

## Processed meat with addition of Preservatives

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### Abstract

The Meat and the meat products are excellent sources of the nutrients for the humans. The Meat and the meat products also provide a favorable environment for the microbial growth. In order to prevent the microbiological contamination of the livestock foods, the synthetic preservatives, including the nitrites, the nitrates, and the sorbates, have been widely used in the food industry due to their low cost and the strong antibacterial activity. The Use of the synthetic chemical preservatives is recently being considered by the customers due to the concerns related to the negative health issues. The demand for the natural substances as the food preservatives has increased with the use of the plant-derived and the animal origin products, and the microbial metabolites. The natural preservatives inhibit the growth of the spoilage microorganisms or the food-borne pathogens by increasing the permeability of the microbial cell membranes, the interruption of the protein synthesis, and the cell metabolism. The Natural preservatives can extend the shelf-life and inhibit the growth of the microorganisms. The natural preservatives can influence the food sensory properties, including the flavor, the taste, the color, the texture, and the acceptability of the food. To increase the applicability of the natural preservatives, a number of the strategies, including the combinations of different preservatives or the food preservation methods, such as the active packaging systems and the encapsulation, have been explored. The applications of the natural preservatives for the meat and the meat products.

**Keywords:** the Meat and the meat products, the food preservation, the packaging, the food-borne pathogens.

### Introduction

The Food-borne pathogens, including the *Listeria monocytogenes*, the *Staphylococcus aureus*, the pathogenic *Escherichia coli*, the *Clostridium perfringens*, the *Campylobacter* spp., and the *Vibrio* spp., cause a large number of the illnesses, with substantial damage to the human health and the economy. The World Health Organization (the WHO), the food contaminated with the food-borne pathogens, the chemicals, and the allergens results in 600 million cases of the food-borne illness and approximately four hundred thousand deaths worldwide/year. Moreover, fifty-six million people die /year and approximately 7.7% of people worldwide suffer from the foodborne diseases. The Meat and the meat products are essential nutrient sources for the humans due to their excellent protein content, the essential amino acids, the vitamin B groups, and the minerals. The meat and the meat products also provide an appropriate environment for the spoilage microorganisms or the food-borne pathogens due to their high water activity and the nutrient factors (1,2,3,4,5,6 and 7). The food industry has advanced

worldwide, resulting in an enhanced threat of the food contamination by the pathogenic microorganisms, the chemical residues, the harmful food additives, and the toxins. The multiplication of the spoilage and the pathogenic microorganisms should be controlled to ensure the food safety. The food preservation techniques for protecting the food from the pathogenic microorganisms and extending the shelf-life include the chemical methods, such as the use of the preservatives; the physical methods, such as the heat treatment, the drying, the freezing, and the packaging; and the biological methods using the microorganisms that have an antagonistic effect on the pathogenic microorganisms and produce the bacteriocins. Among them, the addition of the food preservatives that inhibit the growth of the microorganisms is a widely used food protection technique. The countries in the world has different regulations for the food preservatives (8,9,10,11,12,13 and 14). The Synthetic preservatives have the advantage for the meat processing due to low cost, guaranteed the antibacterial effect or the shelf-life extending activity, and the little effect on the taste, the

