Review Article

The Use of Starter Cultures in Traditional Meat Products

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Starter cultures could play an essential role in the manufacture of traditional cured meat products. In order to achieve objectives related to meat products’ quality and safety improvement, the selection of particular strains constituting a starter culture should be carried out in the context of its application, since its functionality will depend on the type of sausage and process conditions. Also, strain selection should comply with particular requirements to warrant safety. The aim of the current review is to update the knowledge on the use of starter cultures in traditional meat products, with focus on dry-fermented products. In this manuscript, we will try to give answers to some relevant questions: Which starter cultures are used and why? Why are LAB used? What are their role and their specific mode of action? Which other groups of microorganisms (bacteria and fungi) are used as starter cultures and how do they act? A particular revision of omics approach regarding starter cultures is made since the use of these techniques allows rapid screening of promising wild strains with desirable functional characteristics, enabling the development of starter cultures better adapted to the meat matrix.

1. Introduction

Starter cultures or starters are individual or mixed formulations of selected strains with a particular enzymatic activity that when added in a defined concentration to a substrate transform it into a food product with specific characteristics [1]. This concept applied to meat products could be described as viable microorganisms that are able to multiply themselves inside meat products, increasing their preservation, controlling their hygienic safety, and potentiating their acceptability by consumers, maintaining or improving their nutritional quality [1].

The preliminary use of starters in meat products resulted from adding a portion of the final meat products to their raw materials, meaning that part of the already fermented batch of sausage was thrown back into the new mix. This already fermented product contained the necessary microorganisms to start the fermentation of the new batch. This is known as back-slopping or back-inoculation [2].

Fermented meat products may be manufactured without the use of starter cultures, although their use can help to ensure safety, standardising product properties (including flavour and colour), and shorten the ripening period. Nevertheless, well-adapted and qualified presumption of safety (QPS) strains must be used and the establishment of the starter culture must be verified in order to guarantee the expected performance.

Probiotics are live microorganisms that confer a health benefit to the host when administered in adequate amounts [3]. Probiotics have been used in food products, food supplements, and pharmaceutical products. Due to increasing concerns over health, probiotic foods (e.g., probiotic dairy products) are now accepted in the world market. Recently, the possibility of developing probiotic meat products has been discussed [4]. By using probiotic starter microorganisms, potential health benefits can be introduced to meat products and it is already possible to produce probiotic meat products [5, 6]. Nevertheless, the potentially beneficial effects on
human health from eating a probiotic sausage still need confirmation [7, 8].

The starter groups used nowadays in meat industry are, by order of importance, lactic acid bacteria (LAB), Gram-positive catalase-positive cocci (GCC+) (mainly staphylococci), moulds, and yeasts.

Lactic acid bacteria (LAB) are a group of Gram-positive bacteria belonging to the Firmicutes. They are catalase-negative, either rod-shaped (bacilli) or spherical (cocci), characterised by an increased tolerance to acidity (low pH range), and have a low GC (guanine-cytosine) content. Although many genera of bacteria produce lactic acid as a primary or secondary end-product of fermentation, the term lactic acid bacteria (LAB) is conventionally reserved for genera in the order Lactobacillales, which includes *Aerococcus*, *Carnobacterium*, *Enterococcus*, *Lactococcus*, *Lactobacillus*, *Leuconostoc*, *Oenococcus*, *Pediococcus*, *Streptococcus*, *Tetragenococcus*, *Vagococcus*, and *Weissella* [9]. As food fermentation agents LAB are involved in making yogurt, cheese, cultured butter, sour cream, sausage, cucumber pickles, olives, and sauerkraut, some species may spoil beer, wine, and processed meats [10].

Gram-positive catalase-positive cocci (GCC+) are the second most important group of meat starters and are composed of nonpathogenic coagulase-negative staphylococci (CNS). The most important starters from this group are strains belonging to the genera *Staphylococcus* and *Kocuria* [11].

At the beginning of the ripening process, the surface mycobiota is mainly composed of yeasts; however, as fermentation proceeds, moulds outcompete yeasts and predominate in the final product [12]. Moulds colonise the surface of fermented meat products, in some cases conferring particular characteristics, however, in other cases being considered signs of spoilage.

Yeasts are characteristic components of the mycobiota growing on fermented sausages. Their origin is mainly related to the environment and to the meat used as raw material, since yeasts are naturally found on fresh meat. The most common genera are *Candida*, *Rhodotorula*, *Debaryomyces*, and *Trichosporon*. In fermented meats, the lactic acid produced by LAB changes the environment, favouring the development of yeasts, which use all of the nutrients and energy and grow fast [13].

Meat preservation by fermentation has been carried out for thousands of years, but the idea of starter cultures was first introduced for dry sausages in the 1940s with Patent US 2225783 A [14]. The first commercial starter culture was a strain of *Pediococcus acidilactici* that was made available in the US in 1957 [15]. In Europe, the first starter culture to be introduced was strain M53 from the genus *Kocuria*, isolated from a Finnish sausage, which was used to prevent colour and aroma defects [16].

Starter cultures play an essential role in the manufacture of fermented food products. Starters composed of LAB strains produce the lactic acid that acts on meat proteins modifying their water binding capacity, thus contributing to texture, moisture content, flavour, and aroma of the products, and definitively acts on its microbiological safety. Additionally, microbial substances, namely, bacteriocins, produced by Gram-positive species of the LAB group, such as, for example, nisin and other lantibiotics or pediocin-like bacteriocins, have an antimicrobial role with an effect on preservation and safety.

Starter cultures have a number of advantages:

(i) They are of known quantity and quality.
(ii) They reduce the ripening time.
(iii) They increase safety by outcompeting undesirable microorganisms.
(iv) They enable the manufacture of a product of constant quality all year round in any climatic zone, as long as proper natural conditions or fermenting/drying chambers are available.

The aim of the current review is to update the knowledge on the use of starter cultures in traditional meat products, with focus on dry-fermented products.

In this manuscript, we will try to give answers to some relevant questions on this subject, through the analysis of published studies with some applied results. Which starter cultures are used and why? Why are LAB used? What is their role and their specific mode of action? Which other groups of microorganisms (bacteria and fungi) are also used as starter cultures and how do they act? What is their function? A revision related to omics methods applied to the screening of autochthonous strains with desirable functional characteristics, allowing the development of well adapted starter cultures to the meat matrix, will be done.

