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LACTOBACILLI IN FOOD HYGIENE

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SUMMARY

In hygiene and technology of foods, almost all the species within the group of lactic acid bacteria have their meaning, as in the positive (fermentation, antimicrobial activity, sensory characteristics of products, probiotic effect), so in the negative sense spoilage bacteria, biogenic amines producers, antimicrobial resistance). Certain species of lactobacilli are "specialized" for use in some fermented dairy or meat products, where they display their positive effect in terms of creating aimed finished products. Except for that, by synthesizing substances which act competitively to other bacteria, lactobacilli improve microbiological stability of products. These mechanisms of microbial inhibition can also be used in conserving "unfermented" food, which significantly prolongs its expiration date.

Key words: *lactobacilli, food, hygiene*

INTRODUCTION

Lactic acid bacteria are constituent part of natural microflora of different food of animal origin. During fermentation they produce metabolites and reductive products which can significantly affect sensory characteristics, but also microbiological safety of products. Regarding microbiological safety, it is known that lactobacilli are those which most often make up the majority of all lactic acid bacteria, and they are competitive towards many spoilage bacteria and pathogens. Thus different authors have reported on antimicrobial effect of lactobacilli toward the bacteria *Listeria monocytogenes*, *Escherichia coli*, *Salmonella spp.*, *Clostridium spp.*, *Staphylococcus aureus* and others (Leroy et al., 2006). Researches have confirmed inhibitory effect toward the listed bacteria under laboratory conditions, but similar results were also got in some of our researches which apply to course of food production or storage, e.g. in fermented sausages, fish or raw meat (Zdolec et al., 2007). Such cognitions nowadays lead to development of an increasing number of the new so- called functional starter cultures comprised of species with known metabolic profile and proved antimicrobial effect.

Antimicrobial activity of lactobacilli is expressed through synthetized organic acids (lactic, acetic), hydrogen peroxide, carbon dioxide, enzymes, reuterine, dyacethil and bacteriocins (Holzapfel et al., 1995). Creating higher quantities of some of the listed substances still isn't desirable in some food, despite the possible microbial activity. It mostly applies to hydrogen peroxide, CO_2, dyacethil or acetic acid in fermented meat products (Kozačinski et al., 2006). The spectrum of their activity will depend on genetic- phenotypic characteristics of species, i.e. on ability of synthetizing products with antimicrobial activity.

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BACTERIA OF LACTOBACILLUS SPECIES

As far as in 1919 Orla-Jensen claimed that the group of lactic acid bacteria is made up of gram-positive, immobile, non-sporogenous bacilli and cocci which ferment carbohydrates and higher alcohols, thus creating lactic acid. As the time passed, new species were found and taxonomic changes were intensive, which was conditioned by a constant progress in analytical methods. In hygiene and technology of food, almost all the species within the group of lactic acid bacteria have their role, as in the positive (fermentation, antimicrobial activity, sensory characteristics of a product, probiotic effect, etc.), so in the negative sense (spoilage bacteria, biogenic amines producers and other). Lactobacilli are often dominant lactic acid bacteria in different foodstuff, thus contributing the development of specific sensory characteristics and affecting the microbiological quality of a product. They can be found in dairy products, cereal products, meat or fish products, water, silage, beer, wine, fruit and fruit juices, marinated vegetables, sauerkraut, waste waters, sour dough; they are part of natural microflora of oral cavity and digestive tract of people and animals; they rarely show pathogen characteristics.

They can be classified to three groups (Stiles and HolzapfeL, 1997), and these are: 1. obligatory homofermentative - they ferment glucose to lactic acid, and they don't ferment pentoses and gluconate. Some of the representatives are *Lactobacillus (L.) acidophilus*, *L. delbrueckii*, *L. helveticus, L. gasseri, L. ruminis*; 2. facultative heterofermentative – they ferment hexoses to lactic acid and can produce gas from gluconate but not from glucose, and they ferment pentoses to lactic and acetic acid. In foodstuff we find facultative fermentative *L. casei, L. plantarum, L. curvatus, L. paracasei, L. pentosus, L. plantarum, L. rhamnosus* and *L. sakei;* and 3. obligatory heterofermentative – they ferment hexoses to lactic acid, acetic acid, ethanol and CO_2. Creating gas from glucose is characteristic for this group of lactobacilli. Some of the representatives are *L. kefir, L. brevis, L. fermentum* and *L. reuteri.*

