycotoxin Contamination of Food and Animal Feed

Many strategies to prevent mycotoxin contamination of food and animal feed have been developed (Rustom, 1997; Yilmaz, 2001). It is clear that mycotoxins can contaminate agricultural produce, both in the field as well as during storage. The use of pre-harvest control strategies for such resistance varieties, field management, the use of biological and chemical agents, harvest management and post-harvest applications, including improving drying and storage conditions, together with the use of natural and chemical agents and irradiation have clearly been shown to be important in the prevention of mycotoxigenic mould growth and mycotoxin formation (CAC, 2002). The importance of drying and moisture control during storage is generally well understood by the industry, in terms of the importance of prevention of fungal contamination. Interesting results have been reported on the potential use of biocompetitive agents in different biological control strategies to prevent the pre-harvest aflatoxin contamination of crops, such as peanuts, rice, maize, and cottonseed. It is clear that much more work must be conducted to identify various crop genotypes which are resistant to mycotoxigenic fungus infection and subsequently mycotoxin formation. It is also clear that a combination of the development of crop species with resistance to toxigenic fungi and biocompetitive non-mycotoxigenic strain technologies may yield one of the most effective strategies for prevention of mycotoxin contamination (Gendloff, 1986; Reid, 1994). Several natural plant extract and spice oils of eugenol, cinnamon, oregano, oni-

ons, lemongrass, (Yin, 1998, Juglal, 2002) tumeric, mint, and chemical compounds (fungicide, herbicide, and surfactant) are known to prevent both mycotoxigenic mould growth and mycotoxin formation during post-harvest season. In addition to application of plant extracts and chemical agents as well as antagonistic microorganisms, such as lactic acid bacteria with their antifungal properties, seem to be potentially very effective in the prevention of mycotoxin formation. The precise antifungal properties of lactic acid bacteria are still largely unresolved but may involve microbial competition (El-Gendy, 1981), as well as extracellular metabolites which are heat-stable and of low molecular weight. Again, further investigations are clearly needed to gain a better understanding of this antifungal action. Various physical and chemical strategies have also been developed to help prevent mycotoxin contamination, including physical separation, extraction with sorbents, and adsorption (Sinha, 1998). The fluorescence sorting of maize, cottonseed and figs by examination under UV light is known to be the cheapest and the simplest acceptable way for the screening of aflatoxins. It is clear that no single currently available physical or chemical detoxification method will be suitable for all foods and animal feeds. The effectiveness of a method in the detoxification of mycotoxins depends on the nature of the food, environmental conditions such as moisture content, temperature, as well as the type of mycotoxin, its concentration and the extent of binding between mycotoxin and constituents. While a range of chemical compounds, including hydrochloric acid, ammonia, hydrogen peroxide, O₃, sodium bisulfite, and chlorine seem to hold great potential in the detoxification of mycotoxins, unfortunately their use significantly decreases the nutritional value of the foods or produces toxic derivatives in the treated product with undesirable sensory properties. This will severely limit their widespread use. At the same time it should be noted that chemical treatment is not allowed within the EC for commodities destined for human consumption. Recently, there has been an increasing interest in the use of bacteria, yeast, and fungi to help reduce the toxic effect of mycotoxins (Bata, 1999). While most studies to date on mycotoxin detoxification by microorganisms have been undertaken under laboratory conditions, there is data on the effective use of *F. aurantiacum* in the detoxifying AFB₁ from various food products, including milk, peanuts, maize, and red pepper without leaving toxic end products. One potential drawback here is the production of a bright orange pigment by the organism which restricts its use in the detoxification of food and in feed fermentations.

The most recent approach to the problem has been the use of mycotoxin-binding agents in the diet that sequester the mycotoxin in the gastrointestinal tract thus reducing their bioavailability. Although AC, HSCAS, aluminosilicate, zeolite, and bentonite have shown good potential for use in the animal feed to help overcome aflatoxicosis, the future *in vivo* investigations must focus on other problematic mycotoxins. Interestingly lactic acid bacteria and bifidobacteria have been shown to bind AFB₁, but mechanistic studies need to be conducted on the precise binding mechanism, while the conditions favoring the release of bound toxin molecules need to be investigated as well.

Concluding comments

Mycotoxins are a food safety risk globally. International risk assessments have been performed by JECFA (1998, 2001) for aflatoxin B₁, aflatoxin M₁, DON, fumonisins, ochratoxin A, T-2 toxin and HT-2 toxin. These analyses indicate that health risks from mycotoxins are generally orders of magnitude lower in developed countries than for populations from developing regions. The scope of the mycotoxin problem is readily understood

when it is appreciated that there are many thousand secondary fungal metabolites (Cole, 2003), the vast majority of which have not been tested for toxicity or associated with disease outbreaks. In developing countries it is likely that consumers will be confronted with a diet that contains a low level of toxin and in many cases, there may be other toxins present. For example, aflatoxins, fumonisins, DON and zearalenone may occur together in the same grain; many fungi produce several mycotoxins simultaneously, especially *Fusarium* species (Cole, 2003). Co-occurrence of mycotoxins is of special concern, for instance, in the case of fumonisins (apotent cancer promoter) and aflatoxin (a potent human carcinogen) where a complimentary toxicity mechanism of action occurs (Riley, 1998). In Africa and Asia the co-occurrence of these mycotoxins is common and a significant percentage of the population is infected with Hepatitis B or C which leads to the conclusion that mycotoxins in these regions can have devastating human health effects. Implicit with these conclusions are the existence of syndromes of apparently unknown aetiology and epidemiology that may involve mycotoxins and the difficulty of establishing „no effect“ levels for mycotoxins.

References

- AOAC International 2005. Official Methods of Analysis of AOAC International, 18th edn. AOAC International, Gaithersburg, USA;
- Bata, A., L'aszity, R., 1999. Detoxification of mycotoxin-contaminated food and feed by microorganisms. Trends Food Sci. Technol., 10:223–228, with permission from Elsevier;
- Benford, D., Boyle, C., Dekant, W., Fuchs, R., Gaylor, D.W., Hard, G., McGregor, D.B., Pitt, J.I., Plestina, R., Shephard, G., Verger, P.J.P., Walker, R., 2001. Ochratoxin A. In: Safety evaluation of certain mycotoxins in food. WHO Food Additives Series No. 47, FAO Food and Nutrition Paper No. 74, Prepared by the 56th Meeting of the Joint FAO/WHO Expert Committee on Food Additives (JECFA), Geneva, WHO, pp 281–415;
- Bennett, J.W., Klich, M., 2003. Mycotoxins. Clin. Microbiol. Rev., 16, 497–516;
- Berthiller, F., Dall'Astra, C., Schumacher, R., Lemmens, M., Adam, G., Kraska, A. R., 2005. Masked mycotoxins: determination of a deoxynivalenol glucoside in artificially and naturally contaminated wheat by liquid chromatography–tandem mass spectrometry. J. Agric. Food Chem. 53 (9), 3421–3425;
- Bhat, R.V., Beedu, S.R., Ramakrishna, Y., Munshi, K. L., 1989. Outbreak of trichothecene mycotoxicosis associated with consumption of mould-damaged wheat production in Kashmir Valley, India, Lancet, 35–37;
- Bhat, R. V., Shetty, P.H., Amruth, R.P., Sudershan, R.V., 1997. A foodborne disease outbreak due to the consumption of moldy sorghum and maize containing fumonisin mycotoxins, Journal of Toxicology Clinical Toxicology, 35, 249–255;
- Bray, F., Sankila, R., Ferlay, J., Parkin, D. M., 2002. Estimates of cancer incidence and mortality in Europe in 1995, European Journal of Cancer, 38, 99–166;
- Bryden, W. L., Logrieco, A., Abbas, H. K., Porter, J. K., Vesonder, R. F., Richard, J. L., Cole, R. J., Other significant *Fusarium* mycotoxins, 2001. In: Summerell B.A., Leslie J.F., Backhouse D., Bryden W.L. and Burgess L.W. eds. *Fusarium*: Paul E. Nelson Memorial Symposium. APS Press, St Paul, Minnesota, pp. 360–392;
- Campbell G. D., 1991. Trichothecene mycotoxicosis B a new entity? South African Medical Journal, 80, 361–362;
- CAST, 1989. Mycotoxins: Economic and Health Risks. Report No. 116. Council for Agricultural Science and Technology, Ames, Iowa, USA.
- CAST, 2003. Mycotoxins: Risks in Plant, Animal and Human Systems, Report No. 139, Council for Agricultural Science and Technology, Ames, Iowa, USA;
- Castegnaro, M., Canadas, D., Vrabcheva, T., Petkova-Boccharova, T., Chernozemsky, I. N., Pfohl-Leschkowicz, A., 2006. Balkan endemic nephropathy: Role of ochratoxin A through biomarkers. Molecular Nutrition and Food Research, 50, 519–529;
- Codex Alimentarius Commission, 2002. Proposed draft code of practice for the prevention (reduction) of mycotoxin contamination in cereals, including annexes on ochratoxin A, zearalenone, fumonisins and trichothecenes, CX/FAC 02/21, Joint FAO/WHO Food Standards Programme, Rotterdam, The Netherlands;

- Cole, R. J., Jarvis, B. B., Schweikert, M. A., 2003.** Handbook of Secondary Fungal Metabolites, vol. III. Academic Press, New York;
- Commission of the European Communities, 2006.** Commission Regulation (EC) No. 401/2006 of 23 February 2006 laying down the methods of sampling and analysis for the official control of the levels of mycotoxins in foodstuffs, Official Journal of the European Union, L70, 12–34;
- Creppy, E. E., Roschenthaler, R., Dirheimer, G., 1984.** Inhibition of protein synthesis in mice by ochratoxin A and its prevention by phenylalanine. *Food Chem Toxicol.* 22(11):883–886;
- Culvenor, C. C. J., Edgar, J. A., Mackay, M. F., Gorst-Allman, C. P., Marasas, W. F. O., Steyn, P. S., Vleggaar, R., Wessels, P. L., 1989.** Structure elucidation and absolute configuration of phomopsisin A, a hexapeptide mycotoxin produced by *Phomopsis leptostromiformis*, *Tetrahedron* 45, 2351–2372;
- Dickens, J. W., Whitaker, T. B., 1982.** Sampling and sample preparation, In: International Agency for Research on Cancer, Selected methods of analysis, Switzerland: International Agency for Research on Cancer, Lyon, p 17;
- Engelhardt, G., Ruhland, M., Wallnöfer, P. R., 1999.** Metabolism of mycotoxins in plants. *Adv. Food Sci.*, 21, 71–78;
- El-Gendy, S. M., Marth, E. H., 1981.** Growth of aflatoxin production by *Aspergillus parasiticus* in the presence of *Lactobacillus casei*, *J. Food Prot.*, 44:211–212, with permission from Journal of Food Protection;
- European Commission (EC), 2006.** Commission Directive No. 401/2006 Sampling method. *Off. J. EU Communities*, L 70/12;
- Fayos, J. D., Lokensgard, D., Clardy, J., Cole, R. J., Kirksey, J. W., 1974.** Structure of verruculogen, a tremor-producing peroxide from *Penicillium verruculostm*, *J. Am. Chem. Soc.* 96, 6785;
- Food and Agriculture Organization, 1997.** Worldwide regulations for mycotoxins 1995, A compendium. *FAO Food and Nutrition Paper* 64. Food and Agriculture Organization of the United Nations, Rome, Italy;
- Food and Agriculture Organization, 2001.** Safety Evaluation of Certain Mycotoxins in Food, Prepared by the Fifty-sixth meeting of the Joint FAO/WHO Expert Committee on Food Additives;
- (JECFA), FAO Food and Nutrition Paper 74,** Food and Agriculture Organization of the United Nations, Rome, Italy, p 705;
- Food and Agriculture Organization, 2004.** Worldwide regulations for mycotoxins in food and feed in 2003, *FAO Food and Nutrition Paper* 81, Food and Agriculture Organization of the United Nations, Rome, Italy;
- Gareis, M., Maskierte Mykotoxine, 1994.** *Übers. Tierernährung*, 22, 104–113;
- Gendloff, E. H., Rossmann, E. C., Casale, W. L., Isleib, T. G., Hart, L. P., 1986.** Components of resistance to *Fusarium* ear rot in field corn, *Phytopathology*, 76, 684–688;
- Gleadle A. E., Morthby, E. M., Hatch, A., Burt, R., 1988.** Scientific Co-operation Task on Aflatoxins: the co-ordinators view. In *Mycotoxins and Phycotoxins: Developments in Chemistry, Toxicology and Food Safety* (M. Miraglia, H. van Egmond, C. Brera, and J. Gilbert, Eds.), Alaken Inc., (in press).
- Glenn, A. E., 2007.** Mycotoxigenic *Fusarium* species in animal feed, In: Morgavi, D.P., Riley, R.T. (Eds.), *Fusarium Toxins: Presence in Feeds and Toxic Effects in Animals*, *Anim. Feed Sci. Technol.*;
- Gorst-Allman, C. P., Steyn, P. S., Vleggaar, R., 1982.** The biosynthesis of roquefortine, An investigation of acetate and mevalonate incorporation using high field NMR spectroscopy, *J. Chem. Soc. Chem. Commun.* 652;
- Hart, L. P., Schabenberger, O., 1998.** Variability of vomitoxin in truckloads of wheat in a wheat scab epidemic year, *Plant Disease*, 82, 625–630;
- Hestbjerg, H., Nielsen, K. F., Thrane, U., Elmholt, S., 2002.** Production of trichothecenes and other secondary metabolites by *Fusarium culmorum* and *Fusarium equiseti* on common laboratory media and a soil organic matter agar: an ecological interpretation, *J. Agric. Food Chem.*, 50, 7593–7599;
- International Agency for Research on Cancer (IARC), 1993a.** Some naturally occurring substances: Food items and constituents, heterocyclic aromatic amines and mycotoxins. Aflatoxins, WHO IARC Monographs on the Evaluation of Carcinogenic Risks to Humans, Vol. 56, Lyon, IARC, pp 245–395;
- International Agency for Research on Cancer (IARC), 1993b.** Some naturally occurring substances: Food items and constituents, heterocyclic aromatic amines and mycotoxins, Ochratoxin A. WHO IARC Monographs on the Evaluation of Carcinogenic Risks to Humans, Vol. 56, Lyon, IARC, pp 489–521;
- International Agency for Research on Cancer (IARC), 2002.** Some traditional herbal medicines, some mycotoxins, naphthalene and styrene, Fumonisin B₁. WHO IARC Monographs on the Evaluation of Carcinogenic Risks to Humans, Vol. 82, Lyon, IARC, pp 301–366;
- Jarvis BB., 2002.** Chemistry and toxicology of molds isolated from water-damaged buildings, *Mycotoxins and Food Safety, Adv. Expt. Med. Biol.*, 504, 43–52;
- JECFA (Joint FAO/WHO Expert Committee on Food Additives), 1998.** Safety evaluation of certain food additives and contaminants, Aflatoxins. WHO Food Additives Series 40, WHO, Geneva Switzerland, pp. 359–468;
- JECFA (Joint FAO/WHO Expert Committee on Food Additives), 2001.** Safety evaluation of certain mycotoxins in food, Aflatoxins. WHO Food Additives Series 47, WHO, Geneva Switzerland, pp. 701p;
- Jimenez, M., Mateo, R., 1997.** Determination of mycotoxins produced by *Fusarium* isolates from banana fruits by capillary gas chromatography and high-performance liquid chromatography, *J. Chromatogr. A* 778, 363–372;
- Juglal, S., Govinden, R., Odhav, B., 2002.** Spice oils for the control of co-occurring mycotoxin-producing fungi, *J. Food Prot.*, 65:683–687, with permission from Journal of Food Protection 683–687;
- Kellerman, T. S., Marasas, W. F. O., Thtel, P. G., Gelderblom, W. C. A., Cawood, M., Coetzer, J. A.W., 1990.** Leukoencephalomalacia in two horses induced by oral dosing of fumonisin B₁, *Onderstepoort J. Vet. Res.*, 57, 269–275;
- Kraska, R., Baumgartner, S., Josephs, R., 2001.** The state-of-the-art in the analysis of type-A and -B trichothecene mycotoxins in cereals, *Fresenius J. Anal. Chem.*, 371, 285–299.
- Kuiper-Goodman, T., 1991.** Risk assessment to humans of mycotoxins in animal derived food products. *Vet. Hum. Toxicol.* 33(4), 325–333;
- Leistner, L., Toxicogenic Penicillia occurring in feeds and foods, 1984.** In: Kurata H. and Ueno Y. eds. *Toxicogenic Fungi – Their Toxins and Health Hazard*, Elsevier Science Publishing Company, Inc., New York, pp. 162–171;
- Luo, X.Y., 1988.** Outbreaks of moldy cereals poisoning in China, In: *Issues in food safety*, Washington, DC: Toxicology Forum, pp 56–63;
- Marasas, W.F.O., Riley, R.T., Hendricks, K.A., Stevens, V.L., Sadler, T.W., Gelineau-van Waes, J., Missmer, S.A.,**

- Cabrera, J., Torres, O., Gelderblom, W.C.A., 2004.** Fumonisin disrupt sphingolipid metabolism, folate transport, and neural tube development in embryo culture and in vivo, A potential risk factor for human neural tube defects among populations consuming fumonisin-contaminated maize, *Journal of Nutrition*, 134, 711–716;
- Marasas, W.F.O., Jaskiewicz, K., Venter, F. S., Van Schalkwyk, D. J., 1988.** *Fusarium moniliforme* contamination of maize in esophageal cancer areas in Transkei, *S. Afr Med. J.* 73, 110–114;
- Miller, J. D., 1998.** Global significance of mycotoxins. In: Miraglia M., van Egmond H., Brera C., Gilbert J. eds. *Mycotoxins and Phycotoxins – Developments in Chemistry, Toxicology and Food Safety* Alaken Inc., Ford Collins, Colorado, pp.3–15;
- Milićević, D., Verica Jurić, Stefanović, S., Jovanović, M., Janković, S., 2008.** Survey of Slaughtered Pigs for Occurrence of Ochratoxin A and Porcine Nephropathy in Serbia, *International Journal of Molecular Sciences*, 9, 2169–2183;
- Nagel, D. W., Pachler, K. G. R., Steyn, P. S., Vleggaar, R. Wessels, P. L., 1976.** The chemistry and ¹³C NMR assignments of oxaline, a novel alkaloid from *Penicillium o.uzlicum*, *Tetrahedron* 32, 2625;
- Omar, R.F., Rahimtula, A.D., 1991.** Role of cytochrome P-450 and in ochratoxin A-stimulated lipid peroxidation, *J Biochem Toxicol.*, 6(3), 203–209;
- Pestka, J. J., 1995.** Fungal toxins in raw and fermented meats, In: Campbell-Platt, G. and Cook P.E eds. *Fermented Meats*. Blackie Academic and Professional, Glasgow, U. K. pp. 194-216;
- Pohland, A. E., Wood, G. E., 1987.** Occurrence of mycotoxins in food, In *Mycotoxins in Food* (P. Krogh, Ed.), pp. 356–65. Academic Press, New York;
- Reid, L. M., Mather, D. E., Bolton, A. T., Hamilton, R. I., 1994.** Evidence for a gene for silk resistance to *Fusarium graminearum* ear rot of maize, *J. Hered.*, 85:118–121;
- Rheeder, J.P., Marasas, W.F.O., Thiel, P.G., Sydenham, E.W., Shephard, G.S., Van Schalkwyk, D. J., 1992.** *Fusarium moniliforme* and fumonisins in corn in relation to human esophageal cancer in Transkei, *Phytopathology*, 82, 353–357;
- Riley, R.T., 1998.** Mechanistic interactions of mycotoxins, Theoretical considerations, In: Sinha K.K. and Bhatnagar D. eds. *Mycotoxins in Agriculture and Food Safety* Marcel Dekker, Inc., New York, pp. 227–253;
- Ross, P. F., Nelson, P. E., Richard, J. L., Osweiler, G. D., Rice, L. G., Plattner, R. D., Wilson, T. M., 1990.** Production of fumonisins by *Fusarium moniliforme* and *Fusarium proliferatum* isolates associated with equine leukoencephalomalacia and a pulmonary edema syndrome in swine, *Appl. Environ Microbiol.*, 56. 3225–3226;
- Rustom, I. Y. S., 1997.** Aflatoxin in food and feed: occurrence, legislation and inactivation by physical methods, *Food Chem.*, 59:57–67, with permission from Elsevier;
- Saenz de Rodrigues, C.A., Bongiovanni, A. M., Conde de Borrego, L., 1985.** An epidemic of precocious development in Puerto Rican children, *Journal of Pediatrics*, 107, 393–396;
- Samarajeewa, U., Sen, A. C., Cohen, M. D., Wei, C. I., 1990.** Detoxification of aflatoxins in foods and feeds by physical and chemical methods, *J. Food Prot.*, 53:489–501, with permission from Journal of Food Protection;
- Schneweis, I., Meyer, K., Engelhardt, G., Bauer, J., 2002.** Occurrence of zearalenone-4- β -D-glucopyranoside in wheat. *J. Agric. Food Chem.*, 50, 1736–1738;
- Schuller, P.L., Van Egmond, H. P., Stoloff, L., 1983.** Limits and regulations on mycotoxins, In: Naguib K, Naguib MM, Park DL, Pohland AE (eds) *Proc Int Symp on Mycotoxins*, 6–8 September 1981, Cairo, Egypt, pp 111–129;
- Scott, P. M., 1998.** Industrial and farm detoxification processes for mycotoxins. In: Le Bars, J., and Galtier, P. Eds., *Mycotox '98 International symposium*, 2-4 July, Toulouse, France, 543–548;
- Shephard, G. S., 2006.** Mycotoxins in the context of food risks and nutrition issues, In: Barug D, Bhatnagar D, Van Egmond HP, Van der Kamp JW, Van Ossenbruggen WA, Visconti A, editors. *The mycotoxin fact book*, Wageningen: Wageningen Academic, pp 21–36;
- Sinha, K. K., 1998.** Detoxification of mycotoxins and food safety. In: Sinha, K.K., and Bhatnagar, D. Eds., *Mycotoxins in Agriculture and Food Safety*. Marcel Dekker, Inc., New York, 381–405;
- South, P, Egan, S. K., Troxell, T., Bolger, P. M., 2004.** U.S. Food and Drug Administration's Dioxin Monitoring Program, Organohalogen compounds, 66, 2117–2121;
- Stevens, V. L., Tang, J., 1997.** Fumonisin B1-induced sphingolipid depletion inhibits vitamin uptake via the glycosylphosphatidylinositol-anchored folate receptor, *Journal of Biological Chemistry* 272, 18020–18025;
- Steyn, P. S., 1973.** The structures of five diketopiperazines from *Aspergillus ustus*, *Tetrahedron* 29, 107;
- Steyn, P. S., Vleggaar, R., Ferreira, N. P., Kriby, G. W., Varley, M. J., 1975.** The steric course of proton removal during the cyclization of P-cyclopirozonic acid in *Penicillium cyclopium*, *J. Chem. Soc. Chem. Commun.*, 465–466;
- Steyn, P. S., Vleggaar, R., 1985.** Tremorgenic Mycotoxins. *Fortschr. Chem. Org. Naturst.*, 48, 1-80;
- Van Egmond, H.P., 1989.** Aflatoxin M1 occurrence, toxicity, regulation. In: van Egmond H.P. eds *Mycotoxins in Dairy Products*. Elsevier Applied Science Publishers, Barking, Essex, England. pp.11-55.
- Van Egmond, H. P., Speijers, G. J. A., 1994.** Survey of data on the incidence of ochratoxin A in food and feed worldwide, *Nat. Toxins*, 3, 125–144.
- Van Egmond, H. P., 1989.** Food Addit Contam 6: 4. Food and Agriculture Organization (1997) Worldwide regulations for mycotoxins 1995. A compendium. FAO Food and Nutrition Paper 64. Food and Agriculture Organization of the United Nations, Rome, Italy 139–188;
- Veldman, A. J., Meijs, A. C., Borggreve, G. J., Heeresvan der Tol, J. J., 1992.** Carry-over of aflatoxins from cows' food to milk, *Anim. Prod.*, 55, 163–168;
- Wallnöfer, P. R., Preiss, U., Ziegler, W., Engelhardt, G., 1996.** Konjugatbildung organischer Schadstoffe in Pflanzen. *UWSF Z. Umweltchem. Ökotox.*, 8, 43–46;
- Whitaker, T.B., Dickens J.W., Monroe, R.J., 1979.** Variability associated with testing corn for aflatoxin. *Journal of the American Oil Chemists Society*, 56, 789–794;
- Whitaker, T. B., Dowell, F.E., Hagler, W. M., Giesbrecht, F. G., Wu, J., 1994.** Variability associated with sampling, sample preparation, and chemically testing farmers' stock peanuts for aflatoxin, *Journal of the Association of Official Analytical Chemists International*, 77, 107–116;
- Whitaker, T. B., Springer, J., Defize, P., de Koe, W. J., Coker, R., 1995.** Evaluation of sampling plans used in the United States, United Kingdom and The Netherlands to test raw shelled peanuts for aflatoxin, *J. Assoc. Off. Anal. Chem. Intl.*, 78, 1010–1018;
- Whitaker, T. B., Truckess, M. W., Johansson A. S., Giesbrecht, F. G., Hagler, W. M., Bowman, D. T., 1998.** Variability associated with testing shelled corn for fumonisin, *Journal of the Association of Official Analytical Chemists International*, 81(6), 1162–1168;
- Whitaker, T. B., Hagler, W. M., Giesbrecht, F. G., Johansson, A. S., 2000.** Sampling, sample preparation, and analytical variability associated with testing wheat for

- deoxynivalenol, Journal of the Association of Official Analytical Chemists International, 83(5), 1285–1292;
- World Health Organization, 1979.** Mycotoxins, In Environmental Health Criteria, Vol. 11;
- World Health Organization, 1990.** Toxicological evaluation of certain food additives and contaminants, Chapter Patulin, The 35th meeting of the Joint FAO/WHO Expert Committee on Food Additives, WHO Geneva, WHO Food Additives Series, 26, 143–165;
- World Health Organization, 1991.** Toxicological evaluation of certain food additives and contaminants, Chapter Ochratoxin A. The 37th meeting of the Joint FAO/WHO Expert Committee on Food Additives, WHO Geneva, WHO Food Additives Series, 28, 365–417;
- World Health Organization, 1995.** Evaluation of certain food additives and contaminants: forty-fourth report of the Joint FAO/WHO Expert Committee on Food Additives, Ochratoxin A and Patulin, WHO Technical Report Series, 859, 35–38;
- World Health Organization, 1996.** Toxicological evaluation of certain food additives and contaminants, Chapters Ochratoxin A and Patulin, The 44th meeting of the Joint FAO/WHO Expert Committee on Food Additives, WHO Geneva, WHO Food Additives Series, 35, 363–402;
- World Health Organization, 1999.** Evaluation of certain food additives and contaminants, Aflatoxins. Forty-ninth report of the Joint FAO/WHO Expert Committee on Food Additives, WHO Geneva, WHO Technical Report Series, 884, 69–77;
- World Health Organization, 2000.** Evaluation of certain food additives and contaminants, Zearalenone. Fifty-third report of the Joint FAO/WHO Expert Committee on Food Additives, WHO Geneva, WHO Technical Report Series, 896, 93–96;
- World Health Organization, 2002.** Evaluation of certain mycotoxins in food. Fifty-sixth report of the Joint FAO/WHO Expert Committee on Food Additives, WHO Technical Report Series 906, World Health Organization, Geneva, Switzerland, p 70;
- Wu, F., 2004.** Mycotoxin risk assessment for the purpose of setting international regulatory standards, Environ. Sc. Technol., 38, 4049–4055;
- Yagen, B., Joffe, A. Z., Horn, P., Mor, N., Lutsky, I. I., 1977.** Toxins from a strain involved in ATA: In: J.V. Rodricks, C.V. Hesseltine and M.A. Mehlman (Eds.), Mycotoxins in Human and Animal Health, Pathotox Publishers, Ill., pp. 327–336;
- Yamazaki, M., Suzuki, S. and Miyaki, K., 1971.** Tremorgenic toxins from *Aspergillus fumigatus* Fres. Chem. Pharm. Bull., (Tokyo) 19, 1739;
- Yang, C. S., 1980.** Research on esophageal cancer in China: a review. Cancer Res. 40, 263332644.
- Yılmaz, A., Ozay, G., 2001.** Gıda ve yemlerde mikotoksinlerin detoksifikasyonu, Gıda Dergisi, 7:80–84;
- Yin, M. C., Cheng, W. S., 1998.** Inhibition of *Aspergillus niger* and *Aspergillus flavus* by some herbs and spices, *J. Food Prot.*, 61: with permission from Journal of Food Protection 123–125;

Paper received: 23.03.2009.

PREVALENCE AND RESISTANCE AGAINST DIFFERENT ANTIMICROBIAL COMPOUNDS OF *Campylobacter* spp. IN/FROM RETAIL POULTRY MEAT*

Smole-Možina Sonja, Kurinčić Marija, Kramar Ana, Uršič Simona, Katalinić Višnja

Abstract: The increasing antimicrobial resistance rates of microorganisms is an urgent world-wide problem, especially microbial multidrug resistance phenotypes, dispersed also among food-related bacteria. A case study could be the resistance of campylobacters, usually transmitted in the food chain by contaminated poultry meat. We tested the resistance of *Campylobacter* chicken meat isolates against a) selected antibiotics used in human and veterinary medicine - erythromycin, ciprofloxacin, tetracycline, b) selected disinfectants used in food processing - benzalkonium chloride (BC), chlorhexidine diacetate (CHA), cetylpyridinium chloride (CPC) and c) alternative group of antimicrobial compounds - phenolic extracts from grape skins of different grape varieties. *Campylobacter* was isolated from retail chicken meat samples by standard (ISO 10272) isolation procedure. Beside classical phenotyping methods of species identification and resistotyping, polymerase chain reaction (PCR) and/or restriction fragment length polymorphisms of specific amplicons (PCR-RFLPs) were used for species identification and determination of mutations in target genes. Antibiotic resistance phenotypes were studied by disk diffusion, agar dilution and broth microdilution method with CellTiter-Blue® reagent and automated fluorescence signal detection. The resistance to disinfectants and phenolic extracts was tested by broth microdilution method and expressed as minimal inhibitory concentrations (MICs). The involvement of efflux pumps in antibiotic and disinfectant resistance was assessed by measurements of MICs with and without addition of chemical efflux pump inhibitors, phenylalanine-arginine β -naphthylamide (PA β N) and 1-(1-naphthylmethyl)-piperazine (NMP) and by testing *Campylobacter jejuni* cmeB mutant strain. High prevalence of antibiotic resistant strains was found among chicken meat isolates. Regarding ciprofloxacin, 66.5% of tested strains were found resistant, including *C. jejuni* and *C. coli* strains, but resistance to erythromycin was much more frequent among *C. coli* isolates (34.5%, but 13.9% in average among 158 tested isolates). Tetracycline resistance was relatively rare, but multidrug resistant strains were found. No significant difference in biocide susceptibility between antibiotic resistant and sensitive *Campylobacter* isolates was confirmed. Finally, the results of initial screening of susceptibility of *Campylobacter* meat isolates against grape skin phenolic extracts tested are promising for further study of such antimicrobial compounds (or their mixtures) for potential use in assuring safety of poultry meat and other products.

Key words: food safety, poultry meat, *Campylobacter*, antibiotic resistance, disinfectant resistance, phenolic extracts

Zastupljenost *Campylobacter* spp i rezistencija na različita antimikrobna jedinjenja u živinskom mesu iz prometa

Sadržaj: povećani nalaz antimikrobne rezistencije kod mikroorganizama je rasprostranjen svetski problem, naročito rezistencija fenotipova mikroorganizama na više antimikrobnih lekova koja se javlja i kod bakterija hrane. Jedna od studija mogla bi da bude rezistencija *Campylobacter* mikroorganizama koja se prenosi putem lanca hrane preko kontaminiranog živinskog mesa. Ispitivali smo rezistenciju izolata *Campylobacter* vrsta iz pilećeg mesa na: a) odabrane antibiotike koji se koriste u humanoj i veterinarskoj medicini – eritromicin, ciprofloksacin, tetraciklin, b) odabrane dezinficijense koji se koriste u preradi hrane – benzalkonijum hlorid (BC), hlorheksidin diacetat (CHA), cetilpiridin hlorid (CPC) i c) alternativnu grupu antimikrobnih jedinjenja – fenolne ekstrakte iz kože različitih vrsta grožđa. *Campylobacter* je izolovan iz uzoraka pilećeg mesa iz prometa standardnom metodom (ISO 10272). Pored klasičnih metoda identifikacije vrsta, fenotipizacije i tipizacije rezistencije, za identifikaciju i utvrđivanje mutacija selektovanih gena korišćene su PCR i/ili PCR-RFLP metoda specifičnih amplikona. Fenotipovi koji pokazuju antimikrobnu rezistenciju su proučavani metodama disk difuzije, agar dilucije i bujon mikrodilucije sa CellTiter-Blue reagensom i automatskom detekcijom fluorescentnog signala. Rezistencija na dezinficijense i fenolne ekstrakte je ispitivana korišćenjem bujon mikrodilucije i izražena je kroz minimalne inhibitorne koncentracije (MIC). Uloga efluks pumpi kod rezistencije na antibiotike i dezinficijense je ispitivana merenjem MIC, sa i bez dodavanja hemijskih inhibitora efluks pumpi kao što su fenilalanin-arginin, β -naftilamid (PA β N) i 1-(1-naftilmetil)-piperazin (NMP), kao

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

i ispitivanje *cmeB* mutiranog soja *Campylobacter jejuni*. Među izolatima pilećeg mesa utvrđen je veliki broj sojeva rezistentnih na antibiotike. 66.5% ispitivanih sojeva su pokazali rezistenciju na ciprofloksacin, uključujući *C. jejuni* i *C. coli* dok je rezistencija na eritromicin bila zastupljenija kod izolata (34.5%, ali prosečno 13.9% od 158 ispitivanih izolata). Rezistencija na tetraciklin je bila relativno retka, ali su pronađeni sojevi sa multiplom rezistencijom. Nije potvrđena značajna razlika u prijemčivosti na biocide između rezistentnih i osetljivih izolata *Campylobacter* vrsta. Konačno, rezultati inicijalnog skrininga prijemčivosti mesnih izolata *Campylobacter* vrsta na fenolne ekstrakte iz kože grožđa su obećavajući za izvođenje daljih studija ovih antimikrobnih jedinjenja (ili njihovih smeša) i ispitivanje mogućnosti njihove upotrebe u osiguranju bezbednosti živinskog mesa i drugih proizvoda.

Ključne reči: bezbednost hrane, živinsko meso, *Campylobacter*, antimikrobna rezistencija, rezistencija na dezinficijense, fenolni ekstrakti

Introduction

Thermotolerant campylobacters as gram-negative, non-sporeforming microaerophilic microorganisms are currently recognized as the leading cause of foodborne illnesses in many developed countries worldwide (Tauxe, 2002; EFSA, 2006; 2008). Identified by case-control studies, the major route of infection in humans is thought to be the consumption of undercooked, contaminated broiler chicken meat, because of the high prevalence of contamination of chicken carcasses with *Campylobacter* and the frequency of poultry consumption. Foodborne exposure is frequent also via cross-contamination during preparation of meat and/or other food. Although most reports based on molecular typing have shown a major contribution of chicken meat to human *Campylobacter* infections (Zorman *et al.*, 2006), there is still much unknown in epidemiology of campylobacters. Since most infections occur sporadically, the sources usually remain unidentified.

Additional problem to frequent contamination of some food types and water is the emergence and spread of antimicrobial resistance among campylobacters from different sources, including multidrug resistant isolates. Macrolides, fluoroquinolones and tetracyclines are the most commonly used antimicrobial agents for the treatment of severe human *Campylobacter* infections, as well as for veterinary purposes including treatment of food animals. The prevalence of resistant *Campylobacter* strains is increasing worldwide and is becoming a major concern for public health (Moore *et al.*, 2006).

The mechanisms of macrolide and fluoroquinolone resistance in *Campylobacter* spp. have been described recently (Gibreel *et al.*, 2005; Moore *et al.*, 2006). Mutations in target genes and efflux pumps activity are both important. For example, mutations in domain V of the 23S rRNA gene at positions 2074 and 2075 have been attributed to high-level erythromycin resistance (Vacher *et al.*, 2003; Payot *et al.*, 2004). The presence of *tetO* gene contributes to tetracycline resistance. In addition, recent studies demonstrated the involvement of

CmeABC efflux pump in both intrinsic and acquired resistance to erythromycin in *C. jejuni* and *C. coli*, mostly by the use of the efflux pump inhibitor (EPI), phenylalanine-arginine β -naphthylamide (PABN) (Payot *et al.*, 2004; Mamelli *et al.*, 2005; Gibreel *et al.*, 2007; Kurinčič *et al.*, 2007).

In comparison to antibiotic resistance, *Campylobacter* resistance to biocides has been described more recently and much less studied. In contrast to antibiotics and bacterial resistance to antibiotics, resistance to disinfectants is thought unlikely to occur because most disinfectants are complexes of antimicrobials that inactivate several target sites in bacterial cells (McDonnell and Russell, 1999; Russell, 2002). Due to multiplicity of cellular targets, bacterial biocide resistance results from changes of envelope permeability or enhanced biocide efflux (Poole, 2002). Antimicrobial resistance in *Campylobacter* may be mediated by different resistance-nodulation-cell division (RND) efflux or non-RND efflux pumps, which could be involved in the extrusion of toxic compounds (Pumbwe *et al.*, 2005). The involvement of efflux mechanisms in bacterial resistance is mostly studied by the use of efflux pump inhibitors (EPIs) like phenylalanine-arginine beta-naphthylamide (PA β N) and 1-(1-naphthylmethyl)-piperazine (NMP) which enhance drug accumulation inside the bacterial cell, thereby increasing bacterial susceptibility to antimicrobials (Marquez, 2005). Those studies can be confirmed by using mutants lacking functional genes of efflux pump proteins (Lin *et al.*, 2002). A possible linkage of biocide and antibiotic resistance in different enteric bacteria via efflux related mechanisms has been recently reported (Thorrold *et al.*, 2007; Karatzas *et al.*, 2007; 2008).

Because of the increasing problem of antimicrobial resistance of bacteria in general and in the food chain, many different types of alternative bioactive compounds have been screened recently for their potential antibacterial effects. Among others, phenolic extracts from different plant materials gave promising results (Cowan, 1999; Moreno *et al.*, 2006).

We studied the prevalence of thermotolerant *Campylobacter* spp. in retail poultry meat samples and included the isolates of *Campylobacter jejuni* and *C. coli* in further testing of their antimicrobial resistance and the mechanisms involved in resistance. In this study, erythromycin, ciprofloxacin, tetracycline as well as benzalkonium chloride, chlorhexidine diacetate and cetylpyridinium chloride resistance is presented and discussed. In addition, an alternative group of antimicrobial compounds - phenolic grape skin extracts of fourteen *Vitis vinifera* varieties grown in Dalmatia (Croatia) have been screened for antimicrobial activity against selected multiresistant chicken meat isolate.

Materials and methods

Isolation of bacterial strains from poultry meat and species identification

A hundred and fifty eight (158) samples of chicken meat from different suppliers on Slovenian market were investigated with the ISO 10272 guideline for the presence of thermotolerant campylobacters in the periods 2002-2003 and 2008-2009. A hundred and eighteen (118) strains isolated in the first period were long-term stored at -80°C in culture collection ZIM, BF, Ljubljana, for further studies.

C. jejuni and *C. coli* were identified by standard phenotyping (ISO) methods and polymerase chain reaction (PCR) procedures with amplification of hippuricase gene in *C. jejuni* and aspartokinase gene in *C. coli* in multiplex PCR as well as with the genus specific primers, as described previously (Zorman and Smole Možina, 2002). Additionally, the identity of most chicken meat isolates from the period 2002-2003 was confirmed by PFGE typing using *Sma*I restriction endonuclease and CHEF mapper XA System (Bio-Rad) (Zorman et al., 2006).

Determination of antibiotic and disinfectant resistance and mechanisms involved

Antimicrobial resistance testing was first performed using disc diffusion method as described previously (Kurinčič et al., 2005). Minimal inhibitory concentrations (MICs) of ciprofloxacin (Fluka Biochemika), erythromycin and tetracycline (both from Sigma-Aldrich) were determined by E-test as well as with broth microdilution method (Kurinčič et al., 2007). CellTiter-Blue[®] reagent and automated fluorescence signal detection by a microplate reader (Tecan, Mannedorf/Zurich, Switzerland) were used. Mutations in bacterial target genes (*23S rRNA*, *tetO*) were studied by PCR-RFLP and PCR, as described previously (Kurinčič et al., 2007). The resistance of

selected 25 strains was tested against benzalkonium chloride (BC), chlorhexidine diacetate (CHA) and cetylpyridinium chloride (CPC), (Sigma-Aldrich, Saint Luis, USA) with broth microdilution method on the same principle as described for antibiotic resistance testing. The involvement of efflux pumps in antibiotic and biocide resistance mechanisms were evaluated by measurements of antimicrobial MICs values with or without chemical efflux pump inhibitors, phenylalanine-arginine β -naphthylamide (PABN), (Sigma-Aldrich Saint Louis, USA) or 1-(1-naphthylmethyl)-piperazine (NMP), (Chess GmbH, Mannheim, Germany) as described by Kurinčič et al. (2007). For this purpose, the Müller Hinton (MH) broth was supplemented with PABN (20 $\mu\text{g}/\text{mL}$) or NMP (80 $\mu\text{g}/\text{mL}$). Two independent experiments were conducted to confirm the reproducibility of MIC data and the ATCC 33559 and ATCC 33560 strains were included as quality control strains. Additionally, another strategy was used to study the mechanisms of resistance. We determined MIC values of selected antibiotics and disinfectants also for *Campylobacter jejuni* NCTC 11168 and its *cmeB* mutant strain, kindly provided by dr. Payot (Institut National de la Recherche Agronomique, UR086 BioAgresseurs, Santé, Environment, Nauzilly, France).