2. Starter Cultures in Dry-Fermented Meat Products

The first generation of meat starter cultures was generally based on microorganisms isolated from vegetable fermentation, such as *L. plantarum* and members of the genus *Pediococcus*. Then, a second generation of starter cultures comprising meat-borne strains, such as *L. sakei* and coagulase-negative staphylococci (CNS), was developed, harbouring phenotypic traits of technological relevance [17]. More recently, efforts have been dedicated to the study of the physiological and technological properties of LAB and CNS isolated from traditional fermented sausages, in order to develop functional starter cultures that enhance safety and nutritional advantages while maintaining industrial performance [5, 18].

The manufacturing of dry-fermented sausages involves spontaneous fermentation commanded by bacteria (LAB) and GCC+ and, less importantly, by fungi, namely, moulds and yeasts [19].

Most meat starter cultures commercially available are combined cultures of LAB (mainly *Lactobacillus* spp. and *Pediococcus* spp.) and GCC+ (primarily *Staphylococcus* spp. and *Kocuria* spp.). These bacteria are responsible for the microbial reactions that occur during meat fermentation, such as acidification, catalase activity, and bacteriocin production [11].

Several studies have addressed the importance of using starter cultures in traditional dry-fermented meat products...
not only for safety or conformity reasons, but also for uniformity purposes [20–22].

Although most studies about the use of starter cultures are on dry-fermented sausages [23–25], a few works on other meat products, such as hams or fresh sausages, have also been reported [26].

Inoculation of starter cultures in dry-fermented meat products may occur either by incorporation as an ingredient in the meat batters or by surface inoculation.

Bacteria are usually incorporated in the meat batters at concentrations between 5 and 8 log colony forming units (cfu)/g [23]. Yeasts may be inoculated either on the surface of the sausage or in the meat batter at a concentration typically between 4 and 6 log cfu/g. Moulds are always surface-inoculated, due to their strictly aerobic character, frequently by dipping in an aqueous solution of spores at concentrations ranging from 3 to 4 log spores/cm².

2.1. Their Role in Quality Improvement of Sausages. The selection of starter cultures for quality improvement of sausages is based on technologically relevant traits. The autochthonous microbiota of sausages and other meat products, as well as the microbiota of the processing environment of the production units, may be a good starting point for the isolation of potential starters, because those strains are well adapted to the meat environment [19].

Bourdichon and coworkers [27] presented a list of microorganisms used in food fermentation in a wide range of food matrices (dairy products, meat, fish, vegetables, legumes, cereals, beverages, and vinegar).

2.1.1. Bacteria: LAB and GCC+. When selecting starter cultures for dry- and semidry-fermented sausages, LAB and CNS strains with useful metabolic activities and benefits during fermentation should be used.

(1) Lactic Acid Bacteria (LAB). Lactic acid bacteria (LAB) are Gram-positive, non-spore-forming cocci or bacilli with a low GC content [28]. They generally are nonrespiratory and lack catalase. They produce lactic acid as one of the main fermentation products of carbohydrates. They lack genuine catalase and do not possess cytochromes. All LAB grow anaerobically, but unlike most anaerobes, they grow in the presence of O₂ as “aerotolerant anaerobes” [9].

According to the current taxonomic classification, they belong to the phylum Firmicutes, class Bacilli, order Lactobacillales. Six different families include all genera, as shown in Table 1 (http://www.uniprot.org/taxonomy/186826).

Lactic acid bacteria are among the most important groups of microorganisms used in food fermentation. They contribute to the taste and texture of fermented products and inhibit food spoilage bacteria by producing growth-inhibiting substances and large amounts of lactic acid.

Based on sugar fermentation patterns, there are two broad metabolic categories of LAB: homofermentative and heterofermentative. The homofermentative pathway produces basically only lactic acid, whereas the heterofermentative pathway produces CO₂ and ethanol or acetate in addition to lactic acid [9]. Homofermentative LAB include some lactobacilli and most enterococci, lactococci, pediococci, streptococci, tetragenococci, and vagococci that ferment hexoses through glycolysis by the Embden-Meyerhof-Parnas pathway. On the other hand, heterofermentative LAB ferment pentoses mainly through the phosphoketolase pathway and include leuconostocs, some lactobacilli, oenococci, and Weissella species.

Relevant technological features for LAB starters include fast production of lactic acid; growth at different temperatures, salt concentrations, and pH values; gas production from carbohydrates; catalase activity and hydrolysis of hydrogen peroxide; nitrate and nitrite reduction; moderate proteolytic and lipolytic enzymatic activities; good performance in combined starters with other microbial components [29].

However, fermentation conditions must be controlled to avoid excessive pinholes, gas pockets, and off-flavours, resulting from gas production from carbohydrates [30]. Additionally, the production of hydrogen peroxide may result in undesirable oxidation, known as greening [30]. Furthermore, it must be taken into account that proteolytic and lipolytic activities should be moderate, to avoid undesirable sensory changes.

As for the role of LAB in the quality of dry-fermented meat products, LAB participate in the coagulation of muscle proteins by acidifying the batters, which results in increased slice stability, firmness, and cohesiveness of the final product [31, 32]. Besides, they contribute to the flavour of the final product through the formation of noticeable acidic and vinegary (acetic acid) tastes. Moreover, the existing acidic conditions may increase the activity of cathepsin D, which is again responsible for muscle proteolysis [33].

Several authors have reported the use of LAB starter cultures for the production of fermented sausages [34–38]. For example, Wang and coworkers reported the inoculation with L. sakei as beneficial for microbiological quality against the growth of foodborne pathogens, also improving sensory characteristics [34].

The use of coagulase-negative staphylococci (CNS) as meat starter cultures contributes to an adequate colour development based on their nitrate reductase activity. On the other hand, their catalase activity reduces oxidative damage and their metabolism contributes to flavour. The flavour-generating potential of CNS is even more important when producing low-salt [47, 48] or low-fat [49, 50] sausages [51]. However, the full metabolic potential of CNS should be further explored, so that we may take advantage of more technological features of CNS [52].
Table 1: Families and genera of LAB.

