USAGE OF STARTER CULTURES AS INHIBITORS OF MICROBIOLOGICAL HAZARDS IN FERMENTED MEAT PRODUCTS

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Abstract: A large number of food poisonings occur worldwide due to hygienic, toxicological and other food contaminants. They imply consequences that endanger the health and safety of consumers and, furthermore, result in economic losses. Food sold on the market must meet the basic criteria for good hygienic and manufacturing pracitices, as well as the food safety requirements. Meat and meat products are one of the most common sources of foodborne diseases due to a large number of possibilities for contamination from foodborne pathogens during processing, storage, transportation, etc. In the contemporary industry of meat, besides the usage of preservatives and good hygienic and manufacturing practices, the usage of starter cultures, as inhibitors of microbiological activities, has become increasingly important, as well. The usage of starter cultures contributes to the inhibition of growth and proliferation of pathogenic microorganisms. Lactic acid bacteria (LAB) create a large number of antimicrobial metabolites. The most common, between them, are: organic acids, bacteriocins, diacetates, hydrogen peroxide, carbon dioxide, etc. They create unfavorable conditions for the activity of unwanted microorganisms. Coagulase-negative staphylococci have the ability to reduce the nitrate to nitrite. Nitrites have antimicrobial properties.
Keywords: pathogenic microorganisms, safety, inhibitors, starter cultures, lactic acid bacteria

1. INTRODUCTION

Food spoilage has always been and still is a problem for human beings today. We constantly strive to find ways to preserve food for a long period of time while keeping it safe and healthy.

It is thought that biological procedures for food preservation were first discovered by the ancient Egyptians quite by accident, by spontaneous fermentation due to the action of the microorganisms present in the food itself. Fermentation leads to microbiological stability, improved sensory characteristics and nutritional values of the food.

The reasons for lactic acid fermentation were discovered by Pasteur in 1857, and a year later, he discovered the causes of alcoholic fermentation. Lister succeeded in identifying and isolating pure cultures of LAB in 1870. Commercial production of pure cultures began in a very short time. Pure cultures started to be used in the food industry as early as the beginning of the 19th century.

LAB reduce the pH value by decomposing sugars and creating organic acids, primarily lactic acid. LAB contribute to lower pH and thus the finished product is safe for consumption.

Larrouture [1] point out that LAB also influence the flavour formation in fermented sausages. In fermented meat products, proteolysis is largely attributed to LAB.

Coagulase negative staphylococci have a major role in the development of flavor, taste and color in meat products [2]. They also have the ability to reduce nitrates to nitrites, which contributes to the production of nitrosomyoglobin, which is responsible for the typical red color of meat products. The main characteristic of staphylococci is the creation of lipase which plays an important role in the aroma formation of fermented meat products. *Staphylococus xylosus*, *Staphylococus carnosus* are the most common lipolytic starter cultures used in fermented meat products [3].

Meat and meat products must not contain micro-organisms, their toxins, and metabolites in an amount that represents an unacceptable risk to human health. Our regulation as well as that of the European Union does not allow unsafe products to be put into circulation.

Today, at the beginning of the 21st century, foodborne diseases caused by poisoning are a global public health problem in industrialized countries. One tenth of the population suffers from them annually [4,5,6].

The spoilage of meat products is caused by microorganisms that are members of the *Pseudomonadaceae* and *Enterobacteriaceae* families. We should also point out the following pathogens: *Salmonella spp., Listeria monocytogenes*, *Escherichia coli O157: H7* and *Staphylococcus aureus* as toxic microorganisms. It is therefore necessary to stimulate the growth of technologically important microorganisms during the ripening of fermented sausages, thereby limiting the growth of undesirable microorganisms [7].

Marriott and Gravani [8] point out that foodborne intoxication occurs when toxin producing bacteria grow in the food and release toxins into the food. The infection can occur as a result of the ingestion of microorganisms from the

food in human organisms, causing diseases. Infectious microorganisms may be the cause of enterotoxin-induced diseases in the gastrointestinal tract or by fusion and integration in the tissues.
The following microorganisms: *Salmonella, Escherichia coli O157: H7, Listeria monocytogenes, Campylobacter jejuni / coli, Yersinia enterocolitica, Aeromonas hydrophila* and *Vibrio parahaemolyticus* or *Vibrio vulnificus*, etc., cause diseases because they settle and multiply in the human body [9,10]. Because the cause of these diseases are the cells of the microorganisms, they are called alimentary(foodborne) infections. On the other hand the following microorganisms: *Staphylococcus aureus, Clostridium perfrigens, Clostridium botulinum, Bacilus cereus, Streptococci* etc., some types of mould such as *Aspergillus, Fusarium, Penicillium* etc. produce toxins in food products. Consuming such products causes diseases, independently of the presence of the cells that produced them, therefore, they are called alimentary (foodborne) intoxication [11].
As for the food in Europe, Sánchez - Ortega et al., [12] point out that the most common cause of food poisoning of the population is the presence of *Salmonella* in meat and meat products. Olson et al., [13] indicate that tefmophilic *Campylobacter spp*. is considered to be one of the leading causes of bacteriological gastroenteritis in humans in developed countries.