Determination of antimicrobial activity of phenolic grape skin extracts

Phenolic extracts of native or introduced* grape varieties (White: Debit, Kuć, Kujundžusa, Maraština, Medna, Rkaciteli*, Zlatarica; Red: Babić, Lasin, Merlot*, Plavina, Rudežusa, Trnjak, Vranac*) were extracted from homogenized grape skins using conventional solvent extraction procedure. They were characterized with determination of total phenols (TPC), total flavonoids (TFLO), total flavanols (TFA) and total anthocyanins (TA), HPLC analysis of phenolic compounds and with different procedures for antioxidant activity as described by Katalinić et al. (2009). Determination of the minimum inhibitory concentration (MIC) of fourteen different extracts was performed for selected multiresistant poultry meat isolate *C. coli* 137 and two susceptible reference strains, *Campylobacter coli* ATCC 33559 and *Staphylococcus aureus* ATCC 25923, with broth microdilution method by a Microplate Reader (Tecan, Mannedorf/Zurich, Switzerland) as described by Klančnik et al. (2009a). Minimum inhibitory concentrations (MICs) of tested phenolic extracts are expressed in mg of gallic acid equivalents (GAE) per mL of growth medium (Katalinić et al., 2009). All measurements of MIC values were repeated three times and the most representative values were used.

Results and discussion

Isolation and identification of thermotolerant *Campylobacter* spp. from chicken meat

In total, 86.7% (137/158) of Slovene fresh retail chicken meat samples were found positive for thermotolerant campylobacters. The rate has not changed significantly during the six-year period (90.0% of tested samples were found positive in the years 2002-2003 and 84.7% in the years 2008-2009). Such results indicate the high extent of chicken meat contamination with campylobacters on retail market in Slovenia. Similar observations were reported from other European countries like Italy, France, Great Britain and Poland (Pezzoti *in sod.*, 2003; Meldrum *in Wilson*, 2007, Maćkiw *in sod.*, 2008), while reports from Scandinavian countries show much lower contamination level (NORM/NORM-VET, 2006; EFSA, 2007). The prevalence of thermotolerant *Campylobacter* in retail poultry meat in reports of official monitorings of food safety in Slovenia in recent years is constantly increasing. In the years 2006 and 2007, the officially tested poultry meat samples were found positive for thermotolerant campylobacters in 59.0 and 67.1%, respectively (EFSA, 2009).

A hundred and twelve (112) chicken meat isolates collected in 2002-2003 have survived long-term freezing and were included in PCR species identification. We found high proportion of *C. coli* among thermotolerant campylobacters from chicken meat (64/112), not reported from other European countries, except in some Balkan countries (Uzunović-Kamberović *et al.*, 2007). This unusual result of classical and molecular identification of strains with species specific PCR primers was confirmed also by molecular typing of strains with macrorestriction analysis with *Sma*I and PFGE typing (Zorman *et al.*, 2006). However, in recently tested chicken meat samples *C. jejuni* was much more frequently isolated than *C. coli* (Table 1).

Antimicrobial resistance and mechanisms involved in *Campylobacter* chicken meat isolates

We tested the occurrence of antimicrobial resistance to ciprofloxacin and erythromycin among 158 chicken meat isolates and tetracycline resistance among 61 isolates identified as *C. jejuni* or *C. coli*. Resistance to ciprofloxacin was most frequent (66.5%) and almost equally distributed among *C. jejuni* and *C. coli* isolates (Table 1). In our previous report, including the chicken meat isolates from the years 2002-2003, the rate of resistant *C. jejuni* isolates was only 38.5% (Kurinčič *et al.*, 2005), but among isolates from the period 2008-2009, 74.6% of *C. jejuni* isolates were resistant to ciprofloxacin.

This indicates still increasing rate of ciprofloxacin resistance. The prevalence of erythromycin resistance was much lower, but in fact very high, in comparison with the reports from some other European countries, USA or Canada (EFSA, 2007; Gyles, 2008). A significant difference in resistance rates was found among *C. jejuni* and *C. coli* isolates. Similarly, in food producing animals, the prevalence of erythromycin resistance is generally reported to be higher in *C. coli* than in *C. jejuni*, particularly among *C. coli* isolates from swine (Belanger and Shryock, 2007). In our study three groups of strains were observed concerning erythromycin resistance: susceptible with MICs, from 0,25 to 2 µg/mL, low-level resistant (LLR) with MICs, from 4 to 16 µg/mL, and high-level resistant (HLR) with MICs, higher than 32 µg/mL.

PCR-RFLP procedure has been used to test the presence of the A2075G mutation in the 23S rRNA gene. Seven HLR *C. coli* strains exhibited the A2075G mutation. Conversely, the A2075G mutation was not identified in any of LLR and susceptible strains. Other studies have also indicated that the mutation at position 2075 is usually responsible for high-level erythromycin resistance (Payot *et al.*, 2004; Mamelli *et al.*, 2005; Gibreel *et al.*, 2005). Interestingly, no A2075G mutation was identified in one HLR *C. coli* isolate originated from chicken meat.

Eight, out of 61 isolates from retail chicken meat samples (13.1%), were found resistant to tetracycline. PCR procedure confirmed the presence of *tetO* gene in all tetracycline resistant strains. No strains susceptible to tetracycline were found to have *tetO*. This is in agreement with other reports, but much more prevalent resistant strains have been found in different countries and from different sources, including farm animal isolates (Alfredson and Korolik, 2007; Mazi *et al.*, 2008; Uzunović-Kamberović *et al.*, 2007; 2009).

Our recent study of antibiotic resistance of *Campylobacter* isolates from animals, food and environmental sources (surface and drinking water) and different geographical regions also revealed interesting differences in resistance patterns of strains from different sources. For comparison with Slovene chicken meat isolates (Table 1), Table 2 includes strains from different sources and two different geographical regions. Beside Slovene samples, animal, food, environmental (water) and human clinical *Campylobacter* isolates from Zenica-Doboj canton, collected during two bilateral research projects in the years 2002-2007, are included. Comparison of these results with our recent testing of antibiotic resistance of chicken meat isolates reveals again the increasing rate of ciprofloxacin resistant isolates.

Table 1. Frequency of isolation of *Campylobacter jejuni* and *C. coli* from retail chicken meat samples and percentage (resistant/tested,%) of resistant isolates against erythromycin, ciprofloxacin and tetracycline
Tabela 1. Učestalost izolovanja *Campylobacter jejuni* i *C. coli* iz uzoraka pilećeg mesa iz prometa i procenat (rezistentnih/ispitanih,%) rezistentnih izolata na eritromicin, ciprofloksacin i tetraciklin

Species isolated from retail chicken meat	Frequency of isolation	Erythromycin -R		Ciprofloxacin -R		Tetracycline -R	
		Count	Percentage	Count	Percentage	Count	Percentage
<i>C. coli</i>	65 (33.5%)	10/29	34.5%	18/30	60.0%	4/28	14.3%
<i>C. jejuni</i>	129 (66.5%)	12/129	9.3%	87/128	68.0%	4/33	12.1%
Σ	194 (100%)	22/158	13.9%	105/158	66.5%	8/61	13.1%

Table 2. Percentage (%) of resistant strains of *C. jejuni* and *C. coli* against erythromycin, ciprofloxacin and tetracycline among the isolates from farm animals, retail chicken meat, surface and drinking water and human clinical samples, collected in the period 2002-2007 in Slovenia and Zanica-Doboj canton (taken from Kurinčič et al., 2009).

Tabela 2. Procenat (%) rezistentnih sojeva *C. jejuni* i *C. coli* na eritromicin, ciprofloksacin i tetraciklin u izolovima sa farmskih životinja, pilećeg mesa iz prometa, sa površina, iz pijaće vode i iz humanih kliničkih uzoraka sakupljenih u periodu od 2002-2007. u Sloveniji u kantonu Zanica-Doboj (preuzeto Kurinčič i sar., 2009).

Source and number of tested isolates	Farm animal (n = 15)	Chicken meat (n = 112)	Water (n = 50)	Human clinical isolates (n = 179)
Antimicrobial agent				
Erythromycin -R (%)	38.5	21.4	44.0	10.6
Ciprofloxacin -R (%)	30.8	43.8	26.0	31.8
Tetracycline - R (%)	61.5	18.8	6.0	6.8

Concerning possible cross-resistance we selected twenty five chicken meat isolates and reference strains *C. jejuni* ATCC 33560 and *C. coli* 33559 to be tested for their resistance against three antibiotics (erythromycin, ciprofloxacin, tetracycline) and three disinfectants (benzalkonium chloride (BC), cetylpyridinium chloride (CPC), chlorhexidine diacetate (CHA). For presentation, the results for sixteen thermotolerant *Campylobacter* chicken meat isolates are included in Table 3.

Strains susceptible and resistant to antibiotics (also multidrug resistant strains like *C. coli* 137, *C. coli* 140, and *C. coli* 171, Table 3) were tested. Two different BC and CPC resistance phenotypes were observed and classified, as described previously for *Listeria monocytogenes* (Aase et al., 2000). Irrespective of antibiotic resistance, all chicken meat isolates were sensitive to BC at concentration 1 µg/ml, or below, and were considered as BC sensitive, but three (12%) of the isolates were considered CPC resistant (MIC 4 µg/mL). Most of the strains were sensitive to CHA concentration 1 µg/ml or below, but four strains were tolerant to CHA concentration 2 µg/ml. However, no significant difference in biocide susceptibility between antibiotic resistant

and sensitive *C. coli* and *C. jejuni* isolates was found. The isolates from the same meat samples usually shared the same resistotype (like strains 53/1 and 53/4 in Table 3).

With the aim to study the mechanisms involved, the resistance of isolates to antibiotics and disinfectants was studied in the absence and presence of efflux pumps inhibitors (EPIs), PAβN and NMP. Both EPIs increased erythromycin susceptibility significantly, wherein PAβN had greater effect than NMP, although both affected the main efflux pump, CmeABC, in *Campylobacter* cells. Both EPIs had much greater effect in *C. coli* than in *C. jejuni*. The results confirm that efflux mechanism mediated by efflux pumps plays an active role in resistance to erythromycin in *Campylobacter*. The presence of efflux pumps activity in HLR isolates with 23 rRNA mutations suggests that the synergistic activity of these two drug resistance mechanisms exist in *Campylobacter*. BC susceptibility was significantly increased by both EPIs. There was no significant difference between NMP and PAβN effect on BC susceptibility. The smaller effect of EPIs was observed when used in the presence of ciprofloxacin, tetracycline, CHA and CPC (data not shown).

Table 3. Antimicrobial activity (expressed as MICs, $\mu\text{g/mL}$) of antibiotics (erythromycin, ERI, tetracycline, TET, ciprofloxacin, CIP), and disinfectants (benzalkonium chloride, BC), cetylpyridinium chloride (CPC), chlorhexidine diacetate (CHA)) against reference strains and sixteen thermotolerant *Campylobacter* chicken meat isolates

Tabela 3. Antimikrobna aktivnost (izražena kao MICs, $\mu\text{g/mL}$) antibiotika (eritromicin, ERI), tetraciklin, TET, ciprofloksacin, CIP) i dezinficijensa (benzaalkonijum hlorid, BC), cetilapiridin hlorid (CPC), hlordexin diacetat (CHA) na referentne sojeve i 16 termotolerantnih izolata *Campylobacter* iz pilećeg mesa

Species	Strain	ERI	TET	CIP	BC	CPC	CHA
<i>C. coli</i>	ATCC 33559	2	0.25	0.063	0.125	2	1
<i>C. jejuni</i>	ATCC 33560	0.5	1	0.125	<0.016	0.125	0.5
<i>C. jejuni</i>	203	0.5	1	0.125	0.25	2	1
<i>C. jejuni</i>	K29/3	0.25	0.5	0.063	0.5	4	2
<i>C. jejuni</i>	K45/4	0.5	0.5	8	0.063	4	1
<i>C. jejuni</i>	K49/4	0.5	0.125	0.063	1	2	0.125
<i>C. spp.</i>	K31/2	2	0.5	8	0.25	1	2
<i>C. spp.</i>	K37/4	128	0.5	0.25	0.25	2	0.5
<i>C. spp.</i>	K40/2	0.25	1	8	0.125	2	1
<i>C. coli</i>	128	0.25	0.25	32	0.25	4	1
<i>C. coli</i>	137	> 512	256	16	1	1	0.25
<i>C. coli</i>	140	512	128	16	0.063	1	0.25
<i>C. coli</i>	171	512	32	16	1	0.5	0.125
<i>C. coli</i>	K31/4	1	0.5	8	0.125	1	2
<i>C. coli</i>	K32/3	2	128	16	0.25	2	0.5
<i>C. coli</i>	K39/3	1	1	64	0.125	1	2
<i>C. coli</i>	K53/1	1	128	16	0.063	0.5	0.5
<i>C. coli</i>	K53/4	1	128	16	0.063	0.5	0.5

In the study of efflux involvement in antibiotic and disinfectant resistance of *Campylobacter* strains, the reference strain *C. jejuni* NCTC 11168 and its *cmeB* mutant were also assessed - by using the EPIs PABN and NMP. Both inhibitors, PABN and NMP, increased the susceptibility of the wild-type strain to erythromycin by 4-fold. Additionally, PABN was also able to reduce the MIC of ciprofloxacin, BC, CPC and CHA by 2-fold. In case of tetracycline, no effect of the inhibitors was observed. Insertional inactivation of *cmeB* gene increased the susceptibility to antibiotics erythromycin, ciprofloxacin and tetracycline by 8-fold, 4-fold, 8-fold, respectively, and to disinfectant BC by 4-fold. Additionally, both inhibitors were also able to reduce the MIC of ciprofloxacin and all disinfectants tested by at least 2-fold in *cmeB* mutant, suggesting that resistance mechanisms are very complex and another efflux pump(s) are involved in *Campylobacter jejuni* resistance to these antimicrobials.

Antimicrobial activity of phenolic grape skin extracts against antibiotic resistant chicken meat isolate

In our recent studies of antimicrobial activity of phenolic extracts from different plant sources (Klančnik *et al.*, 2009a,b;) we got an evidence that campylobacters, although gram-negative organisms, could be quite sensitive to different phenolic compounds or their mixtures (Katalinić *et al.*, 2009). For this reason we tested also the extracts from grape skins of white and red cultivars against different antibiotic resistant chicken meat isolates. The results for the strain *C. coli* 137 (MDR meat isolate, see Table 3) and two antibiotic susceptible reference strains *Campylobacter coli* ATCC 33559 and *Staphylococcus aureus* ATCC 25923 are collected in Table 4. Staphylococci are known as susceptible microorganisms to different antimicrobials and thus usually used as reference material in screening tests of antimicrobial activity of new compounds, so they were used for comparison

also in these experiments. We found very low MICs for both *Campylobacter* strains, although the outer membrane surrounding the cell wall in gram-negative bacteria could restrict diffusion of compounds through its lipopolysaccharide covering. In our previous work gram-positive bacteria were more sensitive than gram-negative bacteria, especially for oil-soluble extracts with carnosic acid as the major phenolic compound (Klančnik *et al.*, 2009a). However, in this initial screening of the activity of grape skin phenolic extracts against *Campylobacter* chicken meat isolates we got very promising results, especially in antimicrobial activity of grape skin extracts of white cultivars, which on average gave even better results. It is important that they leave the wine processing still rich in biologically valuable components. Further tests are needed to confirm the screening results in *in vitro* and *in vivo* assays to confirm the potential use as additives for reduction of bacterial load of fresh chicken meat and products and thus in lowering the risk of bacterial transmission via this important route.

Conclusion

The emergence and dissemination of resistant bacteria is an inevitable side effect of the use of antimicrobials. We need a monitoring system of the prevalence and antibiotic resistance of zoonotic bacteria from human, animal, food and environmental samples to understand the epidemiology of resistant strains to assure food safety and consumers health. In our studies we confirmed high prevalence and also antibiotic resistance of *C. jejuni* and *C. coli* in/from retail chicken meat. Ciprofloxacin is one of the critically important antimicrobial agents in human medicine. It is often used for treatment of human gastroenteritis because of its activity against enteric bacterial pathogens. However, agricultural use of some fluoroquinolones, including food producing animals, contribute to selection of resistant *Campylobacter* spp. which are transmitted into the food chain. We have confirmed high and still increasing rate of ciprofloxacin resistant isolates

Table 4. Antimicrobial activity of phenolic extracts (expressed as MICs of total phenols, e.g. mg GAE* per mL of growth medium in broth microdilution test) from grape skins of white and red cultivars against *C. coli* 137 (MDR chicken meat isolate) and two reference strains

Tabela 4. Antimikrobna aktivnost fenolnih ekstrata (izražena kao MIC ukupnih fenola, npr. GAE* po mL podloge u bujonskom mikrodilucionom testu) iz kože grožđa belih i crvenih sorti na *C. coli* 137 (MDR izolat iz pilećeg mesa) i dva referentna soja

Testing organisms	<i>C. coli</i> 137 (MDR chicken meat isolate)	<i>Campylobacter coli</i> ATCC 33559 (susceptible reference strain)	<i>Staphylococcus aureus</i> ATCC 25923 (susceptible reference strain)
Cultivars			
Kujundžuša	0.076 ± 0.020	0.032 ± 0.005	0.15 ± 0.02
Rkaciteli	0.023 ± 0.000	0.014 ± 0.002	0.20 ± 0.03
Zlatarica	0.051 ± 0.020	0.042 ± 0.005	0.21 ± 0.03
Medna	0.024 ± 0.005	0.014 ± 0.002	0.21 ± 0.03
Kuč	0.023 ± 0.010	0.019 ± 0.002	0.26 ± 0.04
Maraština	0.036 ± 0.010	0.015 ± 0.002	0.21 ± 0.03
Debit	0.060 ± 0.010	0.025 ± 0.005	0.25 ± 0.05
<i>Average for white cultivars</i>	<i>0.042 ± 0.02</i>	<i>0.023 ± 0.01</i>	<i>0.22 ± 0.04</i>
Vranac	0.10 ± 0.03	0.20 ± 0.03	0.23 ± 0.03
Trnjak	0.21 ± 0.04	0.14 ± 0.03	0.22 ± 0.04
Rudežuša	0.13 ± 0.02	0.25 ± 0.06	0.29 ± 0.04
Merlot	0.07 ± 0.01	0.13 ± 0.03	0.44 ± 0.08
Babić	0.10 ± 0.02	0.08 ± 0.01	0.42 ± 0.08
Lasin	0.03 ± 0.01	0.04 ± 0.01	0.34 ± 0.07
Plavina	0.05 ± 0.01	0.09 ± 0.02	0.29 ± 0.06
<i>Average for red cultivars</i>	<i>0.10 ± 0.05</i>	<i>0.13 ± 0.07</i>	<i>0.32 ± 0.09</i>

*Total phenols are expressed as gallic acid equivalents (GAE) in grape skin extract

from retail chicken meat. However, so far we have not found any evidence suggesting that tolerance to disinfectants or other potential antimicrobials (like plant phenolic extracts) is connected to antibiotic resistance of *Campylobacter* isolates.

Acknowledgement

The authors would like to thank the Ministry of Higher Education, Science and Technology of the

Republic of Slovenia for financing their projects, especially PhD grants for M.K. and A.K., to dr. Tina Zorman for her contribution in early testing of *Campylobacter* in Laboratory for Food Microbiology, BF, and to dr. Alenka Štorman for enabling the part of experimental work that was performed at ZZV Celje in 2009.

References

- Aase, B., Sundheim, G., Langsrud, S., Rorvik, L. M., 2000. Occurrence of and a possible mechanism for resistance to a quaternary ammonium compound in *Listeria monocytogenes*. *International Journal of Food Microbiology* 62, 57–63;
- Alfredson, D. A., Korolik, V., 2007. Antibiotic resistance and resistance mechanisms in *Campylobacter jejuni* and *Campylobacter coli*. *FEMS Microbiology Letters*, 277, 2, 123–32;
- Belanger, A. E., Shryock, T. R., 2007. Macrolide-resistant *Campylobacter*: the meat of the matter. *Journal of Antimicrobial Chemotherapy*, 60: 715–723;
- Cowan, M. M., 1999. Plant products as antimicrobial agents. *Clinical Microbiology Reviews* 12, 654–582;
- EFSA, 2006. *Campylobacteriosis overtakes salmonellosis as the most reported animal infection transmitted to humans in the EU. 02.03. 2009 on <http://www.efsa.europa.eu/EFSA/>*;
- EFSA, 2007. The Community Summary Report on Trends and Sources of Zoonoses, Zoonotic Agents, Antimicrobial Resistance and Foodborne Outbreaks in the European Union in 2006. (24. 03.2009); (http://www.efsa.europa.eu/EFSA/efsa_locale1178620753812_1178671312912.htm).
- EFSA, 2008. **Scientific opinion of the panel on biological hazards on a request from the European Food Safety Authority on foodborne antimicrobial resistance as a biological hazard, 2008.** *The EFSA Journal* 765, 1–87;
- EFSA, 2009. **The community summary report on trends and sources of zoonoses and zoonotic agents in the European Union in 2007,** *The EFSA Journal*, 223, 1–313;
- Gibreel, A., Kos, V. N., Keelan, M., Trieber, C. A., Levesque, S., Michaud, S., Taylor, D. E., 2005. Macrolide resistance in *Campylobacter jejuni* and *Campylobacter coli*: molecular mechanism and stability of the resistance phenotype. *Antimicrobial Agents and Chemotherapy* 49, 2753–2759;
- Gibreel, A., Wetsch, N. M., Taylor, D. E., 2007. Contribution of the CmeABC efflux pump to macrolide and tetracycline resistance in *Campylobacter jejuni*. *Antimicrobial Agents and Chemotherapy* 51, 9, 3212–3216;
- Gyles, C. L. 2008. Antimicrobial resistance in selected bacteria from poultry. *Animal Health Research Reviews*, 9, 2, 149–158;
- Karatzas, K. A. G., Randall, L. P., Webber, M., Piddock, L. J. V., Humphrey, T. J., Woodward, M. J., Coldham, N. G., 2008. Phenotypic and proteomic characterization on multiple antibiotic-resistant variants of *Salmonella enterica* Serovar Typhimurium selected following exposure to disinfectants. *Applied and Environmental Microbiology* 74, 4, 1508–1516;
- Karatzas, K. A., Webber, M. A., Jorgensen, F., Woodward, M. J., Piddock L. J., Humphrey, T. J., 2007. Prolonged treatment of *Salmonella enterica* serovar Typhimurium with commercial disinfectants selects for multiple antibiotic resistance, increased efflux and reduced invasiveness. *Journal of Antimicrobial Chemotherapy* 60, 5, 947–955;
- Katalinić Višnja, Smole Možina Sonja, Skroza Danijela, Generalić Ivana, Abramović Helena, Miloš M., Ljubenković I., Piskernik Saša, Pezo I., Terpinc Petra, Boban M., 2009. Polyphenolic profile, antioxidant properties and antimicrobial activity of grape skin extracts of 14 *Vitis vinifera* varieties grown in Dalmatia (Croatia). *Food Chemistry (in press)*;
- Klančnik Anja, Guzej Bernarda, Hadolin Kolar Majda, Abramović Helena, Smole Možina Sonja, 2009a. In vitro antimicrobial and antioxidant activity of commercial rosemary extract formulations. *Journal of Food Protection (in press)*;
- Klančnik Anja, Piskernik Saša, Jeršek Barbara, Smole Možina Sonja, 2009b. Protimikrobno delovanje rastlinskih fenolnih izvlečkov na patogene bakterije. In: Raspor P. (ur.) Protimikrobne snovi. Pomen mikrobiologije in biotehnologije za prihodnost. Biotehniška fakulteta, Ljubljana;
- Kurinčič, M., Berce, I., Zorman, T., Smole Možina, S., 2005. The prevalence of multiple antibiotic resistance in *Campylobacter* spp. from retail poultry meat. *Food Technology and Biotechnology* 43, 157–163;
- Kurinčič Marija, Botteldoorn Nadine, Herman Lieve, Smole Možina Sonja, 2007. Mechanisms of erythromycin resistance *Campylobacter* spp. isolated from food, animals and humans. *International Journal of Food Microbiology*, 120, 186–190;
- Kurinčič Marija, Lušicky Mojca, Uzunović-Kamberović Selma, Smole Možina Sonja, 2009. Epidemiologija in antibiotična odpornost bakterij *Campylobacter* iz vzorcev vod in piščančjega mesa. In: Raspor P. (ur.) Protimikrobne snovi. Pomen mikrobiologije in biotehnologije za prihodnost. Biotehniška fakulteta, Ljubljana;
- Lin, J., Overbye, M., L., Zhang, Q., 2002. CmeABC functions as a multidrug efflux system in *Campylobacter jejuni*. *Antimicrobial Agents and Chemotherapy* 46, 7, 2124–2131;
- Maćkiw, E., Popowski, J., Szponar, L., 2008. Thermotolerant *Campylobacter* spp. – Report on monitoring studies performed in 2004–2005 in Poland. *Food Control*, 19: 219–222;
- Mamelli, L., Prouzet-Mauleon, V., Pages, J.-M., Megraud, F., Bolla, J.-M., 2005. Molecular basis of macrolide resistance in *Campylobacter*: role of efflux pumps and tar-

- get mutations. *Journal of Antimicrobial Chemotherapy* 56, 491–497;
- Marquez, B., 2005.** Bacterial efflux and efflux pumps inhibitors. *Biochimie* 87, 1137–1147;
- Mazi, W., Senok, A., Al-Mahmeed, A., Arzese, A., Bindayna, K., Botta, G., 2008.** Trends in antibiotic sensitivity pattern and molecular detection of tet(O)-mediated tetracycline resistance in *Campylobacter jejuni* isolates from human and poultry sources. *Japanese Journal of Infectious Diseases*, 61, 82–84;
- McDonnell, G., Russell A. D., 1999.** Antiseptics and disinfectants: activity, action, and resistance. *Clinical Microbiology Reviews* 12, 147–179;
- Meldrum, R. J., Wilson I. G., 2007.** *Salmonella* and *Campylobacter* in United Kingdom retail raw chicken in 2005. *Journal of Food Protection* 70, 8, 1937–1939;
- Moore, J. E., Barton, M. D., Blair, J. S., Corcoran, D., Doolley J. S. G., Fanning, S., Krempf, I., Lastovica, A. J., Lowery, C. J., Matsuda, M., McDowell, D. A., McMahon, A., Millar, B. C., Rao, J. R., Rooney, P. J., Sails, A., Seal, B. S., Snelling, W. J., Tolba, O. 2006.** The epidemiology of antibiotic resistance in *Campylobacter*. *Microbes and Infection* 8, 1955–1966;
- Moreno, S., Scheyer, T., Romano, C. S., Vojnov, A. A., 2006.** Antioxidant and antimicrobial activities of rosemary extracts linked to their polyphenol composition. *Free Radical Research*, 40, 223–231;
- NORM/NORM-VET 2006.** Usage of Antimicrobial Agents and Occurrence of Antimicrobial Resistance in Norway. Tromsø/Oslo 2007;
- Payot, S., Avrain, L., Magras, C., Praud, K., Cloeckaert, A., Chaslus-Dancla, E., 2004.** Relative contribution of target gene mutation and efflux to fluoroquinolone and erythromycin resistance, in French poultry and pig isolates of *Campylobacter coli*. *International Journal of Antimicrobial Agents* 23, 468–472;
- Payot, S., Bolla, J.-M., Corcoran, D., Fanning, S., Mégraud, F., Zhang, Q., 2006.** Mechanisms of fluoroquinolone and macrolide resistance in *Campylobacter* spp. *Microbes and Infection*, 8, 7, 1967–1971;
- Pezzotti, G., Serafin, A., Luzzi, I., Mioni, R., Milan, M., Perin, R., 2003.** Occurrence and resistance to antibiotics of *Campylobacter jejuni* and *Campylobacter coli* in animals and meat in northeastern Italy. *International Journal of Food Microbiology*, 15, 281–287;
- Poole, K., 2002.** Mechanisms of bacterial biocide and antibiotic resistance. *Journal of Applied Microbiology Symposium Supplement* 92, 55–64;
- Pumbwe, L., Randall, L. P., Woodward, M. J., Piddock L. J. 2005.** Evidence for multiple-antibiotic resistance in *Campylobacter jejuni* not mediated by CmeB or CmeF. *Antimicrobial Agents and Chemotherapy*, 49, 1289–1293;
- Russell, A. D., 2002.** Introduction of biocides into clinical practice and the impact on antibiotic-resistant bacteria. *Journal of Applied Microbiology Symposium Supplement* 92, 121–135;
- Tauxe, R. V., 2002.** Emerging foodborne pathogens. *International Journal of Food Microbiology* 78, 31–41;
- Thorrold, C. A., Letsoalo, M. E., Duse, A. G., Marais, E., 2007.** Efflux pump activity in fluoroquinolone and tetracycline resistant *Salmonella* and *E. coli* implicated in reduced susceptibility to household antimicrobial cleaning agents. *International Journal of Food Microbiology* 113, 315–320;
- Uzunović-Kamberović Selma, Zorman Tina, Berce Ingrid, Herman Lieve, Smole Možina Sonja, 2009.** Comparison of the frequency and the occurrence of antimicrobial resistance among *C. jejuni* and *C. coli* isolated from human infections, retail poultry meat and poultry in Zenica-Doboj Canton, Bosnia and Herzegovina. *Medicinski glasnik Zeničko-Dobojskog kantona (in press)*;
- Uzunović-Kamberović Selma, Zorman Tina, Heyndrickx M., Smole Možina Sonja, 2007.** Role of poultry meat in sporadic *Campylobacter* infections in Bosnia and Herzegovina: laboratory-based study. *Croatian Medical Journal*, 2007, 48, 844–853;
- Vacher, S., Menard, A., Bernard, E., Megraud, F., 2003.** PCR-restriction fragment length polymorphism analysis for detection of point mutations associated with macrolide resistance in *Campylobacter* spp. *Antimicrobial Agents and Chemotherapy* 47, 1125–1128;
- Zorman Tina, Heyndrickx, M., Uzunović-Kamberović Selma, Smole Možina Sonja, 2006.** Genotyping of *Campylobacter coli* and *C. jejuni* from retail chicken meat and humans with campylobacteriosis in Slovenia and Bosnia and Herzegovina. *International Journal of Food Microbiology*, 110, 24–33;
- Zorman Tina., Smole Možina Sonja, 2002.** Classical and molecular identification of thermotolerant campylobacters from poultry meat. *Food Technology and Biotechnology* 40, 177–183.

Paper received: 28.04.2009.

PARAMETRI I KRITERIJUMI ZA OCENU KVALITETA POLUTKI I MESA SVINJA*

Petrović Ljiljana, Tomović V., Džinić Natalija, Tasić Tatjana, Ikonić P.

Sa dr ž a j: U radu je dat pregled metodologije ocene kvaliteta polutki svinja na liniji klanja po obavezujućim regulativama EU, kao i rezultati koji se, u većini evropskih zemalja, postižu primenom tih procedura u pogledu prosečnog prinosa mesa u polutkama na nivou celih država.

Ukazano je na zastarelost i nepreciznost naših propisa u toj oblasti, te izostanak ocene kvaliteta i klasiranja svinjskih polutki koje su u prometu. Nadalje, detaljno je opisana metodologija razvoja matematičkih modela za dve manuelne metode: metoda dve tačke i invazivna-optička metoda (uređajem FOM), koje su predložene nacrtom budućeg Pravilnika o kvalitetu zaklanih svinja i kategorizaciji svinjskog mesa.

Takođe, predočeni su kriterijumi i parametri (pH_i , pH_v , SVV i boja-L*) za ocenu kvaliteta proizvedenog mesa, radi razvrstavanja po kvalitetu, na meso: BMV/PSE (bledo, meko i vodnjikavo); TCŠ/DFD (tamnocrveno, čvrsto i nevodnjikavo); CČN/RFN (crvenoružičasto, čvrsto i nevodnjikavo); BČN/PFN (bledo, čvrsto i nevodnjikavo) i CMV/RSE (crvenoružičasto, meko i vodnjikavo), svojstava, odnosno parametri i kriterijumi za senzornu ocenu kvaliteta mesa namenjenog preradi ili mikrokonfekciji i pakovanju, odnosno tokom skladištenja upakovanog mesa.

Ključne reči: svinje, kvalitet polutki, kvalitet mesa, parametri, kriterijumi

PARAMETERS AND CRITERIA FOR QUALITY EVALUATION OF PORK CARCASS HALVES

A b s t r a c t: The paper gives a review of assessment methodology of pig carcass quality on the slaughterline according to EU madatory regulations, and also the results achieved applying that procedures in most of the European countries, regarding the average meat yield in carcasses in that countries.

The paper points out that our regulations in this area are not current and precise, therefore the carcass quality on the market is not assessed and the pork carcasses are not classified. Further, the methodology of mathematical model development is described for two methods: manual two-point method and invasive-optical by FOM device, which are proposed in the draft of Regulations on quality of slaughtered pigs and pork categorization.

The criteria and parameters (pH_p , pH_v , WHC and colour – L*) for the quality assessment of produced meat quality are pointed out, for the classification of meat in the following categories: PSE (pale, soft, exudative), DFD (dark red, firm and dry), RFN (red-pink, firm, non-exudative), PFN (pale, firm and non-exudative) and RSE (red-pink, soft and exudative), i.e. parameters and criteria for sensory quality assessment of meat intended for processing or retail cut and packing, i.e. during storage of packed meat.

Key words: pig, carcass quality, meat quality, parameters, criteria

Uvod

Postupak ocenjivanja kvaliteta polutki i mesa svinja na liniji klanja, kao i mesa nakon hlađenja, a u novije vreme i u distribuciji, naročito kada je meso mikrokonfekcionirano i na neki od poznatih načina upakovano, svakodnevna je procedura u savremenom konceptu proizvodnje svinjskog mesa. To je značajna karika u specifičnom lancu proizvodnje

i plasmana svinjskog mesa, od proizvođača svinja, odnosno svinjskog mesa do potrošača. Utvrđivanje kvaliteta polutki i mesa u svim fazama proizvodnje, daje osnov za optimalno iskorišćenje sirovine usmeravanjem polutki, odnosno mesa, na dalju preradu ili u maloprodaju, u skladu sa utvrđenim svojstvima. Sa druge strane, na taj se način iskazuju rezultati mnogobrojnih aktivnosti u uzgoju i selekciji svinja. Povratna informacija proizvođačima svinja o posti-

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTORI: Ljiljana Petrović, ljiljapet@uns.ns.ac.yu, Natalija Džinić, Vladimir Tomović, Tehnološki fakultet, Novi Sad; Tatjana Tasić, Predrag Ikonić, Institut za prehrambene tehnologije, Novi Sad

AUTHORS: Ljiljana Petrovic, ljiljapet@uns.ns.ac.yu, Natalija Dzinic, Vladimir Tomovic, Technological Faculty, Novi Sad; Tatjana Tasic, Predrag Ikonc, Institute for Food Technology, Novi Sad

gnutom kvalitetu omogućava im da sagledaju uspešnost rada kao i smernice za dalje unapređenje u toj oblasti. Pri tome, od najvećeg značaja je što ocena kvaliteta polutke i mesa omogućava odgovarajuće vrednovanje u svim fazama proizvodnje i iskazivanje utvrđene vrednosti kroz cenu. Što, takođe, deluje kao dodatni podsticaj daljem unapređenju kvaliteta i rentabilnosti proizvodnje i plasmana svinjskog mesa.

U našoj zemlji se, nažalost, ni postojeći propis o klasiranju svinjskog mesa u polutkama ne primenjuje, a o oceni kvaliteta mesa radi sistematskog praćenja i preduzimanja korektivnih mera ili radi odgovarajućeg vrednovanja, odnosno cenovnog iskazivanja ostvarenih rezultata u pogledu tehnološkog kvaliteta mesa, za sada, nema ni govora. Stoga je želja autora ovoga rada da predoče domaćoj stručnoj i naučnoj javnosti procedure na osnovu kojih to može da se čini, na koji način se odgovarajući parametri i kriterijumi za ocenu kvaliteta polutke i mesa svinja mogu da ugrade i u naše propise, odnosno šta je, ipak, postignuto, do sada, u toj oblasti kod nas.

Ocena kvaliteta polutke

Od kolikog je značaja ocena kvaliteta trupova i polutke svinja potvrđuje činjenica da su u evropskim zemljama sa razvijenim uzgojem svinja, prvi standardi za ocenu kvaliteta trupova definisani još krajem šezdesetih godina prošlog veka. Na bazi tih nacionalnih standarda Zapadne Nemačke i Holandije oformljen je prvi standard EEZ-a čija je primena u šest zemalja, tadašnjim članicama EEZ-a, započela 1970. godine (Živković, 1985; Srećković i sar., 1985; Nikolić i sar., 1989; Manojlović i Petrović, 1999; Džinić i sar., 2006a; 2006b). Standard je i kao nacionalni, bio zasnovan na stanovištu da se pod kvalitetom trupa podrazumeva prinos, odnosno količina mesa i njegova raspoređenost na trupu. Taj stav je bio dominantan, mada su već sedamdesetih godina prošlog veka postojala i takva mišljenja da kvalitet trupa obuhvata činioce koji se odnose na randman, ali i činioce kvaliteta mišićne mase (Rahelić, 1984; 1987; Rede, 1987), pa osobine trupa treba da se izražavaju odnosom tkiva na trupu (mišićno i masno) i karakteristikama tih tkiva (Živković, 1985; Rede i Petrović, 1997; Petrović i Manojlović, 1999). Standard EEZ-a je uključivao masu polutke, debljinu masnog tkiva lednog dela na dva merna mesta, kao i ocenu tipa i konformacije polutke. Ta merenja su bila osnov za određivanje procenta mesnatosti i trgovačke klase polutke, odnosno trupova. Mada je u početku u nekim zemljama postojao otpor prema oceni konformacije trupa, već 1975. godine je usledila

izmena standarda kojom je usaglašena klasifikacija polutke prema prinosu mesa, a zatim je usvojena regulativa [Commission Regulation (EEC) No 2967/85; 1985]. Posle ova dva standarda, zasnovana na merenju mase, linarnih parametara i vizuelnoj klasifikaciji polutke, januara 1989. godine stupio je na snagu SEUROP standard EEZ-a koji uključuje primenu elektronskih instrumenata i kompjutersku obradu podataka. Standard je, prethodno, punih pet godina bio u fazi provere, testiranja i tehničko-organizacionih ispitivanja.

Rezultati dugogodišnje primene ovih standarda sumirani su u okviru EUPIGCLASS projekta, na osnovu kojih može da se najbolje oceni postignut efekat, najpre u nekim zemljama EU-15, a potom i stanje u zemljama koje su tada trebale da postanu članice EU (NAS), (Hansson, 2003).

Rukovodilac ovog dela istraživanja (Hansson, 2003), u rezimeu, naglašava da su rezultati, koji su u ovom radu predočeni u skraćenom obimu u tabeli 1, zasnovani na podacima dobijenim kroz upitnik poslat svim zemljama članicama EU i NAS zemljama. Kao dopuna korišćeni su i neki oficijelni statistički podaci. Cilj je bio da se prikupe informacije za elaboraciju Programa obezbeđenja kvaliteta (QAP), jednog od zadataka projekta.

U ovom izveštaju je ukratko opisana proizvodnja svinja u Evropi, iz kojeg se vidi da je skoro 200 miliona svinja zaklano u 2001. godini, kada su podaci prikupljeni. U najvećem broju zemalja proizvode se hibridni tovljenici, ukrštanje se obavlja sa belim rasama (VJ i ŠL) u populaciji krmača, a durok, hempšir i pietren su rase nerastova. Koriste se i neke sintetičke linije.

U skoro svim zemljama svinje se kolju sa manje od 125 kilograma žive mase, dajući trup na liniji klanja od 93 kilograma, i manje. Praćena su EU-pravila za pripremu. Klasifikacija je sprovedena u svim zemljama EU, a metode su bazirane na merenju debljine masnog i mišićnog tkiva za izračunavanje procenta mesa, izraženog kao udeo u masi celog trupa, u skladu sa EU zahtevima.

Program klasiranja su kontrolisali zvanični predstavnici državnih organa, u najvećem broju zemalja, u cilju sprovođenja zvanične ocene, sa visokim poverenjem industrije i tržišta. Specijalizovane organizacije, odgovorne za kontrolu, već postoje u većini zemalja, tako da je implementacija Programa obezbeđenja kvaliteta (QAP) bila veoma laka, kako zaključuje Hansson (2003) u ime istraživača tog dela Projekta.

Na našim prostorima, prvi propis o oceni trupova svinja na liniji klanja objavljen je već krajem 1969. godine. Bio je to Jugoslovenski standard za svinje za industrijsku preradu sa oznakom JUS

Tabela 1. Prikaz broja zaklanih svinja u zemljama EU i NAS, prosečne mase toplih trupova na liniji klanja, prosečni prinos mesa u polutkama i najčešće korišćeni uređaji i metode za određivanje prinosa mesa

Table 1. Number of slaughtered pigs in EU and NAS countries, average masses of warm carcasses at the slaughterline, average yield in carcass halves and the most often used devices and methods for meat yield determination

Zemlje članice EU i NAS zemlje	Broj zaklanih svinja 2001. godine	Prosečna masa toplih polutki (kg)	Prinos mesa u polutkama	
(%)	Uređaji koji su u upotrebi			
Danska	21.000.000	78	60	uređaj FOM, AUTOFOM, CC
Belgija	11.000.000	90	60	invazivni CGM ili PG 200 uređaj
Francuska	26.000.000	90	60	uređaj CGM i metoda dve tačke
Španija	36.000.000	79	58	uređaj FOM i HGP, AUTOFOM
Nemačka	40.000.000	93	56.7	uređaj FOM, AUTOFOM
Italija	13.000.000	uglavnom većih masa trupova	nema sistematizovanih podataka	FOM i HGP 4
Holandija	20.000.000	80	57	HGP
Irska	3.400.000	72	58,4	HGP ver 2
Litvanija	800.000	–	–	uređaj FOM – u razvoju
Estonija	500.000	77	56	nije u upotrebi ni jedan uređaj
Slovenija	500.000	82	55,4	metoda dve tačke
Poljska	20.000.000	80	50	različiti tipovi manuelnih testova, AUTOFOM–u razvoju
Mađarska	3.300.000	90	53	FOM
Bugarska	2.000.000	70	~ 45	nije uvedeno klasiranje, HGP za istraživanja
Slovačka	1.800.000	90	52	FOM i metoda dve tačke
Češka	3.600.000	88	54	FOM i metoda dve tačke
Kipar	650.000	75	55	invazivni uređaji i HGP
Švedska	3.300.000	89	57	HGP
Norveška	1.300.000	80	56	HGP 4
Finska	2.200.000	–	–	HGP 2 i 4

E.C1.021 III-1969, s tim što se sa ocenom mesnatosti polutki svinja na linijama klanja u industrijskim klanicama tadašnje SFRJ započelo aprila 1973. godine. Uz određene korekcije, standard je bio u upotrebi sve do donošenja Pravilnika o kvalitetu zaklanih svinja i kategorizaciji svinjskog mesa sa zakonskom primenom od aprila 1985. godine.