<table>
<thead>
<tr>
<th>Family</th>
<th>Genus</th>
<th>Cellular morphology</th>
<th>Sugar fermentation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aerococcaceae</td>
<td>Aerococcus</td>
<td>Cocci-tetrads</td>
<td>Homofermentative</td>
</tr>
<tr>
<td>Carnobacteriaceae</td>
<td>Carnobacterium</td>
<td>Bacilli</td>
<td>Homofermentative</td>
</tr>
<tr>
<td>Enterococcaceae</td>
<td>Enterococcus</td>
<td>Cocci</td>
<td>Homofermentative</td>
</tr>
<tr>
<td>Enterococcaceae</td>
<td>Lactobacillus</td>
<td>Bacilli</td>
<td>Strain-dependent</td>
</tr>
<tr>
<td>Lactobacillaceae</td>
<td>Pediococcus</td>
<td>Cocci-tetrads</td>
<td>Homofermentative</td>
</tr>
<tr>
<td>Leuconostocaceae</td>
<td>Leuconostoc</td>
<td>Cocci</td>
<td>Heterofermentative</td>
</tr>
<tr>
<td></td>
<td>Oenococcus</td>
<td>Cocci</td>
<td>Heterofermentative</td>
</tr>
<tr>
<td></td>
<td>Weissella</td>
<td>Cocci/bacilli</td>
<td>Heterofermentative</td>
</tr>
<tr>
<td>Streptococcaceae</td>
<td>Lactococcus</td>
<td>Cocci</td>
<td>Homofermentative</td>
</tr>
<tr>
<td></td>
<td>Streptococcus</td>
<td>Cocci</td>
<td>Homofermentative</td>
</tr>
</tbody>
</table>

Table 2: Species of GCC+ isolated from fermented sausages and their role in the fermentation process.

<table>
<thead>
<tr>
<th>Family</th>
<th>Genus</th>
<th>Species</th>
<th>Metabolic activities</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Staphylococcaceae</td>
<td>Staphylococcus (CNS)</td>
<td>S. xylosus</td>
<td>(i) Nitrate reductase</td>
<td>[39–42]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>S. carnosus</td>
<td>(ii) Proteolytic</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>S. equorum</td>
<td>(iii) Lipolytic</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>S. succinus</td>
<td>(iv) Catalase</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>S. saprophyticus</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Micrococcaceae</td>
<td>Micrococcus</td>
<td>M. luteus</td>
<td>(i) Nitrate reductase</td>
<td>[27, 43]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>M. lylae</td>
<td>(ii) Antioxidative</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>K. varians</td>
<td>(iii) Catalase</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>K. kristinae</td>
<td>(iv) Lipolytic</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>(v) Proteolytic</td>
<td></td>
</tr>
</tbody>
</table>

Besides contributing to flavour, *Staphylococcus* and *Kocuria* also provide nitrate-reductase and antioxidant activities [53, 54].

Numerous studies addressing the use of starter cultures in meat products have been published, with both single (either LAB or GCC+) and mixed cultures.

Several authors have reported the use of CNS starter cultures for the production of fermented sausages. According to Ravyts et al. [51], the success of CNS in flavour development seems to be determined by acidification.

Hugas and Monfort [31] highlighted the need to use selected strains of GCC+ to ensure sensory quality. Besides, other authors have described the capability of *S. xylosus* and *S. carnosus* strains to modulate aroma through the degradation of amino acids and free fatty acids (FFAs) [55–57].

Autochthonous strains of *S. xylosus* have been recommended for the production of very aromatic sausages in Southern Europe, instead of the less adapted commercial starter cultures [58].

Lusnica and colleagues have studied the effect of an added starter culture (*S. xylosus* and *S. carnosus*) to a frankfurter-type meat emulsion in degrading polychlorinated biphenyls (PCBs) [59]. Furthermore, quite a few works have been published reporting the results obtained by the utilisation of mixed starter cultures (LAB and CNS) [25, 60–66].

Bacteriocinogenic LAB and selected strains of *S. xylosus* and *S. carnosus* are commercially available for use in improving the safety, colour, and flavour of final products. It is also important to assess positive interactions, such as growth and proteolytic activity, among the different starter cultures strains [67–70].

The effect of different starter culture combinations (*Staphylococcus carnosus*, *Pedriococcus pentosaceus*, and *Lactobacillus sakei*) on the quality of Turkish type fermented sausage (*Sucuk*) has been evaluated during ripening and it was concluded that the use of lipolytic starter cultures (*S. carnosus/L. sakei*) would have a positive effect in accelerating ripening and enhancing the quality of dry-fermented sausages [71].

Tremonte and coworkers demonstrated that *S. xylosus* and *Kocuria varians* are able to stimulate the growth of *L.
sakei strains, positively influencing the proteolytic activity of strains in a combined use [66].

Casquete and colleagues have emphasised the importance of autochthonous starter cultures in improving homogeneity and safety of fermented meat products, without depreciating their sensory characteristics [60–62]. Furthermore, they have highlighted the importance of choosing a starter formulation consisting of a combination of strains that is appropriate for each ripening procedure [60].

We may conclude that flavour and aroma of fermented sausages result from the combined action of different bacteria: LAB produce lactic acid and small amounts of acetic acid, ethanol, and acetoin; however, the proteolytic and lipolytic activities of both LAB and GCC+ are essential to the sensory quality of fermented sausages.

2.1.2. Fungi: Yeasts and Moulds. Fungi generally contribute to a characteristic flavour of some fermented meat products. Yeasts may be either inoculated in the meat batters or surface-inoculated, whereas moulds are always inoculated at the surface of sausages. Surface inoculation has a further physical protective role.

(1) Yeasts. The first studies with yeasts in fermented sausages were conducted in the first decades of the 20th century, when the importance of the "fleur du saucisson" was recognized and the use of pure yeast cultures for flavouring in fermented sausages began to be recommended. Later on, it was established that yeasts are part of the microbiota of fermented sausages and their use as starter cultures was suggested, because the addition of selected Debaryomyces strains could improve the colouring, colour, and flavour of sausages [72].

Several studies have tried to understand the role of yeasts as secondary microbiota in fermented meat products. Yeast strains belonging to the genera Debaryomyces, Yarrowia, Pichia, Rhodotorula, Cryptococcus, and Trichosporon have been isolated from meat products [73], with clear predominance of the Debaryomyces genus [13].

Some yeasts have been shown to contribute to flavour and texture development throughout the curing of various products [74–76]. Moreover, some studies have shown that the characteristic flavour of dry-cured meat products may be developed through the influence of yeasts [77–79].