flavor, the color, and the texture. However, the synthetic preservatives tend to be less preferred by the food consumers because of a number of health concerns regarding their side effects. The food consumers selected preservatives as the most concerned food additive owing to their negative impacts on the health. The Sorbic acid, the benzoic acid, and their salts have been reported to promote the mutagenic and the carcinogenic compounds. The Nitrites and the nitrate, used as preservative and coloring agents in the meat, have been associated with the leukemia, the colon cancer, the bladder cancer, and others. The Natural preservatives have emerged as alternatives to the synthetic preservatives. The Natural preservatives have shown potential to provide the effective antimicrobial activity while reducing the negative health effects. The Meat and the meat products containing the synthetic additives, are a major concern for the human health. Hence, the meat manufacturers and the researchers have begun to consider the use of the natural rather than the synthetic preservatives (15,16,17,18,19,20 and 21). Representatively, the 'clean label' food trends, including the meat and the meat products, began and possessed an important source of the food marketing. It includes consumer-friendly characteristics, such as the synthetic additive-free, the least processing, a brief list of the food ingredients, and the procedure of the traditional methods. The clean label food material market, including the natural preservatives, is likely to value, mostly owing to growing consumer requests for all the natural products. The natural preservatives such as the nisin, the natamycin, the  $\epsilon$ -polylysine, and the grapefruit seed extract are registered, but they are not approved for the meat products, or their concentration is not specified. The replacement of synthetic preservatives with the natural preservatives has major positive effects and is being accepted by the customers. The food producers also encounter challenges, including a decrease in price competitiveness due to the relatively high price of the natural preservatives and a decrease in the antibacterial effect due to the food ingredients, such as the carbohydrates, the proteins, and the lipids. In the case of the plant-derived substances, the standardization is problematic because of the influence of the country of origin, the soil, and the harvest seasons. The toxicity evaluation or identification of exact compounds for several plant-derived compounds contained in extracts and the essential oils have been performed. To solve these problems, various studies have been conducted to optimize the extraction process, combine other antimicrobial substances, apply active packaging, and encapsulate antibacterial substances to improve their utilization (22,23,24,25, 26, 27 and 28).

This review summarizes the current knowledge about the application of the natural preservatives for the meat and the meat products against the food-borne pathogens and the spoilage microorganisms.

The Application Technique of the Natural Preservatives to the Meat and the Meat Products

The Natural preservatives are manufactured in a variety of formulations including powder formed by drying methods and liquid forms such as essential oils. The Natural preservatives are directly added to the meat products and extend the shelf-life by inhibiting the bacterial growth. It is possible to increase the antibacterial effect of the natural preservatives through a combination of the other food processing methods (29,30,31,32,33,34 and 35). In the case of the plant-derived natural preservatives, it is necessary to consider the form applied to the food. The Natural preservatives are commonly prepared in the form of extracts using organic solvents, water, and essential oils. The plant extracts obtained from rosemary, chestnut, sage, cranberry, oregano, grape seed, and others have been used as the meat preservatives. Many studies have been conducted to apply the plant-derived substances to the meat products in the form of the essential oil because the antibacterial effect of essential oil type is better than that of extract type. However, it is difficult to apply large amounts of the essential oil to the food because of its distinct organoleptic properties. Recent developments have attempted to solve this problem by applying essential oils with other antibacterial substances. The advantage of this application is that it reduces the amounts of essential oils with strong flavor and increases antioxidant and antibacterial effects through synergistic effects. In terms of industrial perspective, if synthetic preservatives cannot be completely replaced with the natural preservatives, due to the industrial problems, such as increasing the economic costs or the complexity of the product manufacturing process, they could be replaced gradually by composing a mixed formulation of the synthetic preservatives and the natural preservatives (36,37,38,39, 40,41 and 42). The gamma irradiation and the high-pressure processing (the HPP) treatment are the physical food-processing methods that can further increase the antibacterial efficacy of the natural preservatives. The Unlike thermal food processing, these two food processing techniques could be used for the pasteurization of the raw meat because it has a minor effect on the food composition. In 1997, the WHO, the Food and Agricultural Organization (FAO), and the International Atomic Energy Agency (the IAEA) concluded that foods processed in the proper doses of the irradiation are nutritionally sufficient and safe to consume. The irradiation is permitted for the food preservation in more than sixty countries. Recent approaches in the food irradiation have involved the use of combined treatments with the natural preservatives to reduce irradiation doses. The gamma irradiation of medium doses (2–6 kGy) with the natural compounds and active packaging has been applied to extend the shelf-life of the meat and the meat products. The HPP is also a non-thermal technique for the food preservation that inhibits the growth of the microorganisms and maintains the natural properties of the food. The HPP is performed under high pressures (100–800 MPa) at mild temperature or the weak heating. The Previous studies have reported the potential capability of combining