Danas je, u Republici Srbiji, još na snazi i delimično je u upotrebi citirani Pravilnik o kvalitetu zaklanih svinja i kategorizaciji svinjskog mesa („Sl. list SFRJ“ br. 2 i 12 iz 1985. godine). Ovim Pravilnikom propisuju se minimalni uslovi koje, u pogledu kvaliteta, mora da ispunjava meso svinja (svinjsko meso) u trupovima, polutkama i osnovnim delovima polutke i jestivi delovi zaklanih svinja, kao i uslovi držanja, čuvanja, pakovanja i transportovanja tog mesa i tih jestivih delova.

Po odredbama citiranog Pravilnika pod mesnatošću trupa, ili svinjskih polutki podrazumeva se ukupna masa mišićnog tkiva bez mesa trbušno-rebarnog dela i bez mesa glave. Mesnatost polutki mesnatih svinja utvrđuje se na liniji klanja, najkasnije jedan čas posle klanja, a meri se masa toplih polutki i debljina masnog tkiva na leđima. Masno tkivo na leđima, sa kožom, meri se na sredini leđa, gde je masno tkivo najtanje (međurebarni prostor između 13. i 15. lednog pršljena) i na krstima na mestu na kome mišić *M. Gluteus medius* najviše urasta u masno tkivo. Zbir tih mera predstavlja debljinu masnog tkiva na leđima. Za određivanje prinosa mesa mesnatih svinja u polutkama, na osnovu obavljenih merenja, koriste se tabela 1 (prinos u kilogramima) i tabela 2 (prinos u procentima), koje čine sastavni deo Pravilnika.

Opšti izgled trupa, polutke, četvrti, osnovnih delova ili jestivih delova zaklanih svinja, kao i originalno upakovanog mesa, utvrđuje se adspekcijom i palpacijom i obuhvata: oblik i građu polutke, četvrti ili osnovnih delova i jestivih delova, razvijenost mišićnog i masnog tkiva i oštećenja i promene boje mesa, odnosno jestivih delova.

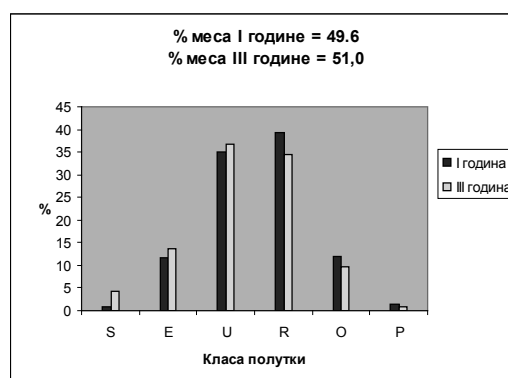
Naš Pravilnik predviđa klasifikaciju svinjskih polutki i osnovnih delova svinjskih polutki samo na osnovu toga da li su polutke i osnovni delovi polutke namenjeni za promet (oznaka „K“), ili za preradu (oznaka „P“).

U domaćoj literaturi, gotovo od momenta donošenja Pravilnika, u velikom broju radova autori iznose negativna iskustva stečena primenom istog (Nikoilić i sar., 1989; Petrović i sar., 1996; Petrović i Manojlović, 1999; Vidović, 1999; Tomović, 2002; Petrović Ljiljana i sar., 2003; Džinić, 2005), te ukazuju da u Pravilniku postoji neusaglašenost između prinosa mesa u polutkama zaklanih svinja, izraženog u kilogramima, i prinosa mesa izraženog u procentima.

Nadalje je, poznato da metodologija za određivanje mesnatosti navedena u Pravilniku daje, u istim polutkama, u proseku, za oko 10 do 12 posto manje vrednosti od vrednosti dobijenih metodom parcijalne disekcije (Vidović, 1999; Petrović i Manojlović, 1999; Džinić i sar., 2001; Tomović, 2002; Džinić, 2005; Okanović i sar., 2006).

Rezimirajući, može se reći, da se brojni istraživači slažu u oceni da su dobijeni rezultati o prinosu mesa (kg, procenat) u polutkama, određeni prema važećem Pravilniku, krajnje nepouzdana, te da je to verovatni uzrok prestanka klasiranja svinjskog mesa na linijama klanja svinja u Srbiji i izostanka prometa klasiranog mesa u polutkama. Takođe, može da se kaže da smo jedna od retkih, ako ne i jedina zemlja u Evropi u kojoj se u prometu nalaze neklasirane polutke svinja, sa svim negativnim posledicama po naše svinjarstvo, ali i industriju mesa (Petrović i sar., 2003; Džinić, 2005; Okanović i sar., 2006).

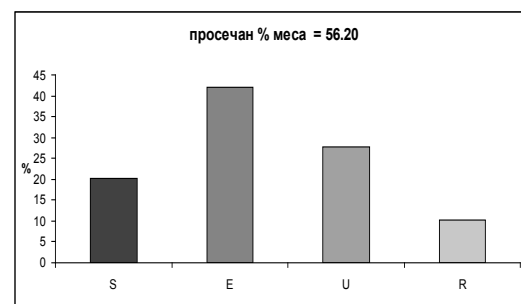
Pošto se proizvodnja u našoj industriji mesa ne prati na način kako je to regulisano u EU, teško je reći čak i koliki je broj svinja zaklan u našim klanicama u poslednjih desetak godina, a prosečne mase toplih polutki, ili prinos mesa u polutkama veoma je teško čak i proceniti. No, u okviru Projekta BTN351008: „Proizvodnja i priprema svinjskog mesa za veleprodaju, maloprodaju, industriju gotove hrane i preradu“ kojeg je finansiralo Ministarstvo za nauku Republike Srbije, u periodu od 2005. do 2008. godine Petrović i sar. (2008) su u nekoliko navrata određivali prosečan kvalitet polutki (procenat mesa) u jednoj našoj industrijskoj klanici, a dobijeni podaci su predočeni na grafiku 1.



Grafikon 1. Prosečan kvalitet polutki (procenat mesa) u I (n = 833) i III (n = 702) godini ispitivanja

Figure 1. Average quality of carcass halves (% of meat) in 1st (n = 833) and 3rd year of investigation

Kako spomenuta industrija mesa poseduje i sopstvenu farmu, u okviru ovog Projekta sprovedene su mnogobrojne aktivnosti radi poboljšanja kvaliteta polutki zaklanih svinja, kako u selekciji i ukrštanjem, tako i u ishrani. Kao rezultat preduzetih mera registrovano je poboljšanje kvaliteta trupova u III-oj u odnosu na I godinu, istraživanja na nivou cele farme sa koje su poticale ispitane svinje. Istovremeno, intenzivno se radilo i na formiranju višerasnih hibrida, uglavnom trorasnih i četvororasnih, a postignuti rezultati, u odnosu na prinos mesa u polutkama, predočeni su na grafikonu 2. (Tomović, 2002; Džinić i sar., 2004; Džinić, 2005; Petrović i sar., 2006; Džinić i sar., 2006a; 2006b)



Grafikon 2. Učestalost komercijalnih klasa (SEUROP) polutki svinja u populaciji trorasnih i četvororasnih hibrida svinja (n = 217) formiranih u programu ukrštanja na jednoj našoj farmi

Figure 2. Frequency of commercial classes (SEUROP) of pig carcasses in the population of threebreed and fourbreed pig hybrids (n = 217) obtained in the breeding programme on one of our farms

Na osnovu iznetih podataka i velikog iskustva autora, može, gotovo sa sigurnošću, da se tvrdi da je to i prosečna slika kvaliteta polutki (procenat mesa ~ 50–52 posto) proizvedenih u našoj zemlji. Dakle, nedvosmisleno može da se zaključi da izostanak sistematskog praćenja kvaliteta polutki na liniji klanja za posledicu ima ovako slab kvalitet polutki koji nas, po predočenim rezultatima, svrstava na samo dno evropskih uzgajivača svinja i proizvođača svinjskog mesa (Petrović i Manojlović, 1999; Vidović, 1999; Tomović, 2002; Petrović i sar., 2003; Džinić Natalija, 2005, Okanović i sar., 2006; Džinić i sar., 2007)

S obzirom na predočene nedostatke našeg Pravilnika, u zemljama nastalim iz bivše SFRJ taj Pravilnik je izmenjen (u Republici Sloveniji 1995, a u Republici Hrvatskoj 1999. godine) i usklađen sa aktuelnim propisima u EU. Sve izneto nameće potrebu usaglašavanja i naših priopisa sa standardima razvijenih zemalja, pre svega zemalja EU i uvođenja savremenih metoda za ocenu kvaliteta polutki na linijama klanja svinja, sa obaveznom primenom.

Razumevajući potrebu da se u procesu usaglašavanja domaćeg zakonodavstva sa zakonodavstvom EU, ali i da se u proizvodnji svinja i svinjskog mesa krene u pravcu unapređenja, Ministarstvo poljoprivrede, šumarstva i vodoprivrede je sa Tehnološkim fakultetom u Novom Sadu sklopilo ugovor radi realizacije projekta: „Definisanje parametara i kriterijuma za ocenu kvaliteta polutki svinja u cilju izrade predloga pravilnika o kvalitetu zaklanih svinja i kategorizaciji svinjskog mesa“. Na projektu je angažovano 18 istraživača iz 6 Naučno-istraživačkih organizacija, uključujući i predstavnika Udruženja republičkog saveza uzgajivača svinja. Projekat je realizovan u periodu od 16. juna 2008. do 16. marta 2009. godine, kada je, na osnovu obavljenih istraživanja, Ministarstvu predat nacrt Pravilnika o kvalitetu zaklanih svinja i kategorizaciji svinjskog mesa (2009).

U nastavku će biti predočena metodologija rada, kao i dati predlozi metoda i postupaka za ocenu kvaliteta polutki predloženih u nacrtu Pravilnika (2009).

U zemljama EU [Council Regulation (EC) No 3513/93..., 1993] pod svinjskim trupom/polutkama podrazumeva se trup zaklane, iskrvarene i eviscerirane svinje, ceo ili rasečen niz središnju liniju, bez jezika, čekinja, papaka, genitalnih organa, sala, bubrega i dijafragme. Posebnim propisom EU [Commission Regulation (EEC) No 2967/85..., 1985] usvojeno je da se masa toplog trupa/polutki i mesnatost odrede, što je moguće pre, odnosno najkasnije 45 minuta nakon klanja, kao i da se masa ohlađenog trupa/polutki dobija umanjivanjem mase toplog trupa/polutki za 2 posto.

Primereno savremenim zahtevima u pogledu kvaliteta, sasvim je razumljivo da se u praksi zemalja, pre svega onih sa tradicionalno razvijenim stočarstvom i proizvodnjom mesa, javila potreba da se u dugom procesu proizvodnje, što je moguće pre, predvidi i/ili utvrdi kvalitet polutki, odnosno trupova. Rezultati tih zahteva, a pre svega multidisciplinarnog pristupa problematici, su savremene metode i vrlo složena tehnička rešenja, čija primena omogućava da se merenjem odabranih pokazatelja kvaliteta, obradom i evidencijom dobijenih podataka precizno utvrdi i objektivno izdiferencira kvalitet, vrednost i klasa polutki/trupova, kako u primarnoj proizvodnji (*invivo*), tako i na liniji klanja. (Radovanović, 1992; 2001; Rede i Petrović, 1997; Petrović, 1999; Tomović, 2002; Džinić, 2005).

Zajednička odlika svih, do sada usavršenih rešenja, je da se radi o vrhunskoj i vrlo osetljivoj bio-medicinskoj opremi, odnosno elektronskim, optičkim, ultrazvučnim i video mernim instrumentima prilagođenim radu u nepovoljnim mikroklimatskim uslovima pogona industrije mesa. Ovi uređaji se, po pravilu, jednostavno montiraju i podešavaju za rad, veoma brzo daju precizne informacije, ispunjavaju sve zahteve u pogledu higijene i bezbednosti, a obučena lica ih veoma lako koriste (Radovanović, 1992; 2001; Rede i Petrović, 1997; Tomović, 2002; Džinić, 2005).

Mnoge od tih novih mernih instrumenata, u EU, SAD, Kanadi, Australiji, Novom Zelandu i drugim razvijenim zemljama priznale su odgovarajuće državne komisije tih zemalja i već su potvrđeni kroz široku primenu u proizvodnim uslovima, odnosno zvanično su uvršćeni u odgovarajuće nacionalne propise o klasiranju svinjskog mesa u trupovima.

Iako se radi o savremenoj instrumentalnoj opremi, primena ovih uređaja zahteva njihovu prethodnu kalibraciju. Metodologija kalibracije merne opreme kao i kriterijumi za utvrđivanje preciznosti, odnosno ponovljivosti merenja definisani su odgovarajućim propisima EU [Commission Regulation (EEC) No 2967/85..., 1985; Commission Regulation (EC) No 3127/94..., 1994]. Naime, svi uređaji, u svom softverskom paketu, imaju ugrađen matematički model za izračunavanje procenta mesa, koji se definiše regresionom analizom na bazi veličina izmerenih instrumentalno (najčešće debljina masnog i mišićnog tkiva) i procenta mesa određenog metodom parcijalne disekcije. Da bi matematički model bio prihvaćen, odnosno da bi rezultati dobijeni instrumentalnim merenjem bili precizni i ponovljivi, usvojeno je da standardna devijacija regresije (RMSE), izračunata između procenata mesa određenih instrumentalnom metodom i metodom parcijalne disekcije, na reprezentativnom uzorku od najmanje 120 polutki, mora da bude manja od 2,5 posto,

s tim da, ukoliko se matematički model definiše dvostrukom regresijom, za proveru varijabilnosti (RMSE < 2,5 posto) je dovoljno izvršiti merenja samo na 50 polutki. S obzirom na to da je disekcija cele polutke na osnovna tkiva vrlo komplikovana i dugotrajna, usvojena je metoda parcijalne disekcije, koju su detaljno opisali Walstra i Merkus (1996). Po ovoj metodi, polutka se, po anatomski precizno definisanoj shemi, raseca na 12 delova, a samo četiri najznačajnija dela (but, leđno-slabinski deo, plečka i rebarno-trbušni deo) polutke, koji sadrže 75 posto svih poprečno-prugastih mišića, se disekiraju na osnovna tkiva. Na osnovu mase mišićnog tkiva u tim delovima i mase podslabinskog mišića (filea) izračunava se procenat mesa u polutki, množenjem tog zbira sa faktorom 1,3.

Na osnovu utvrđenog procenta mesa, u zemljam EU [Council Regulation (EEC) No 3220/84..., 1984], trupovi/polutke se klasiraju u šest komercijalnih klasa prema sledećoj skali: $S \geq 60$; $55 \leq E < 60$; $50 \leq U < 55$; $45 \leq R < 50$; $40 \leq O < 45$; $P \leq 40$.

Detaljnou analizom citiranih evropskih regulativa kao i nacionalnih propisa pojedinih zemalja EU (Engleska, Irska, Slovenija) kojima se uređuje opisana problematika, dolazi se do saznanja da većina nacionalnih propisa daje mogućnost ocene kvaliteta polutki i na bazi ručno (manuelno) uzetih podataka (linearnih mera) i izračunavanja pomoću relativno jednostavnih formula (matematičkih modela), koji su, naravno, utvrđeni i provereni na temelju citiranih zahteva. Takođe, se u citiranim propisima definiše i propisuje za veće objekte, u kojima se nedeljno kolje više od 200 svinja, obavezna upotreba nekog od savremenih uređaja (najčešće FOM-a ili nekog drugog invazivnog optičkog uređaja) i za taj uređaj daje se obavezujuća matematička formula za izračunavanje procenta mesa u polutkama (klase).

Kao rezultat rada na spomenutom Projektu EUPIGCLASS, Daumas (2003) opisuje proceduru koju treba primeniti pri izboru statistički reprezentativnog uzorka od 120 svinja za uzimanje lineranih mera za debljinu masnog tkiva i mišića na toplim polutkama, za odabranu manuelnu ili instrumentalnu metodu, kao i način sprovođenja parcijalne disekcije istih ohlađenih polutki, pri definisanju nacionalnih obavezujućih parametara i kriterijuma za klasiranje svinjskog mesa u skladu sa EU regulativama. Ta procedura je poštovana i pri realizaciji našeg Projekta finansiranog sredstvima Ministarstva poljoprivrede Republike Srbije, odnosno pri odabiru prosečnih uzoraka koji će na najbolji način statistički reprezentovati varijabilnost domaće tovne svinje, a sastojala se u sledećem:

Broj polutki: Za konstruisanje matematičkih izraza $120(147) + 50$

Rase: Treba ravnomerno odabrati rase svinja koje se uzgajaju u celoj zemlji. Odabrani genotipovi svinja za ispitivanja bili su: švedski landras – 29 grla; veliki jorkšir – 14 grla; dvorasni melezi: švedski landras x veliki jorkšir – 12 grla; veliki jorkšir x švedski landras – 9 grla; švedski landras x pijetren – 3 grla; trozasni melezi: (švedski landras x veliki jorkšir) x durok – 12 grla; (švedski landras x veliki jorkšir) x pijetren – 21 grlo; (švedski landras x veliki jorkšir) x hempšir – 14 grla; četvororasni melez: (švedski landras x veliki jorkšir) x (durok x pijetren) – 6 grla. (U svim kombinacijama meleza prvo je prikazan genotip majke pa oca.)

Farme: Svinje treba da potiču sa više farmi. U sprovedenim ispitivanjima svinje su poticale sa farmi: „Čenej“ – Čenej; „Aleksa Šantić“ – Aleksa Šantić; „Nukleus“ – Rača Kragujevačka; „Vizelj“ – Padinska Skela; „Union MZ“ – Požarevac i „Institut za stočarstvo“ – Zemun.

Pol: Oba pola, a muška grla moraju da budu kastrirana najmanje 30 dana pre klanja (cca 50 : 50). U obavljenom ispitivanju bilo je 71 ženskih i 76 muških grla.

Klanice: Veći broj. U obavljenim ispitivanjima: IM „Neoplanta“, Novi Sad; „Imes“, Padinska Skela; „Institut za stočarstvo“, Zemun; „Union MZ“, Požarevac.

Timovi za disekciju: Veći broj. U obavljenim ispitivanjima: mesari klanice „IMES“ prva ekipa (7); mesari klanice „IMES“, druga ekipa (11); mesari klanice „IMES“ plus mesari Instituta za stočarstvo Beograd, Zemun (11); mesari Instituta za stočarstvo Beograd, Zemun (29); mesari klanice „UNION MZ“ plus mesari Instituta za stočarstvo Beograd – Zemun (18); mesari IM „Neoplanta“ prva ekipa (37); mesari IM „Neoplanta“, druga ekipa (34), a pri svakoj disekciji bili su prisutni i istraživači sa Projekta (najmanje 4 uz svaku ekipu mesara).

Merenje mase toplih polutki: 45' post mortem (EU 3220/84) (Tačnost vage $\pm 0,5$ kg)

Masa ohlađenih polutki: Masa toplih polutki umanjena za 2 posto (EU 2967/85). U ovim istraživanjima, zbir 12 osnovnih anatomskih delova dobijenih rasecanjem po definisanom postupku parcijalne disekcije + file.

Definicija polutke: Polutka bez jezika, čekinja, papaka, genitalnih organa, sala, bubrega i dijafragme (EU 3513/93).

Kriterijumi za mase polutki (tovne svinje za klanje): Masa toplih polutki od 50 do 120 kg (EU 3513/93).

Grupe polutki po masama: Odabrati po mogućnosti podjednak broj polutki iz svih težinskih grupa u definisanom rasponu za Tovne svinje (grafikoni 3.a i 4.a).

Grupe polutki po procentu mesa: U svakoj težinskoj grupi i u svakoj grupi po procentu mesa trebalo bi disekcirati podjednak broj polutki (grafikon 3.b i 4.b).

Debljine masnog i mišićnog tkiva na toplim polutkama (merna mesta) na kojima su uzimane linearne mere, radi konstruisanja matematičkih izraza-modela za određivanje procenta mesa:

1. Prema našem važećem Pravilniku (u medijalnoj ravni):

Na sredini leđa gde je slanina najtanja, sa kožom, u milimetrima (između 13. i 15. leđnog pršljena) – merno mesto LEĐA,

Na krstima gde *M. gluteus medius* najviše urasta u slaninu, sa kožom, u milimetrima – merno mesto KRSTA.

2. Prema FOM uređaju, ali u medijalnoj ravni (za manuelnu metodu dve tačke):

Između 3. i 4. lumbalnog pršljena (gledano kaudo-kranijalno) – debljina masnog tkiva sa kožom (MT1), u milimetrima; između 3. i 4. poslednjeg rebra – debljina masnog tkiva sa kožom (MT2) u milimetrima.

3. Prema FOM uređaju (za instrumentalnu invazivnu metodu):

Između 3. i 4. poslednjeg rebra, 7 santimetara od medijalne ravni – debljina masnog tkiva sa kožom (RF) i debljina *M. longissimus dorsi* (RM), u milimetrima; između 3. i 4. lumbalnog pršljena (gledano kaudo-kranijalno), 8 santimetara od medijalne ravni – debljina masnog tkiva sa kožom (LF), u milimetrima.

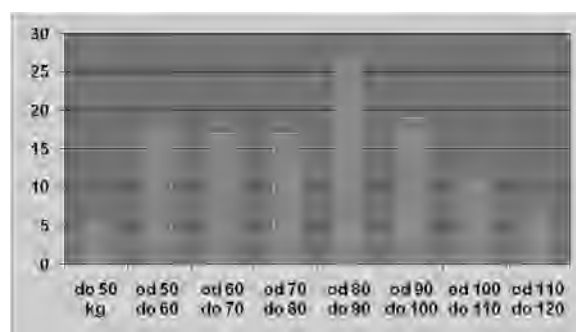
4. Prema francuskoj, slovenačkoj, hrvatskoj, itd. metodi dve tačke (manuelna metoda):

Debljina masnog tkiva sa kožom, u milimetrima, izmerene na krstima na najtanjem mestu, odnosno gde *M. gluteus medius* najviše urasta u slaninu (S),

Debljina *M. longissimus dorsi* (slabinskog mišića), mereno kao najkraća veza prednjeg (kranijalnog) završetka *M. gluteus medius* sa gornjim (dorzalnim) rubom kičmenog kanala (M).

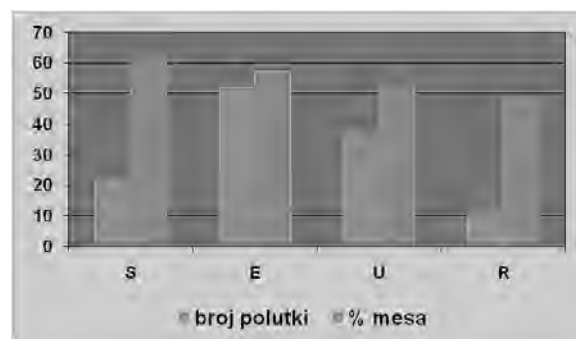
5. Postupak disekcije: Leve polutke (EU 3127/94) disekcirane su po proceduri koju su opisali Walstra i Merkus (1996). Disekcija se but, plećka, kare i rebarno-trbušni deo + file.

Od ukupno 147 obavljenih disekcija za **metodu FOM uređaja** uzeti su podaci za 120 polutki na kojima su izmerene linearne mere bile validne po metodologiji rada uređaja FOM, odnosno postojali su svi podaci za disekciju i linerne mere.



Grafikon 3.a Broj polutki po težinskim grupama za metodu FOM uređaja

Figure 3.a. Number of carcass halves by weight groups for the method FOM device



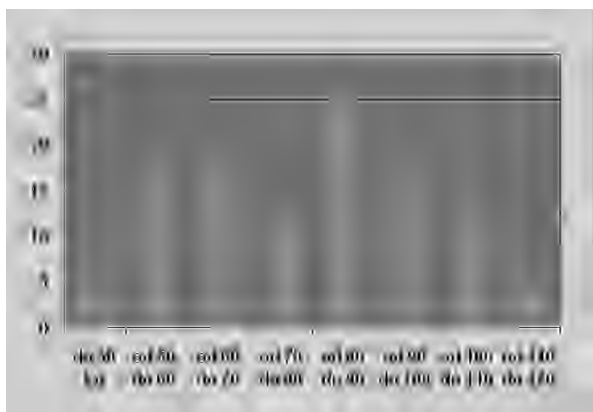
Grafikon 3.b Broj polutki po klasama i prosečan prinos mesa po klasi za metodu FOM uređaja

Figure 3.b. Number of carcass halves by classes and average yield in the class for the method of FOM device

Od ukupno 147 obavljenih disekcija za **metodu dve tačke** uzeti su podaci za 120 polutki na kojima su izmerene linearne mere bile validne po metodi dve tačke, odnosno postojali su svi podaci za disekciju i linerne mere (na 107 istih polutki su uzete linearne mere za obe metode, odnosno podaci za disekciju, a na po 13 različitih polutki su uzete linerne mere samo za metodu uređaja FOM, odnosno za metodu dve tačke, sa odgovarajućim podacima za disekciju).

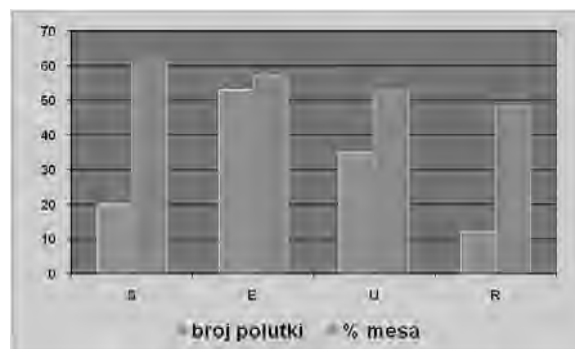
Linearne mere uzete po metodologiji važećeg Pravilnika i metodologiji uređaja FOM, ali u medijalnoj ravni – za metodu dve tačke nisu dalje razmatrane jer ni pojedinačno ni na ukupnom uzorku (n=120) u poređenju sa rezultatima disekcije nije dobijena tražena pouzdanost (RMSE < 2,5 posto).

Statistička obrada dobijenih podataka radi konstruisanja matematičkih modela urađena je prema dokumentaciji koja se šalje u Brisel za zemlje EU radi sertifikacije metoda za ocenu mesnatosti na liniji klanja iz „EUIPIGCLASS“ projekta (*Causeur i sar.*, 2006).



Grafikon 4.a Broj polutki po težinskim grupama za metodu dve tačke

Figure 4. a. Number of carcasses halves by weight groups for the „two points” method



Grafikon 4.b Broj polutki po klasama i prosečan prinos mesa po klasi za metodu dve tačke

Figure 4. b. Number of carcass halves by class for „two points” method

Utvrđena je jednačina višestruke linearne regresije za ocenu mesnatosti, RMSE, RMSEP (Root Mean Squared Error of Prediction), (Causeur i sar., 2006), pri čemu je vrednost RMSEP preporučena kao validacioni kriterijum umesto vrednosti RMSE [Commission Regulation (EC) No 3127/94..., 1994)].

Konstruisani matematički model –Metoda FOM uređaja

$$Y = 55,6925 - 0,2402LF - 0,4575RF + 0,1578RM$$

$$RMSE = 2,12$$

$$RMSEP = 2,16$$

(videti Grafikon 5)

Konstruisani matematički model - Metoda dve tačke

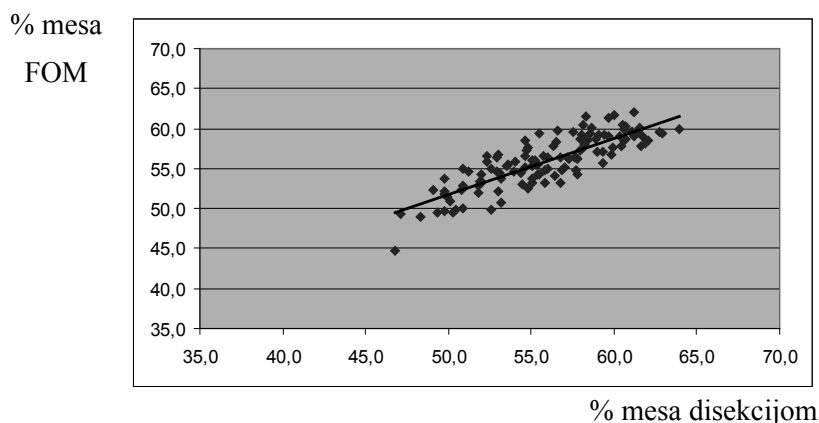
$$Y = 49,6358 - 0,5667S + 0,2069M$$

$$RMSE = 2,12$$

$$RMSEP = 2,16$$

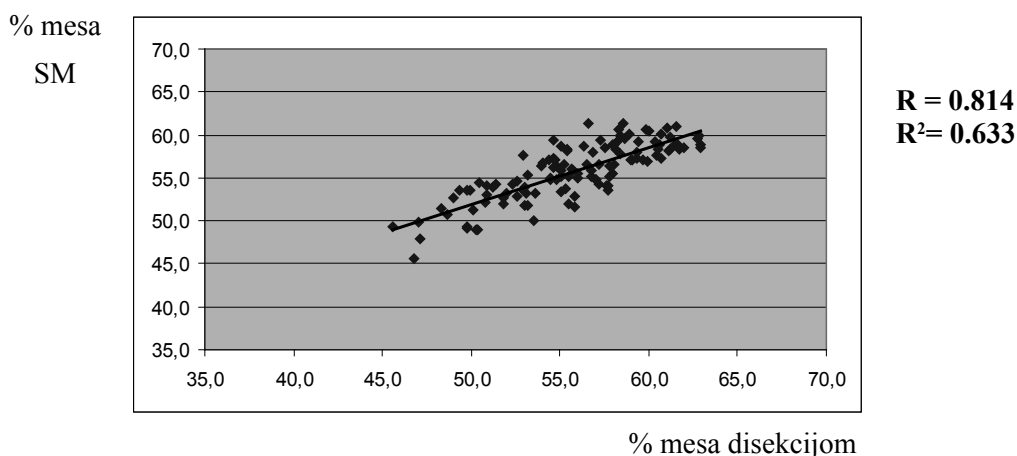
(videti Grafikon 6)

Disekcija i uporedno uzimanje linearnih mera sa 50 novih polutki za metodu dve tačke i instrumentalno određivanje % mesa i klase polutki, te dokazivanje da su izrađeni matematički modeli validni u poređenju sa rezultatima disekcije (RMSE < 2,5 posto), obavljena je na slučajnom uzorku, kako je predočeno na grafiku 7.a i 7.b

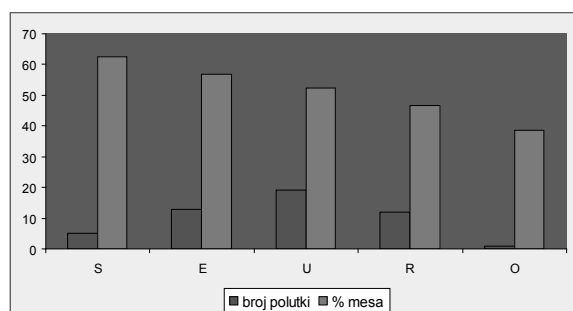


Grafikon 5. Linearna korelacija između prinosa mesa utvrđenog uređajem FOM i metodom disekcije

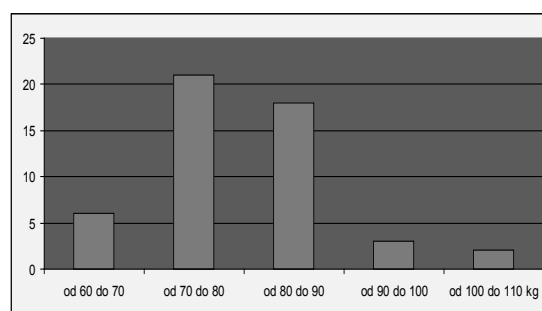
Figure 5. Linear correlation between the yield determined by FOM and dissection method



Grafikon 6. Linearna korelacija između prinosa mesa utvrđenog metodom dve tačke i metodom disekcije
Figure 6. Linear correlation between yield determined by „two points” method and dissection method



Grafikon 7.a Broj polutki po težinskim grupama
Figure 7a. Number of carcass halves by weight groups



Grafikon 7.b. Broj polutki po klasama i prosečan prinos mesa po klasi
Figure 7b. Number of carcass halves and average yield by class

Metoda FOM uređaja

$$Y = 55,6925 - 0,2402LF - 0,4575RF + 0,1578RM$$

$$RMSE = 2,27$$

$$RMSEP = 2,41$$

(videti Grafikon 8)

Konstruisan matematički model je validan.

$$Y = 49,6358 - 0,5667S + 0,2069M$$

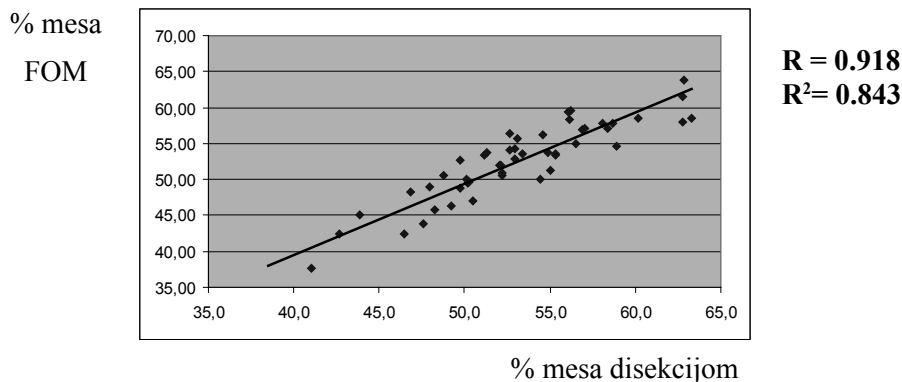
Metoda dve tačke

$$RMSE = 2,38$$

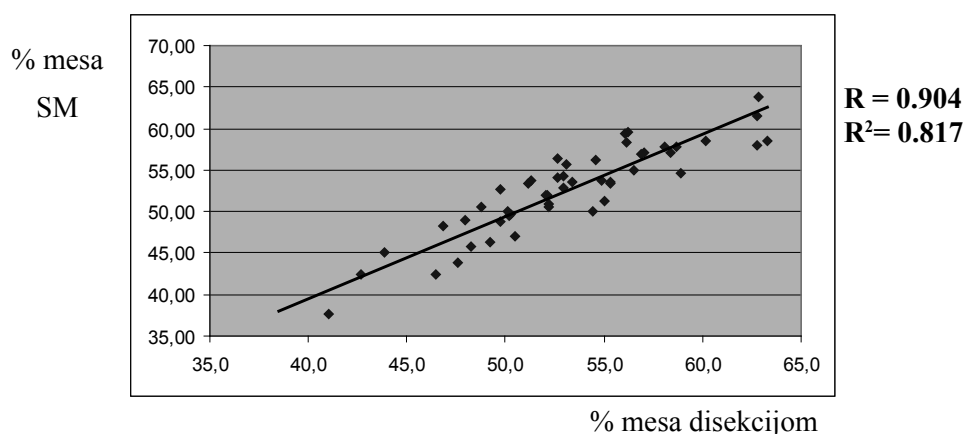
$$RMSEP = 2,39$$

(videti Grafikon 9)

Konstruisan matematički model je validan.



Grafik 8. Linearna korelacija između prinosa mesa utvrđenog uređajem FOM i metodom disekcije
Figure 8. Linear correlation between the meat yield determined by FOM and dissection method



Grafikon 9. Linearna korelacija između prinosa mesa utvrđenog metodom dve tačke i metodom disekcije
Figure 9. Linear correlation between meat yield determined by „two points” method and dissection method

ne validacije dobijeni matematički modeli, odnosno definisani parametri (za manuelnu metodu dve tačke i instrumentalnu, uređajem FOM) predloženi su u nacrtu Pravilnika (2009) za oficijelne, pri određivanju klase polutke po SEUROP kriterijumima. Naravno, da svi zainteresovani subjekti mogu da koriste i druge metode, odnosno uređaje, ali moraju da dokumentuju validnost korišćene metodologije po opisanoj proceduri iz nacrtu Pravilnika. Nacrtom Pravilnika predviđeno je da klasiranje sprovodi, za tu delatnost, imenovana kontrolna organizacija, od strane nadležnog ministarstva, ili po dogovoru zainteresovanih subjekata, u skladu sa ISO EN 45 004 standardima.

Ocena kvaliteta mesa

Pored mesnatosti trupova, čime se obezbeđuje kvantitet sirovine, za kvalitet proizvoda od mesa ili sam plasman svežeg mesa, od izuzetnog je značaja kvalitet mesa. Odnosno, kako ističe Radovanović (2001), zahtev za mesom vrhunskog kvaliteta ne predstavlja samo deo „navike” potrošača u ambijentu tržišne ekonomije, zavidne kupovne moći stanovništva i visokog obima potrošnje, primeren dostignutom nivou životnog standarda, već je to, u uslovima izražene konkurencije, imperativ uspešnog opstanka na tržištu. Dakle, u razvijenom delu sveta postoji saglasnost u pogledu jasno definisane strategije prema kojoj *kvalitet* proizvoda ima primarni značaj i nalazi se u centru pažnje svih aktivnosti, budući da predstavlja konkurentsku prednost i čini ciljanu osnovu razvoja, dok je *kvantitet* samo jedan od integralnih elemenata kvaliteta.

Kvalitet mesa je termin koji sveobuhvatno opisuje biohemijske, hemijske i fizičko-hemijske karakteristike mesa (Honikel, 1999).

Kvalitet mesa je rezultat složenih i osetljivih biohemijskih procesa i promena koje se u mišiću odvijaju nakon klanja. Skup faktora koji utiču na tok i intenzitet postmortalnih procesa i promena je veoma širok, a složeni biohemijski procesi rezultiraju formiranjem kompleksa svojstava koje obuhvataju pojmom „kvalitet” (Rede i Petrović, 1997).

Pri određivanju kvaliteta od presudnog su značaja dva momenta i to: definisanje faktora (parametara) kvaliteta na osnovu kojih se izražavaju pojedinačna svojstva kvaliteta i kvantitativno izražavanje tih karakterističnih svojstava (kriterijuma) u odnosu na opšti kvalitet. Ocena kvaliteta je potpunija, ukoliko je ispitani i definisan veći broj svojstava (Joksimović, 1977).

Različite grupe potrošača imaju različite zahteve u pogledu kvaliteta. Neki su zainteresovani pre svega, za dobra senzorna svojstva, odnosno izgled mesa. Velike razlike u boji, bez obzira da li su posledica razlike u SVV, pH ili hemijskog stanja pigmenta, nisu poželjne. Odrezak u prodaji treba da je krto meso; veće količine masnog tkiva su nepoželjne, čak i ako povoljno utiču na ukus. Drugi potrošači mogu da budu zainteresovani za uslove pod kojima je meso proizvedeno, za koje smatraju da su etički prihvatljivi. Potrošači koji su spremni da plate posebnu cenu za takvo meso bili su, do sada, uglavnom koncentrisani na način uzgoja, ali nema sumnje da će da budu zainteresovani i za postupak sa životinjama pre klanja. Proizvođači svežeg mesa su zainteresovani za tačno određen kvalitet mesa, tj. da nije BMV ili TČS meso. Prerađivače interesuje da je meso dobrih osobina za prerađivanje radi što boljeg iskorišćenja sirovine i kvaliteta proizvoda (Barton, Gade, 1985).

Honikel (1999) pod kvalitetom mesa podrazumeva zbir svih objektivno izmerenih (n) svojstava,

odnosno prema Hofmannu (1986), Honikel (1999) definiše kvalitet mesa kao skup svih tehnoloških, nutritivnih (hranljivih), senzornih i higijenskih (odnosno higijensko-toksikoloških) svojstava, odnosno faktora kvaliteta.

U poslednje vreme sve veća pažnja se posvećuje i tzv. „etičkom kvalitetu mesa“ koji podrazumeva „organski“, nasuprot „neorganskom“ uzgoju životinja, zatim religijske zahteve, dobrobit životinja („Animal Welfare“), kao i odobravanje, odnosno neodobravanje, genetske modifikacije životinja i stočne hrane. Takođe, velika pažnja se posvećuje i ispunjenju ekoloških standarda u uzgoju životinja i proizvodnji i preradi mesa (Murray, www.ccsi.ca/Meetings/ACM_Pork_Quality).

Merenje svojstava mora da se preduzme u pravo vreme, na način koji nije destruktivan i u reprezentativnim mišićima koji su lako dostupni (*M. semimembranosus* i *M. longissimus dorsi*) (Honikel, 1999).

Tradicionalno, govori se o tri sasvim izdiferencirana tehnološka kvaliteta svinjskog mesa. Proizvedeno meso (posle završenog hlađenja 24 časa *post mortem*) može da bude sledećeg kvaliteta: „normalno“ (crveno ružičasto, čvrsto i nevodnjikavo – CČN), BMV (bledo, meko i vodnjikavo) i TČS (tamno, čvrsto i suvo), a poznat je još jedan kvalitet mesa koji nastaje u uslovima intenzivnog hlađenja („cold shortening“). Pomenuti kvaliteti mesa međusobno se razlikuju prema makroskopskim, mikroskopskim i fizičko-hemijskim svojstvima svežeg mesa, kao i prema senzornim i tehnološkim svojstvima konačnih proizvoda u toku i posle kulinarne pripreme, odnosno prerade (Rede i Petrović, 1997). Od 1992. godine u literaturi (Kauffman i sar., 1992; Warner i sar., 1993; Van Laack i sar., 1996; Warner i sar., 1997; Joo i sar., 1999; Toldra i Flores, 2000; Kušec i sar., 2004; Džinić, 2005; Xing i sar., 2007; Qiao i sar., 2007a; Qiao i sar., 2007b; Fischer, 2007) se navode, odnosno opisuju još dva, intermedijarna, kvaliteta svinjskog mesa koji su označeni kao CMV i BČN kvaliteti. CMV (crveno ružičast, mek i vodnjikav) kvalitet svinjskog mesa je prihvatljiv po boji, ali je meso meko i slabe sposobnosti vezivanja vode, dok se BČN (bled, čvrst i nevodnjikav) kvalitet odlikuje bledom bojom, ali dobrom čvrstinom i sposobnošću vezivanja vode.