Furthermore, the manufacture of dry-fermented sausages with optimised concentrations of Debaryomyces spp. in the presence of LAB and CNS has been demonstrated to have a positive effect on the final flavour and sensory quality by inhibiting the development of rancidity and generating ethyl esters that contribute to the proper sausage aroma [78].

(2) Moulds. Surface moulding of fermented meat products is considered a desirable event in most European countries, which include Italy, Romania, Bulgaria, France, Hungary, Switzerland, Southern Germany, Spain, Austria, and Belgium [12]. In fact, the presence of mycelium at the surface of sausages has several main advantages:

(i) It prevents excessive drying, allowing homogeneous dehydration of the product [12].

(ii) It metabolizes peroxides, protecting fat from oxidation, thus preventing rancidity [12].

(iii) It reduces O$_3$ levels on the product surface, thus avoiding oxidative processes and improving meat colour [80].

(iv) It contributes to the flavour of the final product, by breaking up fats, proteins, and lactic acid, thus favouring pH increase [12].

The use of moulds as a seasoning for sausage can have both desirable and undesirable consequences. The desirable consequences are the creation of a successful product that appeals to consumers. The undesirable consequences are health risks associated with the growth of undesirable moulds that produce highly toxic secondary metabolites, mycotoxins, such as ochratoxin A (OTA), or penicillin produced by species of Penicillium [81].

Furthermore, surface moulding of fermented meat products was observed during storage and can be a quality problem, because of the undesirable effects, mainly connected to the production of off-flavours [81].

Surface mould inoculations were traditionally done with the autochthonous mycobacteria, which was mainly composed of Penicillium spp., Aspergillus spp., or Scopulariopsis spp. The first toxicologically and technologically suitable mould starter culture for meat products, P. nalgiovense strain, was selected by Mintzlaff and Leistner in 1972 [82]. However, nowadays, a wide assortment of industrialised starter cultures is commercially available as an alternative to the inoculating mixtures composed of autochthonous strains.

Some studies on the use of mould starter cultures have already been performed [80,83]. For example, quality traits of wild boar mould-ripened salami manufactured with different selections of meat and fat tissue and with and without commercial bacterial starter cultures have been investigated [84]. The use of a bacterial starter culture in the manufacture of mould-ripened wild boar salami resulted in significantly lower peroxide values, lower TBARS concentrations, and lower amounts of biogenic amines, namely, histamine, cadaverine, and putrescine, associated with better sensory evaluation scores.

Application of commercial moulds to sausage surfaces improves primarily the safety towards regarding mycotoxin production. Moreover, the production of antibiotics, namely, penicillin, also needs to be controlled [82]. Additionally, sausage producers achieve more consistent flavour, taste, and drying rate and a more uniform appearance.

Table 3 shows a list of moulds found in fermented meat products.

Among the species mentioned in Table 3, P. nalgiovense and P. gladioli are currently considered safe and are commercially available to be used as starter cultures in meat products [12].

2.2. Antimicrobial Activity of Starter Cultures. Bacteriocins, natural antimicrobial peptides, and the acid lactic produced from glucose could be used to improve the quality and safety of meat products by avoiding the presence of pathogens, such as Listeria monocytogenes and spoilage microorganisms, and
Table 3: Species of moulds usually found in dry-fermented sausages.

<table>
<thead>
<tr>
<th>Common species</th>
<th>Uncommon species</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Penicillium nalgiovense</strong></td>
<td>P. waksmanii</td>
</tr>
<tr>
<td>P. gladioli</td>
<td>Aspergillus ochraceus</td>
</tr>
<tr>
<td>P. camemberti</td>
<td>E. herbariorum</td>
</tr>
<tr>
<td>P. chrysogenum</td>
<td>E. repens</td>
</tr>
<tr>
<td>P. aurantiognicenum</td>
<td>A. niveus</td>
</tr>
<tr>
<td>P. brevicompactum</td>
<td>P. citrinum</td>
</tr>
<tr>
<td>P. nordicum</td>
<td>A. candidus</td>
</tr>
<tr>
<td>P. phoenicicum</td>
<td>P. crustosum</td>
</tr>
<tr>
<td>Eurotium rubrum</td>
<td>P. commune</td>
</tr>
<tr>
<td>P. griseofulvum</td>
<td>A. sclerotiorum</td>
</tr>
<tr>
<td>P. olsonii</td>
<td>A. versicolor</td>
</tr>
<tr>
<td>P. implicatum</td>
<td>P. alii</td>
</tr>
<tr>
<td>Scopulariopsis candida</td>
<td>P. fellutanum</td>
</tr>
</tbody>
</table>

improving the competitiveness of their producers for survival [85].

A list of the main bacteriocins produced by LAB along with a list of bacteria they are effective against is summarised in Table 4.

Several L. sakei and L. curvatus have been reported as bacteriocin producers and have been used as protective cultures, and their activity against L. monocytogenes has been proved in meat products [87–90].

Lactococcus lactis and Enterococcus spp. strains isolated from different food matrices have been shown to produce bacteriocins [91–93].

Pediococcus acidilactici MCH14 pediocin-producing strain and the pediocin PA-1 itself have been demonstrated to inhibit the growth of the foodborne pathogens L. monocytogenes and Clostridium perfringens in Spanish dry-fermented sausages and frankfurters [94].

Bacteriocins produced by strains of L. plantarum isolated from Portuguese traditional pork products have been shown to have a broad spectrum of activity [95].

LAB starter cultures have been used in the production of Nham, which is a Thai-style fermented pork sausage, for their antilisterial activity in order to reduce the severity of postacidification and increase the shelf life of Nham at ambient temperature [96, 97].

Additionally, also S. xylosus strain SX S03/1 M/1/2 has been shown to produce a thermostable bacteriocin which could be used as starter culture or meat additive to prevent possible handling or meat processing contamination [98].

2.3. Competitiveness of Starter Cultures. One of the most important properties of meat starter cultures is the ability to colonize the meat environment, in competition with the autochthonous microbiota and dominating the microbial community of fermented products. The starter culture must compete with the natural microbiota of the raw material, which carries out the expected metabolic activities through its growth rate and survival under the prevailing conditions during sausage production. Low temperatures, high salt concentrations, and, to a lesser extent, oxygen availability are among the most important preservative conditions during meat fermentation [17].