Bernardeau et al. (2008) indicate that lactobacilli are heterogeneous microbial group which consists of 135 species and 27 subspecies, and their classification keeps changing. By applying modern molecular methods it is suggested that extreme diversity of genome of this bacteria genus can lead to new sub generic classifications. The combination of genotypic and phenotypic tests e.g. DNA based techniques, and conventional tests of fermentation profiles (using carbohydrates) is necessary for determination of species. Pulsed Field Gel Electrophoresis (PFGE) has been successfully applied on species from milk, and at the same time this is the most discriminative and repeatable method. Data from professional literature support the hypothesis that taking in lactobacilli by food isn't risky since lactobacillemia caused by the food, especially by fermented dairy products, is extremely rare and appears only with predisposed patients. Some metabolic characteristics, like creating biogenic amines in fermented products, can lead to nonacceptance of some lactobacilli in food production. A small number of lactobacilli in starter cultures and probiotics can show antimicrobial resistance spread. Still, it can be underestimated since researches of spreading resistance aren't systematic. The authors consider the spread of antimicrobial resistance to be the only relevant reason to be cautious, which demands monitoring of sensitivity to antibiotics because such species could be the carriers of resistant genes and present a potential risk for spreading genes to other bacteria. Still, as a general rule, lactobacilli have high natural resistance which can't be spread, and it is resistance to most antibiotics, especially vancomycin. Safety estimation of lactobacilli which are significant from the technological aspect should be limited to antibiotic profiles and researches of transmissibility of antimicrobial resistance.

ANTIMICROBIAL EFFECT OF LACTOBACILLI BACTERIOCINS

Bacteriocins are antimicrobial substances, peptides or proteins of different antimicrobial spectrum and mode of activity, different molecular masses, genetic and biochemical characteristics, which in the case of bacteriocinogenic lactobacilli damage the membranes of related gram- positive bacteria (Tagg et al., 1976; Jack et al., 1995). There were found among lactobacilli many species which, except for the regular antimicrobial metabolites, synthesize bacteriocins which inhibit the growth of pathogen bacteria from the food, such as *Listeria monocytogenes, Staphylococcus spp., Clostridium spp., Enterococcus spp.* and *Bacillus spp* (Holzapfel et al., 1995). Some of the bacteriocinogenic species of lactobacilli are *L. sakei* (bacteriocin sakacin), *L. curvatus* (curvacin), *L. plantarum* (plantaricin), *L. acidophilus* (lactacin, acidocin) and *L. bavaricus* (bavaricin). Although the researches of bacteriocins and their effect on food safety have been very extensive, up to this day only one bacteriocin (nizin; Federal Register, 1998) is used commercially in milk industry.

ORGANIC ACIDS

It is considered that direct antimicrobial effect of organic acids as metabolitic products of lactobacilli and other bac-

teria of lactic acid appears by their effect on cytoplasmatic membrane by exchanging potentials and inhibiting active transport (De Vuyst and Vandamme, 1994). Lactic acid, as the final product of homofermentative activity of lactobacilli, lowers the pH- value of the substrate (food, nutritional medium, etc.) which destroys or inhibits pathogen bacteria or bacteria of decay. Professional literature especially emphasizes the sensitivity of gram- negative bacteria which are significant for food safety (like *Salmonella* spp. and *Escherichia coli*) on conditions of acidification (Adams and Nicolaides, 1997). Acetic acid shows stronger antimicrobial activity than lactic acid, and it appears in heterofermentative activity of decomposing carbohydrates, e.g. by the activity of species of the *Leuconostoc* genus or heterofermentative lactobacilli (e.g. *L. brevis*). Larger quantities of acetic acid can be expected in fermentation of foodstuff in the presence of autochthonous microflora with predominance of heterofermentative species. Our experiences show that, except for the desirable antimicrobial effect, acetic acid has also the unwanted effect to sensory characteristics of some foodstuff, like fermented sausages, which can be prevented by adding competitive homofermentative lactobacilli (Zdolec, 2007).