the HPP and the natural preservatives including the essential oil and the antibacterial peptides in alleviating both the processing conditions of the HPP and the concentration of the natural preservatives while maintaining antibacterial effects (43,44,45,46,47,48 and 49). The Encapsulation is one of the effective approaches for expanding the applicability of the natural preservatives to the food. The encapsulation was performed with GRAS (generally recognized as safe) materials such as the alginate, the chitosan, the starch, the dextrin, and the proteins using the various techniques including the spray-drying, the extrusion, the freeze-drying, the coacervation, and the emulsification. The application of the natural preservatives to the meat is limited due to their characteristics, such as low solubility and the bioavailability, the rapid release, and the easy degradation. Moreover, the environmental conditions, such as the pH, the storage temperature and the time, the oxygen and the light exposures could influence the efficacy of the natural preservatives. Through encapsulation, the natural preservatives, especially hydrophobic compounds (e.g., the essential oil), could improve its stability and expand the versatility of the food processing while maintaining the antibacterial effect (50,51,52,53, 54,55 and 56). The Active packaging is an innovative packaging technology that allows for an interaction with the product and its environment to extend the shelf-life and to ensure its microbial safety while keeping the original properties of the packaged food. In relation to the European Union Guidance to the Commission Regulation (the EUGCR), active packaging is a type of the food packaging with a further beneficial function, while providing a protective barrier against the external influence. In the meat industry, the antimicrobial active packaging could be applied in several methods which are the incorporation of the natural preservatives into a sachet inside the packaging, the packaging film composition with the natural preservatives, the packaging coated with the natural preservatives onto the surface of the food, and use of the antimicrobial polymers as the packaging materials (57,58,59,60,61,62 and 63). The application of the microorganism-derived natural preservatives, known as the bio-preservation, in which the useful microorganisms or their antibacterial substances have antagonistic effects on pathogenic or spoilage microorganisms, are used is also a meat preservation method in the spotlight. This method is mainly involved in the lactic acid bacteria, the *Lactobacillus* spp., the *Leuconostoc* spp., the *Pediococcus* spp., and the *Lactococcus* spp., that have a GRAS status, widely participate in the fermentation processes, and produce the various antibacterial metabolites such as the organic acids, the hydrogen peroxide, and the bacteriocins. In terms of the application to the meat products, the bio-preservation methods included the direct inoculation with the lactic acid bacteria, which has an inhibitory effect on the spoilage or the pathogenic bacteria, the inclusion of bacterial strains producing antimicrobial substances in the fermentation starter, and treatment with the purified bacteriocins

(64,65,66,67,68,69 and 70).

### **The Natural Preservatives from the Plants and Their Application for the Meat and the Meat Products**

The antibacterial effect of the plant-derived natural preservatives is closely related to the polyphenols, the phenolics, and the flavonoids. The Plant-derived polyphenols have various classifications and structures, as the phenolic acids (the caffeic acid, the rosmarinic acid, the gallic acid, the ellagic acid, the cinnamic acid), the flavones (the luteolin, the apigenin, the chrysoeriol), the flavanols (the catechin, the epicatechin, the epigallocatechin, the galocatechin, and their gallate derivatives), the flavanones (the hesperidin, the hesperetin, the heridictyol, the naringenin), the flavonols (the quercetin, the kaempferol, the myricetin), the isoflavones (the geinstein, the daidzin, the formononetin), the coumarins (the coumarin, the warfarin, the 7-hydroxycoumarin), the anthocyanins (the pelargonidin, the delphinidin, the cyanidin, the malvidin), the quinones (the naphthoquinones, the hypericin), the alkaloids (the caffeine, the berberine, the harmaline), and the terpenoids (the menthol, the thymol, the lycopene, the capsaicin, the linalool) (71,72,73,74,75,76 and 77). The Polyphenols have been recognized for their effective antimicrobial properties. Although the antimicrobial mechanism has not yet been clearly elucidated, the cell membrane-disturbing molecules, such as the hydroxy group (OH-), which induces the leakage of intracellular components, inactivation of metabolic enzymes, and extinction of the adenosine triphosphate (the ATP) structure; direct pH change in the environment by the improvement in proton concentration, reduction of the intracellular pH by separation of acid molecules, and modification of the bacterial membrane permeability; an organic acid in the plant extracts may influence the oxidation of the nicotinamide adenine dinucleotide (the NADH), the eliminating, the reducing agent used in the electron transport system (78,79,80,81,82,83 and 84).