Osim „normalnog“, ostali kvaliteti se smatraju, manje ili više nepoželjnim, jer pored nekih pozitivnih svojstava koja mogu da budu od značaja samo u nekim tehnološkim operacijama prerade mesa, kod BMV i TČS mesa uglavnom prevladavaju nepoželjna senzorna i tehnološka svojstva (Rahelić, 1984; 1987; Rede i Petrović, 1997; Petrović i sar., 2003).

Kod svinjskog mesa mnogo je veća učestalost pojavljivanja mesa BMV kvaliteta, dok se TČS meso mnogo češće javlja kod govedeg i jagnječeg mesa (Rede i Petrović, 1997; Honikel, 1999; Tomović i sar., 2008; Tomović, 2009).

Podložnost promenama toka postmortalnih procesa u mišićima, a time i promena kvaliteta mišića, odnosno proizvedenog mesa, uslovljena je genetski (endogeni faktori), a aktivirana je i spoljašnjim nadražajima iz okoline u kojoj se životinja nalazi (egzogeni faktori) (Rede i Petrović, 1997; Tomović, 2002; 2009; Tomović i sar., 2004; 2006; 2008; Džinić, 2005; Džinić i sar., 2007). Dakle, kvalitet mesa zavisi od brojnih endogenih (genetskih) i egzogenih (spoljašnjih) faktora (Rosenvold i Andersen, 2003).

Po mišljenju većine autora uticaj egzogenih faktora na kvalitet mesa je značajniji od uticaja endogenih faktora, pri čemu autori (Rede, 1987; Čepin i Čepin, 2001; Džinić, 2005) navode da dominantni uticaj na kvalitet mesa ima ishrana i način držanja životinja. Pored toga, brojnim ispitivanjima (Rahelić, 1984; 1987; Manojlović i Rahelić, 1987; Wiktor, 1987; Petrović i Manojlović, 1999; Rosenvold i Andersen, 2003) utvrđena je mogućnost smanjenja pojavljivanja mesa izmenjenog kvaliteta optimizacijom premortalnih i postmortalnih faktora proizvodnje, odnosno smanjenjem stresa u operacijama pretklanja (smeštaj na farmi, utovar, prevoz, istovar, odmaranje u depou klanice, otpremanje iz depoa) i klanja (omamljivanje, iskrvarenje), zatim operacija na liniji klanja (šurenje, opaljivanje, vađenje unutrašnjih organa), kao i intenziviranjem hlađenja mesa, pri čemu Honikel (1999) posebno ističe da kvalitet proizvodnje utiče na kvalitet mesa, ali da faktori proizvodnje nisu karakteristike kvaliteta mesa.

Tehnološka svojstva mesa, pre svega, imaju značaj za industrijsku proizvodnju i preradu mesa na svim nivoima (Radovanović, 1992; Honikel, 1999). Većina karakteristika izmerenih na polutkama i otkoštenom mesu služi upravo ovoj svrsi (Honikel, 1999), ali i u razvojnim istraživanjima kada se dobijeni podaci koriste za analizu uspešnosti primenjenih postupaka–operacija (Petrović i Manojlović, 1999)

Objektivno predviđanje i/ili utvrđivanje tehnološkog kvaliteta mesa najčešće podrazumeva merenje navedenih faktora kvaliteta: temperature, vrednosti pH, sposobnosti vezivanja vode (gubitak mase ceđenjem) i boje.

Merenje temperature je prema Honikel-u (2002), za svinjsko meso sa sertifikatom u Nemačkoj obavezno. Prilikom smeštaja svinja u klanicu rektalna temperatura mora da bude niža od 39,2°C (kriterijum za dobrobit životinja), odnosno da bi se dobio pečat kontrolisanog kvaliteta svinjskog mesa, pre

hlađenja, odnosno 45 minuta *post mortem*, u dubini buta temperatura mora da bude niža od 40,0°C.

Naši podaci govore da je u brojnim merenjima u našim pogonima industrije mesa registrovana prosečna T_i , po pravilu, znatno viša od 40 °C (Petrović, 2005; 2008) i kreće se od 41,6 °C (Tomović i sar., 2008) do 42,7 °C (Janković, 2008), što govori o izostanku bilo kakvih korektivnih mera u postupku sa životinjama u operacijama pretklanja (dobrobit životinja).

Vremenom je vrednost pH postala nezaobilazan podatak u ocenjivanju kvaliteta mesa pa se određuje, može se reći, pri svakom ispitivanju kvaliteta mesa (Rahelić, 1987). Merenje vrednosti pH je najdirektniji način da se dobiju informacije o svojstvima kvaliteta mesa (Honikel, 1999).

Vrednost pH kao faktor kvaliteta mesa je vrlo značajna, jer, direktno ili indirektno, utiče i na druga svojstva mesa kao što su: sposobnost vezivanja vode, boja, mekoća, ukus, održivost i dr. Vrednost

pH treba meriti u raznim fazama tokom, pre i *post rigor* perioda. Izuzetan značaj pridaje se vrednosti pH utvrđenoj u prvom satu *post mortem* (Hofmann, 1986; Manojlović i Rahelić, 1987; Honikel, 1999).

Posle 24 časa vrednost pH_k ne bi smela da bude niža od 5,4. Izuzetno niske vrednosti pH_k uzrokuju veliki gubitak mase ceđenjem, dok, s druge strane, vrednost pH_k viša od 5,85 skraćuje održivost svinjskog mesa (Rede i Petrović, 1997).

Istraživači, međutim, ne koriste uvek iste granične vrednosti pH_i i pH_k za utvrđivanje kategorija kvaliteta svinjskog mesa (BMV, CMV, CČN, BČN i TČS). Primera graničnih vrednosti pH u literaturi ima mnogo, a samo neki od njih su prikazani u tabeli 2.

Sposobnost vezivanja vode, koja se uglavnom određuje 24 časa *post mortem*, odnosno kada je proizvodnja svinjskog mesa završena, u kombinaciji sa ostalim faktorima kvaliteta (vrednost pH, boja) često se koristi kao faktor kvaliteta mesa (Manojlović i Rahelić, 1987; Honikel, 1999).

Tabela 2. Kriterijumi za vrednost pH prema kojima se svinjsko meso razvrstava u različite kategorije kvaliteta

Table 2. Criteria for pH values according to which pork is classified into various categories

Autori	Kvalitet mesa	pH _i (pH _{1h})	pH _{2h}	pH _k (pH _{24h})
Honikel i Fischer (1977)	BMV	< 5,9		
Kellner i sar. (1979)	BMV	< 5,7		
Manojlović (1982)	BMV TČS	≤ 5,9		≥ 6,3
Kauffman i sar. (1992)	BMV			< 6,0
	CMV			< 6,0
	CČN			< 6,0
	BČN			< 6,0
	TČS			> 6,0
Warner i sar. (1997)	BMV			< 6,0
	CMV			< 6,0
	CČN			< 6,0
	TČS			≥ 6,0
Toldra i Flores (2000)	BMV		< 5,8	
	CMV		< 5,8	
	CČN		> 5,8	
	TČS			> 6,0
Tomović (2002)	BMV	< 5,8		
	CMV	5,8 – 6,0		
	CČN	> 6,0		
Džinić (2005) Petrović (2008)	BMV	< 5,8		< 6,2
	CMV	< 5,8		< 6,2
	CČN	> 5,8		< 6,2
	BČN	> 5,8		< 6,2
	TČS	> 5,8		> 6,2

S obzirom na činjenicu da se za određivanje sposobnosti vezivanja vode koristi više metoda i više načina izražavanja dobijenih rezultata, u tabeli 3.

su prikazani samo neki od kriterijuma za sposobnost vezivanja vode prema kojima se svinjsko meso razvrstava u različite kategorije kvaliteta.

Tabela 3. Kriterijumi za sposobnost vezivanja vode prema kojima se svinjsko meso razvrstava u različite kategorije kvaliteta

Table 3. Criteria for water holding capacity according to which pork is classified into various quality categories

Autori	Kvalitet mesa	“bag” metod (“drip loss”) (%)*	Metoda kompresije	
			% vezane vode	cm ² –površina ovlažena sokom
<i>Honikel i Fischer (1977)</i>	BMV			> 5
<i>Kellner i sar. (1979)</i>	BMV			> 10
<i>Kauffman i sar. (1992)</i>	BMV	> 5		
	CMV	> 5		
	CČN	< 5		
	BČN	< 5		
	TČS	< 5		
<i>Kim i sar. (1996)</i>	BMV	> 7,5		
	CMV	> 7,5		
	CČN	< 7,5		
	TČS	< 5,5		
<i>Warner i sar. (1997)</i>	BMV	> 5		
	CMV	> 5		
	CČN	< 5		
	TČS	< 5		
<i>Joo i sar. (1999)</i>	BMV	> 6		
	CMV	> 6		
	CČN	≤ 6		
	TČS	< 6		
<i>Toldra i Flores (2000)</i>	BMV	> 6		
	CMV	> 6		
	CČN	< 6		
	TČS	< 3		
<i>Tomović (2002)</i>	BMV		< 50	
	CMV		50–60	
	CČN		> 60	
<i>Petrović (2002)</i>	BMV		< 50	>12
	CMV		50–60	12–10
	CČN		60–70	10–5
	TČS		> 70	<5
<i>Džinić (2005)</i>	BMV		< 50	
	CMV		< 50	
	CČN		> 50	
	BČN		> 50	
	TČS		> 50	
<i>Petrović (2008)</i>	BMV		< 50	
	CMV		< 50	
	CČN		50–60	
	BČN		> 60	
	TČS		> 65	

Od mnogobrojnih faktora koji uslovljavaju boju svinjskog mesa najznačajniji je sadržaj pigmentata u momentu smrti životinje. Osnovni nosilac boje je sarkoplazmatski protein – pigment mioglobin (Mb), koji mišić boji crveno, a funkcija mu je reverzibilno vezivanje kiseonika (Rede i Petrović, 1997; Mancini i Hunt, 2005).

Međutim, pored sadržaja mioglobina i ostalih proteina (hemoglobin i citohrom C), na boju mesa utiče i niz drugih pre- (vrsta i rasa životinje, uslovi držanja – ishrana, starost, godišnje doba, operacije pretklanja, vrsta mišića) i postmortalnih faktora (Mancini i Hunt, 2005, Džinić, 2005; Tomović i sar., 2008; Tomović, 2009).

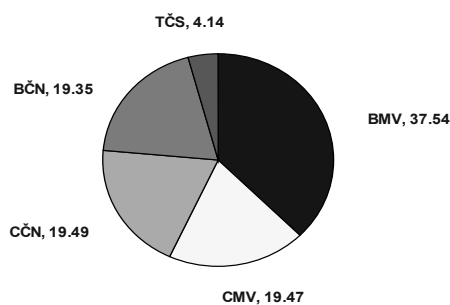
Boja mesa može da se odredi senzorno i instrumentalno. Instrumentalno određivanje boje zasniva se na merenju refleksije svetlosti određenih talasnih dužina sa površine mesa (Manojlović i Rahelić, 1987). Za instrumentalno određivanje boje

danas je najviše u upotrebi uređaj „Chroma Meter” Japanskog proizvođača „Minolta” kojim se, u različitim sistemima (CIEL*a*b* sistem, CIE sistem; CIE, 1976), mogu meriti različite karakteristike boje. U CIEL*a*b* sistemu boja se iskazuje preko: L* (svetloća), a* (udeo crvene i zelene boje) i b* (udeo žute i plave boje) vrednosti, dok se u CIE sistemu boja iskazuje preko: Y (sjajnost, procenat), Č (čistoća, procenat) i λ (dominantna talasna dužina, nm) vrednosti. Svetloća boje (L* vrednost – CIEL*a*b* sistem) se, najčešće, izmerena 24 časa *post mortem*, u kombinaciji sa ostalim faktorima kvaliteta (vrednost pH, sposobnost vezivanja vode), koristi kao pokazatelj kvaliteta mesa (Manojlović i Rahelić, 1987; Honikel, 1999). U tabeli 4. prikazani su neki od kriterijuma za svetloću (L* vrednost) prema kojima se svinjsko meso razvrstava u različite kategorije kvaliteta.

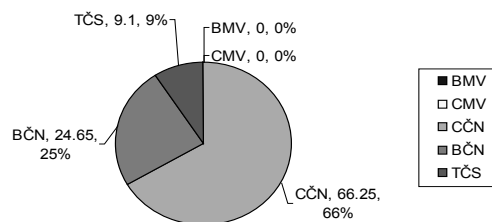
Tabela 4. Kriterijumi za boju prema kojima se svinjsko meso razvrstava u različite kategorije kvaliteta

Table 4. Criteria for water holding capacity according to which pork is classified into various quality categories

Autori	Kvalitet mesa	L* vrednost (boja)
Kauffman i sar. (1992)	BMV	> 50
	CMV	42–50
	CČN	42–50
	BČN	>50
	TČS	< 42
Kim i sar. (1996)	BMV	> 55
	CMV	49–55
	CČN	49–55
	TČS	< 49
Warner i sar. (1997)	BMV	> 50
	CMV	42–50
	CČN	42–50
	TČS	< 42
Joo i sar. (1999)	BMV	> 50
	CMV	≤ 50
	CČN	≤ 50
	TČS	≤ 43
Toldra i Flores (2000)	BMV	> 50
	CMV	44–50
	CČN	44–50
	TČS	<44
Tomović (2002)	BMV	> 55
	CMV	≤ 55
	CČN	< 55
Džinić (2005) Petrović (2008)	BMV	> 50
	CMV	43–50
	CČN	43–50
	BČN	>50
	TČS	< 43



Grafikon 10. Uticaj genotipa (trorasni i četvororasni hibridi) na učestalost pojavljivanja različitog kvaliteta *M. semimembranosus* (n=217)
Figure 10. Influence of genotype (three-breed and four-breed hybrids) on frequency of various quality occurrence *M. semimembranosus* (n=217)



Grafikon 11. Prosečan kvalitet mesa ispitanih tovljenika sa farme „Čenej“ (n=77) kao rezultat brojnih korekcija u uzgoju, ishrani i operacijama predklanja

Figure 11. Average meat quality of examined fattening pigs from the „Čenej“ farm (n=77) as the results of numerous corrections in breeding, nutrition and pre slaughtering

Na Tehnološkom fakultetu u Novom Sadu, već više decenija se na Katedri za tehnologiju mesa izučava kvalitet svinjskog i drugih vrsta mesa i radi na razvoju objektivnih kriterijuma i definisanju parametara za ocenu kvaliteta mesa. Iz predočenih tabela 2, 3. i 4. jasno se uočava da su već više puta naši kriterijumi za predočene parametre za ocenu kvaliteta mesa redefinisani, naravno u skladu sa mnogobrojnim prikupljenim podacima merenja i novim saznanjima iz literature.

Na grafikonima 10 i 11 predočeni su rezultati mnogobrojnih preduzetih mera tokom rada na realizaciji projekta BTN 351008 (Petrović i sar., 2008) na farmi „Čenej“ radi poboljšanja kvaliteta mesa. Izmenama u rasnom sastavu svinja za klanje, odnosno povećanjem udela višelinijjskih hibrida radi povećanja prinosa mesa uočen je značajan pad kvaliteta mesa (grafikon 10). No, korekcijom ishrane i postupaka u operacijama predklanja postignuto je vidno poboljšanje kvaliteta mesa (grafikon 11).

Iako to mnogi potrošači ne priznaju, senzorni faktori kvaliteta su odlučujući u potrošnji mesa (Honikel, 1999).

Honikel (1999), (prema Hofmann, 1986) senzorni kvalitet mesa definiše preko sledećih faktora kvaliteta: čvrstine i mekoće, boje, mramoriranosti, mirisa, ukusa i sočnosti.

Ove faktore kvaliteta je teško izmeriti objektivno, ali čitave armije naučnika pokušavaju da razviju pouzdane i ponovljive senzorne metode (Honikel, 1999).

Gotovo svaki istraživački centar, koji se bavi ispitivanjem kvaliteta svinjskog mesa, razvio je sopstveni deskriptivni sistem za senzorno ocenjivanje svojstava mesa, koje postaje sve značajnije sa povećanjem obima plasmana mikrokonfencioniranog upakovanog mesa.

Za senzorno ocenjivanje čvrstine svežeg svinjskog mesa koriste se analitički deskriptivni testovi (linearne skale) sa različitim brojem nivoa gradacije (uglavnom sa 3 i 5 nivoa gradacije). Zajedno sa čvrstinom, gotovo, uvek se ocenjuje i vlažnost (senzorna ocena sposobnosti vezivanja vode) (Carr i McKeith, 1998).

Boja je kombinacija vizuelno shvaćene informacije sadržane u svetlosti koju reflektuje ili rasipa uzorak (MacDougall, 1982).

Boja svinjskog mesa je svetloružičasta (Briskey i Kauffman, 1971), svetlocrvenoružičasta (Lawrie, 1998), odnosno svetlocrvena (Mancini i Hunt, 2005).

Boja je, verovatno, najznačajnije svojstvo kvaliteta mesa, jer se primećuje i ocenjuje na prvi pogled, te je od interesa da meso bude što prihvatljivije boje, kako bi ga primetili i prihvatili potrošači (Rede i Petrović, 1997).

Tabela 5. Skale za senzorno ocenjivanje čvrstine i vlažnosti svinjskog mesa
Table 5. Scales for sensory for sensoric evaluation of firmness and juceness of pork

Ocena	NPPC standard za čvrstinu i vlažnost (1991)	NPPC standard za čvrstinu i vlažnost (2000)	
		Konzistencija (čvrstina)	Vlažnost
1	Veoma meka i veoma vodnjikava	Meko – površine preseka se lako deformišu i vidljivo su mekane	Vodnjikavo – na površini preseka se prekomerno nakuplja voda
2	Meka i vodnjikava	Čvrsto – površine preseka teže da zadrže oblik	Vlažno – površine preseka se čine vlažnim, sa malo ili bez slobodne vode
3	Neznatno čvrsta i vlažna	Veoma čvrsto – površine preseka teže da budu veoma glatke, bez promene oblika	Suvo – na površini preseka nema slobodne vode
4	Čvrsta i umereno suva		
5	Veoma čvrsta i suva		

Tabela 6. Skale za senzorno ocenjivanje boje svinjskog mesa
Table 6. Scales for sensory evaluation of pork colour

Ocena	NPPC standard za boju (1991)	NPPC standard za boju (2000)	Tehnologija mesa, Tehnološki fakultet Novi Sad
1	Bledoružičasto siva	Bledoružičastosiva do bela	Veoma bleđa
2	Sivoružičasta	Sivoružičasta	Bleđa
3	Crvenoružičasta	Crvenoružičasta	Umereno ružičasta
4	Purpurnocrvena	Tamnocrvenoružičasta	Crvenoružičasta
5	Tamnopurpurnocrvena	Purpurnocrvena	Tamnije crvenoružičasta
6		Tamno purpurnocrvena	Tamno crvena
7			Veoma tamna

Mramoriranost je pojava manjih ili većih nakupina masnog tkiva (intramuskularno masno tkivo) u rastresitom vezivnom tkivu između snopića mišićnih vlakana, a doprinosi poboljšanju jestivog kvaliteta mesa, odnosno doprinosi boljem ukusu i poboljšava mekoću i sočnost mesa (tabela 7). Mast daje mesu specifičan poželjan ukus. Pošto se masne ćelije razvijaju između slojeva vezivnog tkiva, one ga razlabavljaju, što rezultira u boljoj mekoći mesa. Prisustvo masti u mesu pojačava salivaciju pri žvakanju, pa se stiče utisak veće sočnosti (*Eikelenboom i sar.*, 1996; *Rede i Petrović*, 1997; *Jeremiah i Miller*, 1998; *Jeleníková i sar.*, 2008).

Za senzorno ocenjivanje mekoće i sočnosti koriste se analitički deskriptivni testovi (linearne skale) sa različitim brojem nivoa gradacije (uglavnom sa 8, odnosno 9 nivoa gradacije).

S obzirom da je nacrtom Pravilnika o kvalitetu zaklanih svinja i kategorizaciji svinjskog mesa (2009) definisano da se pod senzornim ispitivanjem kvaliteta svinjskog mesa i jestivih delova zaklanih svinja podrazumeva utvrđivanje faktora kvaliteta koji se ispituju čulom vida, čulom mirisa, čulom ukusa i prstima (palpacijom) u toku proizvodnje ili u sklopu utvrđivanja usaglašenosti kvaliteta originalno upakovanog mesa sa ovim i drugim propisima,

Tabela 7. Skale za senzorno ocenjivanje mramoriranosti svinjskog mesa
Table 7. Scales for sensory evaluation of marbledness pork

Ocena	NPPC standard za mramoriranost (1991)	NPPC standard za mramoriranost (1999)	NPPC standard za mramoriranost (2000)
1	Bez mramoriranosti do praktično bez mramoriranosti	Bez mramoriranosti	Bez mramoriranosti
2	Tragovi do neznatna	Praktično bez mramoriranosti	Tragovi
3	Mala do skromna	Tragovi	Neznatna
4	Umerena do neznatno obilna	Neznatna	Mala
5	Umereno obilna do velika	Mala	Skromna
6		Skromna	Umerena
7		Umerena	Obilna
8		Neznatno obilna	
9		Umereno obilna	
10		Velika	

Senzornom ocenom sočnosti kuvanog mesa manifestuju se dva senzorna doživljaja. Prvi je utisak vlažnosti tokom žvakanja i rezultat je brzog otpuštanja tečnosti iz mesa, dok je drugi zadržana sočnost, uglavnom zbog stimulatornog efekta masti na salivaciju (*Weir*, 1960).

definisani su i parametri koji se ocenjuju i data je skala za ocenjivanje.

Ovim Pravilnikom senzorna ocena opšteg izgleda obuhvata ocenu stanja ambalaže i načina obrade, sečenja i oblikovanja upakovanog komada u skladu sa propisanim zahtevima ovog Pravilnika za taj anatomski deo.

Tabela 8. Skale za senzorno ocenjivanje nežnosti (mekoće) i sočnosti svinjskog mesa
Table 8. Scales for sensoric evaluation of tenderness (softnes) and juiceness of pork

Ocena	AMSA standard za mekoću i sočnost (1995)		Tehnologija mesa, Tehnološki fakultet Novi Sad	
	Mekoća	Sočnost	Mekoća	Sočnost
1	Ekstremno grubo	Ekstremno suvo	Ekstremno grubo	Ekstremno suvo
2	Veoma grubo	Veoma suvo	Veoma grubo	Veoma suvo
3	Umereno grubo	Umereno suvo	Grubo	Suvo
4	Neznatno grubo	Neznatno suvo	Umereno grubo	Umereno suvo
5	Neznatno meko	Neznatno sočno	Nedovoljno meko	Nedovoljno sočno
6	Umereno meko	Umereno sočno	Umereno meko	Umereno sočno
7	Veoma meko	Veoma sočno	Meko	Sočno
8	Ekstremno meko	Ekstremno sočno	Veoma meko	Veoma sočno
9			Ekstremno meko	Ekstremno sočno

Vlažnost komada mesa može da bude: veoma vodnjikava, vodnjikava, umereno vodnjikava, neznatno vlažna, umereno vlažna, neznatno suva, umereno suva i veoma suva.

Boja komada mesa može biti: veoma bleđa, bleđa, umereno ružičasta, crvenoružičasta, tamnije crvenoružičasta, tamnocrvena i veoma tamnocrvena.

Konzistencija mesa može da bude: veoma meka, odnosno površina preseka se može lako defor-

misati, meka, neznatno čvrsta, umereno čvrsta, čvrsta, odnosno površina preseka teži da zadrži oblik, veoma čvrsta, odnosno površine preseka teže da budu veoma glatke, bez promene oblika.

Na osnovu datih ocena senzornog kvaliteta meso može da se okarakterise kao normalno, BMV i TČS, pri čemu originalno upakovano meso u prometu ne sme da bude BMV i TČS svojstava.

Literatura

- AMSA, 1995. Research guidelines for cookery, sensory evaluation and instrumental tenderness measurements of fresh meat. American meat science association, National live stock and meat board, pp. 1–47, Chicago, Illinois, USA;
- Barton-Gade Patricia, 1985. Karakteristike kvaliteta mesa i njihov značaj za proizvode od svinjskog mesa. Tehnologija mesa, XXVI, 9, 250–253;
- Briskey E. J., Kauffman R. G., 1971. Quality characteristics of muscle as a food – The Science of Meat and Meat Products. W. H. Freeman and Company, San Francisco, USA;
- Čepin S., Čepin M., 2001. Uticaj genetike i sredine na kvalitet junećeg trupa i mesa. Tehnologija mesa, 42, 5–6, 283–294;
- Carr S. N., McKeith F. K., 1998. Impact of paylean™ on pork quality. Facts, National Pork Board, pp. 1–4, Des Moines, Iowa, USA;
- Causeur D., Daumas G., Dhorne T., Engel B., Font L., Furnols M., Højsgaard S., 2006. Statistical handbook for assessing pig classification methods: Recommendation from the “EUIPIGCLASS” project group;
- CIE, 1976. International Commission on Illumination, Colorimetry: Official Recommendation of the International Commission on Illumination. Publication CIE No. (E-1.31) Bureau Central de la CIE, Paris, France;
- Commission Regulation (EEC) No 2967/85 of 24 October 1985 laying down detailed rules for the application of the Community scale for grading pig carcasses (1985). Official Journal of the European Communities No L 285, 25/10/1985, 39–40;
- Council Regulation (EC) No 3513/93 of 14 December 1993 amending Regulation (EEC) No 3220/84 determining the Community scale for grading pig carcass (1993). Official Journal of the European Communities No L 320, 22/12/1993, 5–6;
- Commission Regulation (EC) No 3127/94 of 20 December 1994 amending Regulation (EC) No 2967/85 laying down detailed rules for the application of the Community scale for grading pig carcasses (1994). Official Journal of the European Communities No L 330, 21/12/1994, 43–44;
- Daumas D., 2003. A description of the European slaughtering populations and their classification. EUIPIGCLASS report, 42 p;
- Džinić Natalija, Petrović Ljiljana, Manojlović Danica, Tomović V., Timanović S., Trišić-Ilić Svetlana, Kurjakov Nada, 2001. Carcass and pork quality of purebred and four-race hybrids. Proc. 47th ICoMST “Future of Meat”, Krakow, Poland, Vol. I, 2-P19, 146–147.;
- Džinić Natalija, Petrović Ljiljana, Tomović V., Manojlović Danica, Timanović S., Trišić-Ilić Svetlana, Kurjakov Nada, 2004. Influence of seasons on incidence of different *M. semimembranosus* quality of pig halves of three-race hybrids. Proc. 50th ICoMST “1st ... 50th”, Helsinki, Finland, Vol. I, 146–149;
- Džinić Natalija, 2005. Uticaj endogenih i egzogenih faktora na kvalitet svinjskog mesa. Doktorska disertacija, Tehnološki fakultet, Univerzitet u Banjoj Luci, Banja Luka;
- Džinić Natalija, Petrović Ljiljana, Tomović V., Manojlović Danica, Timanović S., Vidarić Dragica, 2006a. Uticaj dužine odmaranja u depou klanice na kvalitet *M. semimembranosus* sa polutki svinja dvorasnih hibrida. Tehnologija mesa, 47, 1–2, 20–26;
- Džinić Natalija, Petrović Ljiljana, Tomović V., Manojlović Danica, Timanović S., Vidarić Dragica, 2006b. Kvalitet polutki i *M. semimembranosus* dvorasnih i četvororasnih hibrida svinja. Tehnologija mesa, 47, 5–6, 175–182.
- Džinić Natalija, Petrović Ljiljana, Tomović V., Manojlović Danica, Timanović S., Vidarić Danica, Kurjakov N., 2007. Quality of halves and pork of F1 descendants of tested Large Yorkshire race boars. Proceeding of the I International Congress, Food Technology, Quality and Safety, XI Symposium NODA, Novi Sad, Serbia, 13–15;
- Eikelenboom G., Hoving-Bolink A. H., van der Wal P. G., 1996. The eating quality of pork. 2. The influence of intramuscular fat. Fleischwirtschaft, 76, 4, 517–518;
- Fischer K., 2007. Drip loss in pork: influencing factors and relation to further meat quality traits. Journal of Animal Breeding and Genetics, 124, 1, 12–18;
- Hansson I., 2003. Pork production and classification of pig carcass in European countries. EUIPIGCLASS GROWTH Project GRD-1999-10914. Anex 9;
- Hofmann K., 1986. Ist Fleischqualität messbare – Chemisch-physikalische Merkmale der Fleischqualität. Kulmbacher Reihe, Band 6, ss. 1–17, Kulmbach, Germany;
- Honikel K. O., Fischer C., 1977. A rapid method for detection of PSE and DFD porcine muscle. Journal of Food Science, 42, 6, 1633–1636;
- Honikel K. O., 1999. Biohemijske i fizičko-hemijske karakteristike kvaliteta mesa. Tehnologija mesa, 40, 3 – 5, 105–123;
- Honikel K. O., 2002. Nova dostignuća i sistemi za proizvodnju mesa visokog kvaliteta. Tehnologija mesa, 43, 3–6, 146–156;
- ISO EN 45 004: General Criteria for the Performance of Various Types of Inspecting Bodies.
- Janković Sanela, 2008. Uticaj dodatka adsorbenata mikotoksina u ishrani svinja na tehnološki kvalitet proizvedenog mesa. Diplomski rad, Tehnološki fakultet, Novi Sad;
- Jeleníková J, Pipek P., Miyahara M., 2008. The effects of breed, sex, intramuscular fat and ultimate pH on pork tenderness. European Food Research and Technology, 227, 4, 989–994;
- Jeremiah L. E., Miller R., 1998. Marbling and Pork Tenderness. Facts, National Pork Board, pp. 1–4, Des Moines, Iowa, USA;

- Joksimović J., 1997.** Osnovi kontrole i upravljanja kvalitetom u proizvodnji hrane. Privredni pregled, Beograd;
- Joo S. T., Kauffman R. G., Kim B. C., Park G. B., 1999.** The relationship of sarcoplasmic and myofibrillar protein solubility to colour and water-holding capacity in porcine *longissimus* muscle. *Meat Science*, 52, 3, 291–297;
- Kauffman R. G., Cassens R. G., Scherer A., Meeker D. L., 1992.** Variation in pork quality. National Pork Producers Council Publication, Des Moines, IA, USA;
- Kellner A. I., Sandor I., Takacs J. 1979.** Occurrence of exudative (PSE) meat alteration on some inland swine races., In: Proceedings 25th European Meeting of Meat Research Workers, pp. 115 – 118, Budapest, Hungary;
- Kim C. J., Lee E. S., Joo S. T., Kim B. C., Kang J. O., Kauffman R. G., Yoo I. J., Ko W. S., Choi D. Y., 1996.** Chemical, physical and structural characteristics of pork loins from four quality groups. In: Proceedings 42nd International Congress of Meat Science and Technology, pp. 312 – 313, Lillehammer, Norway;
- Kušec G., Kralik G., Petričević A., Margeta V., Gajčević Z., Gutzmirtl D., Pešo M., 2004.** Differences in slaughtering characteristics between crossbred pigs with Pietrain and Duroc as terminal sire. In: Proceedings 12th International Symposium "Animal Science Days", pp. 121–127, Bled, Slovenia;
- Lawrie R. A., 1998.** *Lawries Meat Science*, Woodhead Publishing Limited, Cambridge, England;
- MacDougall D. B., 1982.** Changes in the colour and capacity of meat. *Food Chemistry*, 9, 1–2, 75–88;
- Mancini R. A., Hunt M. C., 2005.** Current research in meat color. *Meat Science*, 71, 1, 100–121;
- Manojlović Danica, 1982.** Učestalost pojavljivanja bleđih, mekih i vodnjikavih, kao i tamnih, čvrstih i suvih mišića svinja zaklanih u SAP Vojvodini i značaj tih pojava. Magistarski rad, Tehnološki fakultet, Univerzitet u Novom Sadu, Novi Sad;
- Manojlović Danica, Rahelić S., 1987.** Tehnološki i ekonomski značaj kvaliteta svinjskog mesa u proizvodnji i preradi – Tehnologija proizvodnje i kvalitet svinjskog mesa. U: Novosadski dani industrije mesa – NODA '87, Zbornik radova, ss. 1–24, Tehnološki fakultet, Univerzitet u Novom Sadu, Novi Sad;
- Manojlović Danica, Petrović Ljiljana, Džinić Natalija, Kurjakov Nada, 1999.** Kvalitet trupa i mesa – Osnov kvaliteta proizvoda. Monografija: "Tehnologija proizvodnje i kvalitet konzervi od mesa u komadima", urednik Ljiljana Petrović, izdavač Tehnološki fakultet, Novi Sad, str. 66-90, katalogizacija u publikaciji Biblioteke Matice Srpske, 637,5 (082);
- Murray A., 1997.** *Pork Quality. A Researcher's Perspective*. Agriculture and Agriculture and Agri-Food, Lacombe, Alberta, Canada. Available: www.ccsi.ca/Meetings/ACM_Pork_Quality;
- Nacrt Pravilnika o kvalitetu zaklanih svinja i kategorizaciji svinjskog mesa, 2009;**
- Nikolić M., Brundza V., Petrović D., Petričević A., 1989.** Primena pravilnika o kvalitetu (mesnatosti) svinja na liniji klanja u nas i osvrt na novi EUOP standard EEZ-a. Zbornik IX jugoslovenskog savjetovanja o kvalitetu i standardizaciji mesa stoke za klanj, peradi, divljači i riba, Donji Milanovac, 147–155;
- NPPC (National Pork Producers Council), 1991.** Procedures to evaluate market hogs, 3rd edition. National Pork Producers Council, Des Moines, Iowa, USA;
- NPPC (National Pork Producers Council), 1999.** *Pork Quality Standards*. National Pork Producers Council. Des Moines, Iowa, USA;
- NPPC (National Pork Producers Council), 2000.** *Pork composition and quality assessment procedures*. E. Berg (Ed.), pp. 1 – 38, National Pork Producers Council, Des Moines, Iowa, USA.
- Okanović Đ., Zekić V., Petrović Ljiljana, Tomović V., Džinić Natalija, 2006.** Ekonomičnost proizvodnje svinjskog mesa u polutkama, *Tehnologija mesa*, 47, 5–6, 237–241;
- Petrović Ljiljana, Manojlović Danica, Džinić Natalija, Latkovska Elena, Velemir Jovanka, Adamović Jasminka, 1996.** Evaluation of carcass and meat quality on the slaughterline of pigs with FOM device. Proc 42nd International Congress of Meat science and Technology, Lillehammer, Norway, G-7, 246–247;
- Petrović Ljiljana, 1999.** Razvoj sistema za ocenu kvaliteta trupova na liniji klanja svinja prema zahtevima standarda serije ISO i JUS 9000(EN29000) i EN 45000, Strateški projekat S. 4. 28. 50. 0037, finansiran od Republičkog ministarstva za nauku i tehnologiju, Nosilac zadatka: Ljiljana Petrović, Tehnološki fakultet, Univerzitet u Novom Sadu, Novi Sad;
- Petrović Ljiljana, Manojlović Danica, 1999.** Ocena kvaliteta trupova i mesa na liniji klanja svinja. *Tehnologija mesa*, 40, 3-5, 145–158;
- Petrović Ljiljana, 2002.** Fazni izveštaj o radu na Projektu: Proizvodnja svinjske šunke u konzervi (BTN.5.2.1.7101. B). Rukovodilac projekta: Ljiljana Petrović, Tehnološki fakultet, Univerzitet u Novom Sadu, Novi Sad.
- Petrović Ljiljana, Tomović V., Džinić Natalija, Manojlović Danica, 2003.** Proizvodnja svinjskog mesa sa sertifikatom u Srbiji – stanje i perspektive. Glasnik hemičara i tehnologa Republike Srpske, Banja Luka, Republika Srpska, 44, 39–55;
- Petrović Ljiljana, 2005.** Završni izveštaj o radu na Projektu: „Proizvodnja svinjske šunke u konzervi“ (BNT.5.2.1.7101.B). Razvijeno je 9 novih linija hibrida svinja i to: VJxŠL, ŠLxVJ, (ŠLxVJ)xVJ, (VJxŠL)xD, (ŠLxVJ)xD, (ŠLxVJ)xH, (ŠLxVJ)x(PxD), (VJxŠL)x(PxH), (ŠLxVJ)x(PxH). Rukovodilac projekta: Ljiljana Petrović, Tehnološki fakultet, Univerzitet u Novom Sadu, Novi Sad.
- Petrović Ljiljana, Džinić Natalija, Tomović V., Timanović S., Ikonić P., Tasić Tatjana, 2006.** Quality of halves and meat of pigs obtained in different models of crossbreeding with large yorkshire, Proc. 52nd ICOMST "Harnessing and Exploiting Global Opportunities", 13-18 August, Dublin, Ireland, 425–426;
- Petrović Ljiljana, 2008.** Završni izveštaj o radu na Projektu: „Proizvodnja i priprema svinjskog mesa za maloprodaju, veleprodaju, industriju gotove hrane i preradu“ (BTN.351008). Rukovodilac projekta: Lj. Petrović, Tehnološki fakultet, Univerzitet u Novom Sadu, Novi Sad.
- Pravilnik o kvalitetu zaklanih svinja i kategorizaciji svinjskog mesa.** Sl. list SFRJ br. 2 i 12, 1985;
- Qiao J., Ngadi M. O., Wang N., Gariépy C., Prasher S. O., 2007a.** Pork quality and marbling level assessment using a hyperspectral imaging system. *Journal of Food Engineering*, 83, 1, 10–16;
- Qiao J., Wang N., Ngadi M. O., Gunenc A., Monroy M., Gariépy C., Prasher S. O., 2007b.** Prediction of drip-loss, pH, and color for pork using a hyperspectral imaging technique. *Meat Science*, 76, 1, 1–8;
- Radovanović R., 1992.** Ocena kvaliteta trupova na liniji klanja – Savremeni zahtevi, mogućnosti i perspektive. *Tehnologija mesa*, XXXIII, 5, 169–178;
- Radovanović R., 2001.** Utvrđivanje kvaliteta trupova na liniji klanja: mogućnosti merne opreme nove generacije. *Tehnologija mesa*, 42, 5–6, 309–326;
- Rahelić S., 1984.** *Uzgoj svinje i meso*. Školska knjiga, Zagreb;
- Rahelić S., 1987.** Kvalitet mesa plemenite svinje. Tehnološki fakultet, Univerzitet u Novom Sadu, Novi Sad;

- Rede R., 1987.** Postupci klanja svinja i obrada trupova i njihov uticaj na kvalitet mesa – Tehnologija proizvodnje i kvalitet svinjskog mesa. U: Novosadski dani industrije mesa – NODA '87, Zbornik radova, ss. 69–77, Tehnološki fakultet, Univerzitet u Novom Sadu, Novi Sad;
- Rede R. R., Petrović Ljiljana, 1997.** Tehnologija mesa i nauka o mesu. Tehnološki fakultet, Novi Sad;
- Rosenvold K., Andersen H. J., 2003.** Factors of significant for pork quality – a review. *Meat Science*, 64, 3, 219–237;
- Srećković A., Nikolić A., 1985.** Stanje i perspektive proizvodnje svinja i svinjskog mesa u SFRJ do 1999. godine. Kvalitet mesa i standardizacija, Zbornik VIII jugoslovenskog savjetovanja o problemima kvalitete mesa i standardizacije, Osijek, 49 – 65.
- Toldrá F., Flores M., 2000.** The use of muscle enzymes as predictors of pork meat quality. *Food Chemistry*, 69, 4, 387–395;
- Tomović V., 2002.** Uticaj selekcije i višerasnog ukrštanja svinja na kvalitet polutke i tehnološki, nutritivni i senzorni kvalitet mesa, Magistarski rad, Tehnološki fakultet, Univerzitet u Novom Sadu;
- Tomović V., Petrović Ljiljana, Džinić Natalija, Manojlović Danica, Timanović S., Vidarić Dragica, 2004.** Effect of lairage time on incidence of different quality of *M. semimembranosus* from pig halves of multi-race hybrids. Proc. 50th ICoMST ‘‘1st ... 50th’’, Helsinki, Finland, Vol. I, 285–288;
- Tomović V., Petrović Ljiljana, Džinić Natalija, Tasić Tadjana, Ikonić P., 2006.** The effect of accelerated chilling of carcasses on pork semimembranosus muscle colour, Proc. 52nd ICoMST ‘‘Harnessing and Exploiting Global Opportunities’’, 13-18 August, Dublin, Ireland, 597–598;
- Tomović V., Petrović Ljiljana, Džinić Natalija, 2008.** Effects of rapid chilling of carcasses and time of deboning on weight loss and technological quality of pork semimembranosus muscle. *Meat Science*, Vol. 80, 4, 1188–1193;
- Tomović, V., 2009.** Uticaj brzine hlađenja polutke, vremena otkoštavanja *post mortem* i postupka salamurenja na kvalitet i bezbednost kuvane šunke. Doktorska disertacija, Tehnološki fakultet, Univerzitet u Novom Sadu;
- Van Laack R. L. J. M., Solomon M. B., Warner R. D., Kauffman R. G., 1996.** A comparison of procedures for measurement of pigment concentration in pork. *Journal of Muscle Foods*, 7, 2, 149–163;
- Vidović S. V., 1999.** Selekcija i namenski uzgoj svinja. Monografija: ‘‘Tehnologija proizvodnje i kvalitet konzervi od mesa u komadima’’, urednik Ljiljana Petrović, izdavač Tehnološki fakultet, Novi Sad, Univerzitet u Novom Sadu, Novi Sad, 31–65, katalogizacija u publikaciji Biblioteke Matice Srpske, 637,5 (082);
- Walstra P., Merkus G. S. M., 1996.** Procedure for assessment of the lean meat percentage as a consequence of the new EU reference dissection method in rig carcass classification. DLO – Research Institute for Animal Science and Health (ID – DLO), Research Branch, Zeist, The Netherlands;
- Warner R. D., Kauffman R. G., Russell R. L., 1993.** Quality attributes of major porcine muscles: A comparison with *longissimus lumborum*. *Meat Science*, 33, 3, 359–372;
- Warner R. D., Kauffman R. G., Greaser M. L., 1997.** Muscle protein changes *post mortem* in relation to pork quality traits. *Meat Science*, 45, 3, 339–352;
- Weir C. E., 1960.** The Science of Meat and Meat Products. American Meat Institute Foundation (Eds.), pp. 212–221, Reinhold Publishing Company, New York, USA;
- Wiktor J., 1987.** Premortalni faktori koji utiču na pojavu BMV mesa – Tehnologija proizvodnje i kvalitet svinjskog mesa. U: Novosadski dani industrije mesa – NODA '87, Zbornik radova, ss. 52–58, Tehnološki fakultet, Univerzitet u Novom Sadu, Novi Sad.
- Xing J., Ngadi M., Gunenc A., Prasher S., Garipey C., 2007.** Use of visible spectroscopy for quality classification of intact pork meat. *Journal of Food Engineering*, 82, 2, 135–141;
- Živković Ž., 1985.** Uticaj prinosa na unapređenje kvaliteta i povećanje proizvodnje svinjskog mesa u našoj zemlji. Kvalitet mesa i standardizacija, Zbornik VIII jugoslovenskog savjetovanja o problemima kvalitete mesa i standardizacije, Osijek, 67–71.