2. STARTER CULTURES AS INHIBITORS OF MICROBIOLOGICAL HAZARDS IN THE PRODUCTION OF FERMENTED MEAT PRODUCTS

The use of starter cultures is typical for the production of fermented sausages under industrial conditions, in order to provide the desired course of biochemical changes, which affect the development of the inherent sensory and physico-chemical characteristics and to increase the safety of the sausage [14]. In contrast, the production of traditional dry sausages is based solely on the activity of the natural microflora present in the raw material and the premises where they are produced, the fermentation and ripening process being carried out at low temperature over a long period of time [15].

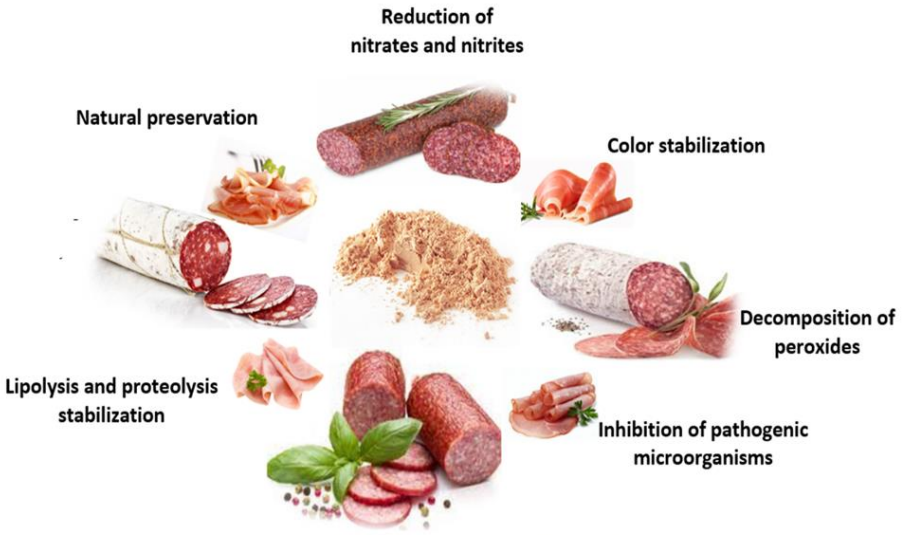

Figure 2. Quality changes in fermented meat products

Starter cultures used in the production of meat products are: LAB, and coagulase - negative staphylococci [16]. The most common bacteria in the starter cultures are: *Lactobacillus (Lactobacillus plantarum, Lactobacillus. casei, Lactobacillus cumates, Lactobacillus pentosus, Lactobacillus alimentarius*), *Pediococcus* (*Pediococcus acidilactici, Pediococcus pentosaceus*), S*taphylococus* (S*taphylococus xylosus, Staphylococus carnosus*), *Micrococcus* (*Micrococcus varians*), *Streptomyces* (*Streptomyces griseus*), as for the yeasts, they are the following: *Debaryomyces hansenii* and the moulds are: *Penicillium chrysogenum, Penicillium nalgiovense* [17].
The most commonly used LAB in the production of fermented meat products are from the genus *Lactobacillus*. Visessanguan et al., [18] also mention the following: *Lactobacillus plantarum*, *Lactobacillus sakei*, *Lactobacillus curvatus* and *Lactobacillus pentosus*.
During fermentation, LAB produce a significant amount of organic acids. They inhibit the growth and proliferation of microorganisms [19]. Baird - Parker [20] points out that lactic and acetic acid are lipophilic acids. They have the

ability to penetrate into the microbial cell in an unassisted form, thereby integrating with the metabolism and lowering the intra cellular pH.

Cleveland et al., [21] report that most LAB produce bacteriocins, i.e. proteins or peptides which have antimicrobial activity against other microorganisms. Bacteriocins are extracellular substances of protein nature that act on strains of the same or related species [22].

Cocolin et al., [23] point out that bacteriocins produced by LAB, especially in the exponential phase, may be an additional inhibiting factor for *Listeria monocytogenes*. Leistner [24] indicates that some species of *Lactobacillus sakei* best inhibit the growth of *Listeria monocytogenes*. Bacteriocins that synthesize gram-positive bacteria act on a wider or narrower spectrum of gram-positive bacteria, their action may be either bacteriocidal with or without cell lysis, or bacteriostatic [25].