### **The Rosemary**

The Rosemary (the *Rosmarinus officinalis* L.) is a perennial herb with the woody, the aromatic, and the evergreen needle-like leaves. Originally from the Mediterranean region, it is broadly distributed throughout the globe. The Rosemary has been used as a spice and the flavoring agent in the food. The Rosemary essential oil is known to contain approximately fifteen kinds of the bioactive compounds. The principal compound was 1,8-cineole (35.32%). Other major compounds were the camphor, the  $\alpha$ -pinene, the trans-caryophyllene, the  $\alpha$ -thujone, and the borneol (85,86,87,88,89,90 and 91). The antibacterial effect of the rosemary ethanol extracts against the *L. monocytogenes* in the beef. The application of 45% rosemary ethanol extract



for the *L. monocytogenes* on the beef led to a 2 log colony-forming unit (CFU)/ gram reduction in the incubation at 4 °C for 9 days. In the chicken meat, the effect of the rosemary essential oil on the inhibition of the *Salmonella* Enteritidis and the spoilage protective effects at 4 and 18 °C was investigated. The 5 mg/mL of the rosemary essential oil induced the decrease in the coliform, the aerobic microorganisms, the lactic acid bacteria, and the anaerobic microorganisms at 18 °C for 24 hours. In Comparing with the untreated chicken meat, the reductions of 1.75 log CFU/ gram (the coliform), 0.87 log CFU/ gram (the aerobic microorganisms), 1.05 log CFU/ gram (the lactic acid microorganisms) and 1.28 log CFU/ gram (the anaerobic microorganisms) were observed in the group treated with rosemary essential oil at 18 °C. The Rosemary oil reduced the *S. Enteritidis* by more than 2 log CFU/ gram at 18 °C, but less than 1 log CFU/ gram at 4 °C (92,93,94,95,96,97 and 98). The rosemary essential oil applied with modified atmosphere packaging for the inhibition of the food-borne pathogens (the *S. Typhimurium* and the *L. monocytogenes*) in the poultry filets under the refrigerated conditions for 7 d was investigated. The 0.2% rosemary essential oil did not affect the sensory profile and inhibited the growth of both pathogens in laboratory media within 24 hours Treatment with 0.2% rosemary essential oil did not affect the reduction in the *S. Typhimurium*, but showed weak antibacterial activity against the *L. monocytogenes* until the first day of the storage (approximately 0.1 log CFU/ gram reduction compared to control) (99,100,101,102,103,104 and 105).