Rad primljen: 14.04.2009.

ZAHVALNOST

Ovaj rad je nastao kao rezultat rada na projektima 20037TR i BTN 351008 koji su finansirani sredstvima MNTR RS.

SUSTAINABILITY OF FOOD PRODUCTION CHAIN*

Okanovic Dj., Mastilovic Jasna, Ristic M.

Abstract: Based on the insight of into the comprehensive actual and current investigations in the area of food production chain sustainability in Europe and in the world, and comparative insight of situation in Serbia, this study presents structure of investigations which have to be realized in order to enable the creation of prerequisites for technological development of food production, as a significant and important branch of Serbian economy, applying sustainable principles from economic, social, and ecological points of view.

Key words: food production chain, sustainability, objectives

Održivost lanca proizvodnje hrane*

Sadržaj: Na osnovu sagledavanja sveobuhvatnosti aktuelnih istraživanja u oblasti održivosti lanca proizvodnje hrane u Evropi i svetu i poredeći situaciju u Srbiji, ovaj rad predstavlja istraživanja koja će biti realizovana da se omogući stvaranje boljih preduslova za tehnološki razvoj proizvodnje hrane, važne i prosperitetne oblasti srpske ekonomije, primenjujući održive zahteve sa ekonomske, društvene i ekološke tačke gledišta.

Ključne reči: lanac proizvodnje hrane, održivost

Introduction

The fundamental task of agriculture is the production of adequate quantities of high quality foods and raw materials of organic origin for the existing world population and its increase of about 93 million people per year (Kennedy, 1993). Ever growing demands for food production impose the needs for more efficient managing of economic resources that such production follows. Management of agricultural resources is crucial for the survival of mankind, i.e. for the economic, cultural and social development of the society.

In Serbia, as a country with exceptional natural resources for agricultural production, production of food is one of the supports of technological development. Structure of natural resources, and also market capacities generated from demands of particular categories of products, caused locations of nearly 90% of food production in the structure of chain of production and processing in segments cha-

racterized with mass production and consumption. In the light of environmental conditions and consumer habits in region of Balkans, the food production chain that could be considered as mass production, can be divided into:

- basic field crops and basic products of their processing (wheat, corn, sunflower, soy),
- mass-produced animal species and products of their processing (pigs, cattle, poultry).

High participation of the mentioned products in the gross production of agro-industrial sector, as well as high degree of exploitation of natural resources through the realization of the mass production of food, sets as imperative serious approach to the realization of all necessary activities in the shortest possible time. On the level of mass production of food, this should follow steps of sustainable technological development that are going to solve the existing problems, introduce necessary developmental solutions and provide conditions for strategic approach to projecting and managing this enormous

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa održanom od 15-17. juna 2009. na Tari

AUTHORS: Djordje Okanovic, djordje.okanovic@fins.uns.ac.rs, Jasna Mastilovic, Milutin Ristic, Institut for Food Technology in Novi Sad, Bulevar cara Lazara 1, 21000 Novi Sad

AUTORI: Djordje Okanovic, djordje.okanovic@fins.uns.ac.rs, Jasna Mastilović, Milutin Ristić, Institut za prehrambene tehnologije u Novom Sadu, Bulevar cara Lazara 1, 21000 Novi Sad.

segment of the agro-industrial which is, from the aspects of characteristics of natural resources and market demands, irreplaceable and obligatory. (Meyer, 2007).

Large volume of production, which is concentrated in the chain of mass production of food is, at the same time, a source of significant losses characterised by weak links in chains of production, processing and distribution. However, there are potential points of significant improvement and contribution to the national economy, environmental protection and competitiveness of this group of goods at the world market (Rowe et. al., 2007).

Investigations and general practice in Serbia and in the world

Trends of investigations concerned with technological development of food production in Europe and world-wide evolved during the past several decades: beginning with investigations predominantly oriented to solutions aimed at production of adequate quantities of quality food that characterized first half of the last century; followed by studies oriented to improvements of food quality and safety during the last decades of 20th century, to the shift of focus on sustainability of food production chain as a whole at the beginning of the new millennium (Risku and Maenpaa, 2007).

The word „sustainability“ in its broadest sense, which is used in the present study, means „the production which ensures that demands of the inhabitants and the market set towards natural environment are achievable, without diminishing the capacities of the environment to satisfy the needs of future generations“, where sustainability of each system and also of system of the food production chain demands equal consideration of economic, ecological and social aspects of sustainability.

The structure of European technological platform „Food for life“ and priorities of investigations and the technological development that are there defined demonstrates the evident shift of focus of European investigations, as well as of European and even world processes of technological development from domains of development of new and improvement of the existing singular technologies to the trend of recognition of food production chain as the whole, from points of view of efficient management with tendencies in all segments and especially the trend of the all-including investigation and defining of all aspects of sustainability of food production chain and mutual interconnection and dependence of all of its segments. European technology platform FOOD FOR LIFE (<http://etp.ciaa.be>) is developed by teams

of the most eminent European experts and it makes the basis for determining the structure of future activities in R&D projects, as well as in orientation of trends of technological developments in production of foods in general. Sustainable production of food, which is in the European technological platform stated as one of its principal aims, defines diversification of focuses of investigations on:

- development of sustainability of food production and distribution chains in Europe;
- elaboration of scenarios of future sustainable food production and provision systems;
- development of systems of sustainable production, preservation, packaging and distribution;
- ensuring sustainable primary production of food in Europe;
- development of consumers' understanding and their relations with sustainability in the food production.

On the basis of European technology platform FOOD FOR LIFE, in almost all European countries teams of the most eminent experts have defined, on multidisciplinary foundations, national technological platforms, where aspect of sustainability of food production chain was positioned in light of the existing problems and developmental potentials of each country.

In Serbia, there is awareness of the necessities of establishing technological development on the sustainable basis at the highest level, put together in the Strategy of Sustainable Development of the Republic of Serbia (<http://www.odrzivi-razvoj.sr.gov.yu>), which went through the public discussion phase and which is expected to be adopted soon. The text of this document does not apply directly on sustainability of chain of mass production of food, but deals with respect on the explained imperatives connected with natural resources, the place of mass production of food in national economy and its significance for each individual as consumer. High quality, applicable and all-including investigations in this domain will be necessary for the implementation of this strategy.

Protection and improvement of environment, as well as rational use of natural resources, appear as one of priorities of this strategy, what is, to a great degree, directly linked with the mentioned problems concerning mass food production chain. Insisting on protection and improvement of system of environmental protection, decreasing pollution and pressures on the environment, use of natural resources in a way which will assure their availability for future generations indicate that establishing a system of protection and sustainable use of natural resources, including soil as starting resource of mass

production of food, will have unequivocal priority, but also the unequivocal necessity for intensive research activities.

Considering agriculture and production of foods, general objective of sustainable development is the creation of economically feasible and ecologically acceptable production, which is capable of entering the European market, including introduction of organic agriculture. Among many important priorities, Strategy of Sustainable Development of Serbia insists on the following priorities:

- investigations of potentials of renewable energy resources, with aims of their verification and more real balancing;
- defining the optimal approach to construction and/or reconstruction of the industrial infrastructure oriented to environmental protection;
- introducing „cleaner“ production and improvements of energy and raw materials efficiency, with simultaneous decrease of quantities of wastes.

The degree of mutual effects of individual links in chain of mass production of food and their mutual interactions with market and with the environment is shown in Figure 1.

Chain of the mass production of food

Chain of mass production of food, perceived in the manner and with segments which have to be recognized, analyzed and synthesized as integral sustainable system is shown in Figure 2. On the basis of experiences of multidisciplinary and highly specialized team of researchers, conception of project named “Sustainability of chain of mass production of food” was realized, is on going and was accepted for financing by the Ministry of Science and Technological Development for the period 2008–2010. Within this project critical points are addressed, which represent objects of investigating activities that have to assure optimal effects on technological development, with respects of individual improvements, but also with respect of improvement of sustainability of chain of mass production of food as the whole.

The very first link in the chain of mass production of food is primary agricultural production. Optimization of conjunction between primary agricultural production and processing of primary agricultural products represents first focus point for realization of significant improvements of sustainability of mass production of food.

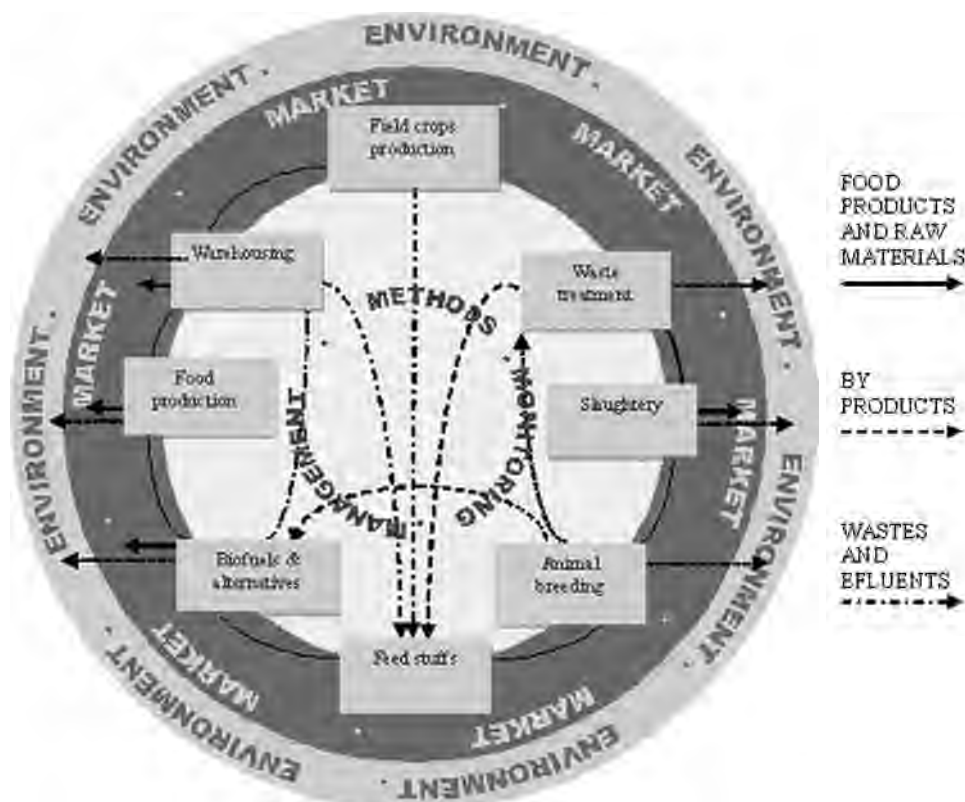


Figure 1. Large scale food production chain in the environment with flows of raw materials, by-products, products and wastes

Slika 1. Lanac masovne proizvodnje hrane u životnoj sredini sa šematskim prikazom tokova sirovina, sporednih i finalnih proizvoda

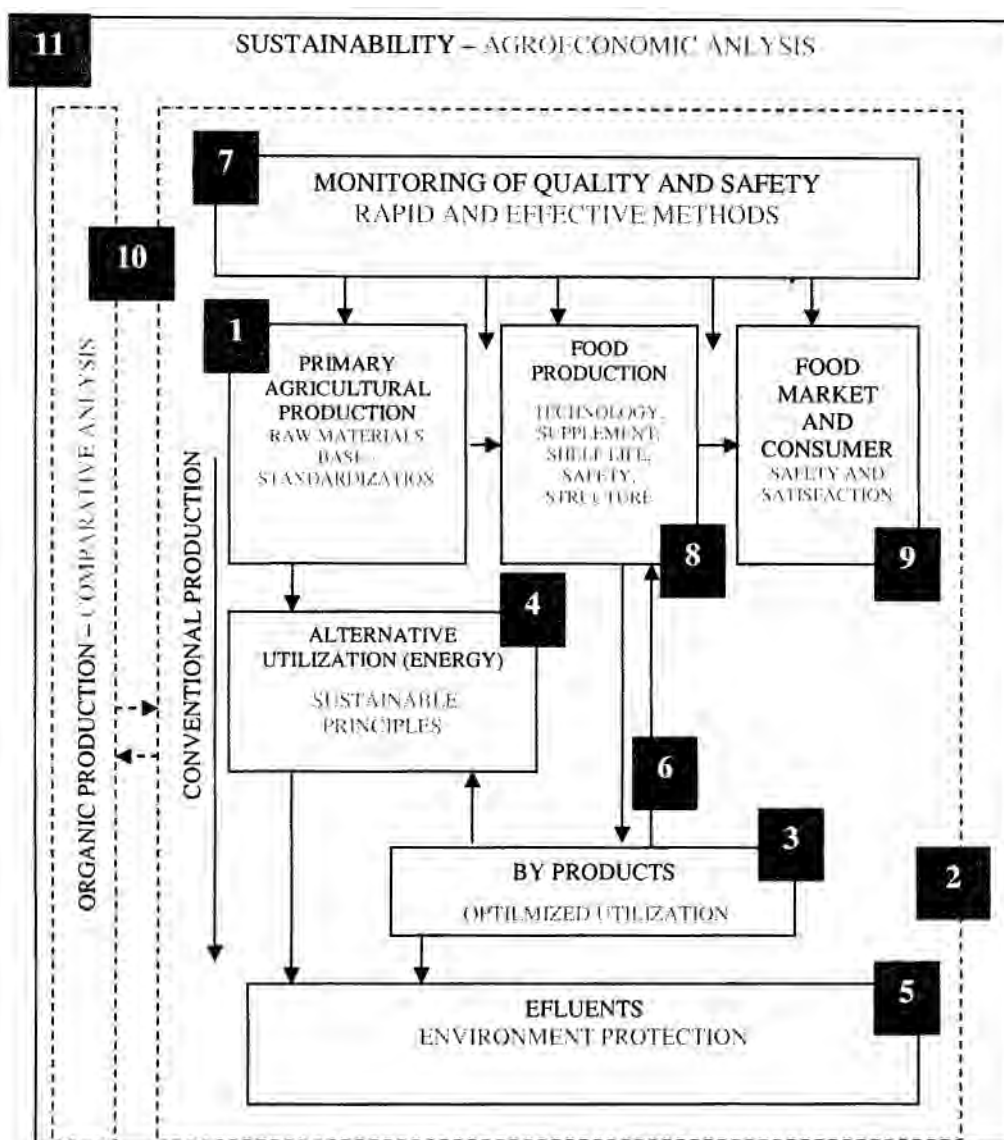


Figure 2. Large scale food production chain with focused critical points
Slika 2. Lanac masovne proizvodnje hrane sa fokusiranim kritičnim tačkama

Chain of mass production of food includes great losses, but also the huge possibilities for potential improvements in optimization of the use of by products that appear in primary agricultural production, as well as in plants for mass processing of foods (Green and Foster, 2005). Two directions of investigations are oriented to improvements of safety, and nutritive properties of animal feeds on one, and improvement of assortment and quality of products and by-products of slaughterhouse industry from the other side.

Sustainable management with the use of by-products, and especially of primary agricultural products for alternative purposes, meaning predominantly the production of biofuels which represents backbone of numerous investigations in the world, in this project are covered by investigational task

AGRICULTURAL PRODUCTS AND BYPRODUCTS FROM PRODUCTION AND PROCESSING OF FOOD AS RAW MATERIAL IN ENERGY AND OTHER ALTERNATIVE FORMS OF PROCESSING, where they will be evaluated.

Important aspects of sustainability of mass production of food chain, which must be recognized through integral research of the effects of chain of mass production of food on the environment (Henningsson, 2004), effluents in the chain of mass production of food will be realized, through investigations of quantities and compositions of the most significant effluents, analyze of potential risks for the environment and development of sustainable solutions of registered problems on their macro level.

Sustainability of system of management, safety, quality and environmental protection processes

belonging to the chain of mass production of food, depends mostly on the availability of research methods (Gerbens *et al.*, 2003), which enable realization of the corresponding parameter measurements during entire production, processing and distribution phases, with application of investigation methods, whose application is sustainable regarding efficiency of obtaining results, costs of the performed tests and scientific impact. In modern research, sustainable production of food is often connected with production of foods based on principles of organic production.

Intended investigations, focused on individual aspects of sustainability of mass food production chain, can be positioned in implementation of the obtained, results so that they veritably contribute to the technological development over integral contribution to sustainability of chain of mass production of food only after their mutual complementarities have been assured through agro-economic analysis of model of sustainable chain of the mass production of foods.

Meat industry – part of the food mass production chain

Meat industry is an important link in the food production chain. Together with intensification of the production process and with production of the even larger quantities of meats, problems with dead animals and accumulation of slaughterhouse wastes also emerge (Table 1).

Solution of the problem of harmless removal of waste products of animal origin, are of exceptional economic importance, today, it is irreplaceable veterinary-sanitary and preventive usage in the suppression of cattle infections and zoonoses and special attention is paid to environmental protection and rehabilitation.

Table 1. Slaughter and Animal Wastes Quantities in Serbia in 2007 (Statistical Office of RS)

Tabela 1. Klanje stoke i količina animalnog otpada u Srbiji u 2007 godini (RZ za statistiku)

Origin of wastes	Slaughter	Wastes, metric tons
Cattle	491 000	21 990
Swines	6 553 000	47 068
Sheep	1 066 000	7 627
Poultry	45 942	27 565
TOTAL		104 250

Quantity of animal waste which appear in circulation of goods (raw meat, intestines, cured products, sausages, fat) as well as quantity of animal

corpses which can be collected, should be added to this quantity. If the production of livestock and meat industry is not going to change drastically, there are 125.000 t of animal by-products annually, or cca 496 t daily, which should be harmlessly removed.

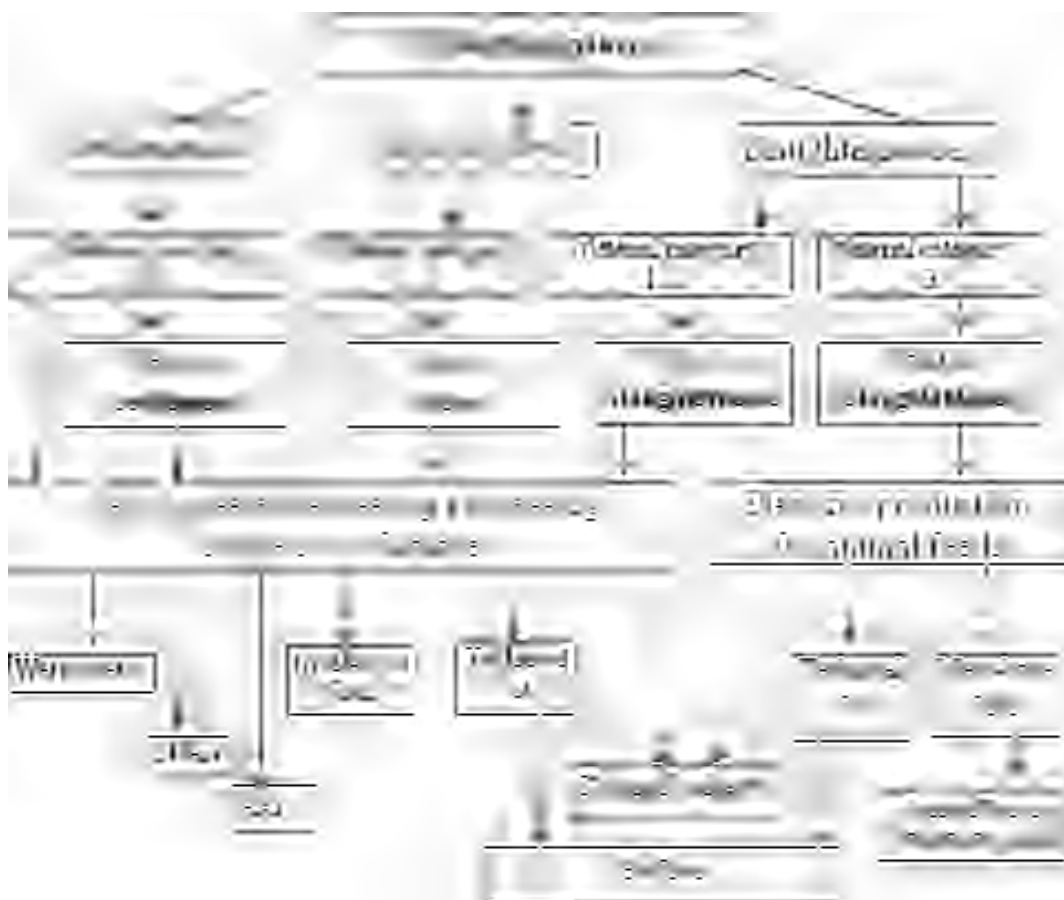
Importance of safe disposal of animal by products

Necessity of solution of safe disposal of animal by-products by their utilization with processing into animal feed and bioenergetics, grows with the intensification of animal growing and the increasing of capacities of industrial slaughterhouses, construction of new small slaughterhouses, building of plants for meat processing and increasing of the volume of international trade of commercial animal products (Okanović *et al.*, 2006). Correct solution of safe disposal of animal by-products can be seen from three key aspects that should fulfill the technological solutions for solving of disposal of such materials by their processing, namely: from the epidemiologic-epizootiologic aspect, with the aspect of environment protection, and the economic aspect.

According to Ristić *et al.*, (1996; 2000), without any doubt, the newest and the best method of safe disposal of animal wastes is their technical processing in separate categories into products for chemical industry, bio-fuels and feed for specific animals.

Prerequisite for safe disposal of animal wastes, using one of the described methods is organized collection and delivery of raw materials. Modern disposal of waste materials demands orderly constructed plants with adequate capacities, which should assure permanent and continuous supply of raw materials. This confirms the importance of recognizing the raw materials fundamentals for each object, i.e. organizing of epizootiologically and economically acceptable region, which should enable obtaining adequate quantities of animal wastes leading to designing and construction of modern object for their safe disposal (Okanović *et al.*, 2008a).

In such collecting circle, organizing of collection of animal wastes represents a very delicate problem, from whose solution to a large extent depends the successful operation of the plant that is going to process such raw materials. This problem, in any case, has to take into consideration both, plant that processes raw materials of animal origin or cattle-growing farms, and slaughterhouses that generate such raw materials. Also, important role in solving of the problem have local municipal communities. They are, according to the existing legislative rules on suppression of contagious diseases, obliged to organize safe disposal



Scheme 1. Organizing of collection, storage and safe disposal of animal waste.

Shema 1. Organizacija sakupljanja, skladištenja, i neškodljivog uklanjanja animalnog otpada

of animal wastes in their region. In other words, organizing the collection of mentioned raw materials should be based on contractual linking of plants for safe disposal and processing of animal wastes and local municipal communities or their corresponding organizations (slaughterhouses, animal farms etc.) (Ristić *et al.*, 2003).

The emphasis on the necessity of transferring of animal wastes from the place where they were generated to the storing place as fast as possible, is of grate importance, as well as the necessity of rapid performing procedure of their processing. This is very significant, not only from the epidemiologic-epizootologic aspect, or from environmental protection aspects, but also from the aspect of their technical processing. Namely, fresh raw materials are processed more easily with generation of lower quantities of waste gases and obtaining of better quality products (Ristić *et al.*, 2007).

Safe disposal of the described animal waste (material Category 1) by combustion on high temperatures (over 850°C) enables obtaining of warm water or steam, as an energent for processing plant that use warm water or steam and ash as construction material for roads.

We shall mention only that, with the respecting procedures of blood collection and its technological processing, various articles for human use can be obtained, primarily products which are used as functional additives in manufacture of meat products. Special processing procedures enable their use as raw materials in pharmaceutical industry or for production of functional foods (Matekalo-Sverak *et al.* 2007).

On the other hand, industrial waste blood can be collected and processed using corresponding technological procedure in a plant for processing of other animal by-products, using special processing unit. Such a one procedure enables obtaining of feed with high protein content, which contains, mostly, high quantities of essential amino acids, vitamins and mineral substances, and, particularly, iron (Okanović *et al.*, 2008b, Ristić *et al.*, 2008).

Articles (meat- and bone meal and fat) obtained by processing of Category 1 of materials are suitable for use as fuel, i.e. as fuel for direct combustion in architecturally separated objects, respecting the corresponding legislative rules.

Conclusions

Economic and general development of the Republic of Serbia should be more based on the organized investigations and development that should produce permanent technological development through the improvement of the existing and creation of new technologies, as well as of new products, processes and services on sustainable foundations which implies their economic, social and above of all, ecological feasibility.

In order to achieve these goals is it necessary to concentrate on:

1. Multidisciplinary research oriented on solving realistic problems which represent brakes for technological development in numerous points of the mass production of food and the necessity of extraordinary large number of economy subjects;
2. Systematic investigations, integrated with all their interrelations and reservations, which assure that these goals are not going to be performed through partial skips in technological development, but through the sustainable solutions, which should bring long-lasting technological development and prosperity;
3. Focused investigations oriented above all on already existing problems.

4. Agricultural and food industry by-products, if not valorized, are disposed on landfills, in waste disposal landfill, buried in arid terrains or in open water courses, thus contaminating the environment.
5. If all mentioned ecological and economical aspects are recognized properly, it becomes clear that organized solving of safe disposal of inedible by-products obtained from slaughtered or died animals by their technical processing is a valuable task. This contributes to prevention of spreading of contagious diseases, and rehabilitation of the environment and rational use of waste materials.
6. The most rational solutions of its disposal is its processing into feed, or raw materials for chemical industry and production of biofuels. Manufacturing of feed from sanitary safe raw materials is multiply valorized, with assurance of the rational development of cattle growing and of protection of the environment. Application of biofuels contributes to reduction of oil consumption (i.e. of imports), reduction of emissions of detrimental gases, stimulation of sustainable development of rural regions.

References

- Gerbens-Leenes P.W., Moll H. C., Schoot Uiterkamp A. J. M., 2003.** Design and development of a measuring method for environmental sustainability in food production system. *Ecological Economics*, 46 (2) p 231–248;
- Green K., Foster. C., 2005.** Give peas a chance: Transformations in food consumption and production systems. *Technological Forecasting and Social Change*, 72 (6) p 663–679;
- Henningsson S., Hyde K., Smith A., Cambel M., 2004.** The value of resource efficiency in the food industry: a waste minimisation project in East Anglia, UK. *Journal of Cleaner Production*, 12 (5) p 505–512;
- Kennedy, P., 1993.** *Arh. Magazin*, 3–8;
- Mastilović Jasna, 2008.** Održivost lanca masovne proizvodnje hrane. XII Internacional *ECO-conference*, *Ecological Movement of the City of Novi Sad*, Proceedings 23–29, Novi Sad;
- Matekalo-Sverak, Vesna, Turubatović, L., Babić, J., Trbović, D., Milićević, D., 2007.** Utilization of powdered hemoglobin in formed meat products. *Proceedings, 53rd ICoMST, Beijing, China*, 431–432;
- Meyer R., 2007.** Comparison of scenarios on futures of European food chains. *Trends in Food Science & Technology*, 18 (11) p 540–545;
- Odredba (EC) 1774/2002 Evropskog Parlamenta i Saveta Evrope;**
- Okanović Đ., Zekić V., Petrović Ljiljana, Tomović V., Đžinić Natalija, 2006.** Ekonomičnost proizvodnje svinjskog mesa u polutkama, *Tehnologija mesa*, (XLVII), 5-6, 237–241;
- Okanović, Đ., Ristić M, Delić, Stanislava, 2008a.** Sporedni proizvodi poljoprivrede i prehrambene industrije i kvalitet životne sredine, *Kvalitet*, 65–68;
- Okanović Đ., Ristić M., Delić Stanislava, Lilić S., 2008b.** Ekonomska analiza opravdanosti investiranja u pogon za preradu krvi, *Biotehnologija u stočarstvu*, vol. 24, (spec. issue), 635–641;
- Risku-Norja H., Mäenpää I., 2007.** MFA model to assess economic and environmental consequences of food production and consumption, *Ecological Economics*, 60 (4) p 700–711;
- Ristić, M., Filipović, S., Sakač Marijana, Kormanjoš, Š., 1996.** *Tehnologija proizvodnje proteinsko-energetskih hraniva od nejestivih sporednih proizvoda zaklane živine*, Monografija, Matica Srpska – Tiski cvet, Novi Sad;
- Ristić M., Radenković Brana, Đorđević, M., 2000.** Monografija „*Neškodljivo uklanjanje uginulih životinja i nejestivih sporednih proizvoda zaklanih životinja*“, Triton-Public, Beograd;
- Ristić, M., Sakač Marijana, Filipović, S., 2003.** Animalni otpaci i njihova sanacija u Srbiji, *Međunarodna eko-konferencija: Zaštita životne sredine gradova i prigradskih naselja*, 397–401, Novi Sad;
- Ristić, M., Filipović, S., Sakač Marijana, 2007.** Usaglašavanje postupaka sakupljanja, transportovanja, prerade, upotrebe i uklanjanja sporednih proizvoda životinjskog porekla

koji nisu namenjeni za ishranu ljudi, sa propisima Evropske Unije. *Projekat*, Institut za prehrambene tehnologije, Novi Sad, str. 13–25 i 30–34;

Ristic M., Okanović Đ., 2008. Processing of animal wastes and environment, XII Internacional *ECO-conference, Ecological Movement of the City of Novi, Proceedings* 321–326, Novi Sad;

Rowe R. L., Street N. R., Taylor G., 2009. Identifying potential environmental impacts of large-scale deployment of dedicated bioenergy crops in the UK, *Renewable and Sustainable Energy Reviews*, 13 (1) p 271–290;

<http://www.etp.ciaa.be>

<http://www.odrzivi-razvoj.sr.gov.yu>

<http://www.rzs.statserb.sr.gov.yu>

Paper recieved: 15.04.2009.

Note: This investigation was carried out within the project: “Sustainability of the chain of mass food production” funded by Ministry of Science and Technological development RS, TR-20066

DIE BEDEUTUNG DER SENSORIK ALS KRITERIUM DER FLEISCHQUALITÄT – EIN VERGLEICH ZWISCHEN VERSCHIEDENEN FLEISCHARTEN UND -ERZEUGNISSEN*

Ristic, M.

Kurzer ueberblick: Für die Erfassung des Genusswertes ist der Mensch das wichtigste „Messinstrument“, der mit seinen Sinnen, wie z.B. Sehsinn, Geruch, Geschmack, Tastsinn zu einer sensorischen Prüfung beitragen kann. Bei verschiedenen Tierarten sind bestimmte Einflussfaktoren für die Fleischqualität wichtig. Beim Geflügelfleisch spielt die Mastintensität und –dauer eine tragende Rolle. Die Rasse und Rassenkreuzungen, sowie Endmastgewichte sind beim Schweinefleisch entscheidend und bei der Rindfleischqualität sind es ebenfalls die Rasse, Mastintensität, -verfahren (Stall- und Weidemast), sowie Schlachtalter und Kategorien. Für die sensorische Analyse stand eine semantisch-nummerische Intervallskala zur Verfügung (1 bis 6), in der eine höhere Punktzahl Ausdruck für eine bessere Bewertung ist. Dabei wurde mit geschulten Prüfern auf Saftigkeit, Zartheit, Aroma und Gesamteindruck geprüft.

Schlüsselwörter: Genusswert, Sensorik, Geflügel, Schwein, Rind, Lamm, Fleischerzeugnisse

The meaning of sensory evaluation as a criterion for meat quality - A comparison of different meat (products)

Abstract: Man is the most important measure for the recording of taste value because he can perform a sensory evaluation with his senses (sense of vision, aroma, taste, sense of touch). For different animal species, certain factors are important for meat quality. For poultry meat, feeding intensity and feeding duration play an important role. Breed and crossbreeding as well as live weight class are decisive for pork < for beef again it is breed, feeding intensity, method of feeding (stable or range land), slaughter age and categories. Semantic-numeric interval scale ranging from 1 to 6 is available, for sensory analysis where a higher number of stands stands for a better evaluation. An expert panel surveyed with respect to juiciness, tenderness, flavour and overall impression.

Key words: taste value, sensory evaluation, poultry, pork, beef, lamb, meat products

Značaj senzorne ocene kao kriterijuma kvaliteta mesa – poređenje između različitih vrsta mesa i proizvoda od mesa

Sadržaj: Za shvatanje prihvatljivosti, čovek je najvažniji „instrument merenja“, koji svojim čulima, kao što su, na primer, čulo vida, mirisa, ukusa i dodira može doprineti senzorskom ispitivanju. Kod različitih vrsta životinja, od značaja su različiti faktori koji utiču na kvalitet mesa. U slučaju mesa živine, osnovnu ulogu imaju intenzitet i trajanje tova. Odlučujući faktori za kvalitet svinjskog mesa su rase i melezi i masa na kraju tova, a kod goveđeg mesa, osim rase, intenzitet tova, postupak gajenja (stajski ili pašnjački), kao i starost i kategorija prilikom klanja. Za senzorsku analizu koriste se semantičko-numeričke skale sa intervalima od 1 do 6, pri čemu veći broj predstavlja bolju vrednost.

Ključne reči: prihvatljivost, senzorika, živina, svinja, goveče, jagnje, proizvodi od mesa

Einleitung

Der Genusswert umfasst alle Kriterien, die beim Genießen eines Lebensmittels zum Tragen kommen und mit den Sinnen erfasst werden. Dabei treten sämtliche Sinne in Aktion: der Gesichtssinn

im Hinblick auf Farbe und Form, der Geruchsinn, der Geschmackssinn und der Tastsinn. Nach Hoffmann (1973, 1995) wird die Fleischqualität definiert als die „Summe aller sensorischen, ernährungs-physiologischen, hygienisch-toxikologischen und verarbeitungstechnologischen Eigenschaften des

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mauntain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

Fleisches“. Die physikalischen Kriterien gleich nach der Schlachtung spielen dabei eine wichtige Rolle zur Erfassung der Fleischqualität (Honikel, 2006). Für die Verbraucher sind die sensorischen Kriterien des Fleisches von großer Bedeutung. Hierbei werden verschiedene Prüfverfahren angewandt (Hammer, 2006; DIN, DLG). Für die Erfassung des Genusswertes ist der Mensch das wichtigste „Messinstrument“, da der entscheidende Teil der Prüfung mit seinen Sinnen (Augen, Geruch, Geschmack, Tastsinn, Temperatur- und Schmerzempfinden, sowie weiteren Sinnen) durchgeführt wird (Ristic, 1988).

Material und Methoden

Geflügelfleisch

Als Versuchsmaterial standen Proben von Broilern (n=2154) aus verschiedenen Versuchsreihen, die sich über einen Zeitraum von 15 Jahren erstreckten, verschiedener Herkünfte und beider Geschlechter zur Verfügung. Ebenfalls wurden die sensorischen Daten von Broilern mit verschiedenen Herkünften (ASA, AA, Hybro, Lohmann, Ross, Shaver, Pilch, Peterson, Cobb) aus den bayerischen Mastleistungsprüfungen in Kitzingen erfasst (n=1000). Die Mastdauer betrug 5-6 Wochen. Vergleichsweise wurden die Daten aus der konventionellen (Ross 308, 5 Wochen) und aus der ökologischen Produktion (RedBro/Shaver, 10 Wochen) herangezogen. Vor der sensorischen Prüfung wurden die Proben des Brust- und Schenkelfleisches in Alufolie verpackt und im Plattenkontaktgrill bis zu einer Kerntemperatur von 75°C erhitzt. Jeweils wurden 10 Proben aus verschiedenen Versuchsgruppen in zufälliger Reihenfolge von einem geschulten Testpanel, bestehend aus 6 Prüfern, bewertet. Der sensorischen Prüfung lag eine semantisch-nummerische Intervallskala von 6 bis 1 zugrunde, in der eine höhere Punktzahl Ausdruck für die bessere Bewertung ist. Die Proben wurden auf Saftigkeit, Zartheit, Aroma und Gesamteindruck geprüft (Ristic, 1983). Proben von Schwein- und Rindfleisch wurden der gleichen Behandlung unterzogen. Weitere ausführliche Informationen über Lebensmittelsensorik sind bei *Hildebrandt* (2008) zu finden.

Bei der statistischen Auswertung werden die einzelnen Punkte von Prüfern zuerst mittels des arithmetischen Mittelwertes oder des Zentralwertes (Median) berechnet. Für die weitere Berechnung kann die Varianzanalyse herangezogen werden. Der multiple Mittelwertvergleich erfolgt durch den Tukey-Test. Signifikante Unterschiede ($p \leq 0,05$) werden mit unterschiedlichen Buchstaben gekennzeichnet.

Versuchsergebnisse und Diskussion

Geflügelfleisch

Broiler

Tabelle 1 gibt Überblick über die sensorischen Daten des Brustfleisches aus verschiedenen Versuchsreihen von Broilern (n=2154). Bei der Saftigkeit ergab sich eine Intervallskala von 3,8 bis 4,9, d.h. die geprüften Proben waren zwischen etwas saftig bis saftig. Der Gesamtmittelwert aller 13 Versuchsreihen lag bei 4,4. Die Bewertungsnoten der Zartheit erreichten ein Qualitätsniveau zwischen 5,0 und 5,7, was sehr zartem Fleisch entspricht. Bei Aroma und Gesamteindruck lag die Bewertung zwischen 3,9 bis 4,7, bzw. zwischen 4,1 bis 4,7. Werden die Gesamtmittelwerte der oben genannten Daten mit den Daten aus der heutigen konventionellen bzw. ökologischen Produktion verglichen (n=200), so ergibt sich eine Verbesserung bei den Daten der konventionellen Produktion, die von Broilern (Ross 308) stammen. Dagegen trat bei der ökologischen Produktion (RedBro) eine Verschlechterung dieser Daten auf. Die Saftigkeit des Schenkelfleisches führte zu einer günstigeren Bewertung (4,3 bis 5,0, Tab. 2). Die Zartheit lag in einem Messbereich zwischen 4,9 bis 5,4. Die Noten für Aroma und Gesamteindruck bewegten sich auf fast gleichem Niveau (3,5 bis 4,4 bzw. 3,7 bis 4,5). Die sensorischen Noten des Schenkelfleisches aus der konventionellen Produktion waren günstiger im Vergleich zum Gesamtmittelwert. Bei der ökologischen Produktion ergab sich wiederum eine schlechtere Bewertung. Werden die sensorischen Daten des Brustfleisches mit denen des Schenkelfleisches verglichen, so kann man feststellen, dass das Schenkelfleisch eine bessere Saftigkeit aufwies, das Brustfleisch dagegen bei Zartheit, Aroma und Gesamteindruck besser abschnitt.