The main metabolic activities and their corresponding technological roles for the main microbial starter groups are shown in Table 5.

In general, CNS are poorly competitive in the presence of acidifying LAB strains [99]. On the other hand, strains of L. sakei have shown superior competitiveness, which could probably be explained by their specialised metabolic repertoire well adapted to the sausage environment, including the arginine deiminase (ADI) pathway [100] and the utilisation of nucleosides [101].

Genus-specific and species-specific PCR and real-time RT-PCR methods have been used to monitor and quantify the populations of the inoculated starter cultures [24]. Moreover, RT-PCR-DGGE and RNA-based pyrosequencing of the 16S rRNA gene have also been used to monitor the microbiota of fermented sausages [102].

2.4. Safety of Selected Meat Starter Cultures. Meat starter cultures or food cultures (FC) are safe live bacteria, yeasts, or moulds used in food production, and they are in themselves a characteristic food ingredient (http://www.effca.org/content/food-culture). Food starter cultures (microorganisms) used directly in food production are regarded as food ingredients in the European Union (EU). Starters enter in a category of food ingredients with a very long history of use in a great variety of food products. If a starter is added to a food product, the requirements established in the General Food Law should be accomplished by the food operator. The food cultures used as starters in the fermentation of foods are not subject to EU premarketing regulation, unless they are regarded as being novel to the EU market and their consumers. Many starters were selected from fermented foods and several microorganisms are present in spontaneously fermented foods. However, regarding safety concerns, any food cultures to be introduced in a food should be evaluated. The approaches for assessing the safety of microorganisms...
### Table 4: LAB bacteriocins, bacteriocin producers, and susceptible pathogenic bacteria.

<table>
<thead>
<tr>
<th>Bacteriocin</th>
<th>Bacteriocin producer</th>
<th>Susceptible pathogenic bacteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sakacin</td>
<td><em>Lactobacillus sakei</em></td>
<td><em>Listeria monocytogenes</em>&lt;br&gt;<em>Staphylococcus aureus</em>&lt;br&gt;<em>Enterococcus</em> spp.&lt;br&gt;<em>Brochothrix thermosphacta</em>&lt;br&gt;<em>Pseudomonas</em> spp.&lt;br&gt;<em>Campylobacter</em> spp.&lt;br&gt;<em>Escherichia coli</em>&lt;br&gt;<em>Klebsiella</em> spp.&lt;br&gt;Other LAB</td>
</tr>
<tr>
<td>Plantaricin</td>
<td><em>L. plantarum</em></td>
<td><em>Listeria monocytogenes</em>&lt;br&gt;<em>Staphylococcus aureus</em>&lt;br&gt;<em>Clostridium perfringens</em>&lt;br&gt;<em>Clostridium tyrobutyricum</em>&lt;br&gt;<em>Bacillus cereus</em>&lt;br&gt;<em>Enterococcus</em> spp.&lt;br&gt;<em>Brochothrix thermosphacta</em>&lt;br&gt;<em>Pseudomonas</em> spp.&lt;br&gt;<em>Salmonella</em> spp.&lt;br&gt;<em>Escherichia coli</em>&lt;br&gt;Other LAB</td>
</tr>
<tr>
<td>Curvacin</td>
<td><em>L. curvatus</em></td>
<td><em>Listeria monocytogenes</em>&lt;br&gt;<em>Staphylococcus aureus</em>&lt;br&gt;<em>Clostridium tyrobutyricum</em>&lt;br&gt;Other LAB</td>
</tr>
<tr>
<td>Nisin</td>
<td><em>Lactococcus lactis</em></td>
<td><em>Listeria monocytogenes</em>&lt;br&gt;<em>Staphylococcus aureus</em>&lt;br&gt;<em>Clostridium tyrobutyricum</em>&lt;br&gt;Other LAB</td>
</tr>
<tr>
<td>Pediocins</td>
<td><em>Pediococcus</em> spp.</td>
<td><em>Listeria monocytogenes</em>&lt;br&gt;<em>Enterococcus</em> spp.&lt;br&gt;Other LAB</td>
</tr>
</tbody>
</table>

Adapted from Fraqueza et al. [86].

entering the human food chain differ considerably depending on the applicable legislation, if any.

Several approaches have been delineated in order to consider the starter cultures safe. The *Qualified Presumption of Safety* (QPS) list is the EFSA fast track risk assessment tool that is used by EFSA panels when evaluating products with microorganisms that require a premarket authorisation (e.g., feed additive cultures, cell factories producing enzymes/additives/vitamins, novel microorganisms, and plant protection). This approach is restricted only to the microorganisms related to regulated food and feed products and is based on history of use, body of knowledge, and the absence of adverse effects at the taxonomic unit level [103, 104].

The *Generally Recognized as Safe* (GRAS) status is open to all types of food additives, which include food cultures. The determination of GRAS status is made by the FDA and/or external experts and is based on the history of use, body of knowledge, and the absence of adverse effects at the strain level.

Food cultures with a long history of safe use in food are considered as traditional food ingredients and are legally permitted for use in foods in the EU without premarket authorisation, as described earlier. As a consequence, EFSA panels do not evaluate microbial strains of food cultures. Nevertheless, the QPS list can be consulted when safety evaluations of food culture are made.

Microorganisms, which are not on the QPS list, are not necessarily considered to be unsafe and their assessment regarding antibioresistance, virulence, and biogenic amine characterization should be done.

The *International Dairy Federation* (IDF) and the *European Food and Feed Cultures Association* (EFFCA) have proposed additional tools and methods to evaluate the safety...
Table 5: Requirements for starter LAB, GCC+, yeasts, and moulds.