### The Sage

The Sage (*Salvia officinalis* L.), belonging to the Lamiaceae family, has been used since prehistoric eras because of its flavor, taste, therapeutic, and preservative properties. The Sage is known to contain considerable amounts of the rosemary acid, the *p*-coumaric acid, and the benzoic acid. The Sage essential oils, camphor, carvacrol, R(+) limonene, and linalool are the major components in terms of content (106,107,108, 109,110 and 111). The antibacterial effects of various sage preparations were assessed for low-pressure mechanically separated meat (MSM) in vacuum packaging stored at -18 °C for 9 months. The MSM from the chickens with the addition of the sage extracts inhibited the growth of all groups of the microorganisms (the mesophilic aerobic microorganisms, the psychrotrophic microorganisms, the *Enterobacteriaceae*, the coliforms, and the enterococci). The most effective antibacterial effect was exhibited by the 0.1% sage essential oil-treated groups (112,113,114,115,116 and 117). The antibacterial effect of the sage essential oil (0.625%) on the survival of the *L. monocytogenes* in the Sous-vide cook-chill beef stored in

the refrigerated storage (2 or 8 °C) for 28 days. The decrease of 1 log CFU/ gram of the *L. monocytogenes* was detected in the sage essential oil-treated groups compared to the control at 2 °C. Although exponential growth was observed from day 14, lower *L. monocytogenes* counts of approximately 1 log CFU/ gram were detected in the sage essential oil-treated samples stored at 8 °C (118,119,120,121,122 and 123).

### The Thyme

The Thyme (*Thymus vulgaris*) is a representative herb used together with the meat and the meat products. The application of the thyme in the meat products can elevate the antioxidant, the antibacterial, the shelf-life extension, and the sensory properties. In the meat sausage, the thyme essential oil inhibited 2.69 log CFU/ gram of coagulase-positive *Staphylococcus* and 4.41 log CFU/ gram of aerobic mesophilic microorganisms, respectively, at a concentration of 0.95% by mixing with 1% (w/w) powdered beet juice. Moreover, the sensory properties, odor, flavor, and overall acceptability improved (124,125,126,127,128 and 129). The 1% thyme oil led to the reduction in the *S. enterica* by 3 log CFU/ gram during the margination process with lemon juice and 0.5% *Yucca schidigera* extract in the raw chicken breast. The major composition of the thyme oil revealed 51.1% and 24.1% thymol and O-cymene, respectively. The antibacterial effects of thyme may be due to additive or synergistic effects with its major and/or minor components. The Thymol and its synergistic effect with other phenolic compounds, such as the carvacrol, the *p*-cymene, and the  $\gamma$ -terpinene, can change the permeability of the bacterial cell wall, leading to cell death (130,131,132,133,134 and 135). The Thyme essential oil encapsulated with the casein and the maltodextrin was evaluated for its antibacterial potential in the vitro and in the situ (the hamburger-like meat products). The encapsulated thyme essential oil showed the same minimum inhibitory concentration (0.1 mg/mL) against the *E. coli*, the *S. Typhimurium*, the *S. aureus*, and the *L. monocytogenes* as that of the unencapsulated thyme essential. In the treated groups with 1% (v/v) of the encapsulated thyme essential oil for the meat, the *E. coli* counts were decreased from 23 most probable number (MPN)/ gram to 0 MPN/ gram, which was similar to the conventional preservative (the sodium nitrate) used as a control until 14 days of the refrigerated storage (4 °C) (136,137,138,139,140 and 141).

### The Oregano

The Oregano (the *Origanum vulgare*) is regularly used in the Mediterranean foods. The oregano essential oil has recognized antibacterial and antioxidant properties for the extension of shelf-life. The antibacterial effects of the