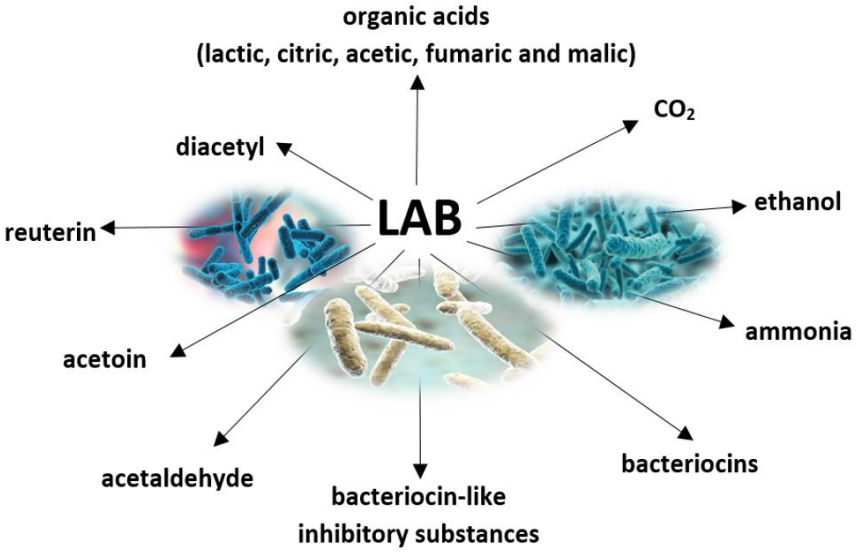

Figure 1. Antimicrobial substances by LAB

There has been an extensive study of the action of bacteriocins against gram-positive bacteria: *Listeria, Staphylococci, Clostridium* and Bacillus strains. Gram-negative bacteria are less sensitive to the effect of bacteriocins. Bacteriocins have much less effect on the sausages themselves, as opposed to in vitro systems. The reason for this is the reaction of bacteriocins to molecules of the food components, primarily fats. This destabilizes the action of the protease and other enzymes. Another reason for their poorer performance is precisely the uneven distribution of bacteriocins in the sausage itself [26].

Hydrogen peroxide produced by LAB has an inhibitory effect on *Pseudomonas sp*. and *Staphylococcus aureus*.

Hydrogen peroxide has a very strong oxidizing effect on cells and cellular proteins. It binds to the cell membrane of the organelles and destroys it. This results in the oxidation of the SH group in enzymes involved in metabolism such as hexonase, aldolase and glyceraldehyde-3-phosphate dehydrogenase [27].

During fermentation, all genera of LAB produce the diacetyl metabolite which has an inhibitory effect on a large number of gram negative bacteria [28].

Holck et al., [29] point out that the synergistic action of various antimicrobial factors ("hurdle concept") creates unfavorable conditions for the survival of microorganisms that cause spoilage-pathogens and toxicogenic microorganisms. This ensures product stability and security.

The importance of microbial succession is enormous during the ripening of traditional fermented sausages, it is a complex process conditioned by complex interactions between biotic (composition of the microflora and microbial relationships) and abiotic factors (physico-chemical properties of the charge) which significantly affect the dynamic changes that occur during the product ripening process [30].

Apatogenic *Staphlococcus* strains (*Staphlococcus carnosus* and *Staphlococcus xylosus*) and micrococci (*Micrococcus aurantiacus M 53* and *Micrococcus varians*) used in starter cultures have the ability to reduce nitrates to nitrites [31]. The pH of the batter should be 6.0 to 5.4 for nitrate reductase activity. When the pH value is lower

their activity is prevented [32]. Nitrite is added to the product for quality maintenance (color, flavor, prevention of lipid oxidation) and product safety [33].

In addition to bacteria, the yeasts (*Debaryomyces* and *Streptomyces)* also participate in the fermentation process. They produce enzymes that ferment the sugars in the lactic acid [34]. Moulds are important in the ripening process of salami, their role having multiple purposes. They affect: aroma, prevention of oxidation, gradual reduction of moisture and prevent the development of mycotoxic molds [35].

3. CONCLUSION

Today, food safety is an important factor in maintaining consumer health. The modern approach to food safety is achieved by the application of the HACCP principles, and the use of preservatives. Furthermore, the use of starter cultures as inhibitors of microbiological hazards in the last few decades has given highly significant results. Starter cultures have great technological significance as well: they control the fermentation process, accelerate fermentation thereby making production more cost-effective, enhance the sensory characteristics and increase the viability of the finished product.

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