In mehreren bayerischen Mastleistungsprüfungen wurden 9 Herkünfte (n=1000) verglichen. Gleichzeitig führte man bei diesem Material auch eine sensorische Analyse durch (Tab. 3). Die höchste Bewertung der Saftigkeit des Brustfleisches bekam die Herkunft Cobb 500 mit 4,7. Die Herkünfte Shaver und Cobb 500 erreichten für die Zartheit Noten von 5,5. Die günstigsten Noten für Aroma lagen bei 4,5 (Lohmann, Shaver) und beim Gesamteindruck ebenfalls bei 4,5 (AA, Lohmann, Shaver, Cobb 500). Die Herkunft Peterson erzielte die höchste Bewertung bei der Saftigkeit (4,9) und der Zartheit (5,4) des Schenkelfleisches (Tab. 4). Die Broiler Cobb 500 schnitten bei Aroma mit 4,2 und dem Gesamteindruck 4,3 als beste ab. Auch hier ließ sich feststellen, dass die Saftigkeit des Schenkelfleisches

Tab. 1: Sensorische Daten des Brustfleisches¹⁾ (n=2154 bzw. 200; *Ristic* 2009)**Tabela 1.** Senzorski podaci o mesu grudi (n=2154 bzw. 200; *Ristic* 2009)

Versuchsreihe	Saftigkeit		Zartheit		Aroma		Gesamteindruck	
	0	s	0	s	0	s	0	s
A	4,6	0,5	5,0	0,4	4,7	0,5	4,7	0,4
B	4,4	0,4	5,1	0,4	4,2	0,6	4,3	0,5
C	3,8	0,5	5,1	0,4	4,1	0,4	4,1	0,4
D	4,2	0,5	5,2	0,4	4,2	0,5	4,3	0,5
E	4,4	0,5	5,3	0,3	4,3	0,4	4,4	0,4
F	4,6	0,5	5,4	0,3	4,2	0,5	4,4	0,4
G	4,7	0,5	5,3	0,3	4,4	0,4	4,5	0,4
H	4,1	0,5	5,3	0,3	3,9	0,5	4,1	0,5
I	4,8	0,6	5,5	0,4	4,0	0,8	4,2	0,8
J	4,9	0,4	5,7	0,2	4,5	0,6	4,6	0,5
K	4,6	0,4	5,3	0,3	4,3	0,4	4,5	0,3
L	4,4	0,7	5,4	0,3	4,4	0,6	4,5	0,5
M	4,0	0,6	5,0	0,5	4,2	0,6	4,3	0,5
0 Gesamt	4,4	0,6	5,2	0,4	4,3	0,6	4,4	0,5
Konvention. Produktion	4,5	0,5	5,2	0,4	4,7	0,5	4,7	0,4
Ökolog. Produktion	3,8	0,6	5,0	0,5	4,2	0,4	4,3	0,5

¹⁾ Semantisch-nummerische Intervallskala von 1 (sehr unbefriedigend) bis 6 (hervorragend)

Tab. 2: Sensorische Daten des Schenkelfleisches (n=2154 bzw. 200; *Ristic*, 2009)**Tabela 2.** Senzorski podaci o mesu bataka (n=2154 bzw. 200; *Ristic*, 2009)

Versuchsreihe	Saftigkeit		Zartheit		Aroma		Gesamteindruck	
	0	s	0	s	0	s	0	s
A	4,7	0,4	5,0	0,3	4,3	0,5	4,5	0,4
B	4,9	0,4	5,0	0,3	3,8	0,7	3,9	0,6
C	4,3	0,4	5,0	0,3	3,5	0,5	3,7	0,5
D	4,8	0,4	5,2	0,4	3,9	0,7	4,1	0,7
E	4,4	0,4	5,0	0,3	3,9	0,4	4,0	0,4
F	4,9	0,4	5,2	0,4	3,9	0,6	4,1	0,5
G	5,0	0,4	5,4	0,3	4,4	0,5	4,5	0,4
H	4,9	0,4	5,4	0,3	4,0	0,5	4,2	0,5
I	4,7	0,4	5,2	0,4	3,8	0,6	4,0	0,5
J	4,8	0,4	5,3	0,3	4,2	0,5	4,3	0,5
K	4,7	0,4	5,1	0,3	4,2	0,5	4,3	0,4
L	4,6	0,4	4,9	0,4	4,2	0,5	4,3	0,5
M	4,3	0,6	4,5	0,6	3,9	0,6	4,0	0,6
0 Gesamt	4,7	0,4	5,1	0,4	4,0	0,6	4,2	0,6
Konvention. Produktion	4,9	0,6	5,2	0,5	4,3	0,4	4,7	0,5
Ökolog. Produktion	4,3	0,5	4,4	0,6	3,8	0,5	4,2	0,5

günstiger bewertet wurde im Vergleich zum Brustfleisch; dagegen waren die Zartheit, Aroma und Gesamteindruck beim Brustfleisch besser.

der Firma Grimaud und Brinkmann) und Mularden (HYTOP 42) wurden Unterschiede gefunden (Ristic *et al.*, 2006). Eine sehr deutliche Abstufung fand bei

Tab. 3: Sensorische Daten des Brustfleisches verschiedener Herkünfte (n=1000; Ristic, 2009)

Tabela 3. Senzorski podaci za meso grudi različitih provenijencija (n=1000; Ristic, 2009)

Herkunft	Saftigkeit		Zartheit		Aroma		Gesamteindruck	
	0	s	0	s	0	s	0	s
ASA	4,4	0,5	5,3	0,4	4,3	0,5	4,4	0,4
AA	4,5	0,6	5,2	0,3	4,4	0,5	4,5	0,5
Hybro	4,4	0,6	5,2	0,4	4,2	0,5	4,3	0,4
Lohmann	4,5	0,7	5,2	0,4	4,5	0,6	4,5	0,6
Ross	4,4	0,6	5,4	0,4	4,3	0,5	4,4	0,5
Shaver	4,6	0,6	5,5	0,4	4,5	0,5	4,5	0,5
Pilch	4,5	0,5	5,4	0,3	4,0	0,6	4,1	0,5
Peterson	4,4	0,5	5,3	0,4	4,0	0,5	4,2	0,6
Cobb 500	4,7	0,5	5,5	0,3	4,4	0,5	4,5	0,4
F-Wert	***		***		***		***	

Tab. 4: Sensorische Daten des Schenkelfleisches verschiedener Herkünfte (n=1000; Ristic, 2009)

Tabela 4. Senzorski podaci za meso bataka (n=1000; Ristic, 2009)

Herkunft	Saftigkeit		Zartheit		Aroma		Gesamteindruck	
	0	s	0	s	0	s	0	s
ASA	4,7	0,4	5,2	0,4	4,1	0,5	4,2	0,4
AA	4,8	0,5	5,2	0,3	4,0	0,6	4,0	0,6
Hybro	4,7	0,4	5,1	0,4	3,9	0,6	4,0	0,6
Lohmann	4,7	0,4	5,1	0,4	3,8	0,5	4,0	0,5
Ross	4,7	0,5	5,2	0,4	4,0	0,6	4,1	0,6
Shaver	4,6	0,4	5,1	0,2	4,1	0,5	4,2	0,5
Pilch	4,8	0,5	5,3	0,4	3,9	0,5	4,1	0,5
Peterson	4,9	0,3	5,4	0,3	3,7	0,4	3,9	0,4
Cobb 500	4,7	0,3	5,3	0,3	4,2	0,6	4,3	0,5
F-Wert	*		***		***		***	

Enten und Gänse

Bei Überprüfung von verschiedenen Mastverfahren (Schnell-, Intensiv- und Weidemast) bei Gänsen wurde neben dem Schlachtkörperwert noch die sensorische Qualität der Brustmuskulatur untersucht (Ristic, 1991). Die beste Bewertung der Saftigkeit und der Zartheit wurde bei der Schnellmast nach 9 Wochen erreicht (Tab. 5). Die Weidemast führte zu einer schlechteren Bewertung bei allen sensorischen Kriterien. Bei einer weiteren Überprüfung bezüglich der sensorischen Qualität von Pekingenten (Cherry Valley) nach unterschiedlichem Mastalter (42, 47 und 54 Tagen), sowie Flugenten (CANEDINS R 61

der Bewertung der sensorischen Kriterien zwischen den einzelnen Altersstufen von Enten statt (Tab. 6). Die günstigste Bewertung von Saftigkeit, Aroma und Gesamteindruck erzielten die Pekingenten nach einem Alter von 47 Tagen. Die Zartheit von Pekingenten (54 Tage) erreichte die höchste Note von 5,4. Die Flugenten, sowie die Mularden schnitten etwas schlechter ab.

Schweinefleisch

Für die Bewertung der sensorischen Kriterien wurden 2,5 cm dicke Scheiben aus dem Teilstücken

Tab. 5: Sensorische Kriterien des Brustfleisches von Gänsen (n=72; *Ristic*, 1991)**Tabela 5.** Senzorski kriterijumi za meso grudi gusaka (n=72; *Ristic*, 1991)

Mastverfahren ¹	Saftigkeit	Zartheit	Aroma	Gesamteindruck
Schnellmast	4,3	4,4	4,0	4,1
Intensivmast	3,8	4,2	4,0	4,0
Weidemast	2,8	3,2	3,7	3,3

¹Schnellmast 9 Wochen, Intensivmast 23 Wochen, Weidemast 33 Wochen

Tab. 6: Sensorische Kriterien des Brustfleisches von Enten (n=80; *Ristic et al.*, 2006)**Tabela 6.** Senzorski kriterijumi za meso grudi pataka (n=80; *Ristic i sar.*, 2006)

Herkunft	Saftigkeit	Zartheit	Aroma	Gesamteindruck
Pekingente (42 Tage)	4,8	5,0	5,0	5,0
Pekingente (47 Tage)	5,4	5,2	5,6	5,4
Pekingente (54 Tage)	4,8	5,4	4,9	4,9
Flugente (84 Tage)	4,6	4,4	4,6	4,6
Mularde (84 Tage)	3,9	3,8	4,1	3,9

Kotelett (*M. longissimus dorsi*) und Kamm in Abhängigkeit von der Mastendgewichtsstufe mit Alufolie abgedeckt und im Plattenkontaktgrill bis zu einer Kerntemperatur von ca. 75°C gegrillt. Die Proben stammen von Pietrain-NN* Landrasse-Kreuzungen mit einem Lebendgewicht von 110, 135 und 160 kg (*Fischer et al.*, 2006). Bei den gegrillten Rückensteaks (LD) schnitten die Proben aus der 135 kg-Gruppe am besten ab und die aus der 160 kg-Gruppe am schlechtesten (Tab. 7). Signifikante

Differenzen gab es jedoch nur bei der Saftigkeit und dem Gesamteindruck. Bei den Kammsteaks änderten sich die Bewertungen mit zunehmendem Mastendgewicht nur geringfügig und für die einzelnen Prüfkriterien in unterschiedlicher Richtung. In einer weiteren Untersuchung von Schweinefleisch bei ausgewählten Rassenkreuzungen (Hampshire (Ha), Duroc (Du), Pietrain (Pi-nn), sowie Pi-NN und den Kreuzungskombinationen Du*Ha und Ha*Pi-nn) wurde der Genusswert am *M. longissimus*

Tab. 7: LSQ-Mittelwerte von sensorischen Merkmalen bei gegrillten Steaks aus den Teilstücken Kotelett (*M. longissimus dorsi*) und Kamm (*Fischer et al.*, 2006, mod.)**Tabela 7.** LSQ – srednje vrednosti senzornih osobina odrezaka pečenih na roštilju (kotlet - *M. longissimus dorsi*) i grebena (*Fischer i sar.*, 2006, mod.)

Merkmal ¹	Muskel ²	Mastendgewicht		
		110 kg n=36	135 kg n=54	160 kg n=33
Saftigkeit	LD	3,0 ^{ab}	3,4 ^a	2,9 ^b
Zartheit		3,6	3,8	3,5
Aroma/Geschmack		3,4	3,4	3,2
Gesamteindruck		3,3 ^{ab}	3,5 ^a	3,1 ^b
Saftigkeit	Kamm	4,0	3,9	3,9
Zartheit		4,1	4,3	4,0
Aroma/Geschmack		4,0	3,7	3,9
Gesamteindruck		3,9	3,8	3,8

¹ Beurteilung nach 6-Punkte-Skala: 1 = schlechteste, 6 = beste Bewertung

² LD=*M. longissimus dorsi* (3.-4. Lendenwirbel), Kamm = Querschnitt der Muskulatur aus Teilstück Kamm über 3.-4. Halswirbel

^{a,b} Ungleiche Indices kennzeichnen signifikante Differenzen (P<0,05) zwischen den Mastend-gewichtsstufen

dorsi ermittelt (Fischer et al., 2000). Bei allen genannten Prüfmerkmalen lagen die Nachkommen der reinrassigen Du-Eber an der Spitze (Tab. 8). Signifikante Unterschiede bestanden bei Zartheit und Saftigkeit zur Pi-nn-Gruppe, die durch einige PSE-Fälle belastet ist, ebenso bei Aroma/Geschmack und Gesamteindruck, aber auch zusätzlich zur Pi-NN-Gruppe. Inwieweit die Freilandhaltung von Mastschweinen als Beitrag zur Landschaftspflege von Schweinen auf stillgelegten landwirtschaftlichen Nutzflächen dienen könnte, wurde am Beispiel

Tab. 8: Mittelwerte von Merkmalen der sensorischen Qualität im *M. longissimus dorsi* (n=30-34; Fischer et al., 2000, mod.)

Tabela 8. Srednje vrednosti osobina senzornog kvaliteta *M. longissimus dorsi* (n=30-34; Fischer et al., 2000, mod.)

Eberrasse	Saftigkeit	Zartheit	Aroma	Gesamteindruck
Ha	3,7 ^a	3,9	3,4	3,5
Du	3,7 ^a	4,2 ^a	3,7 ^a	3,8 ^a
Pi-NN	3,5	3,8	3,3 ^b	3,4 ^b
Pi-nn	3,3 ^b	3,4 ^b	3,2 ^b	3,3 ^b
Ha*Pi-nn	3,6	3,9	3,5	3,6
Du*Ha	3,6	3,8	3,4	3,5
F-Test	**	***	**	***

nur mit ungleichen Buchstaben gekennzeichnete Mittelwerte sind signifikant ($P < 0,05$) verschieden

des *Düppeler Weideschweins* untersucht (Fischer, Beinlich, 2005). Die im Mai mit ca. 9 Monaten geschlachteten Tiere erhielten bis Ende Februar erhöhte Getreiderationen, weil die Weidefläche aufgrund der Jahreszeit nur geringen Aufwuchs bot. Bei der sensorischen Prüfung lagen die Messwerte in einem Bereich von 3,5 bei der Saftigkeit und 3,9 bei der Zartheit (Tab. 9). Allerdings ergab sich zwischen Minimum- und Maximumwerten bei den einzelnen sensorischen Kriterien eine große Spannbreite.

Tab. 9: Merkmale der sensorischen Qualität im *M. longissimus dorsi* (Mittelwert, Standardabweichung und Spannweite; Fischer und Beinlich, 2005, mod.)

Tabela 9. Senzorske osobine kvaliteta *M. longissimus dorsi* (srednja vrednost, standardna devijacija, raspon Fischer, Beinlich2005.)

Merkmal	0	s	min	max
Saftigkeit	3,5	0,5	2,8	4,3
Zartheit	3,9	0,6	3,0	5,0
Aroma	3,6	0,4	3,2	4,5
Gesamteindruck	3,6	0,5	3,0	4,7

Übersicht 1. Bewertungsschemata verschiedener Autoren

Pregled 1. Šeme ocenjivanja prema različitim autorima

Punkte	Prädikat	Kriterien	Autor
0-10	verdorben - vollkommen	Geschmack, Geruch, Farbe, Aussehen, Konsistenz, Formerhaltung	Gutschmidt (1951)
1-8	extrem weich, extrem fest	Festigkeit, Zartheit, Krümeligkeit, Klebrigkeit, Saftigkeit	Fischer (1990)
1-8	außerordentlich schlecht, außerordentlich gut	Zartheit, Aroma/Geschmack, Gesamteindruck	Branscheid et al. (2006)
1-6	unbefriedigend, ausgezeichnet	Saftigkeit, Zartheit, Aroma, Gesamteindruck	Ristic (1983)
1-6	geringste- bzw. höchste Merkmalsintensität	Festigkeit, Saftigkeit, Kauphase, Zartheit, Krümeligkeit, unzerkaubare Bestandteile	Augustini (1996)
	beschreibende Begriffe	Aussehen, Geruch, Geschmack, Textur/ Mundgefühl	DIN 10964
0-5	ungenügend - sehr gut	Äußeres, Aussehen, Farbe, Farbhaltung, Zusammensetzung, Konsistenz, Geruch, Geschmack	DLG (2009)

Übersicht 2. Bewertungsschema (*Ristic, 1983*)
Pregled 2. Šema ocenjivanja (*Ristic, 1983*)

Punktezahl	Saftigkeit	Zartheit	Aroma	Gesamteindruck
6	sehr saftig	sehr zart	ausgezeichnet	ausgezeichnet
5	saftig	zart	sehr gut	sehr gut
4	etwas saftig	etwas zart	gut	gut
3	etwas trocken	etwas zäh	befriedigend	befriedigend
2	trocken	zäh	ausreichend	ausreichend
1	sehr trocken	sehr zäh	wenig ausreichend	unbefriedigend

Übersicht 3. Bewertungsschema nach DLG (2009)
Pregled 3. Šema ocenjivanja prema DLG (2009)

Punkte	Qualitätsbeschreibung	Allgemeine Eigenschaften
5	sehr gut	keine Abweichung von den Qualitätserwartungen
4	gut	geringfügige Abweichungen
3	zufriedenstellend	leichte Abweichungen
2	weniger zufriedenstellend	deutliche Abweichungen
1	nicht zufriedenstellend	starke Abweichungen
0	ungenügend	nicht bewertbar

Übersicht 4: Bewertungsschema für Zartheit, Aroma/Geschmack und Gesamteindruck (*Branscheid et al, 2006, mod.*)

Pregled 4. Šema ocenjivanja za mekoću, ukus i ukupan utisak (*Branscheid i sar., 2006, mod.*)

Bewertung	Punkte
außerordentlich gut	8
sehr gut	7
gut	6
noch gut	5
eher schlecht	4
schlecht	3
sehr schlecht	2
außerordentlich schlecht	1

Rind- und Lammfleisch

Für diese Untersuchung standen 2- und 3-jährige Ochsen der Rasse Hereford aus Uruguay aus ganzjähriger Weidehaltung mit einem mittleren Schlachtgewicht von 225 bzw. 282 kg zur Verfügung. Zu einem Vergleich wurden Proben aus Deutschland aus einem Qualitätsfleischprogramm von Jungbulln der Rasse Fleckvieh und Kreuzungen Fleckvieh x Limousin im Alter von 19 bis 23 Monaten mit einem mittleren Schlachtgewicht von 383 kg aus der Intensivmast herangezogen (*Branscheid et*

al., 2006). Die uruguayischen Proben wurden 20 Tage, die deutschen Proben von jedem Tier je zur Hälfte 7 bzw. 20 Tage gereift. Tab. 10 gibt Information über die Zartheitsbewertung der Rindfleischproben durch die Verbraucher. Prüfer mittleren Alters (26-40 Jahre) bewerteten die Proben kritischer als die jüngeren und älteren Altersgruppen. Zwischen den verschiedenen Tierarten wurden bezüglich des Alters statistische Unterschiede gefunden. Für die Bewertung des Gesamteindrucks standen Fleischproben kastrierter männlicher Lämmer der Rasse Corriedale aus Uruguay, die in Weidehaltung gemästet wurden, zur Verfügung. Dabei handelte es sich um leichte (3-4 Monate, Schlachtgewicht 11,1 kg) und schwere Tiere (12-13 Monate, Schlachtgewicht 19,4 kg). Parallel dazu wurden wiederum Tiere aus Deutschland herangezogen, nämlich unkastrierte männliche Lämmer der Kreuzungen schwarzköpfiges Fleischschaf bzw. Suffolk x Merinolandschaf mit einem Alter von 4-6 Monaten (Schlachtgewicht von 23,2 kg). Auch hier wurden die uruguayischen Proben 20 Tage, die deutschen je zur Hälfte 7 und 20 Tage gereift. Hierbei zeigte sich wiederum, dass die Prüfer unterschiedlicher Altersgruppen auch unterschiedlich bewerteten. Bei den Proben aus Uruguay lag die Bewertung des Gesamteindrucks bei den Noten zwischen 5,5 bis 6,7 und bei den Proben aus Deutschland zwischen 5,8 bis 7,4 (Tab. 11). Die Grenze für „noch gute“ Bewertung lag bei Note 5. Die Rindermast wird in den Ländern mit

intensiver Landwirtschaft aufgrund der höheren wirtschaftlichen Effizienz überwiegend mit Bullen durchgeführt. Unter vergleichbaren Bedingungen ist der Fettgehalt des Fleisches niedriger und die Fleischfarbe dunkler, sind Bullen stressempfindlicher und streuen die Qualitätsmerkmale stärker. Dadurch ist das Fleisch zäher und im Aroma flacher (Augustini, 2001). Tabelle 12 gibt Auskunft über die sensorischen Eigenschaften von Bullen und Färsen (Rasse Aubrac). Hierbei zeigte sich, dass das Färsenfleisch eine günstigere Bewertung erhielt. Eine niedrige Energiekonzentration des Futters wirkt sich besonders negativ bei Jungbullen aus (Tab. 13). Bei der Bewertung der Fleischqualität von Schwarzbuntbullen unterschiedlicher Mastintensität wurden die besten Noten für Zartheit und Aroma bei einer

höheren Mastintensität erreicht. Das gleiche gilt für den Vergleich bezüglich des Mastverfahrens, nämlich zwischen Stall- und Weidemast, für die Stallmast (Tab. 14). Die im Alter zwischen 16,8 und 23 Monaten Mastbullenschlachtkörper wurden in 4 Altersgruppen mit je 14-16 Tieren aufgeteilt (KÖGEL et al., 2002). Die Scherkraftwerte waren in der ersten Gruppe am höchsten, gingen dann bis zur Gruppe 3 in etwa linear zurück und stiegen bis zur Gruppe 4 wieder etwas an. Dieser Verlauf in der Fleischzartheit (Scherkraftwert) deckt sich weitgehend mit dem Verlauf der Zartheit – sensorisch ermittelt (Tab. 15). Es liegt die Vermutung nahe, dass ein höherer intramuskulärer Fettgehalt der Gruppe 3 eine positive Einwirkung auf die Zartheit hatte.

Tab. 10: Zartheitsbewertung der Rindfleischproben (n=100; Branscheid et al., 2006, mod.)

Tabela 10. Ocena mekoće govedeg mesa (n=100; Branscheid i sar., 2006, mod.)

Alter der Prüfer	Uruguay 2 Jahre	Uruguay 3 Jahre	Deutschland 7 Tage	Deutschland 20 Tage
18-25 Jahre	5,8 ^b	5,2 ^{ab}	5,1 ^a	6,1 ^a
26-40 Jahre	4,9 ^b	5,1 ^{ab}	4,3 ^{bc}	5,0 ^b
41-60 Jahre	5,9 ^a	4,9 ^b	4,2 ^c	4,9 ^b
61-75 Jahre	5,6 ^b	5,7 ^a	5,1 ^{ab}	6,7 ^a

Tab. 11: Gesamteindruck der Lammfleischproben (n=100; nach Branscheid et al., 2006, mod.)

Tabela 11. Ukupan utisak probe jagnječeg mesa (n=100; nach Branscheid i sar., 2006, mod.)

Alter der Prüfer	Uruguay leicht	Uruguay schwer	Deutschland 7 Tage	Deutschland 20 Tage
18-25 Jahre	5,7 ^{ab}	5,8 ^b	6,2 ^b	6,4 ^b
26-40 Jahre	5,9 ^{ab}	6,1 ^{ab}	5,8 ^b	5,8 ^b
41-60 Jahre	5,5 ^b	5,7 ^b	5,8 ^b	6,2 ^b
61-75 Jahre	6,4 ^a	6,7 ^a	7,2 ^a	7,4 ^a

Tab. 12: Fleischqualität verschiedener Kategorien (*M. long. dorsi*, Aubrac; Augustini, 2001, mod.)

Tabela 12. Kvalitet mesa različitih kategorija (*M. long. dorsi*, Aubrac; Augustini, 2001, mod.)

Merkmal	Bulle (n=15)	Färsen (n=6)
Anzahl	15	6
Zartheit	4,0 ± 0,8	4,8 ± 0,5
Saftigkeit	4,1 ± 0,8	4,7 ± 0,6
Aroma	4,0 ± 0,5	4,2 ± 0,4

Tab. 13: Einfluss der Mastintensität auf die Fleischqualität (Schwarzbuntbullen; Augustini, 2001, mod.)

Tabela 13. Uticaj intenziteta tova na kvalitet mesa (Augustini, 2001, mod.)

Mastintensität	hoch	mittel	niedrig
n	18	24	8
Zartheit	4,9 ± 0,4	4,3 ± 0,9	3,4 ± 0,9
Aroma	4,2 ± 0,6	3,9 ± 0,6	3,2 ± 0,8

Tab. 14: Fleischqualität von Färsen einer Blonde d'Aquitaine x Braunviehkreuzung nach Stall- und Weidemast; (Augustini, 2001, mod.)

Tabela 14. Kvalitet mesa junećeg meleža Blonde d'Aquitaine x Braunvieh (Augustini, 2001, mod.)

Mastverfahren	Stallmast	Weidemast
n	10	11
Zartheit	4,2	3,7
Aroma	3,8	2,9

Tab. 15: Merkmale der Fleischqualität bei Jungbullen, nach Schlachtersklassen (n=60; Kögel et al., 2002, mod.)

Tabela 15. Osobin kvaliteta mesa mladih bikova prema starosnoj kategoriji ((n=60; Kögel i sar., 2002, mod.)

Schlachalter (Monate)	n	Scherkraft (kg)	Zartheit (Punkte)	intramuskulärer Fettgehalt (%)
16,8	15	6,0	3,6	2,85
18,8	15	5,8	3,8	2,58
20,7	14	5,3	4,0	2,89
23,0	16	5,4	3,9	2,30

Tab. 16: Sensorische Bewertung der Rohschinken nach DLG-5-Punkte-Schema (Troeger et al., 2006, mod.)

Tabela 16. Senzorna ocena sirovih šunki prema DLG šemi sa 5 tačaka (Troeger i sar., 2006, mod.)

Produkt	Qualitätsabweichung	Bewertung ¹ (Punkte)	Qualitätszahl ²
Knochenschinken I	Farbfehler (Vergrauung im Kern)	4	3,6
	Speck rötlich	4	
	beginnende Fettveränderung	4	
	salzig	3	
Knochenschinken II	Farbfehler	4	3,6
	Blutpunkte	4	
	salzig	3	
Lachsschinken I	salzig	4	4,2
	phenolisch	3	
Lachsschinken II	ohne Abweichungen		5,0
Kammschinken	Speck rötlich	4	4,5
	leimig	4	

¹ maximale Punktzahl: 5

² maximale Qualitätszahl: 5,00

Goldener DLG-Preis = 5,00

Silberner DLG-Preis = 4,50-4,99

Bronzener DLG-Preis = 4,00-4,49

Fleischerzeugnisse

Im Rahmen der sensorischen Prüfung wurden die Fleischerzeugnisse von 5 Sachverständigen bezüglich Aussehen, Konsistenz, Geruch und Geschmack nach dem DLG-5-Punkte-Schema (ausgezeichnet = Qualitätszahl 5,0; sehr gut = Qualitätszahl 4,5-4,9; gut = Qualitätszahl 4,0-4,4; ohne Prämierung = Qualitätszahl < 4,0) bewertet. Hierbei ergab sich eine Bewertung von Rohschinken mit den Qualitätszahlen in einem Bereich zwischen 3,6-5,0 (Troeger et al., 2006). Das Produkt Lachsschinken erhielt mit 5,0 die beste Note (Tab. 16). Bei einer weiteren Untersuchung von Rinder- und Schweineschinken haben die Rinderschinken eine Bewertung zwischen 3,9-5,0 erreicht, die Schweineschinken zwischen 4,2-4,8 (Tab. 17). Bei den Knochenschinken wurde „salzig“ in geringerer Ausprägung beanstandet, sowie bei dem 20 Monate gereiften Schweineschinken „beginnende Fettveränderung“ ebenfalls in geringerer Ausprägung (Troeger et al., 2007).

Tab. 17: Sensorische Bewertung der Produkte nach dem DLG-5-Punkte-Schema (Troeger et al., 2007, mod.)
Tabela 17. Senzorska ocena proizvoda (Troeger i sar., 2007, mod.)

Produkt	Qualitätsabweichung	Bewertung (Punkte)	Qualitätszahl
Rinderschinken, roh I	Farbe zu dunkel (DFD)	4	3,9
	Rauch zu stark	3	
	säuerlich	4	
	dampfzig	4	
	Fluoreszenz im Kern	ohne Punktabzug	
Rinderschinken, roh II	ohne Abweichungen		5,0
Rinderschinken, roh III	Fluoreszenz im Kern (deutlich)	ohne Punktabzug	4,2
	Rauch zu stark	3	
	salzig	4	
	säuerlich	4	
Schweineschinken, roh I	beginnende Fettveränderung	4 4	4,2
	salzig		
Schweineschinken, roh II	Speck rötlich	4	4,8
	salzig	4	
	Fluoreszenz	ohne Punktabzug	

Zusammenfassung

An einem umfangreichen Versuchsmaterial wurde der Genusswert des Brust- und Schenkel-fleisches von Broilern erfasst (n=3154). Die höchste Bewertung erreichte die Zartheit (5,4-5,7), gefolgt von Saftigkeit (4,9-5,0), Aroma und Gesamteindruck (4,4-4,7). Beim Vergleich zwischen verschiedenen Herkünften wurde durchaus eine sensorische Bewertung in einem oberen Qualitätsniveau gefunden. Die Schnellmast bei Gänsen führte zu einer besseren Bewertung der Saftigkeit und der Zartheit. Bei den Enten ergab die Mastdauer von 47 Tagen die günstigsten sensorischen Noten. Die Bewertungsnoten von Schweinefleisch in Abhängigkeit von den verschiedenen Einflussfaktoren lagen in einem Messbereich zwischen 3,0 bis 4,3, die

einem mittleren Qualitätsniveau entsprechen. Das Rindfleisch erreichte eine sensorische Bewertung im Durchschnitt von 2,9 bis 4,2 und lag somit auf gleichem Qualitätsniveau wie das Schweinefleisch. Vergleicht man die Bewertungsnoten zwischen den verschiedenen Fleischarten, ließ sich feststellen, dass das Geflügelfleisch im oberen Qualitätsniveau (4 bis 6) lag, wohingegen Schweine- und Rindfleisch ein mittleres Qualitätsniveau (3 bis 4) erreichten. Die Fleischerzeugnisse werden nach der DLG eigenen Prüfmethode „Beschreibende Prüfung mit integrierter Bewertung“ mit der Qualitätsbeschreibung von sehr gut (=5) bis ungenügend (=0) bewertet. Dabei werden die Abweichungen registriert und daraus wird die Qualitätszahl abgeleitet, anschließend wird die Prämierung des Produkts vorgenommen.

Literatur

- Hoffmann, K., 1973.** Was ist Fleischqualität? Fleischwirtschaft 53, 485;
- Hoffmann, K., 1995.** Der Qualitätsbegriff bei Fleisch – Inhalt und Anwendung. Kulmbacher Reihe Bd. 14, Bundesanstalt für Fleischforschung, 169–193;
- Honikel, K., O., 2006.** Physikalische Messmethoden zur Erfassung der Fleischqualität. In: Qualität von Fleisch und Fleischwaren, Bd. 2, Frankfurt a.M., Fleischerfachverlag; 855–881;
- Hammer, G., 2006.** Methodik der sensorischen Analyse. In: Qualität von Fleisch und Fleischwaren, Bd. 2, Frankfurt a.M., Fleischerfachverlag 882-889;
- DIN 10 969.** Sensorische Prüfverfahren, beschreibende Prüfung mit anschließender Qualitätsbewertung. Beuth Verlag GmbH, Berlin, 2000;
- DLG-Qualitätswettbewerb: Prüfbestimmungen für Fleischerzeugnisse (Schinken und Wurst), 2009.** Hrsg.: Deutsche Landwirtschafts-Gesellschaft e.V., Zertifizierungsstelle. Frankfurt/Main, 51. Auflage;
- Hildebrandt, G., 2008.** Geschmackswelten – Grundlagen der Lebensmittelsensorik. DLG-Verlag, Frankfurt a.M.;
- Fischer, K., J., Lindner, J., P., Judas, M., Höreth, R., 2006.** Schlachtkörperzusammensetzung und Gewebebeschaffenheit von schweren Schweinen. II. Mitteilung;

- Merkmale der Fleisch- und Fettqualität. Arch. Tierz., Dummerstorf 3, 279–292;
- Branscheid, W., Dobrowolski, A., Spindler, M., San Julian, C., Font Furnols, M., Angels Oliver, M., Caneque, V., Montossi, F., Wicke, M., 2006.** Verbraucherakzeptanz von uruguayischem und deutschem Rind- und Lammfleisch. Fleischwirtschaft 86 (8), 101–106;
- Fischer, K., 1990.** Sensorische Prüfung in der Qualitätsbewertung von Schweinefleisch. Manuskript zu Workshop „Schweinefleischbeschaffenheit nach der Halothansanierung“, 17./18. Dezember 1990, Nordhausen;
- Fischer, K., Wicke, M., Lindner, J. P., Reichel, M., 2000.** Der Genusswert von Schweinefleisch bei ausgewählten Rassenkreuzungen. Mitteilungsblatt der BAFF (148), 669–677;
- Fischer, K., Beinlich, B., 2005.** Freilandhaltung von Maatschweinen als Beitrag zur Landschaftspflege – Realisierte Schlachtkörper- und Fleischqualität am Beispiel des *Düppeler Weideschweins*. Mitteilungsblatt der Fleischforschung Kulmbach 44 (170), 295–303;
- Augustini, C., 1996.** Bewertungsschema „Texturmerkmale von Fleisch“, 22. Kulmbacher Fortbildungstage 14.-16. und 16.-18. Oktober 1996 der BAFF Kulmbach;
- Augustini, C., 2001.** Qualitätsrindfleischerzeugung zwischen extensiver und intensiver Produktion. Fleischwirtschaft 81 (4), 134–138;
- Kögel, J., C., Augustini, Petautschnig, A., 2002.** Einfluss des Schlachalters auf die Rindfleischqualität: Untersuchungen der Arge Alpen-Adria führen zu neuen Erkenntnissen. Schule und Beratung Nr. 12 IV 4–12;
- Troeger, K., Irina Dederer, M., Ristic, P., Radetic, L., Turubatovic, D., Cavor, 2006.** Qualität von Rohschinken aus Montenegro, hergestellt nach traditionellem Verfahren. Fleischwirtschaft 86 (4), 100–103;
- Troeger, K., Dederer Irina, Ristic, M., Turubatovic, L., Beric, M., Stojanovic A., 2007.** Rohpökelwaren und Rohwurst aus Serbien – Qualität der nach traditionellen Verfahren hergestellten Produkte. Fleischwirtschaft 87 (8), 95–100;
- Ristic, M., 1988.** Genusswert von Rindfleisch. Fleischwirtschaft 68 (9), 1130–1138;
- Ristic, M., 1983.** Einfluss von Geschlecht und Alter auf sensorische Daten von Broiler verschiedener Herkunft. Mitteilungsblatt der BAFF 81, 5586–5600;
- Ristic, M., 1991.** Schlachtkörperwert von Gänsen verschiedener Herkünfte und Mastverfahren. Mitteilungsblatt der BAFF 30, Nr. 111, 5–10;
- Ristic, M., Damme, K., Freudenreich, P., 2006.** Schlachtkörperwert von Enten und Gänsen. Abhängigkeit von Herkunft und Alter der Tiere. Fleischwirtschaft 86 (2), 107–110;
- Ristic, M., 2009.** Sensorische Eigenschaften des Broilerfleisches – Ein Vergleich zwischen verschiedenen Versuchsreihen und Herkünften. Mitteilungsblatt der Fleischforschung Kulmbach 48, Nr. 183, 7–13;
- Gutschmidt, J., 1951.** Über die organoleptische Bewertung von Lebensmitteln mit Hilfe des Karlsruher Bewertungsschemas. Dt. Lebensmittel-Rundschau 47, 244–251;
- Schön, L., Ristic, M., Reuter, H., 1974.** Über die Haltbarkeit von (tief)gefrorenem Schlachtgeflügel. Die Fleischwirtschaft 54 (5), 909–912.

Paper received: 1.04.2009.

Сенсорные системы «электронный нос» для контроля качества мяса*

Чернуха Ирина М., Кузнецова Татьяна Г., Селиванова Екатерина Б.

Р е ф е р а т: Научные исследования показали целесообразность применения электронного носа «VOCmeter» для инструментального контроля качества свежести мяса и мясных продуктов. Результаты подтвердили возможность использования системы для определения видовой принадлежности мяса.

Сравнительные исследования показали перспективность применения инструментальных систем для объективной оценки запаха и аромата мяса и мясных продуктов, что позволит в дальнейшем избежать влияния человеческого фактора при органолептических исследованиях. Преимуществами этих методов являются простота и скорость выполнения анализов.

Ключевые слова: мясо, электронный нос, свежесть, аромат, видовая принадлежность

Electronic nose sensory systems for meat quality control

A b s t r a c t: Researches confirmed VOCmeter (electronic nose) to be an advanced meat quality control instrument, in particular to analyze the freshness of meat and meat products. Results show the possibility of VOCmeter to be used for meat species identification, as well.

Comparative experiments demonstrated the perspectives of instrumental systems for objective flavor evaluation of meat and various meat products in order to avoid the "human factor" in panel testing.

Simplicity, sensitivity and rapidity are the advantages of the above method.

Key words: sensory systems, electronic nose, product freshness, identification, flavor evaluation

Senzorni sistem elektronskog nosa za kontrolu kvaliteta mesa

S a d r ž a j: Autori su potvrdili da je VOCmetar (elektronski nos) savremen kontrolni instrument za određivanje kvaliteta mesa, posebno za vršenje ispitivanja svežine mesa i mesnih proizvoda. Rezultati ukazuju na mogućnost upotrebe VOSmetra u cilju određivanja životinjskog porekla mesa.

U poređni ogledi su pokazali perspektive instrumentalnih sistema za objektivnu evaluaciju ukusa mesa i mesnih prerađevina u cilju izbegavanja "ljudskog faktora" tokom testiranja.

Jednostavnost, osetljivost i brzina su glavne prednosti prikazane metode.

Ključne reči: senzorni sistemi, elektronski nos, svežina proizvoda, identifikacija, procena ukusa

Введение

Для контроля качества пищевых продуктов традиционно используются органолептические показатели, оцениваемые в основном с помощью органов зрения, вкуса и обоняния. Большую ценность органолептические показатели приобретают при дополнении качественной информации количественной оценкой, получаемой с помощью аналитических методов.

Запах - один из основных показателей качества пищевых продуктов, который формируется комплексом летучих веществ. Анализ

запаха осложнен тем, что его составляют разнообразные легколетучие вещества с относительно небольшой молекулярной массой (Анисимкин и сат. 1998). В связи с чем, применение средств и технологий современной техники и электроники для решения задач, связанных с установлением качества запаха, является, без сомнения, чрезвычайно актуальным. Аналитические возможности современных газовых и жидкостных хроматографов и масс-спектрометров позволяют получить разнообразную информацию о качественном и количественном составе запахов пищевых про-

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

АВТОРЫ: Ирина М. Чернуха, kd-7@mail.ru, Татьяна Г. Кузнецова, Екатерина Б. Селиванова, ГНУ ВНИИМП им. В.М. Горбатова Россельхозакадемии, 109316, Россия, г. Москва, ул. Талалихина, 26.

AUTORI: Irina M. Černuha, kd-7@mail.ru, Tatjana Kuznecova, Ekatarina B. Selivanova, Sveruski naučnoistraživački institut industrije mesa, VNIIMP, Gorbatova Roselholcakademi, Talalihina 26, 109316, Moskva, Rusija.

дуктов. Однако такие исследования являются зачастую неоправданно дорогостоящими, требуют сложной подготовки проб, больших затрат времени и химических реактивов. Именно по этой причине становятся приоритетными разработки более простых, дешевых и, самое главное, быстрых анализаторов для экспрессной оценки состава запахов пищевых продуктов в практической работе лабораторий предприятий.

В конце 1980 годов была разработана схема одновременной обработки аналитических сигналов от группы неселективных сенсоров (*Грень и сат* 1985). Возможность реализации такого технического решения основана, в первую очередь, на опережающем развитии средств вычислительной техники, обеспечивающей обработку многопараметрической информации в режиме реального времени. При использовании мультисенсорной системы можно получать с известной точностью информацию, как о составе, так и о концентрации отдельных составляющих многокомпонентных газовых смесей. Итогом этих исследований стал новый тип искусственных аналитических систем - «электронный нос» (*Грень и сат* 1985).

«Электронный нос» - это анализатор паров или газов на основе разнородных сенсоров, имитирующих работу органов обоняния человека. Подобная сенсорная система обеспечивает получение узнаваемого образа анализируемой смеси паров пахучих веществ, которая может содержать сотни различных химических соединений. «Электронный нос» состоит из сенсоров, которые подбираются по их химическому средству к отдельным компонентам анализируемой смеси газов и паров. Каждый сенсор обладает различной чувствительностью к анализируемым веществам и имеет свой специфический профиль откликов в ответ на тестируемые запахи (*Чернуха и сат* 2008).

В свете решения задач оценки качества мясного сырья и вспомогательных материалов, а также идентификации и установления факта фальсификации пищевых продуктов при входном контроле на предприятиях, использование «электронного носа» представляется достаточно перспективным. Применение сенсорных систем позволяет обойти массу проблем, связанных с использованием в оценке качества пищевых продуктов специально обученных дегустаторов. К числу таких проблем относят: особенности сенсорной чувствительности каждого из дегустаторов; адаптацию чувствительности обонятельного органа при длительном воздействии стимула; влияние на остроту обо-

няния усталости, различных инфекций, токсических веществ, физического состояния человека; субъективности в оценках восприятия и ряд других факторов.

В представленной статье показаны возможности использования мультисенсорной системы «VOCmeter» для оценки качества мясного сырья и готовой продукции, с целью повышения объективности получаемых результатов, внедрения экспресс-методик, позволяющих сократить время и затраты на проведение испытаний, а также избежать противоречий, возникающих при использовании традиционных методов исследования.

Материалы и методы

Объектами исследования являлось мясное сырьё (свинина охлаждённая и размороженная); охлаждённое мясное сырьё различных видов убойных животных и птицы; свинина запечённая, полученная из охлаждённого и размороженного мясного сырья; свинина и говядина после варки; ветчинные консервы: из свинины (ветчина «Рубленая») и из говядины (ветчина «Любительская»). Сенсорную оценку проводили органолептическим методом и на приборе «VOCmeter» фирмы «AppliedSensor» (Германия), включающем восемь сенсоров QMB и четыре сенсора MOS (рис.1).



Рис. 1. Прибор „VOCmeter”
Slika 1. Instrument „VOCmeter”

Результаты и обсуждение

Как показали проведённые исследования, немаловажное значение «электронный нос» может иметь при оценке свежести пищевой продукции, тем более что на сегодняшний день обонятельные тесты не позволяют решить данную задачу в требуемом объеме.

Калибровочные графики мультисенсорной оценки свежести мышечной и жировой тканей на примере свинины представлены на рис.2 и 3.

Границы показаний сенсоров для соответствующих категорий свежести мясного сырья (мышечной и жировой тканей отдельно) устанавливали согласно результатам принятых физико-химических и органолептического методов исследования. Критерии оценки свежести мяса инструментальным сенсорным методом определяли путём обработки показаний сенсоров методом главных компонент.