oregano were due to two bioactive the polyphenols, the thymol and the carvacrol (142,143,144,145,146 and 147). The component of the oregano essential oil and its impact on the shelf-life of black wildebeest Biceps femoris muscles was investigated at 2.6 °C. The components of the oregano oil were the thymol, the carvacrol, the  $p$ -cymene, the  $\beta$ -caryophyllene, the  $\gamma$ -terpinene, the  $\alpha$ -humulene, and the  $\alpha$ -pinene; among them, the carvacrol (42.94%) and the thymol (17.40%) were the highest. The total viable counts and the lactic acid bacteria reached the spoilage limit (7 log CFU/ gram) after 3 days. The growth rates for the total viable counts and the lactic acid microorganisms in the treated group were 40% higher than those in the untreated groups (148,149,150,151,152 and 153). The combinatorial effect of the oregano essential oil with the caprylic acid was studied in the vacuum-packed minced beef. The addition of 0.2% oregano essential oil with 0.5% caprylic acid and 0.1% citric acid in the minced beef reduced the counts of the lactic acid microorganisms by 1.5 log CFU/ gram in vacuum packaging. The cell counts of the psychrotrophic microorganisms and the *L. monocytogenes* were reduced by more than 2.5 log CFU/ gram at 3 °C for 10 days. The Oregano essential oil inhibits the growth of the microorganisms by releasing volatile components during the drying process. It was reported that the addition of the oregano essential oil composed of the carvacrol (64.5%), the  $p$ -cymene (5.2%), and the thymol (2.9%) inhibited *S. Enteritidis* and *E. coli* in the beef drying process. For drying, a filter paper was soaked with oregano essential oil and placed in front of the fan of the drier. The beef samples were dried at 55 °C for 6 hours. Consequently, both microorganisms (*S. Enteritidis* and *E. coli*) were not detected after treatment with 3 mL of oregano essential oil (154,155,156,157,158 and 159).

### The Chestnut

The *Castanea crenata* was classified into the *Castanea* family and is a woody plant native to the East Asia, including the Korea and the Japan. The *Castanea sativa* is one of the most important *Castanea* families and the food resources of the European areas for long periods. Chestnut shells contain abundant phenols and hydrolyzable tannins. The chestnut inner shell extracts using ethanol exhibited antimicrobial effects against *C. jejuni* in the chicken meat at a concentration of 2 mg/mL. The polyphenol and flavonoid contents of chestnut inner shell ethanol extracts were  $532.96 \pm 3.75$  mg gallic acid/100 g and  $12.28 \pm 0.03$  mg quercetin/100 g, respectively (160,161,162,163,164 and 165). The influence of the chestnut extracts (the *Castanea sativa*) on the leaf, the bur, and the hull of the beef patties under refrigerated conditions ( $2 \pm 1$  °C) for 18 days to extend shelf-life. Among the

chestnut extracts from the leaf, the bur, and the hull, only the leaf extract at a concentration of 1000 mg/kg had weak antimicrobial activity. The lactic acid microorganisms and the *Pseudomonas* spp. were reduced by 0.37 log CFU/ gram and 0.33 log CFU/ gram at 7 days, respectively (166,167,168,169,170 and 171).

### The Grapefruit Seed Extract (the GSE)

The GSE is a by-product of the *Citrus paradise*. The GSE contains the various phenolic compounds and the flavonoids, such as the catechin, the citric acid, the naringenin, the procyanidin, and the epicatechin gallate. The GSE has been described to have a wide-ranging spectrum antimicrobial, the antiparasitic, and the antifungal activities. The Polyphenols in the GSE are unstable but can be chemically modified to become more stable using quaternary ammonium compounds, such as the benzethonium chloride, during the industrial procedure of the commercial GSE preparations (172,173,174,175,176 and 177). The bacteriostatic effect of the commercial GSE (the Citricidal) on the sous-vide chicken products against the *C. perfringens*. The cell numbers of the *C. perfringens* were consistently approximately 2.5 log CFU/ gram regardless of the treatment or the control groups until 9.5 h of stored at 19 °C; however, the storage of the control and 50 or 100 ppm GSE treated groups at 25 °C for more than 6 h resulted in fast growth rates of *C. perfringens*, showing 2–3 log CFU/ gram. GSE concentrations at 200 ppm inhibited the growth of the *C. perfringens* stored at 19 and 25 °C. The active packaging system for the inhibition of the food-borne pathogens used the mixed natural preservatives consisting of the GSE (80 mg/m<sup>2</sup>) with the cinnamaldehyde (200 mg/m<sup>2</sup>) and the nisin (60 mg/m<sup>2</sup>) was assessed for the beef storage. The Active packaging showed lower counts of the psychrotrophic and the anaerobic microorganisms compared to the control groups at 1–2 log CFU/ gram. The packaged beef samples with mixed natural preservatives showed a decrease in the *L. monocytogenes*, the *S. aureus*, and the *C. jejuni* for approximately 4.7 log CFU/ gram, 0.81 log CFU/ gram, and 3.1 log CFU/ gram compared to wrapped packaging at 28 days of the refrigerated storage, respectively. The *C. jejuni* was observed below the detection limit after 21 days of the storage (184,185,186,187,188 and 189).