<table>
<thead>
<tr>
<th>Microbial group</th>
<th>Metabolic activity</th>
<th>Technological role</th>
</tr>
</thead>
<tbody>
<tr>
<td>LAB</td>
<td>Acidification</td>
<td>Modulate flavour (acid/tangy)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Inhibit pathogens</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Develop texture</td>
</tr>
<tr>
<td></td>
<td>Proteolysis</td>
<td>Accelerate drying</td>
</tr>
<tr>
<td></td>
<td>Antimicrobial</td>
<td>Develop flavour</td>
</tr>
<tr>
<td></td>
<td>Antioxidant</td>
<td>Inhibit pathogens</td>
</tr>
<tr>
<td></td>
<td>Probiotic</td>
<td>Extend shelf life</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Protect colour</td>
</tr>
<tr>
<td>GCC+</td>
<td>Nitrate reductase</td>
<td>Compete in the gastrointestinal tract</td>
</tr>
<tr>
<td></td>
<td>Degradation of amino acids and FFAs</td>
<td></td>
</tr>
<tr>
<td>Yeasts</td>
<td>Antioxidant</td>
<td>Develop typical red (cured) colour</td>
</tr>
<tr>
<td></td>
<td>Proteolytic</td>
<td>Develop flavour</td>
</tr>
<tr>
<td></td>
<td>Lipolytic</td>
<td>Prevent rancidification</td>
</tr>
<tr>
<td>Moulds</td>
<td>Antioxidant</td>
<td>Prevent rancidification</td>
</tr>
</tbody>
</table>

Adapted from [17].

of food cultures with the unique target of keeping a high level of food safety and to protect human life and health. According to Laulund et al. [105], whatever the strategy applied, it is imperative to have an evaluation of the food cultures’ safety at three levels: (a) at the strain level, (b) during production, and (c) in the process it is applied to and throughout the shelf life of the food.

2.4.1. Assessment of Antibioresistance. The One Health concept recognises that the health of people is connected to the health of animals and the environment. The food chain has been recognized as one of the main routes for the transmission of antibiotic-resistant bacteria between animal and human populations [106]. Antibiotic resistant bacterial strains may be a potential direct link between the indigenous microbiota of animals and the human gastrointestinal tract.

Bacterial strains selected as starters with technological or food protective characteristics to be introduced in food always need to be phenotypically assessed for antibiotic resistance to clinically relevant antibiotics. The phenotypic testing based on determination of a minimum inhibitory concentration (MIC) for a selected group of antimicrobials should be performed. The absence of phenotypic antibiotic resistance is preferred, but if a resistance profile is observed, a proper analysis of the whole genome potentially combined with information that the observed resistance is not transferable is needed; only then can the strain(s) be considered safe for use in food culture [107].

The possibility of antimicrobial resistance transfer from viable microorganisms to other microorganisms is related to the genetic basis of the resistance being considered most plausible, when the resistance is mediated by added/acquired genes. Regarding this possibility, several safety assessments have been done by several authors on the species usually selected for starters, such as CNS or LAB.

Safety hazards associated with CNS were mostly limited to the presence of antibiotic resistance [108]. CNS strains resistant to multiple antibiotics have been reported [109]. Kastner et al. [110] detected the tetracycline resistance genes tetK in Staphylococcus spp. starter cultures.

The detection of antibiotic resistant (AR) strains among LAB has resulted in their recognition as a reservoir of AR genes horizontally transmissible to pathogens through the food chain, which constitutes a problem [111, 112]. Antibiotic multiresistant strains of lactobacilli and other LAB have been isolated from dry-fermented meat products [113–120]. LAB possesses a broad spectrum of natural (intrinsic) and acquired antibiotic resistance. However, only resistance acquired by mutation or horizontal gene transfer poses a risk for public health [121].

The most common resistance genes detected in LAB isolated from dry-fermented sausages are the tetracycline resistance genes tetM, tetW, and tetS and the genes coding for erythromycin resistance, ermB and ermC [117, 120]. These are genes linked to mobile elements, and if the phenotypic expression of antibiotic resistance is expressed, their presence is considered a hazard.

2.4.2. Detection of Strains Producers of Biogenic Amines. Any strains to be incorporated as starters in fermented meat products should be assessed for their (in)ability to mediate the production of biogenic amines. Strategically, the use of Lactobacillus spp. or Pediococcus spp. non-BA producer strains could dominate and avoid the presence of high contents of BA in meat products. Several authors have reported the important role of starter cultures in decreasing the content in biogenic amines [47, 48, 122–126].

2.4.3. Toxigenic Potential. Among LAB, enterococci play an important role in food fermentation and may contribute to
the organoleptic uniqueness of some products, but they are also responsible for community-acquired and nosocomial infections [118]. Some of the most important virulence factors include the production of hydrolytic enzymes, namely, gelatinase, lipase, and DNase, haemolytic activity and the production of cytolsyn, the presence of adhesins, and the ability to form biofilms [127].

Two studies with enterococci strains isolated from several Portuguese dry-fermented sausages revealed that although meat enterococci harbour antibiotic resistance and produce biofilms, a reduced number of virulence factors were detected [118, 128]. However, a third study with Portuguese dry-fermented products from northern Portugal has detected phenotypic and genotypic evidence of potential virulence factors among *Enterococcus* spp. isolates, which is a reason of concern [129].

Some members of the CNS group, primarily *S. epidermidis*, are common nosocomial pathogens, and the presence of regulatory elements involved in the control of virulence-factor synthesis has recently been identified. Remarkably, strains of *S. xylosus* were isolated from patients who had an underlying disease, while the same species has been reported to be involved in infections of poultry [130].

Although CNS of food origin have not been found to produce nosocomial infections, some strains that produce enterotoxins have been described. Vernozy-Rozand et al. [131] reported enterotoxin E to be the most common enterotoxin in *S. equorum* and *S. xylosus*, although it is reported that the occurrence of staphylococcal enterotoxin genes in CNS from slightly fermented sausages was rare, detecting only *entC* in *S. epidermidis* [132].

Absence of genes coding for staphylococcal enterotoxins or enterotoxin-like superantigens is a requirement for strains selected as starter cultures, and the *S. xylosus* and *S. carnosus* strains currently used as starter cultures or isolated from fermented meat products generally lack toxin genes [11].

The analysis of virulence factors in strains of *S. epidermidis*, *S. simulans*, *S. xylosus*, *S. kloosii*, and *S. caprae* revealed sometimes high percentage of incidence of the following virulence traits: production of slime, α- and β-haemolysins, DNase, TNase, hyaluronidase, and TSST-1 and production of enterotoxins SEA, SEB, SEC, and SED [118].

### 2.4.4. Strains with Ability of Biofilm Formation

In food industry, biofilm formation is undesirable for hygienic and safety reasons, as it can allow the attachment of food-spoilage or pathogenic microorganisms to food or food surfaces [134]. Nevertheless, several authors believe that colonization of food surfaces by starters could be desirable, as it would inhibit colonization by pathogenic or spoilage bacteria [135].