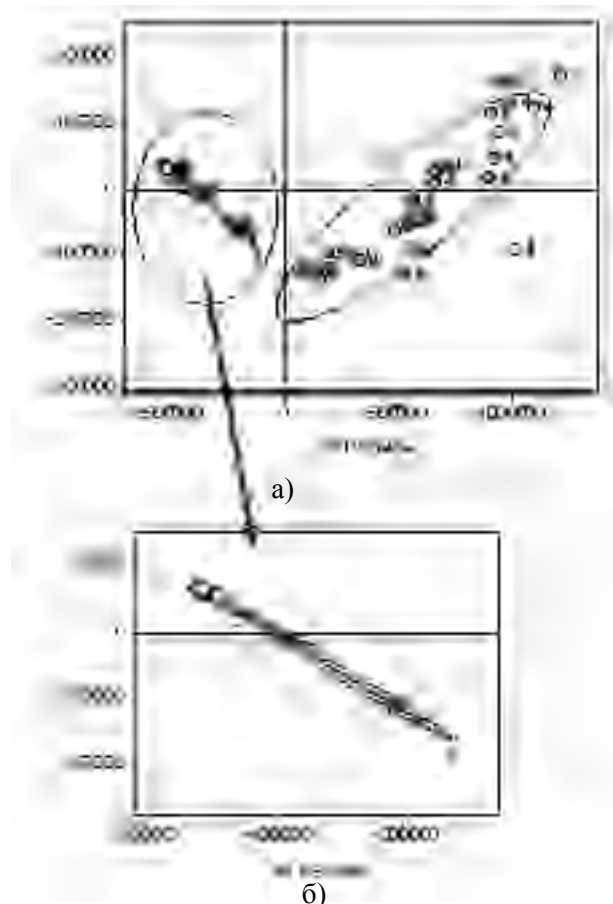


Рис. 2: а) График пространственного расположения точек мультисенсорного анализа мышечной ткани четырёх категорий свежести (1 – свежее мясо, 2 – свежее, не подлежащее длительному хранению, 3- сомнительной свежести, 4 – несвежее); б) Фрагмент графика а) для мышечной ткани трёх категорий свежести (свежее; свежее, не подлежащее длительному хранению; сомнительной свежести).

Slika 2. a) grafikon prostornog razmeštanja tačaka multisenzorne analize mišićnog tkiva četiri kategorije svežosti (1 – sveže meso, 2 – sveže, koje nije dugo skladišteno, 3 – sumnjive svežine, 4 – nije sveže);

b) deo grafikona a) za mišićna tkiva tri kategorije svežine (sveže, sveže koje nije dugo skladišteno, sumnjive svežine)

На рис. 2 видно, что с увеличением количества летучих веществ, образующихся в процессе порчи мяса, увеличивается размер кластера (область расположения точек, характеризующая каждую из категорий свежести сырья). Например, область точек, характеризующая свежие образцы, располагается в четвёртой четверти системы координат и её размеры невелики. Следует отметить, что анализ полученных данных позволил выявить группу точек, значения первой главной компоненты которых имели большие величины, чем область, характеризующая свежее мясо. Таким образом, была идентифицирована категория мяса «свежее, не подлежащее длительному хранению». Кластер, характеризующий образцы «сомнительной свежести», имеет большие размеры и располагается в третьей четверти системы координат. Кластер, характеризующий несвежие образцы, имеет наибольшие размеры и располагается в первой и второй четвертях системы координат.

Следует отметить, что с накоплением продуктов порчи мяса увеличивается значение первой главной компоненты.

На рис. 3 видно, что расположение кластеров, свойственных жировой ткани, аналогично расположению кластеров мышечной ткани соответствующих категорий свежести.

Мясо различных видов животных характеризуется специфическим запахом, но в большинстве случаев идентифицировать видовую принадлежность мясного сырья органолептическим методом сложно. В отличие от существующих на сегодняшний день методов определения вида мяса (метод полимеразной цепной реакции, иммуно-ферментного анализа и др.), использование мультисенсорных систем не требует высоких затрат материалов, а также длительной и трудоёмкой подготовки проб.

Проведение сравнительного анализа показаний сенсоров прибора «VOCmeter», полученных при исследовании летучих компонентов говядины, свинины, баранины, мяса кур, страуса и индейки, и обработка их методом главных компонент, позволили получить график, представленный на рис.4.

На рисунке видно, что области точек, характеризующие каждый вид мяса, объединены в кластеры, свойственные каждому из видов мясного сырья. На основании комплекса проведённых работ также установлена возможность применения мультисенсорных инструментальных систем для определения видовой принадлежности мясного сырья. Применение сен-

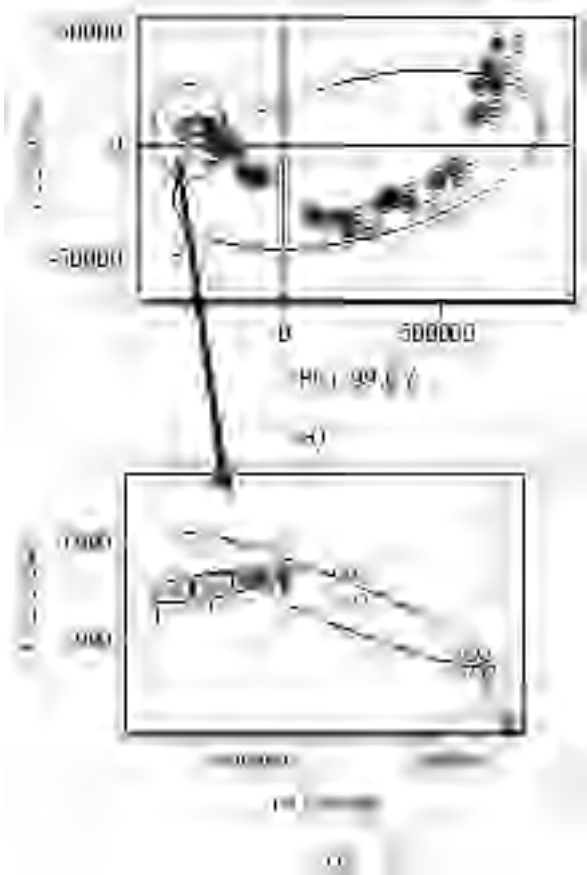


Рис. 3: а) График пространственного расположения точек мультисенсорного анализа жировой ткани трёх категорий свежести (1 – свежее мясо, 2 – сомнительной свежести, 3 – несвежее); б) Фрагмент графика а) для жировой ткани двух категорий свежести (свежее и сомнительной свежести).

Slika 3. а) grafikon prostornog rasporeda tački multisenzorne analize živog tkiva tri kategorije svežine (1 – sveže meso, 2- sumnjive svežine, 3 – koje nije sveže) б) deo grafikona а) za masno tkivo dve kategorije svežine (sveže i sumnjive svežine)

сорных инструментальных методов позволит проводить оперативную идентификацию мясного сырья при входном контроле на предприятиях, таможнях, рынках и т.д.

Инструментальный метод позволяет также проводить дифференциацию охлаждённого и размороженного мясного сырья. Например, на рис. 5 приведены данные показаний сенсоров исследования охлаждённого (0) и размороженного мяса (2), обработанные методом главных компонент. Следует отметить, что кластеры, характеризующие охлаждённое и размороженное мясное сырьё, располагаются в различных

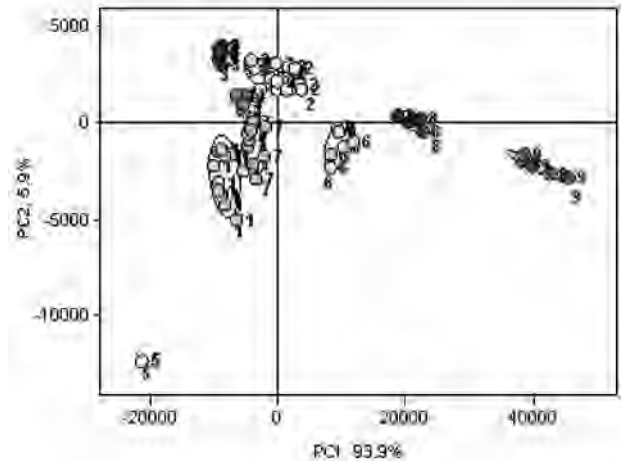


Рис. 4. Пространственное расположение точек мультисенсорного анализа мясного сырья различного вида (1 – свинина, 2 – говядина, 3 – мясо кур, 4 – баранина, 5 – телятина, 6 – мясо страуса, 7 - мясо индейки, 8 – мясо кролика, 9 – оленина).

Slika 4. prostorni raspored tačaka multisenzorne analize mesa razlišitih vrsta (1 – svinjetina, 2 – govedina, 3 – meso kokošaka, 4 – ovčetina, 5 – teletina, 6 – meso nojeva, 7 – meso ćuraka, 8 – meso zečeva i 9 – meso jelena)

четвертях системы координат на значительном расстоянии друг от друга, что обусловлено изменением белковой системы мышечной ткани в процессе замораживания.

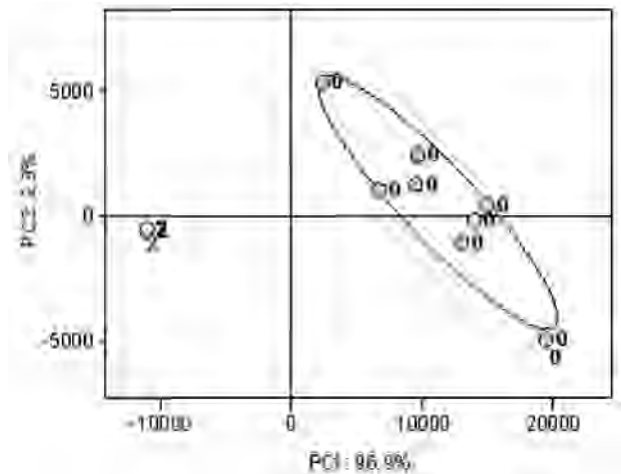


Рис. 5. Пространственное расположение точек мультисенсорного анализа охлаждённого (0) и размороженного мясного сырья (свинина и говядина).

Slika 5. Prostorni raspored tačaka multisenzorne analize ohlađenog (0) i odmrznutog (2) mesa (svinja i goveda)

Большой научный и практический интерес представляет изучение возможности использования мультисенсорных аналитических систем для анализа аромата готовой продукции. С этой целью на приборе «VOCmeter» провели анализ летучих компонентов запеченного мяса (свинина), полученного из охлаждённого (обр. А и В) и размороженного (обр. С) мясного сырья.

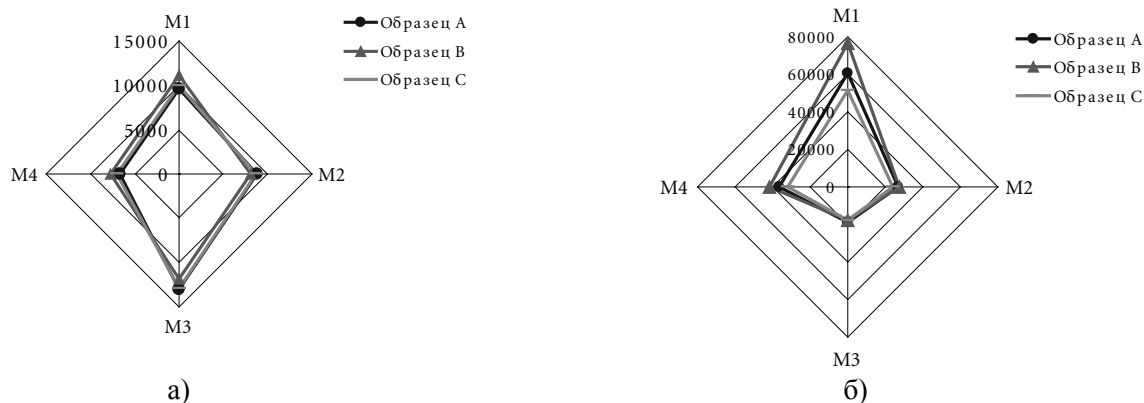


Рис. 6. „Визуальные отпечатки“ мультисенсорного анализа образцов мясного сырья (а) и запеченного мяса (б).

Slika 6. „Vizuelni otisci“ multisenzorne analize uzoraka sirovog mesa (a) i pečenog mesa (b)

На рис.6а видно, что формы и площади «визуальных отпечатков» охлаждённого и размороженного мясного сырья не имеют существенных отличий ($S_{vo}(\text{обр.А})=17,5 \cdot 10^7$; $S_{vo}(\text{обр.В})=18,2 \cdot 10^7$; $S_{vo}(\text{обр.С})=17,9 \cdot 10^7$) и являются характерными для данного вида мяса (свинина).

Известно, что при термической обработке мяса его компоненты (аминокислоты, углеводы и др.) вступают в различного рода превращения, давая новую гамму веществ и соединений, формирующую аромат мясных изделий. Усло-

На рис.6б представлены «визуальные отпечатки» исследования образцов запеченного мяса. Следует отметить, что после термической обработки образцов полученные «отпечатки» характеризуются большими площадями по сравнению с «отпечатками» исходного мясного сырья. При этом площадь «отпечатка» готового продукта, полученного из охлаждённого сырья,

на $56 \cdot 10^7 \div 136 \cdot 10^7$ единиц больше «отпечатка» образца из размороженного мяса, что обусловлено, по-видимому, различной реакционной способностью веществ-предшественников аромата в мясном сырье.

В табл.1 приведены сравнительные данные органолептического анализа, полученные по результатам оценки образцов дегустационной комиссией по 9-балльной шкале, и площадей «визуальных отпечатков» сенсоров прибора «VOCmeter».

Таблица 1. Результаты оценки запаха образцов запеченного мяса органолептическим и инструментальными методами

Tabela 1. Rezultati ocene mirisa uzoraka pečenog mesa organoleptičkim i instrumentalnim metodama

Результаты оценки	Образец А	Образец В	Образец С
Средний балл	7,20±0,05	8,70±0,06	5,80±0,05
Площадь «визуального отпечатка», S_{vo}	249,4*10 ⁷	329,5*10 ⁷	193,0*10 ⁷

вия кулинарной обработки, вид используемого сырья оказывают существенное влияние на конечные результаты превращений, т.е. состав ароматобразующих сложных смесей веществ, определяющих специфический запах готового продукта.

На основе данных сравнительного анализа пока зана прямая зависимость изменения площади «визуального отпечатка» и балльной органолептической оценки запаха. Полученные результаты подтвердили возможность применения мультисенсорных систем для инструментальной

оценки запаха готовой продукции с целью повышения объективности анализа.

Мультисенсорные системы также могут быть успешно использованы для оценки аромата мясных продуктов при совершенствовании технологий, разработке рецептур с использованием различных видов сырья, ароматизаторов, пряностей и т.д. В качестве примера на рис.7 представлены «визуальные отпечатки» показаний сенсоров, полученные при исследовании говядины и свинины после варки.

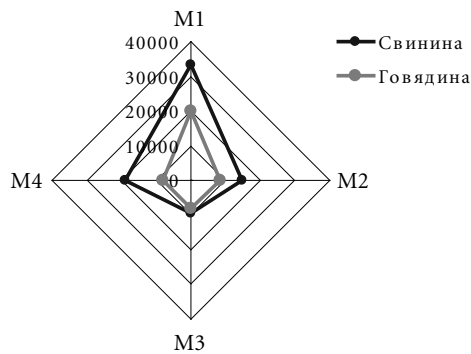


Рис.7. „Визуальные отпечатки“ мультисенсорного анализа образцов говядины и свинины после варки.

Slika 7. „Vizuelni otisak“ multisenzorne analize uzoraka govedine i svinjetine posle kuvanja

Сравнительный анализ «визуальных отпечатков», приведённых на рис.7, позволил установить аналогичную динамику накопления ароматобразующих веществ в говядине и свинине, однако свинина обладала большей интенсивностью аромата, чем говядина, при тех же режимах термической обработки мясного сырья.

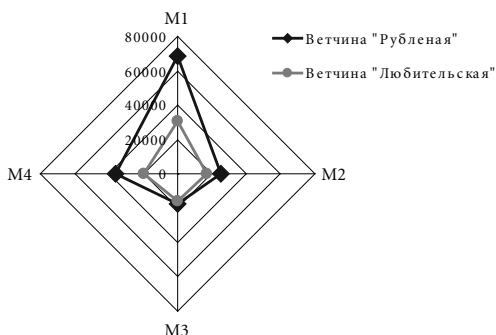


Рис.8. „Визуальные отпечатки“ мультисенсорного анализа консервов ветчинных, выработанных из свинины (ветчина «Рубленая») и говядины (ветчина «Любительская»).

Slika 8. „Vizuelni otisak“ multisenzorne analize šunke u konzervi izrađene od svinjskog mesa (šunka „isečena“ i govedina) (šunka „Любительская“)

Таблица 2. Площади «визуальных отпечатков» ветчинных консервов

Tabela 2. Površina „vizuelnih otisaka“ konzervi šunke

Результаты оценки	Ветчина «Рубленая»	Ветчина «Любительская»
Площадь «визуального отпечатка», S _{во}	263,3*10 ⁷	86,2*10 ⁷

На рис.8 и табл.2 приведены результаты инструментального исследования разработанных во ВНИИМП ветчинных консервов, выработанных из различных видов мясного сырья (Чернуха и соавт. 2008).

В результате проведённых исследований установлено, что площадь «визуального отпечатка» ветчины «Рубленая» из свинины больше «визуального отпечатка» ветчины «Любительская» из говядины (табл.2).

Полученные различия площадей «визуальных отпечатков», характеризующих интенсивность аромата мяса после варки (рис. 7) и консервов (рис.8) из говядины и свинины, обусловлены различным содержанием компонентов мясного сырья, являющихся предшественниками ароматобразующих соединений.

Выводы

Полученные результаты органолептического и инструментального исследования мяса подтвердили перспективность использования мультисенсорных систем для анализа качества продукции мясной промышленности, в том числе для оценки:

- свежести мясного сырья;
- идентификации видовой принадлежности мяса;
- запаха и аромата мяса и мясопродуктов.

В дальнейшем в институте планируется проведение исследований по разработке методик объективного анализа интенсивности аромата готовой продукции на приборе «VOCmeter», а также методик идентификации посторонних запахов и фальсификации мясного сырья. Сенсорные системы займут достойное место среди аналитических методов исследования качества пищевой продукции.

Библиография

Анисимкин В.И., Верона Э., Земляков В.Е., Крышталь Р.Г., Медведь А.В., 1998. Интегральная решетка датчиков для анализа многокомпонентных газовых смесей. Письма в ЖТФ, том 24, №16;

Грень А.И., Высоцкая Л.Е., Михайлова Т.В., 1985. Химия вкуса и запаха мясных продуктов. Киев, Наук. Думка, 100 с;

Чернуха И.М., Кузнецова Т.Г., Селиванова Е.Б., Иванкин А.Н., 2008. Исследование возможностей исполь-

зования прибора «VOCmeter» для оценки свежести мяса. Мясная индустрия, №3, с.49-51;

Чернуха И.М., Сметанина Л.Б., Захаров А.Н., Анисимова И.Г., Воробьева О.В., 2008. Современные аспекты технологий ветчинных консервов. 11-я Международная научно-практическая конференция памяти В.М Горбатова. Сборник докладов, с.163–168.

Paper recieved: 6.05.2009.

Činioci od značaja za održivost ribe i odabranih proizvoda od ribe u prometu*

Baltić Ž. M., Kilibarda Nataša, Dimitrijević Mirjana

Sadržaj: Riba je jedna od najvrednijih namirnica životinjskog porekla u ishrani ljudi. Ulov ribe u svetu je dostigao svoj maksimum krajem prošlog veka. Međutim, tržište se rastućim potrebama za ribom podmiruje proizvodnjom ribe u akvakulturi. U prometu se više od 50 posto ribe nalazi kao sveža riba, manje od jedne četvrtine kao zamrznuta riba a približno ista količina ribe (oko 11 posto) u prometu se nalazi kao konzerva od ribe i kao dimljena riba. Kako je riba lako kvarljiva namirnica načinu njenog stavljanja u promet i činionicima od značaja za njenu održivost u prometu posvećuje se posebna pažnja.

Cljučne reči: riba, promet, održivost

FACTORS SIGNIFICANT FOR THE SHELF-LIFE OF FISH AND SELECTED FISH PRODUCTS IN RETAIL

Abstract: Fish is one of most nutritive valuable food of animal origin in human nutrition. World fish landing reached its own maximum at the end of the past century. However, increasing market needs for food products are compensated with fish production in aquacultures. Over 50% of fish market is covered with fresh fish, less than one quarter is frozen fish and approximately 11% of market is covered with canned and smoked fish. Fish is very perishable food, so modalities of trading with fish and fish products and aspects of shelf life are things of major importance in fishing industry.

Key words: fish, trade, shelf-life

Uvod

Riba je, nema sumnje, veoma cenjena i tražena hrana na tržištu. Tržište se ribom snabdeva iz dva izvora, odnosno, ribom koja se izlovljava iz prirodnih resursa (okeani, mora, jezera i reke) i ribom koja se gaji u akvakulturi. Vodena sredina se odlikuje raznovrsnošću živog sveta i smatra se da samo riba ima oko 30.000 vrsta. Ekonomski i komercijalni značaj za ishranu ljudi ima, međutim, svega oko 65 vrsta riba. Od toga najveći broj vrsta u prometu se nalazi kao sveža i zamrznuta riba, a manja količina ribe se koristi za izradu proizvoda od ribe. Na našem tržištu nalazi se živa riba, na različite načine obrađena sveža i zamrznuta riba (može i neobrađena) kao i proizvodi od riba, uglavnom konzerve od ribe, a retko dimljena i soljena riba.

Riba u ishrani ljudi

Riba u ishrani ljudi ima veliki značaj i njena potrošnja naročito se povećala od 1995. godine, kada je svet počeo da shvata značaj hranljive vrednosti ribe. Razlozi povećane potrošnje ove namirnice su saznanja da je meso ribe u mnogo manjoj meri uzrok zoonoza, u odnosu na meso stoke za klanje, zatim da je značajno manje opterećeno različitim aditivima koji se u savremenoj proizvodnji koriste u svinjarstvu i živinarstvu. Stoga, meso ribe predstavlja značajan, a u mnogim zemljama sveta i dominantan izvor proteina (od 15–24 posto) Procenjuje se da se blizu 15 posto potreba za životinjskim proteinima u svetu podmiruje potrošnjom ribe (Baltić i Tadić, 2001). U mesu ribe, ukupna količina aminokiselina proteina ne razlikuje se značajno od aminokiselina proteina mesa stoke za klanje. Mišići ribe sadrže manje vezivnog tkiva od mišića stoke za klanje,

*Plenary paper on International 55th Meat Industry Conference held from June 15-17th 2009 on Tara mountain

*Plenarno predavanje na Međunarodnom 55. savetovanju industrije mesa, održanom 15-17. juna 2009. na Tari

AUTORI: Milan Baltić, baltic@vet.bg.ac.rs, Mirjana Dimitrijević, Fakultet Veterinarske medicine, Univerzitet u Beogradu, Bulevar Oslobođenja 18, Beograd, Srbija; Nataša Kilibarda, Veterinarski specijalistički institut „Subotica“, Segedinski put 88, Subotica, Srbija;

AUTHORS: Milan Baltić, baltic@bg.ac.rs, Mirjana Dimitrijevic, Faculty of Veterinary Medicine, Univerzity in Belgrade, Bulevar Oslobođenja 18, Belgrade, Serbia; Natasa Kilibarda, Veterinary specialize institute “Subotica”, Segedinski put 88, Subotica, Serbia;

pa se, samim tim, meso ribe brže i lakše resorbuje, odnosno ima visok koeficijent svarljivosti (Baltić i Teodorović, 1997). Stručnjaci, naročito, preporučuju korišćenje ribe i plodova voda u ishrani ljudi zbog povoljanog sadržaja proteina, minerala, vitamina, a posebno esencijalnih masnih kiselina u mesu ribe, za koje je dokazano da pogoduju u prevenciji mnogobrojnih oboljenja (Kilibarda Nataša, 2006; Connor, 2000). Zbog velikog značaja polinezasićenih masnih kiselina n-3 klase, u Evropi su date i preporuke o optimalnom dnevnom unosu. Stručnjaci u Velikoj Britaniji predlažu da unošenje pojedinih masnih kiselina bude od 200 mg do 1250 mg dnevno. U Danskoj preporučeni unos je 300 mg dnevno, dok u Nemačkoj, optimalni unos polinezasićenih masnih kiselina iznosi 1500 mg dnevno (Mason Pamela, 2000).

Ulov i proizvodnja ribe

Tragovi iz istorije ljudskog roda ukazuju na to da su ribu u ishrani koristili već odavnina. Ribolovom je čovek lako i jednostavno dolazio do hrane. Lov ostalih životinjskih vrsta, sisara, zahtevao je više okretности, umešnosti i lukavstva, a uz to bio je i znatno opasniji. Još u kamenom dobu čovek se bavio ribolovom, odnosno koristio je različite vrste udica za ribolov. Vremenom se tehnika ribolova poboljšavala, pa su u bakarnom i gvozdenu dobu korišćeni, pored udica, i drugi ribarski alati (mreže, harpuni). Ribolov je nastao u različitim vremenskim periodima u različitim krajevima sveta. U Mesopotamiji ribolovom su se bavili 5000 godina pre nove ere. Gajenje riba u akvakulturi bilo je poznato u Asiriji 2000 godina pre Hrista. I Kinezi su gajili ribu u akvakulturi pre rođenja Hrista. Stanovnici Lepenskog vira takođe su koristili ribu u ishrani. Kostur ribe pronađen pri arheološkim iskopavanjima na ovom lokalitetu govori o tome da su stanovnici Lepenskog vira izlovljavali ribu čija je masa bila oko 200 kilograma. Stari Grci su bili dobri poznavaoци ribe i ribolova, a Aristotel se i naučno bavio proučavanjem riba i ribolova, posebno tuna (Baltić i Teodorović, 1997; Kilibarda, 2006; Chazistefanou, 2008).

Riba je oduvek bila posebno cenjena u zemljama koje su imale izlaz na more, a ako su uz to postojali i oskudni uslovi za razvoj poljoprivrede, tada je razumljiv i značaj ribarstva za te zemlje. Ulov ribe u svetu u 20. veku porastao je od početka veka skoro za dvadeset puta. Naime, 1900. godine ulov ribe u svetu bio je oko pet miliona tona, da bi na kraju 20. veka bio blizu 100 miliona tona. Ovaj obim ulova nije ostao bez posledica, odnosno ugrozio je opstanak najčešće lovljenih vrsta. Ukupan ulov ribe početkom 21. veka dostigao je svoj maksimum od 95,61 milion tona (2000. godine) i od tada se nije povećavao. Prosečan ulov ribe od 2000. do 2005.

godine bio je 93,31 milion tona. Najveći ulov ribe i plodova voda u svetu ostvaruje u poslednjih pet godina Kina (2000.–2005. godina) i on iznosi 16,60 miliona tona. Među deset zemalja sa najvećim ulovom ribe i plodova voda u svetu su pored Kine, Peru, SAD, Japan, Indonezija, Čile, Indija, Ruska Federacija, Tajland i Norveška (Kilibarda i sar., 2008).

Poslednjih godina proizvodnja ribe u akvakulturi ima prosečni godišnji porast između 9 i 10 posto. Toliko povećavanje proizvodnje nema nijedna grana stočarstva. Ima mišljenja da će za 30 do 40 godina proizvodnja ribe u akvakulturi, zajedno sa ulovom ribe iz prirodnih resursa, biti, po količini, ista kao što je to proizvodnja mesa stoke za klanje. Akvakultura je jedini način da se zadovolje rastuće potrebe za ribom. Ulov ribe, od 1950. do 2000. godine, se stalno povećavao, a od tada stagnira, dok proizvodnja plodova voda u akvakulturi stalno raste. Proizvodnja ribe u akvakulturi nije se znatnije menjala od 1950. do 1980. godine. Od 1980. do 2005. godine proizvodnja ribe u akvakulturi povećala se za više od deset puta, tako da je 2005. godine bila oko 48 miliona tona. U akvakulturi, se najčešće, gaje šaranske vrste riba (tostolobik, šaran i amur), (Kilibarda i sar., 2008; Mitrović- Tutundžić i Baltić, 2000). U ukupnoj proizvodnji ribe i plodova voda 1950. godine bilo je najveće učešće mekušaca (46,53 posto) a zatim slatkovodne ribe (41,72 posto). Posle 30 godina odnosno, 1980. godine u proizvodnji plodova voda slatkovodna riba učestvovala je sa 44,61 posto, a mekušci sa 39,11 posto. Učešće slatkovodne ribe proizvedene u akvakulturi se i dalje povećavalo, tako da je 2005. godine, u ukupnoj proizvodnji imala udeo od 54,03 posto. Proizvodnja mekušaca je 2005. godine bila 28,19 posto. Riba u akvakulturi može da se proizvodi u slatkim, morskim i bočatnim vodama. Proizvodnja ribe najveća je u slatkim vodama i ona je 2005. godine iznosila 57,52 posto od ukupne proizvodnje ribe u akvakulturi. Učešće proizvodnje ribe u morskim vodama, u akvakulturi bilo je 34,72 posto, a učešće proizvodnje ribe u bočatnim vodama 2005. godine bilo je 7,76 posto (Kilibarda i sar., 2008).

Namena ulovljene i proizvedene ribe

Ulovljena riba kao i riba proizvedena u akvakulturi iskorišćava se na različite načine, što zavisi od mnogobrojnih činilaca (vrste ribe, obima ulova različitih vrsta, mogućnosti prerade, zahteva tržišta i drugog). Najosnovnija podela ribe, po nameni, zasniva se na tome da li je ulovljena, odnosno proizvedena riba namenjena za ishranu ljudi ili se koristi u druge svrhe. Od ukupno ulovljene i proizvedene ribe od 2000. do 2005. godine za ishranu ljudi koristilo se od 97 037 do 108 009 miliona tona, ili od 74,00 posto do 76,40 posto. Za ostale svrhe koristilo se

od 30 824 do 34 675 miliona tona ribe, ili od 22,40 posto do 26,00 posto. Riba namenjena ishrani ljudi, najčešće, se koristi kao sveža riba (više od 50 posto), a nešto manje od jedne četvrtine se stavlja u promet kao zamrznuta riba. Približno ista količina ribe (od 10 do 11 posto) koristi se za proizvodnju konzervi, odnosno za druge vidove konzervisanja (dimljena, soljena i sušena riba). Riba koja nije namenjena za ishranu ljudi, uglavnom, se koristi za proizvodnju ribljeg brašna (od 70,40 posto do 82,00 posto), ali i za druge svrhe (ishrana riba u akvakulturi, ishrana pasa i drugih karnivora, tehničko ulje, đubrenje zemljišta, galanterija i drugo) (*Mirilović i sar.*, 2008). Od ukupno ulovljene i proizvedene ribe u svetu, od 2000. do 2005. godine, između 36,6 i 44,4 posto bilo je namenjeno izvozu, a ostali veći deo je bio je namenjen domaćoj (sopstvenoj) potrošnji. Najveći uvoznici ribe su Japan i SAD, čija je vrednost uvezene ribe za 2005. godinu iznosila skoro 12 milijardi dolara. U svetu je 18 zemalja sa vrednošću uvezene ribe većom od milijardu dolara. Najveći izvoz ribe u svetu ostvaruje Kina, koja je 2005. godine izvezla ribe u vrednosti od 7,5 milijardi dolara. U svetu su još 23 zemlje čija je vrednost izvoza 2005. godine bila veća od milijardu dolara. Za pojedine zemlje ribarstvo je značajna privredna grana. O tome govori podatak o učešću ribarstva u ukupnom izvozu ribe, kao posebno vrednog proizvoda. Tako, 99,1 posto od ukupne vrednosti poljoprivredne proizvodnje Maldiva čini riba. Izvoz ribe sa Islanda u vrednosti ukupnog izvoza poljoprivrednih proizvoda, učestvuje sa 94,9 posto. U Norveškoj je taj procenat nešto manji (87,50 posto). Zbog velike potražnje mnoge zemlje su i značajni uvoznici ribe. U Japanu, od vrednosti uvoza ukupnih poljoprivrednih proizvoda riba učestvuje sa više od jedne petine (21,20 posto). Riba u ukupnoj vrednosti uvoza poljoprivrednih proizvoda značajnog udela ima i u Portugaliji, Koreji, Švedskoj, Hong Kongu, SAD itd. Srbija uvozi znatne količine ribe; tako je vrednost uvoza bila, u proseku, za period od 2001. do 2006. blizu 40 miliona dolara, a obim, u proseku 24,4 hiljade tona (*Radosavljević i sar.*, 2008).

Potrošnja ribe

Prosečna godišnja potrošnja ribe u svetu, od 2003. do 2005. godine, bila je 16,4 kilograma po stanovniku. Prosečna potrošnja ribe, u istom periodu, u zemljama u tranziciji, bila je 10,8 kilograma, a u industrijski razvijenim zemljama 29,5 kilograma. Posmatrano po regionima, najveća potrošnja ribe je u Okeaniji i iznosi 22,3 kg, zatim u Evropi, sa 20,2 kg i u Severnoj Americi, sa 17,9 kg po stanovniku go-

dišnje. Najveći svetski potrošač ribe je ostrvska država Maldivi, sa potrošnjom od 202,3 kg po stanovniku, zatim slede, takođe, ostrvske države Island (91,0 kilograma), Grenland (85,0 kilograma) i Farska ostrva (87,0 kilograma). Prosečna godišnja potrošnja ribe u zemljama EU (EU-15), u navedenom periodu, bila je 25,7 kilograma. Od zemalja Evropske unije najmanju potrošnju ribe ima Austrija (11,0 kg), a najveću Portugalija (57,0 kilograma). Prosečna godišnja potrošnja ribe po stanovniku u novoprimitivnim zemljama Evropske unije (EU-12) je 8,4 kilograma. Od ovih zemalja najmanja potrošnja ribe je u Rumuniji (3,5 kg), a najveća u Litvaniji (41,0 kilograma). Od evropskih zemalja izvan Evropske Unije prosečna godišnja potrošnja po stanovniku u Švajcarskoj je 15,0 kilograma, a u Norveškoj 49,0 kg. Od zemalja bivših članica SFRJ najmanju potrošnju ima Srbija (više od 5,0 kilograma), a najveću Hrvatska (13,2 kilograma). U Ruskoj Federaciji prosečna potrošnja ribe po stanovniku je 17,3 kilograma. U zemlji sa najvećim ulovom i proizvodnjom ribe u akvakulturi u svetu, Kini, prosečna godišnja potrošnja ribe po stanovniku je 26,0 kilograma. U Africi prosečna godišnja potrošnja ribe po stanovniku je najmanja u Etiopiji (0,2 kilograma), a najveća u Gabonu (37,2 kilograma). Iz navedenih podataka može da se zaključiti da je potrošnja ribe u svetu veoma različita od zemlje do zemlje, što je uslovljeno, pre svega geografskim položajem, tradicijom, ekonomskim razvojem, navikama itd. (*Lekić-Arandelović i sar.*, 2008).

Potrošnja ribe je kod nas prema podacima o ulovu, proizvodnji u akvakulturi i uvozu ribe, nešto veća od 5 kilograma po stanovniku godišnje. U našoj zemlji potrošnja ribe ne zadovoljava se domaćom proizvodnjom, već se i uvozi. Dok proizvodnja i ulov beleže pad poslednjih godina, uvoz drastično raste. Tako je uvoz ribe od 2001. sa 17 hiljada tona porastao na 29 hiljada tona 2006. godine. Riba se u našoj zemlji jede najviše za vreme tradicionalnih praznika i u dane posta. Smatra se da nepoljoprivredna domaćinstva troše 4,1 kilogram ribe, mešovita 3 kilogram a poljoprivredna 2,9 kilograma godišnje, a da se meso ribe koristi u 95,07 posto domaćinstva, dok 57,3 posto domaćinstava koristi ribu jednom nedeljno, a 39,55 posto samo u vreme posta. Razlog relativno niske potrošnje mesa riba kod nas je slaba kupovna moć stanovništva, ali i ograničena i neadekvatna ponuda na tržištu, kao i nedostatak navike korišćenja ribe u ishrani. Asortiman ponude ribe na našem tržištu je ograničen, odnosno, mali broj vrsta riba se nudi potrošaču, koji uvek želi raznovrsnu ribu u ponudi. Kada je u pitanju ponuda morske ribe, na našem tržištu se, od

plave ribe, mogu da nađu skuša, sardela, papalina, haringa, a od bele ribe oslić, šarpina, brancin, zubatac, orada i losos. Kada je u pitanju slatkovodna riba, u ponudi je najzastupljenija riba iz akvakulture, odnosno šaranske i pastrmske vrste riba (šaran, amur, tolstolobik, pastrmka). Ponuda ribe na našem tržištu je neadekvatna. U njoj se, često, može da nađe riba koja je živa ili zamrznuta, što nije povoljno za kupca, jer on traži ribu koja je očišćena, konfekcionirana i delimično pripremljena, ili spremljena za upotrebu. Prodaja žive ribe je najnepovoljniji način ponude za potrošača. Toplovodne ribe se kod nas, uglavnom, prodaju žive u ribarnicama i kao takve nisu pogodno za brzu pripremu (Kilibarda, 2006; Baltić i Teodorović, 1997; Milanović, 2000).

Način obrade ribe za promet

U odnosu na način obrade, riba u promet može da se stavi u različitim oblicima: a) živa i mrtva riba (ona koja nije egzenterirana i očišćena); b) primarno obrađen trup, što podrazumeva trup ribe bez krljušti i unutrašnjih organa); c) obrađen trup, što podrazumeva trup ribe bez krljušti, peraja, unutrašnjih organa i glave; d) naresci od ribe, pod kojim se podrazumevaju delovi obrađenog trupa dobijeni poprečnim sečenjem trupa u delove (naresci); e) fileti od riba, što podrazumeva delove obrađenog trupa ribe, odrezane sa obe strane, od grudnog peraja do repa, paralelno sa kičmenim stubom (Baltić i Teodorović, 1997). Riblji fileti i naresci mogu da se pripremaju od sveže i zamrznute ribe. Fileti ne sadrže kosti. Riblji fileti su komadi mesa odrezani sa obe strane ribe, od grudnog peraja do repa. Najčešće vrste fileta su: a) fileti sa kožom; b) fileti bez kože; c) „leptir“ filet (levi i desni filet spojen sa trbušnim delom kože. Fileti mogu da sadrže i trbušni deo mišića, ali su tada manje kvalitetni (cenjeni). Riblji naresci se dobijaju sečenjem ribe okomito na kičmeni stub. Ako se riba filetira pre *rigor mortis*-a ona kasnije prolazi kroz *rigor mortis*, a kako mišići nisu podupreti skeletom oni lako pucaju. Filetiranje, zbog toga, treba da se uradi tek kada popusti *rigor*. Filetiranje ribe je teško u *rigor-u* i ne preporučuje se, bez obzira da li se izvodi ručno, ili mašinski (Baltić i Teodorović, 1997; Roth i sar., 2006).

Način stavljanja sveže ribe u promet

Riba se u promet stavlja živa (drži se u vodi), ohlađena (poledena), ohlađena upakovana u vakuum ili MAP i zamrznuta. Već je napomenuto da je za potrošača najnepovoljnije stavljanje žive ribe u promet. To se, naročito, odnosi na gradsko sta-

novništvo koje je, inače, u našoj zemlji i najveći potrošač ribe (Dorđević, 2008). Na našem tržištu najzastupljenija je, u prometu, živa i zamrznuta riba (oslić, skuša). Zamrznuta riba je i najčešći predmet uvoza ribe. U svetu je u prometu sveža riba zastupljena sa više od 50 posto od ukupne ponude ribe (Mirilović i sar., 2008). Riba se najčešće stavlja u promet ohlađena (poledena), odnosno izmešana sa ledom u odnosu 1 : 1, ili je količina leda veća od količine ribe, čak do odnosa 2 : 1. Ovaj način stavljanja ribe u promet najčešće se nalazi u velikoprodaji, odnosno specijalizovanim velikoprodajnim centrima namenjenim samo prodaji ribe.

Održivost sveže ohlađene ribe zavisi od mnogobrojnih činilaca (kvaliteta i temperature vode, odnosno njenog bakteriološkog statusa, gladovanja ribe pre izlova, postupaka sa ribom posle izlova, uslova transportovanja, izloženosti stresu, postupka omamljivanja, iskrvarenja, evisceracije, pranja, obrade trupa, i drugo). Održivost sveže slatkovodne ribe, kao što je navedeno, zavisi od mnogobrojnih činilaca, koji mogu da se podele na premortalne i postmortalne. Od premortalnih poseban značaj ima bakteriološki status vode iz koje je riba izlovljena, njen kvalitet i temperatura, odnosno kod šaranske ribe godišnje doba izlova, gladovanje pre izlova, postupaka sa ribom u toku samog izlova i posle izlova (pranje, posebno pranje škrge), načina i dužine transportovanja, poštovanje dobrobiti radi smanjenja stresa, itd. Postupci koji su vezani za obradu ribe, a uključuju omamljivanje, iskrvarenje, uklanjanje krljušti i sluzi, evisceraciju i pranje, a koji se ubrajaju u posmortalne postupke sa ribom, mogu značajno da utiču na bakteriološki status i kontaminaciju ribe, kako bakterijama koje potiču od vodene sredine, odnosno koje riba nosi sa sobom, tako i bakterijama koje nisu karakteristične za ribu, a kontaminiraju je u toku obrade, bilo zbog toga što su prisutne na rukama radnika, opremi, ili površinama sa kojima riba dolazi u kontakt. Stepent kontaminacije ribe u toku obrade može da se smanji poštovanjem principa GMP (Dobra proizvođačka praksa – Good Manufacturing Practice), GHP (Dobra higijenska praksa – Good Hygiene Practice) i SOP (Standardni operativni procesi – Standard Operative Procedure). Od posebnog značaja je da se proces obrade ribe odvija dovoljno brzo i bez nepotrebnog zadržavanja. U stvari, brzinu toka operacija obrade treba definisati kao meru dobre proizvođačke prakse. Ribu posle završene obrade treba odmah polediti, odnosno izmešati sa ledom (najbolje ljuspice leda) dobijenim od vode za piće. Za skladištenje poledene ribe koriste se prostorije u kojima je temperatura 0°C. U literaturi, podaci o održivosti ribe odnose se više na morsku ribu dok su podaci o održivosti pastrmke

i šaranske ribe (šaran, amur, tolostolobik) oskudni (Karabasil i sar., 2005; Dorđević i sar., 2006; Huss, 1995).