### The Cinnamon

The Cinnamon is a native plant in Asia that is acquired from the inner bark of the genus *Cinnamomum*. Cinnamon contains several active compounds, such as the cinnamaldehyde, the eugenol, the cinnamyl acetate, the L-borneol, the  $\beta$ -caryophyllene, the caryophyllene oxide, the camphor, the L-bornyl acetate, the  $\alpha$ -terpineol, the  $\alpha$ -

cubebene, the  $\alpha$ -thujene, and the terpinolene. The cinnamon (*Cinnamomum cassia*) essential oils could inhibit the *L. monocytogenes* in the ground beef at refrigerated (0 and 8 °C) and frozen (−18 °C) conditions. The concentration of 5.0% cinnamon essential oil to decrease by 3.5–4.0 log CFU/ gram of *L. monocytogenes* at 0 and 8 °C for 7 days. Under frozen conditions, *L. monocytogenes* was reduced by 3.5–4.0 log CFU/ gram over 60 days. The antibacterial effect and shelf-life extending activity were evaluated using a chitosan edible coating containing 0.6% cinnamon essential oil on the roast duck slices under modified atmosphere packaging (30% carbon dioxide (CO<sub>2</sub>)/70% nitrogen (N<sub>2</sub>)) at the storage at 2 ± 2 °C for 21 days. The edible coating with cinnamon essential oil showed total viable counts reduced by 1 log CFU/ gram compared to the control after 14 days of storage. This was similar to the results of *Enterobacteriaceae* counts. The number of the lactic acid microorganisms was lower than that of the control until the day 7 of the storage, but there was no significant difference from day 11 of the storage. The growth of the *Vibrio* spp. was delayed using edible coating with the cinnamon essential oil within the earlier period of the storage as a result of the microbial diversity sequencing (196,197,198, 199, 200 and 201).

#### The Turmeric

The Turmeric (*Curcuma longa* L.) has long been used as a flavor and the color agent in the food and traditional medicine to treat the various diseases, mainly in the South and East Asia. The main active compounds of the turmeric originate from its constituents, called the curcuminoids. The Curcuminoids (the curcumin, the demethoxycurcumin, and the bis-demethoxycurcumin) content of the turmeric varies between about 2–9% based on its growth environments, such as the cultivar, the soil, and the climatic conditions. The antibacterial effect of the turmeric on the chicken breast meat was assessed for the *E. coli* and the *S. aureus* stored at 4 °C for 48 hours. When 1% turmeric powder was added, no difference in the *S. aureus* counts was observed between the turmeric treated and the control groups. In the case of the *E. coli*, a reduction of 0.2 log CFU/ gram was observed, but this was not statistically significant (202,203,204,205, 206 and 207). The chicken meat was treated with the turmeric powder and the gamma irradiation to improve the meat quality and stability. The total aerobic microorganisms and the coliforms were completely decontaminated with 3% turmeric powder and 2 kGy of the gamma irradiation at 4 °C for 14 days. The microbial characteristics of the edible coatings using the turmeric starch and the bovine gelatin were examined in the frankfurter sausages. The edible coating was developed with a 5% (w/w) aqueous solution of the turmeric starch and

the gelatin. The microbial growth of the coated sausages stored at 5 °C for 20 days decreased by 2.21, 1.01, and 1.65 log CFU/ gram for the mesophilic microorganisms, the lactic acid microorganisms, and the psychotropic microorganisms, respectively. At 10 °C, the decreases were 1.57, 2.14, and 1.99 log CFU/ gram for the mesophilic microorganisms, the lactic acid microorganisms, and the psychotropic microorganisms, respectively (208,209,210,211,212 and 213).