HYDROGEN PEROXIDE AND CARBON DIOXIDE

Hydrogen peroxide creates many lactic acid bacteria in the presence of oxygen together with lactate, pyruvate and NADH over the flavine enzymes (Holzapfel et al., 1995). Its antimicrobial effect manifests in the effect of oxidation on membrane lipids and cell proteins, but a real contribution to decreasing the unwanted bacteria in food is questionable because of its natural decomposition by peroxidases (Caplice and Fitzgerald, 1999). Carbon dioxide is created heterofermentatively and contributes to the appearance of anaerobic conditions, by which it inhibits aerobic bacteria of decay in interaction with the cell membrane by changing extracellular and intracellular pH (De Vuyst and Vandamme, 1994).

DYACETHIL

Dyacethil is an antimicrobial product synthesized by some lactococci, pediococci and species of the Leuconostoc genus by metabolizing citrates. Creating dyacethil is disabled by overcoming homofermentative processes, so its antimicrobial effect is small in such conditions. It is believed that gram- negative bacteria, yeasts and molds are more sensitive to dyacethil, and the mode of activity is attributed to interference in exploiting arginine (Caplice and Fitzgerald, 1999).

INSTEAD OF CONCLUSION

Bacteria of lactic acid, and within them bacteria of Lactobacillus genus, show different characteristics which can be used in hygiene- technological sense in production of foodstuff. Certain species of lactobacilli are “specialized” for applying in some fermented dairy or meat products where they express their desirable effect regarding creating the aimed sensory characteristics of finished products. Except for that, by synthetizing substances which act competitively to other bacteria, lactobacilli improve microbiological stability of products. These mechanisms of microbial inhibition can also be used in preserving the “unfermented” food, which can prolong its shelf- life.

ZUSAMMENFASSUNG
LAKTOBAZILLE IN DER NAHRUNGSHYGIENE

In Hygiene und Technologie von Nahrungsmitteln haben alle Sorten innerhalb der milchsäuerlichen Bakterien ihre Bedeutung, sowohl eine positive (Fermentation, antimikrobische Wirkung, sensorische Eigenschaften des Erzeugnisses, probiotischer Effekt) und als auch eine negative ((Blutungsbakterien, Schöpfer der biogenen Amine, antimikrobische Resistenz). Einige Sorten von Laktobazillen sind „spezialisiert“ für die Anwendung in einigen fermentierten Milch- und Fleischerzeugnissen, in denen sie den gewünschten Effekt hinsichtlich Bildung der gezielten sensorischen Eigenschaften des Erzeugnisses zum Ausdruck bringen. Außerdem sythetisieren sie Stoffe, die auf andere Bakterien eine kompatitive Wirkung haben, Laktobazille fördern mikrobiologische Stabilität des Erzeugnisses. Diese Mechanismen der mikrobischen Inhibition können bei der Konservierung der „nicht fermentierten“ Nahrung angewendet werden, womit die Dauerfrist bedeutend verlängert wird.

Schlüsselwörter: *Laktobazille, Nahrung, Hygiene*

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QUALITY AND HEALTH SAFETY OF MEAT CANS

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SUMMARY

The paper shows the results of the chemical, microbiological and research of the heavy metals findings of meat cans produced in two different production plants. The producer A had the HACCP system already implemented, as opposed to the producer B who was just starting to implement the system. 3,7% of all the analyzed samples, products of the producer B, were objected to during the control and storing. The reasons were aberration in the quality of a product (increased quantity of water and fat), separation of fat from the content of a product, appearance of hollows in the content and corrosion on the lid, seams and the cape of the can. There has also been determined an increased quantity of added polyphosphates in products of the producer B, which affects the safety of products. There were no objections on products of the producer A during purchase and storing. All the researched samples of both producers were microbiologically safe. According to the results of heavy metals in cans with chopped meat of both producers, zinc, tin, lead and arsenic were determined.

Key words: *meat cans, quality, health safety*

INTRODUCTION

Department of Quality Control of the Service for Reception and Support of the Ministry of Defense of the Republic of Croatia performs controlling of the meat products' quality at centralized program of supply at the producer (supplier), and according to the Regulation on the mode of material providing of the Croatian Armed Forces (National Gazette 179/04) in accordance with the Instruction of controlling the quality of material means and services in the

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