Prodaja poleđene ribe u supermarketima nije neuobičajena. Međutim, supermarketi su danas više zainteresovani za prodaju pakovane ohlađene ribe. Kao i kod svih drugih namirnica tako i kod ribe pakovanje ima značajnu ulogu u očuvanju higijenske ispravnosti i kvaliteta ribe, odnosno ima pretežno protektivnu, a manje funkcionalnu ulogu. Pored toga, pakovanje ima ulogu da privuče potrošača, što znači da treba da izgleda dekorativno i atraktivno. Pri tome je od posebnog značaja mogućnost da se preko originalno upakovanog proizvoda, koji je i deklarisan, potrošač bliže upozna sa podacima koji ga informišu o kupljenom proizvodu, da bi ga više zainteresovao. To su podaci o vrsti ribe, ceni, energetske vrednosti, sadržaju masti, uslovima čuvanja, roku održivosti, načinu upotrebe itd. (Cutter Nettles, 2002.; Singh i Heldman, 2001). Upakovana riba pruža potrošaču i veću sigurnost da se radi o ribi koja je zdravstveno bezbedna. Danas se u naučnoj i stručnoj literaturi vrlo često govori o pakovanju ribe. Pri tom, dva su osnovna vida pakovanja sveže ribe: pakovanje ribe vakuumiranjem i pakovanje ribe u modifikovanoj atmosferi gasova (Cutter Nettles Catherine, 2002). Za pakovanje se može reći da je to jedno od najdinamičnijih područja u tehnologiji hrane, što nije iznenađujuće s obzirom na činjenicu da je to u čitavom lancu proizvodnje hrane, a naročito proizvodnje svežeg mesa i sveže ribe, jedna od najkritičnijih tačaka. Naime, način distribucije i postupci sa ribom u toku čuvanja, u maloprodaji pružaju mnogobrojne mogućnosti kontaminacije ribe različitim biološkim opasnostima, naročito bakterijskim, odnosno pružaju mogućnosti za rast i razmnožavanje bakterija, posledično kvaru kao i stvaranju toksina. Vakuum pakovanje hrane ima relativno dugu tradiciju. Pod vakuumiranjem se podrazumeva postupak izvlačenja vazduha, posebno kiseonika iz pakovanja. Na taj način unutar pakovanja nastaju posebni mikroklimatski uslovi koji koče razvoj gram-negativnih bakterija, pa, zbog toga, u vakuumiranim proizvodima preovladavaju gram-pozitivne bakterije, mlečnokiselinske bakterije, laktobacili, pedikoke i *Brochothrix thermospachta*. Pri tome nije isključena mogućnost rasta i drugih bakterija (salmonele, aeromonade, klostridije, jersinije ilisterije), (Dimitrijević, 2007; Kilibarda, 2006). Razume se da vakuumiranje, samo po sebi, nema duži konzervirajući efekat. Taj efekat se postiže skladištenjem pri temperaturama koje dodatno onemogućavaju, odnosno usporavaju rast bakterija. Zbog toga se vakuumirana riba i skladišti pri temperaturama koje ne prelaze +4°C.

Danas se sve češće govori o pakovanju sveže ribe u modifikovanoj atmosferi (MAP). Delovanje ovog pakovanja je slično delovanju vakuuma. Razlika je u tome što se kod vakuum pakovanja unutrašnji mikroklimatski uslovi koji uzrokuju inhibiciju rasta bakterija (stvaranje ugljen-dioksida, pad pH), razvijaju u samom pakovanju u toku skadištenja proizvoda, dok kod pakovanja u MAP, smeša gasova inicira te uslove. MAP može da se definiše kao „način pakovanja pri kome se iz pakovanja uklanja vazduh i zamenjuje jednim ili smešom gasova“. Izbor gasova pri tom uslovljava, uglavnom, vrsta proizvoda koji se pakuje. Izmenom sastava atmosfere unutar pakovanja održivost proizvoda može značajno da se produži. Kod sveže ribe, kao što je to slučaj i kod vakuumiranja, pakovanje u MAP samo po sebi nema značajniji konzervirajući efekat. Zbog toga se i ono kombinuje sa hlađenjem, pa tako i skladištenje sveže ribe upakovane u MAP zahteva temperaturu ne veću od 3°C. Za pakovanje u modifikovanoj atmosferi najčešće se i, uglavnom, koristi smeša ugljen-dioksida, azota i kiseonika u različitim odnosima. Kiseonik stimuliše rast aerobnih, a inhibira rast striktnih anaerobnih bakterija, ugljen-dioksid inhibira rast mikroorganizama u logaritamskoj fazi rasta i produžava im lag fazu rasta, dok azot inhibira rast aerobnih mikroorganizama istiskujući kiseonik u pakovanju. Kiseonik može da ubrza proces oksidacije masti (užeglost), a azot taj proces usporava. U literaturi postoje mnogobrojni podaci o upotrebi pakovanja ribe u MAP i oni se u najvećem broju slučajeva odnose na pakovanje sveže morske ribe. Retko su opisani postupci da se odmrznuta riba, temperirana na temperaturama hlađenja pakuje u MAP i skladišti u prometu hlađenjem. Može se praktično reći da se ova mogućnost samo pominje (Pavlov, 2007; Gonzales-Rodriguez i sar., 2002; Cutter Nettles, 2002). Prema Odredbi Evropske unije o materijalima i predmetima koji dolaze u dodir s hranom koja je stupila na snagu 2004. godine, dopušteno je uvođenje „aktivne“ i „inteligentne“ ambalaže. Pod pojmom „aktivna“ ambalaža definiše se materijal koji je konstruisan na način da otpušta aktivne komponente u hranu, ili ih apsorbuje iz hrane sa ciljem produženja trajnosti ili održavanja ili poboljšavanja uslova pakovanja (Cutter Nettles, 2002).

Pod „inteligentnom“ ambalažom podrazumeva se materijal koji dolazi u dodir sa hranom i koji ujedno ukazuje na stanje upakovane hrane, pa samim tim daje informaciju o svežini, odnosno kvalitetu proizvoda, a da pri tome nije potrebno otvaranje ambalaže da bi se proverio kvalitet. Tipični primeri „inteligentne“ ambalaže sadrže pokazatelje vremena i temperature, a učvršćuju se na površinu ambalaže.

Na isti način mogu da se upotrebe i pokazatelji prisutnosti kiseonika i ugljen-dioksida. Postoje i pokušaji upotrebe pokazatelja razvoja kvarenja proizvoda, koji reaguju sa isparljivim supstancijama nastalim u hemijskim, enzimskim ili mikrobnim reakcijama razgradnje. Takođe, postoji i mogućnost ispitivanja prisustva i kontrolisanja neželjenih mikroorganizama. U ovoj kategoriji ambalaže posebno mesto zauzima „elektronski papir”. Radi se o tehnologiji papirnog tankog displeja, koji bi mogao da se koristi umesto klasičnih nalepnica, u svakoj vrsti pakovanja i ambalaže. Trenutno problem nije u tehnologiji, već u ceni, koja dostiže i 40 dolara po komadiću od nekoliko kvadratnih santimetara (Cutter Nettles, 2002).

Kroz istoriju, ljudi su se trudili da razviju sredstva koja će da obezbede zaštitu hrane od dejstva vremena i uticaja okoline. Značaj pakovanja hrane je u tome što ono obezbeđuje četiri osnovne funkcije. Prva i osnovna funkcija je ta što pakovanje hrane omogućava da se održi integritet hrane u toku procesa proizvodnje, distribucije i prodaje. Zatim, pakovanjem hrane u različite vrste ambalaže, omogućava se njena zaštita od dejstva bioloških, fizičkih i hemijskih opasnosti; zatim zaštita od oksidacije obezbeđuje održavanje originalnih senzornih svojstava hrane tokom čuvanja, odnosno održava se kvalitet namirnica i bezbednost koji su postignuti nekim od procesa konzervisanja, što, sve zajedno, pruža bolju održivost hrane. Zaštita hrane koja se postiže pakovanjem predstavlja, ujedno, i najznačajniju funkciju pakovanja. Ambalaža koja se koristi pri pakovanju hrane ima za cilj i da potrošačima pruži informacije o hrani, zatim podatke o sastavu, hranljivoj vrednosti, poreklu, datumu proizvodnje i roku upotrebe, kao i da se sa jedinstvenih bar kodova može da utvrditi sledljivost upakovane namirnice. Još jedna od funkcija pakovanja hrane je ta što potrošačima olakšava rukovanje namirnicama i nosi sa sobom niz pogodnosti prilikom korišćenja hrane, što se odnosi na veličinu pakovanja, lakoću otvaranja i mogućnost ponovnog zatvaranja (Singh i Heldman, 2001). Prema tržišnim pokazateljima, industrija ambalaže je, trenutno jedan od najbrže rastućih industrijskih sektora, posebno u prehrani. I danas se unapređuju postojeći i pronalaze novi načini pakovanja hrane, a sve radi produženja njene održivosti, zadržavanja originalnih svojstava, poboljšanja kvaliteta, i pre svega, radi proizvodnje hrane koja je bezbedna po zdravlje potrošača. Tome doprinose razvoj zakona i regulativa, koje, u velikoj meri, osiguravaju bezbednost potrošača, što i predstavlja imperativ u proizvodnji hrane (Singh i Heldman, 2001).

Bezbednost sveže ribe u prometu

Sveža riba je, bez sumnje, lako kvarljiva namirnica, što je posledica njenog specifičnog sastava i građe. Kvar ribe je posledica rasta i razvoja mikroorganizama, aktivnosti enzima ribe i promena na mastima. Za mikroorganizme riba može da se kaže da pripadaju dvema osnovnim grupama. Jednu čine bakterije koje su prirodno, ili indirektno prisutne u vodenoj sredini, a označavaju se kao specifične (domaće) i posledica su kontaminacije vode otpadnim materijalom. Primer ove grupe bakterija koje mogu da budu zdravstveni hazard su *Aeromonas hydrophila*, *Clostridium botulinum*, *Vibrio cholerae*, *Vibrio vulnificus* i *Listeria monocytogenes*. Bakterije koje nisu „domaće“ (nisu svojstvene ribi), a značajne su za zdravlje ljudi, uključuju *Enterobacteriaceae*, kao što su to *Salmonella spp. spp.*, *Shigella spp.*, i *Echerichia coli*. Ostale vrste koje mogu da budu značajne za zdravlje ljudi i koje se retko izoluju iz riba su *Edwardsiella tarda*, *Pleisomonas shigelloides* i *Yersinia enterocolitica*. *Staphylococcus aureus* može, takođe, da se pojavi i može da proizvede termorezistentni toksin. Specifične patogene bakterije, kada su prisutne u svežoj ribi, često se nalaze u sasvim malom broju iako je proizvod adekvatno termički obrađen pre upotrebe (jela). U ovom slučaju opasnost po zdravlje ljudi nije značajna. U toku skladištenja (hlađenja ribe) specifične bakterije koje izazivaju kvar ribe umnožavaju se brže od specifičnih patogenih bakterija, tako da se riba pokvari pre nego što postane toksična pa je, kao takvu (pokvarenu), potrošač odbacuje (ne prihvata). Opasnost od patogenih bakterija može da se kontroliše zadovoljavajućom toplotnom obradom koja „ubija“ bakterije, držanjem ribe na niskim temperaturama i sprečavanjem postprocesne kros kontaminacije. *Vibrio* vrste se, uglavnom, susreću u zalivima i priobalju, a njihova brojnost zavisi od dubine vode i nivoa plime i oseke. One su, češće, u toplim vodama i mogu da se nađu tokom letnjih meseci. *Vibrio* vrste su, takođe, i prirodni kontaminanti „bočatnim“ vodama u tropskim predelima i mogu da se nađu u ribama koje se tu gaje u akvakulturi. Opasnost od *Vibrio* vrsta prisutnih u ribi može da se kontroliše toplotnom obradom i preveniranjem kros kontaminacije gotovih proizvoda (proizvoda pripremljenih za jelo). Opasnost po zdravlje ljudi može, takođe, da se umanji brzim hlađenjem posle ulova, što smanjuje mogućnost razmnožavanja ovih bakterija. Neke vrste *Vibrio parahaemolyticus* mogu da budu patogene. Sveža riba može da bude uzrok oboljenja ljudi zbog toga što u njoj mogu da se nađu patogene bakterije (*C. botulinum* tip E i neproteolitički tip B i F, patogene vrste *Vibrio*, *A.*

hydrophila, *Pleisomonas shigelloides*), ili, češće, *Listeria monocytogenes*, *C. botulinum* tip A i B, *C. perfringens*, *Bacillus* vrste ili bakterije čiji su rezervoar ljudi ili životinje (*Salmonella*, *Shigella*, *E. coli*, *Staph. aureus*). Od navedenih bakterijskih vrsta najveću zabrinutost pri pakovanju u anaerobnim uslovima predstavljaju *C. botulinum* tip E i neproteolitički tip B kao i *Listeria monocytogenes*. Riba, pored bakterija i njihovih toksina, može da sadrži i biogene amine, od kojih je najpoznatiji skombrotoksin (histamin) koji je naročito čest kod plave morske ribe (skuša, haringa). Deo opasnosti po zdravlje ljudi mogu da budu i materijali za pakovanje (alergeni). Materijali za pakovanje moraju da imaju ateste da mogu da se koriste za pakovanje hrane. Pri pakovanju ribe nisu isključene ni fizičke opasnosti (metal, plastika, krljušti, kosti kod fileta bez kosti). Bezbednost sveže ribe u prometu bilo da se radi o poleđenoj ili upakovanoj ribi (vakuum, MAP), kao i bezbednost dimljene ribe zavisi od mnogobrojnih faktora (vrste ribe, stepena kontaminacije, načina obrade, temperature skaldištenja itd). Bezbednost ribe u prometu, može da se osigura savremenim principima koji se primenjuju u bezbednosti hrane, što znači poštovanjem i primenom principa dobre proizvođačke prakse (GMP), dobre higijenske prakse (GHP), standardnih operativnih procedura (SOP) i, konačno, uvođenjem, kako u proizvodnju, tako i u promet HACCP koncepta. Osnovni cilj uvođenja HACCP koncepta je stavljanje pod kontrolu bioloških, hemijskih i fizičkih štetnih agenasa koje mogu da budu opasni po zdravlje ljudi (*Karabasil i sar.*, 2005; *Dorđević Vesna i sar.*, 2006; *Dimitrijević*, 2007; *Joffraud i sar.*, 2001; *Huss*, 1995).

Konzervisanje riba zamrzavanjem

Konzervisanje zamrzavanjem je najčešći i najpraktičniji način čuvanja mesa ribe. Ovakav način konzervisanja najmanje utiče na osobine mesa ribe. On se primenjuje, kako za ribu koja je namenjena potrošačima, tako i za ribu koja je namenjena za preradu (*Baltić i Teodorović*, 1997; *Šoša*, 1989). Promene zamrznutog mesa ribe odnose se na promene nastale na proteinima i promene nastale na mastima. Izraženost tih promena (strukturne, fizičke i fizičko-hemijske) zavisi od načina zamrzavanja (brzo ili sporo), visine temperature, dužine trajanja skladištenja, načina odmrzavanja, vrste ribe, stanja ribe pre zamrzavanja, itd (*Šoša*, 1989; *Ward i sar.*, 2000; *Sigurgisladottiri sar.*, 2000). U toku zamrzavanja mesa ribe nastaju promene u prostornoj strukturi proteina i preuređenju veza između proteinskih molekula, što ima za posledicu njihovu

denaturaciju. Denaturisani proteini se slabije rastvaraju i imaju manju sposobnost zadržavanja tečnosti (soka). Proteini miofibrila zamrznute ribe se slabije ekstrahuju. Iz mesa ribe koje je bilo zamrznuto može da se „iscedi“ oko 25 posto vode. Denaturacija proteina zamrzavanjem je spor, ireverzibilan proces, a ispoljava se posle odmrzavanja gubitkom tečnosti („drip“), promenama izgleda, teksture, mirisa i ukusa. Meso postaje „mutno“, drvenasto, vlaknasto i žilavo, a posle kuvanja je suvo. Meso dobija karakterističan ukus („na ustajalu“ ribu). Riba nije podesna za proizvodnju dimljenih proizvoda, zato što posle dimljenja ne poprima gladak i sjajan izgled koji je svojstven za ovu vrstu proizvoda. Ovi negativni efekti zamrzavanja su mnogo izraženiji kod bele ribe koja sadrži malo masti (*Baltić i Teodorović*, 1997; *Šoša*, 1989; *Einen i sar.*, 2002). Promene nastale na mastima vezane su za procese lipolize i oksidacije. Proces lipolize u toku zamrzavanja ne zaustavljaju se u potpunosti, jer enzimi mesa ribe, koncentrisani u nezamrznutoj vodi i pri niskim temperaturama, zadržavaju izvesnu aktivnost. Oksidativnim promenama podležu, prvenstveno, polinezasićene masne kiseline, kojih u mastima riba ima u velikim količinama. Karakterističan oksidativni produkt razgrađivanja polinezasićenih masnih kiselina, malonaldehid, koristi se za praćenje stepena užeglosti masti. Kiseonik iz vazduha, ako površina ribe nije zaštićena glaziranjem ili načinom pakovanja, dolazi u neposredan dodir sa masnim tkivom, koje odmah oksidiše. Ovo je naročito izraženo u površinskim delovima mesa ribe, koji dehidrišu. Kao posledica nastalih promena na mastima, pre svega, oksidacije, javljaju se užegao miris, užegao ukus i žuta boja masnog tkiva. Užegla riba, ponekad, ima miris osoben za uljanu boju (*Baltić i Teodorović*, 1997). Kod zamrznute ribe koja nije zaštićena na odgovarajući način (glazirana, pogodno upakovana), u toku skladištenja nastaje dehidratacija (sublimacija leda) njenih površinskih delova. Stepem nastalih promena zavisi od dužine (trajanja) skladištenja, brzine cirkulacije vazduha, vlažnosti vazduha, fluktuacije temperature, zamrzavanja, itd. Promenjeni delovi mesa su sunderaste strukture, a nastale promene se opisuju terminima „opekotine od zamrzavanja“ („freezburn“) ili „sušenje zamrzavanjem“. Naročito su ove promene izražene na uglovima i rubovima blokova zamrznute ribe. Dehidratacija površinskih delova može da bude izražena u tolikoj meri da u mesu ribe nastaju rupice koje podsećaju na „crvotočinu“ drveta („Hones combin“). Posle odmrzavanja ovi delovi mesa su drvenasti i žilavi, često su užegli, pa meso riba ne može da se preradi. Usled dehidratacije gubitak vode u mesu može biti i do 5 posto (*Baltić i Teodorović*, 1997; *Šoša*, 1989).

Način zamrzavanja mesa ribe

Riba može da se zamrzne u „struji“ hladnog vazduha, u slanom rastvoru ili u „blok“ zamrzivačima (horizontalni ili vertikalni). Bez obzira koji se od ovih postupaka koristi, osnovno je da on obezbeđuje brzo snižavanje temperature, naročito u zoni kristalizacije i da je „spusti“ na temperaturu od -18°C , ili nižu. U zavisnosti od tehničko-tehnološkog rešenja, postupci zamrzavanja ribe mogu da budu kontinualni i diskontinualni. Zamrzavanje mesa ribe u „struji“ hladnog vazduha primenjuje se kod celih (velikih) riba i riba koje su upakovane u sanduke (blokove). Pri zamrzavanju temperatura vazduha je od -38°C do -42°C , cirkulacija vazduha 300–1000 m/min. Za pravilno hlađenje mesa ribe mora da se obezbedi da vazduh kruži oko svih delova ribe (bloka) (Šoša, 1989). Zamrzavanje mesa ribe u „blok“ zamrzivačima (kontaktne ploče) je takav način zamrzavanja pri kome se ribe u bloku (fileti, sitna riba, i drugo) zamrzavaju između dve pomične ploče koje se hlade stalnim protokom rashladnog sredstva. Temperatura kontaktnih ploča je -42°C . Ove ploče mogu da budu u horizontalnom ili vertikalnom položaju. Postoji mogućnost zamrzavanja ribe potapanjem u slani rastvor, glicerol, glikol i propilenglikol koji nisu toksični (Baltić i Teodorović, 1997; Šoša, 1989).

Glaziranje i pakovanje mesa ribe

Kvalitet zamrznute ribe i proizvoda od mesa zamrznute ribe brzo se menja u toku skladištenja i distribuiranja ukoliko se riba na odgovarajući način ne zaštiti od efekata dehidracije, oksidacije, od fizičkih oštećenja i kontaminacije stranim materijama. Površina zamrznute ribe može da se zaštiti glaziranjem (oblaganjem slojem leda), glaziranjem i pakovanjem, ili pakovanjem zamrznute ribe u materijal koji ima svojstva da potpuno prione uz ribu, bez obzira na oblik (pravilnost) bloka ili ribe. Postoje materijali koji imaju ova svojstva (retraktivna), kao i dobra protektivna svojstva (nepropustljivost za gasove i tečnost). Glaziranje se primenjuje prvenstveno, na ribu koja je namenjena za preradu, ili ribu koja je namenjena velikim potrošačima (restorani, vojska, itd), a ređe se primenjuje za ribu namenjenu potrošnji u domaćinstvu. Voda koja se stvara posle otapanja površinskog sloja leda negativno utiče na potrošača, kada je u pitanju prihvatljivost proizvoda. Glaziranje mesa ribe se primenjuje kod blokova ribe i velikih pakovanja, a pakovanje mesa ribe bez glaziranja se koristi za manja pakovanja koja su namenjena domaćinstvima (Baltić i Teodorović, 1997; Šoša, 1989). Kod gla-

zirane ribe difuzija kiseonika iz vazduha je veoma usporena, što znatno usporava oksidaciju masti. Ovo je naročito značajno kada su u pitanju „masne“ ribe (haringa, sardina, skuša, losos, tuna i drugo) koje su podložne oksidaciji. Glaziranje ribe treba da se obavi što je pre moguće, posle zamrzavanja ribe. Ono može da se izvede potapanjem ribe u vodu, ili zalivanjem ribe vodom (sprej). Voda za glaziranje mora da bude higijenski ispravna. Temperatura vode za glaziranje ne sme da bude viša od $+5^{\circ}\text{C}$. Da bi sloj leda bio otporniji na mehaničke udare, ponekad, ako je to dozvoljeno propisom, u vodu za glaziranje mogu da se dodaju aditivi koji nisu štetni po zdravlje ljudi (šećer, skrob, natrijum-aktinat i karboksimetilceluloza). Sloj leda na površini „bloka“ ribe, ili velikih riba, treba da bude podjednake debljine. Ukoliko se želi da sloj leda bude deblji, riba se u vodu potapa više puta uzastopno, ili se riba zaliva više puta. Glaziranje ribe se kontroliše da bi se utvrdila debljina leda. Led na svim površinama treba da bude ujednačene debljine, a količina leda (izražena procentualno) na svim blokovima treba da bude približno ista (Baltić i Teodorović, 1997; Šoša, 1989). Odmah posle završenog glaziranja riba se prenosi u komore (prethodno može da se upakuje u plastičnu i kartonsku ambalažu), koje su namenjene za skladištenje. Prenos ribe u komore za skladištenje treba da se obavi brzo. Ne sme da se dozvoli da se temperatura „podigne“, niti da se ošteti glazura ribe. Oštećenje glazure umanjuje njeno zaštitno delovanje. Ako se riba duže čuva, stanje glazure treba povremeno proveravati zato što se usled isparavanja (sublimacije), ili kondenzacije vodene pare, oštećuje glazura. Ukoliko je glazura oštećena riba ponovo može da se glazira (Baltić i Teodorović, 1997; Šoša, 1989).

Skladištenje zamrznute ribe

Zamrznuta riba se ne stavlja u komore za skladištenje pre nego što se temperatura ribe ne spusti na željeni nivo. Prostor za skladištenje se ne koristi za sam proces zamrzavanja, niti za dalje snižavanje temperature. Ako se pri pretovaru, ili iz nekih drugih razloga riba delimično odmrzne, ona se zamrzava predviđenim postupkom, a zatim se skladišti. Odmrznuta riba ponovo može da se zamrzne samo u slučaju da je namenjena za doradu ili preradu. Na tuni se često, posle pretovara sa broda, pojavljuju znaci površinskog odmrzavanja i njeno ponovno zamrzavanje ne dovodi u pitanje preradu ove ribe u konzerve. Temperatura pri kojoj se riba skladišti, prema našim propisima, ne sme da bude viša od -18°C . U nekim zemljama

se preporučuje temperatura skladištenja od -23°C , -26°C , odnosno -29°C . Temperatura skladištenja je jedan od osnovnih činilaca koji utiče na kvalitet zamrznute ribe. Pri nižim temperaturama usporena je promena parametara kvaliteta ribe. Sa temperaturom zamrzavanja ribe povezana je i vlažnost vazduha. Vazduh, na višim temperaturama može da primi više vlage i da se ne zasiti, tako da je veća i dehidracija ribe na višim temperaturama zamrzavanja. Riba se skladišti tako da se željena temperatura održava stalno. Treba izbegavati fluktuacije temperature, odnosno treba izbegavati promene temperature skladištenja veće od $+2^{\circ}\text{C}$. Fluktuacije temperature negativno utiču na kvalitet ribe (teksturu, pre svega), a i dehidracija mesa pri fluktuaciji temperature je izrazitija. Cirkulacija vazduha pri skladištenju mora da bude umerena i ne veća nego što je potrebno da bi se održala stalna temperatura. Da bi se obezbedilo što ravnomernije kruženje vazduha u svim delovima skladišnog prostora, neophodno je da između zamrznute ribe i zidova, odnosno poda, odnosno tavanice bude 5 do 10 centimetara slobodnog prostora. Ovo je, takođe, neophodno da bi spolja prenetu toplotu apsorbovao vazduh, a ne uskladištena riba. Održivost zamrznute ribe zavisi od velikog broja činilaca (vrsta ribe, način obrade, stanje ribe pre zamrzavanja, način zamrzavanja, „visina“ temperature zamrzavanja, i drugo), (Baltić i Teodorović, 1997; Šoša, 1989; Einen i sar., 2002).

Dimljena riba

Ukupna prosečna proizvodnja dimljene ribe u svetu, za period od 2003. do 2005. godine, bila je 810 798 hiljada tona. U ukupnoj proizvodnji dimljene ribe učešće salmonidnih vrsta bilo je 10,82 posto (8749 hiljada tona), haringe 4,64 posto (37.606 hiljada tona) i ostalih vrsta dimljene ribe 84,54 posto (685.443 hiljada tona). Najveći proizvođač dimljenih salmonidnih vrsta je Francuska sa 23.845 hiljada tona (27,14 posto od ukupne proizvodnje), a zatim slede Nemačka, Danska i Velika Britanija. Kanada je najveći svetski proizvođač dimljene haringe, sa 10 460 tona (27,82 posto od ukupne svetske proizvodnje). Od ostalih vrsta dimljenih riba najveću proizvodnju ima Kina, 268.333 hiljada tona (39,15 posto od svetske proizvodnje). Daleko manju proizvodnju, iza Kine, imaju Tajland, Poljska, Filipini i Indonezija (od 23.000 do 55.000 hiljada tona). Dimljena riba nije tako čest predmet međunarodne trgovine, budući da je najveći proizvođač dimljene ribe, odnosno Kina, praktično ne izvozi. Francuska izvozi oko 10 posto svoje proizvodnje salmonidne ribe; Nemačka, oko jedne trećine; Danska preko 90 posto, a

Velika Britanija 45 posto proizvodnje salmonide dimljene ribe. Kanada izvozi više od 90 posto svoje proizvodnje dimljene haringe. Od ostalih zemalja Indonezija izvozi 5 do 10 posto svoje proizvodnje dimljene ribe, Filipini 3 posto, Tajland 50 posto, a Poljska 10 posto (Popović Ljuba i sar. 2008). U Srbiji, obim proizvodnje dimljene ribe je vrlo mali obzirom na broj objekata koji se bave preradom ribe (oko 10 objekata) i njihove preradne kapacitete, tako da je proizvedena dimljena riba u Srbiji (najčešće hladno dimljena pastrmka) namenjena, uglavnom, specijalizovanim ribljim restoranima (Kilibarda, 2006).

Hladno i toplo dimljena riba sigurno se ubraja među najatraktivnije proizvode od ribe. O tome govori činjenica da od ukupne ponude ribe na francuskom tržištu blizu 20 posto čini dimljena riba. Naročito je zanimljiva hladno dimljena riba, koja, u zavisnosti od vrste, količine soli i visine temperature obrade, može da se koristi bez i sa naknadnom toplotnom obradom. Dimljenje ribe je za naše uslove sigurno jedan od najprihvatljivijih načina prerade ribe, jer ne zahteva skupu opremu, proizvodnja je kratka, a prihvatljivost ovog proizvoda na našem tržištu je, s obzirom na navike (dimljeno svinjsko meso) vrlo dobra. Hladno dimljena riba zahteva zaštitu pakovanjem, bilo vakuumiranjem (danas mnogo češće), bilo pakovanjem u MAP (ređe se primenjuje). Toplo dimljena riba je znatno održivija, ali i ona može da se pakuje i to, uglavnom, vakuumiranjem (Kilibarda, 2006; Espe i sar., 2004). Hladno dimljena pastrmka najčešće se proizvodi od konzumne pastrmke (masa 280- 300 grama), ali i od takozvane lososove pastrmke mase oko jedan kilogram. Prva je uglavnom namenjena dodatnoj toplotnoj obradi na roštilju i često je u ponudi u restoranima, naročito onim specijalizovanim, ribljim. Hladno dimljena lososova pastrmka ne zahteva dodatnu toplotnu obradu, već se jede najčešće, kao predjelo (na primer sa maslacem). S obzirom na način toplotne obrade (hladno dimljenje, temperatura $20-30^{\circ}\text{C}$) za održivost ovih proizvoda neophodno je da su upakovani (vakuum ili MAP) i čuvani pri temperaturama hlađenja (najviše do $+4^{\circ}\text{C}$). U literaturi (Huss i sar., 1995), najčešće, su opisani rezultati ispitivanja održivosti hladno dimljenog lososa kao proizvoda koji je vrlo cenjen i, od dimljene ribe, ima najdužu tradiciju. Ispitivanja održivosti hladno dimljene ribe zasnivaju se na senzornoj oceni (za konzumnju pastrmku toplotnom obradom), hemijskim analizama (ukupni isparljivi azot, trimetilamin, biogeni amini, i etanol) i bakteriološkim analizama (laktobacili, psihrofilne i mezofilne bakterije, enterobakterije, *L. monocytogenes*, *E. coli*, *Salmonella spp.*, i sulfitoredujuće klostridije).

Dobijeni rezultati se međusobno porede i koreliraju (Joffraud i sar., 2006; Leroi i sar., 2001; Dojčinović i sar., 2008; Kilibarda, 2006; Dimitrijević, 2007). Toplo dimljena riba je, sa stanovišta mogućnosti kvara, manje rizična. Kvar bi se ovde mogao vezati, uglavnom, za promene na mastima (užeglost). U toku skladištenja prate se promene senzornih osobina, hemijske osobine (ukupni isparljivi azot, trimetilamin, biogeni amini, malondialdehid, etanol, benzo-a-piren) i bakteriološki status (laktobacili, mezofilne bakterije i sulfitoredujuće klostridije), (Goulas i Kontominas, 2004).

Zaključak

Sa porastom broja stanovnika u svetu i porastom životnog standarda, naročito u zemljama

u razvoju, značajno rastu potrebe za mesom riba i ostalih plodova voda. Ulov ribe iz prirodnih resursa dostigao je svoj maksimum krajem 20. veka tako da dalje povećanje potreba za mesom ribe može da se podmiri gajenjem ribe u akvakulturi. Poslednjih godina proizvodnja ribe u akvakulturi ima prosečan godišnji porast od oko 10 posto, što nema nijedna druga grana stočarstva. Najveći deo ribe se u promet stavlja kao sveža (poledena) riba, zamrznuta riba, kao konzerva od ribe i kao dimljena i sušena riba. Raznovsnošću ponude, naročito pakovanja ribe, da se riba kao namirnica želi što više približiti potrošaču. Zbog toga se u novije vreme pakovanju ribe i produženju održivosti, naročito sveže ribe, posvećuje sve veća pažnja. Održivost ribe u prometu uslovljena je brojnim, često međusobno zavisnim činiocima.

Literatura

- Baltić, M. Ž., Teodorović, V., 1997. Higijena mesa, riba, rakova i školjki, udžbenik, Veterinarski fakultet, Beograd;
- Baltić, M. Ž., Tadić, R., 2001. Proizvodnja i potrošnja mesa riba u svetu i kod nas. Tehnologija mesa, 42, 5-6, 345–357;
- Chatzistefanou Maria, 2008. Ispitivanje obima i strukture ulova i proizvodnje ribe u akvakulturi u Grčkoj na početku 21. veka, Specijalistički rad, Fakultet veterinarske medicine, Univerzitet u Beogradu, 1–89;
- Connor, E. W., 2000. Importance of n-3 fatty acids in health and disease. American Journal of Clinical Nutrition, 71, 171-175;
- Cutter Nettles Catherine, 2002. Microbial Control by Packaging: A Review. Critical Reviews in Food Science and Nutrition, 42 (2), 151–161;
- Dimitrijević Mirjana, 2007. Ispitivanje puteva kontaminacije i preživljavanja različitih sojeva *Listeria monocitogenes* u dimljenom mesu riba, Doktorska disertacija, Fakultet veterinarske medicine, Univerzitet u Beogradu, 1-130.
- Dojčinović, S., Šarić, M., Kilibarda Nataša, Đorđević Vesna, Baltić, M. Ž., 2008. Zbornik radova i kratkih sadržaja, 20. savetovanje veterinara Srbije, Zlatibor, 108–115;
- Đorđević Maja, 2008. Ispitivanje obima i strukture uvoza ribe i proizvoda od ribe u Srbiji od 2001. do 2006. godine, Specijalistički rad, Fakultet veterinarske medicine, Univerzitet u Beogradu, 1–77;
- Đorđević Vesna, Baltić, M. Ž., Kilibarda Nataša, Mitrović Radmila, Karabasil, N., 2006. Evaluation of trout freshness using torrystore. Tehnologija mesa, 47, 1–2, 45–52;
- Einen, O., Guerin, T., Fjaera, S. O., Skjevold, P. O., 2002. Freezing of pre-rigor filets of Atlantic salmon. Aquaculture, 212, 129–140;
- Espe, M., Kiessling, A., Lunestat, B., Torrissen, O., Rora, A. B., 2004. Quality of cold smoked salmon collected in one French hypermarket during a period of 1 year. Lebensmittelwiss. u.-Technol., 37, 627–638;
- Gonzales-Rodríguez, M. N., Sanz, J., Sato, J. A., Otero, A., Garcia-Lopez, M. L., 2002. Numbers and types of microorganisms in vacuum-packed cold-smoked freshwater fish at the retail level. International Journal of Food Microbiology, 77, 161–168;
- Goulas, A. E., Kontominas, M. G., 2004. Effect of salting and smoking-method on the keeping quality of chub mackerel (*Scomber japonicus*): biochemical and sensory attributes. Food Chemistry 93, 511–520;
- Huss, H. H., 1995. Quality and quality changes in fresh fish. FAO Fisheries Technical Paper, Roma;
- Huss, H. H., Karim P., Embarek B., Jeppesen, V. F., 1995. Control of biological hazards in cold smoked salmon production. Food Control, 6, 335–340;
- Joffraud, J. J., Cardinal Mireille, Cornet Josiane, Chasles, J. S., Leon, S., Gigout, F., Leroi F., 2006. Effect of bacterial interactions on the spoilage of cold-smoked salmon. International Journal of Food Microbiology 122, 51–61;
- Joffraud, J. J., Leroi, F., Roy, C., Berdague, J. L., 2001. Characterization of volatile compounds produced by bacteria isolated from the spoilage flora of cold-smoked salmon. International Journal of Food Microbiology, 66, 175–184;
- Karabasil, N., Dimitrijević Mirjana, Teodorović, V., Kilibarda Nataša, Baltić, M. Ž., 2005. Najčešće bakterijske kontaminacije mesa riba, Zbornik predavanja, II Međunarodna konferencija «Ribarstvo», Poljoprivredni fakultet, Beograd;
- Kilibarda Nataša, 2006. Uticaj zamrzavanja na odabrane parametra dimljene pastrmke, Magistraska teza, Fakultet veterinarske medicine, Univerzitet u Beogradu, 1–115;
- Kilibarda Nataša, Baltić Ž. M., Teodorović V., Karabasil N., Dimitrijević Mirjana, 2008. Tama i sjaj ribarstva kao izvora hrane na početku 21. veka, 20. Savetovanje veterinara Srbije, Zbornik radova i kratkih sadržaja, Zlatibor, 34-50;
- Lekić-A-randelović Ivana, Kilibarda Nataša, Dimitrijević Mirjana, Karabasil, N., 2008. Potrošnja ribe u svetu, Evropskoj Uniji i Srbiji, Zbornik radova i kratkih sadržaja, 20. savetovanje veterinara Srbije, Zlatibor, 94–97;
- Leroi, F., Joffraud, J., Chevalier, F., Cardinal Mireille, 2001. Research of quality indices for cold-smoked salmon using a stepwise multiple regression of microbiological counts and physico-chemical parameters. Journal of Applied Microbiology, 90, 578–588;
- Mason Pamela, 2000. Fish oils-an update, The Pharmaceutical Journal, 265, 720–724;
- Milanović, M., 2000. Makroekonomski aspekti ribarstva i nova agrarna politika SR Jugoslavije. Savremeno ribarstvo

- Jugoslavije (Monografija), IV Jugoslovenski simpozijum «Ribarstvo Jugoslavije», 213–223;
- Mirilović, M., Karabasil, N., Teodrović, V., Baltić, M. Ž., Dimitrijević Mirjana, 2008.** Raspored svetske proizvodnje i ulova ribe od 2000. do 2005. godine po obimu. Zbornik radova i kratkih sadržaja, 20. savetovanje veterinarara Srbije, Zlatibor, 98–100;
- Mitrović-Tutundžić Vera, Baltić, M. Ž., 2000.** Stanje slatkovodnog ribarstva u svetu i kod nas i trendovi razvoja. Savremeno ribarstvo Jugoslavije (Monografija), IV Jugoslovenski simpozijum «Ribarstvo Jugoslavije», 1–9;
- Pavlov, A., 2007.** Changes in the meat from aquaculture species during storage at low temperature and attempts for differentiation between thawed-frozen and fresh chilled meat. Bulgarian Journal of Veterinary Medicine, 10, 2, 67–75;
- Popović Ljuba, Kilibarda Nataša, Dimitrijević Mirjana, Dokmanović Marija, Baltić, M. Ž., 2008.** Obim i struktura proizvodnje dimljene ribe u svetu na početku 21. veka. Zbornik radova i kratkih sadržaja, 20. savetovanje veterinarara Srbije, Zlatibor, 104–106;
- Radisavljević Katarina, Tešić, M., Mirilović M., Teodorović, V., Baltić, M. Ž., 2008.** Međunarodni promet ribe i plodova voda na početku 21. veka. Zbornik radova i kratkih sadržaja, 20. savetovanje veterinarara Srbije, Zlatibor, 100–102;
- Roth, B., Slinde, E., Arlidsen, J., 2006.** Pre or post mortem muscle activity in Atlantic salmon (*salmo salar*). The effect on rigor mortis and the physical properties of flash. *Acquaculture*, 257, 504-510;
- Sigurgisladottir, S., Ingvarsdottir, H., Torrissen, O. J., Cardinal Mireille, Hafsteinsson, H., 2000.** Effects of freezing/thawing on the microstructure and the texture of smoked Atlantic salmon (*Salmo salar*). *Food Research International*, 33, 857–865;
- Singh, R. P., Heldman, D. R., 2001.** Introduction to Foods Engineering. Academic Press, London, UK;
- Šoša, B., 1989.** Higijena i tehnologija prerade morske ribe, Školska knjiga, Zagreb;
- Ward, D., Tang, J., Correia, L. R., 2000.** Salt diffusivities and alt diffusion in farmed Atlantic salmon muscle as influenced by rigor mortis. *Journal of Food Engineering*, 43, 115–123;
- Warm, K., Nielsen, J., Hylding G., 2000.** Sensory quality criteria for five fish species. *Journal of Food Quality*, 23 (6), 583–601;

Rad primljen: 23.04.2009.

Zahvalnost

Ovaj rad napisan je u okviru projekta Ministarstva nauke broj 20132, koji finansira Ministarstvo nauke Republike Srbije.



INSTITUT ZA HIGIJENU I TEHNOLOGIJU MESA
INSTITUTE OF MEAT HYGIENE AND TECHNOLOGY



Delatnost Instituta za higijenu i tehnologiju mesa

Naučna i stručna saradnja:

- Stručno-tehnička saradnja u pogonima proizvodnje i prerade mesa;
- Izdavanje sertifikata o autentičnosti za izvoz junećeg mesa na tržište EU;
- Uvođenje HACCP sistema u objekte za klanje, preradu mesa i proizvodnju
- Uvođenje standarda ISO 22 000, 9001, 14 001 u industriju mesa, prehrambenu industriju, ugostiteljske objekte i supermarkete;
- Konsalting u vezi sa propisima zemalja EU, SAD i trećih zemalja koji regulišu klanje stoke i preradu mesa i proizvodnju namirnica životinjskog porekla;
- Tehničko-tehnološko projektovanje i uređenje objekata za klanje stoke i preradu mesa i proizvodnju namirnica životinjskog porekla;
- Određivanje veterinarsko-sanitarnih uslova u objektima za proizvodnju namirnica životinjskog porekla.

U laboratoriji Instituta vrše se ispitivanja namirnica, aditiva, ambalaže, hrane za životinje i predmeta opšte upotrebe. Vrste ispitivanja su:

- Senzorno ocenjivanje;
- Fiziko-hemijska ispitivanja;
- Mikrobiološka ispitivanja;
- Determinacija mikotoksina, rezidua i veterinarskih lekova, toksičnih elemenata, specifičnih proteina i melamina.

Edukacija stručnjaka iz industrije mesa i prehrambene industrije u oblasti bezbednosti hrane.

Institut za higijenu i tehnologiju mesa ima uspostavljen dokumentovan i sertifikovan sistem rada prema zahtevima SRPS ISO 9001:2001.

Laboratorija Instituta je akreditovana prema zahtevima SRPS ISO/IEC 17 025:2006.

Laboratorija Instituta je Nacionalna referentna laboratorija za monitoring program rezidua veterinarskih lekova, koji se prenose iz životne sredine, u organima i tkivima domaćih životinja koje se gaje za ishranu ljudi, primarnim animalnim proizvodima (mleko, jaja i med), farmskoj divljači i ribama —prema programu i uslovima EU.