Among CNS, biofilm formation has been studied in *S. aureus* [136], *S. epidermidis* [136], *S. hominis* [137], *S. sciuri* [135], and *S. equorum* [138]. *S. capitis*, *S. cohnii*, *S. epidermidis*, *S. lentus*, and *S. saprophyticus* have all also been reported to form biofilms [139], though due to different genetic determinants [140]. These studies concluded that, in general, biofilm formation is a strain-dependent characteristic. Furthermore, the capacity of *S. xylosus* to form biofilms may contribute to its survival of food processing [141]. On the other hand, the inability of *S. carnosus* to form biofilms may explain why it is rarely recovered from meat processing environments [142].

LAB biofilms may be used to control the formation of biofilms by the foodborne pathogens *Listeria monocytogenes*, *Salmonella* Typhimurium, and *Escherichia coli* O157:H7 [143].

Genes potentially responsible for biofilm formation and cellular aggregation that may assist the organism to colonize meat surfaces have been identified in *L. sakei* strain 23K [144]. Moreover, the analysis of microenvironments through the scanning electron microscopy (SEM) evidenced the presence of microchannels that favour microbial flow, while the ability of *L. sakei* to form biofilm guarantees the correct colonisation of the different meat niches, throughout the fermentation process (2017).

Biofilm formation in LAB species has been reported to be a stress response and survival strategy in stressful environments [145, 146]. Some reports have also described the genes responsible for quorum sensing, adhesion, and biofilm formation [147–150].

Another possible biocontrol strategy to avoid the presence of pathogens in meat industry could be the use of bacteriocins and enzymes; this is considered important for the maintenance of biofilm-free systems and thus for the quality and safety of foods.

#### 2.5. Functional Starter Cultures

Functional starter cultures are starters that have at least one functional property, which may contribute to food safety and/or offer one or more organoleptic, technological, nutritional, or health advantages [151]. They offer additional functionalities compared to plain starter cultures and are a way of improving the fermentation process of meat products and achieving tastier, safer, and healthier products.

##### 2.5.1. Bioprotective Cultures

Bioprotective cultures in the manufacture of fermented meat products is a well-established technology [86]. Bioprotective starters may contribute to the safety and increase in shelf life of fermented meat products through the release of organic acids [152], the production of bacteriocins against important food pathogens, mainly *L. monocytogenes* [153], and the control of biological hazards [86].

Potential protective starter cultures to use in fermented meat products have been identified [154] and tested [4, 155–157]. The use of bioprotective starter cultures ensures safety, while increasing shelf life, without compromising the nutritional value of fermented meat products or depreciating their sensory quality.

##### 2.5.2. Probiotics

According to the currently adopted definition by the Food and Agriculture Organization/World Health Organization (FAO/WHO) [158], probiotics are defined as “live microorganisms which when administered in adequate amounts confer a health benefit on the host.”

Probiotics are nonpathogenic health-promoting microorganisms that when ingested in defined amounts may have a positive effect on human physiology and health [29]. In 1965,
Lilly and Stillwell proposed probiotics to be "microorganisms promoting the growth of other microorganisms." To act as safe probiotic microorganisms, strains should be of species and genera normally present in the human gastrointestinal tract [159].

Probiotics are LAB (or bifidobacteria), mainly Gram-positive Lactobacillus species.

In general, health benefits of probiotic foods are based on the presence of selected strains of LAB that, having passed through the stomach and the small intestine, survive in the large intestine and confer a health benefit on the host [160].

LAB with probiotic properties may have a positive influence on product taste, flavour, and aroma, as well as on functional and physiological properties [8].

Some LAB strains are able to produce nutraceutical compounds [161]. Studies on Lactococcus lactis highlight the possibility of developing LAB meat starter cultures for in situ production of vitamins, by overexpression and/or disruption of relevant metabolic genes [162–164].

Although dairy products are the most commonly used food vehicles for the delivery of probiotics, several studies dealing with the use of probiotics in fermented meat products to improve their nutritional value as functional foods have been reported [5, 154, 165–167].

The commercial application of probiotics in meat products is not a current procedure, mostly because of technological issues. Although fermented meat products are processed without heating, probiotics may still be inactivated due to low pH or water activity value, as well as by the presence of native microorganisms or curing salts. The most important problem is to find a compromise between technology, safety, quality, and health-beneficial value of food [160]. For recent reviews, please refer to Neffe-Skocińska et al. [168] and Vuyst et al. [8].

Some species involved in sausage fermentation, such as L. plantarum, have been engineered to produce an excess of folate (vitamin B11) [162]. This gives the possibility of fortifying meat products with vitamins and other essential compounds, thus producing healthier meat products [29].

Today, the use of probiotic starters in any fermented food claiming health benefits should be scientifically demonstrated according to the legal requirements of EU for labelling [169, 170].

3. Omics of Meat Starter Cultures

The main bacterial species used in meat fermentation are LAB and CNS. Lactobacillus sakei, Lactobacillus curvatus, Lactobacillus plantarum (mainly in Europe), and Pediococcus pentosaceus and Pediococcus acidilactici (mainly in the US) are the starters commonly used for their fermentative role in dry-sausage production, while Staphylococcus xylosus and Staphylococcus carnosus are known for their involvement in the development and stability of colour and aroma production [171].

Using comparative genomics, transcriptomics, proteomics, and metabolomics, the diversity of strains naturally present in traditional fermented sausages is being explored. These approaches allow rapid high-throughput screening of promising wild strains with desirable functional characteristics and a lack of negative features, enabling the development of starter cultures based on indigenous technological bacteria from traditional sausages, which are thus better adapted to the meat matrix [22, 172].

The first genome sequence of a starter to be published was the one of the LAB L. sakei 23K [144]. Despite the small sized genome (1,883 protein-coding genes), L. sakei contains seven rRNA gene clusters [144]. This redundancy may contribute to its ability to grow in complex microbial ecosystems [173]. With regard to gene products, the L. sakei genome shares the highest level of conservation with Lactobacillus plantarum, which can be used as a starter in fermented meat, dairy, and vegetable products [144, 174, 175]. Genome analysis revealed a specialized metabolic repertoire to adapt and grow on meat products. Important cellular functions are encoded by a redundancy of genes likely to enhance the organism’s robustness and most probably help it to outgrow other competing bacteria. As a unique ability among lactic acid bacteria, L. sakei is able to use meat components, such as purine nucleosides, abundant in meat, upon glucose depletion, to grow and produce energy. Genes possibly responsible for biofilm formation and cellular aggregation, which may assist in colonising meat surfaces, were also identified [144].

The draft genome sequence of L. sakei subsp. sakei strain LS25, a commercial starter culture for fermented sausages, has been released [176]. Slightly larger than the one of L. sakei 23K, this genome has 1,972 predicted protein-coding genes and 7 rRNA operons [176]. Compared to the L. sakei 23K genome [144], 1,618 genes are orthologous, but 250 seem to be unique to LS25, including a set of genes for carbohydrate metabolism, various transporters, and dehydrogenases/oxidoreductases [176].

Complete or draft genome sequences of Pediococcus pentosaceus and Pediococcus acidilactici strains, from diverse Korean fermented food products, have been released, but none isolated from meat products [177–179].

Genomes of several strains of starter CNS have also been published, namely, S. xylosus SMQ-121 [180], S. xylosus S04002 [181], and S. carnosus TM300 [182].

The draft genome sequence of S. xylosus SMQ-121 revealed the absence of genes coding for toxins or virulence factors. Furthermore, only four antibiotic resistance genes were found: two genes encode proteins that belong to the major facilitator superfamilies involved in phenicol and fluoroquinolone resistance; another gene encodes a putative aminoglycoside 3’-phosphotransferase for resistance to aminoglycosides; and the last one encodes trimethoprim resistance. Nevertheless, this strain was found to be sensitive to amikacin, chloramphenicol, ciprofloxacin, and trimethoprim [180].

A genome comparison of several S. xylosus meat starter cultures, including strain S04002, with other S. xylosus strains causing cow and goat mastitis, among others, has shown the presence of aroma compounds in S. xylosus S04002 [181].

S. carnosus TM300 genome has the highest GC content of all sequenced staphylococcal genomes [182]. It contains only one prophage and one genomic island characterised by a mosaic structure composed of species-specific genes. All starter cultures features, such as nitrate/nitrite reduction,
several sugar degradation pathways, two catalases, and nine osmoprotection systems, are present. It lacks most virulence factors, namely, the typical Staphylococcus aureus toxins, as well as biofilm formation genes, highlighting its nonpathogenic status [182].

Following the publication of the genome sequences of several strains, global approaches based on transcriptomics and proteomics have been developed in order to better understand the adaptation of starters to the meat environment and their interactions with the ecosystem and the meat substrate.

Genes involved in safety and technologically relevant properties of food associated CNS, such as antibiotic resistance, haemolysins, toxins, amino acid decarboxylases, binding proteins to extracellular matrix (ECM), lipases, proteases, stress response factors, and nitrate dissimulation, have been detected using DNA microarrays [183].

S. xylosus C2a strain response to nitrosative [184] or nutrients and osmotic stress [185] has been investigated through DNA microarrays. S. xylosus has been shown to counteract nitrosative stress by developing several oxidative stress resistance mechanisms, such as modulation of the expression of genes involved in iron homeostasis, detoxifying enzymes, and DNA and protein repairs [184]. S. xylosus adapted its metabolism to the meat nutrients and anaerobic conditions by simultaneously using glucose and lactate as carbon sources and by using meat peptides and amino acids. S. xylosus responded to the osmotic stress caused by the addition of salt (NaCl) by overexpressing genes involved in transport and synthesis of osmoprotectants, particularly glycine betaine, and Na⁺ and H⁺ extrusion [185]. To overcome the damaging effects of oxidative and nitrosative stress, staphylococci have developed protection, detoxification, and repair mechanisms controlled by a network of regulators [186].

Among the overexpressed proteins in S. xylosus biofilm, several related to exopolysaccharide biosynthesis were reported [187]. Furthermore, with overexpression of some proteins involved in amino acids metabolism, translation, and secretion, nitrogen metabolism appeared as quite active in sessile cells of S. xylosus. Additionally, protein secretion systems were also upregulated in biofilms, suggesting more active protein trafficking in sessile S. xylosus cells [187].

L. sakei 23K strain global transcriptome response during growth on ribose [188] and L. sakei La22 strain transcriptomic response to meat protein environment [189] have been studied using DNA microarrays.

The ribose uptake and catabolism in L. sakei 23K is highly regulated at the transcriptional level, and it is closely related to the catabolism of nucleosides. A global regulation mechanism seems to allow fine tuning of the expression of enzymes, which control the efficient use of available carbon sources [188].

Whole-genome DNA microarrays were used to analyse gene expression related to growth and survival of L. sakei La22, when grown in a sarcoplasmic (S) or myofibrillar (M) protein-supplemented chemically defined medium (CDM). Most genes related to peptides or amino acids metabolism were overexpressed in both mediums. Still, meat proteins do not represent a stressful environment for L. sakei La22 because no stress response genes were induced [189].

Next generation sequencing methods will improve knowledge related to microbiota and strain characterization involved in dry-fermented meat products. Future work must be done regarding these novel approaches and certainly novel vision of starter behaviour on particular products will be given.

4. Conclusions

The increasing knowledge and exigence level of consumers have forced the search for high value traditional meat products. Consequently, the number of production units (meat transforming) has increased, sometimes in low developed regions in a bewildered way.

The production of traditional meat products, namely, dry-fermented, dry-cured sausages, is still a very traditional and laborious process subjected in several cases to uncontrolled natural environmental conditions. This poses a problem to the producers since their meat products will not be uniform throughout time. Thus, it is necessary to find solutions contributing to the reproducibility of products characteristics. The use of starter cultures based on autochthonous microbiota selection may play here an important role. In fact, the use of these starters in sausages production may improve their sensorial characteristics and contribute to their biopreservation and safety, extending their shelf life, and to increased meat products uniformity.

Selected starter cultures provide a powerful tool for driving the fermentation of meat products, allowing desired quality and safety targets to be reached. Their use in meat fermentation results in acceleration of fermentation time, an improvement of safety (by reducing undesirable microorganisms), and a better quality of the final product. The selection of a starter culture should be carried out in the context of its application, since functionality will depend on the type of sausage, the technology applied, the ripening time, and the ingredients and raw materials used. Future knowledge will be gained with omics methods approach.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

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References


M. Bedía, L. Méndez, and S. Bañón, “Evaluation of different starter cultures (Staphylococci plus Lactic Acid Bacteria) in..."