#### The Plant-Derived Antimicrobial Peptides (the AMPs)

The Plant-derived AMPs have been studied for their potential to inhibit the different pathogens, including the food spoilage microorganisms, the food poisoning microorganisms, the mold, and the yeast species. The antibacterial peptide Leg1 from the chickpea legumin has been reported in the meat application of the plant-derived the AMPs. The Raw pork was pretreated with Leg1 and inoculated with the *E. coli* and the *B. subtilis*. The bactericidal activity was measured at 37 °C for 16 hours. The minimum bactericidal concentrations of Leg1 on the pork meat were 125 µM and 15.6 µM for the *E. coli* and the *B. subtilis*, respectively. This was the same concentration as the MBC of the nisin, the bacteriocin from the *Lactococcus lactis*, for the tested strains. The AMPs from pea (the 11SGP) and the red kidney bean (the RBAH) were used to extend the shelf-life of the raw buffalo meat. In the laboratory media, the Gram-positive (the *L. monocytogenes*, the *B. cereus*, and the *Streptococcus pyogenes*) and the Gram-negative (the *E. coli*, the *Pseudomonas aeruginosa*, the *Acinetobacter baumannii*) microorganisms were inhibited by 11GSP (60 µg/mL) and the Gram-negative microorganisms by 60% and the Gram-positive microorganisms by 90%. RBAH (60 µg/mL) alleviated the growth of the Gram-negative microorganisms by 56% and the Gram-positive microorganisms by 85%. In the buffalo meat, the counts of the mesophilic microorganisms of 11SGP (400 µg/ gram) and the RBAG (400 µg/ gram) treated groups decreased by 1.60 log CFU/ gram and 1.94 log CFU/ gram compared to the control groups. The psychophilic microorganisms, 11SGP and the RBAG reduced by 1.10 log CFU/ gram and 1.47 log CFU/ gram, respectively, after 15 d of the refrigerated storage (4 °C) (172,173,174,175 and 176).

#### The Natural Preservatives from the Animals and Their Application for the Meat and the Meat Products

The Various antibacterial systems of the animal sources are associated with the defense mechanisms against external intruders. The preservatives derived from the animal sources include the lysozymes, the lactoferrin, the ovotransferrin, the lactoperoxidase, the AMPs from the



livestock animals, and the polysaccharides. The Lysozyme can suppress several Gram-positive microorganisms because of the Lysozyme distinctive ability to injure bacterial membranes by hydrolyzing the 1,4- $\beta$ -linkage between the N-acetyl-D-glucosamine and the N-acetylmuramic acid of the peptidoglycan in the bacterial membrane. The Peptide-based antibacterial substances containing the AMPs from the animal sources, the ovotransferrin, and the lactoferrin could influence the cell membranes or the synthesize ATP, the peptides, and the enzymes. The antibacterial mechanism of the AMP has been reported to attach to the bacterial cell membrane and disturb its integrity, resulting in the cell lysis. The AMPs may also exert more complex activities that inhibit the metabolic and the translational systems. The ovotransferrin isolated from the eggs increased the cell membrane permeabilization of the Gram-positive and the Gram-negative microorganisms. The ovotransferrin destroyed the cell membrane integrity, increased the permeability of the pathogen membranes, and induced morphological changes. The Lactoferrin has antibacterial effects related to the large cationic patches present on the surface and the iron impoverishment. The Lactoferrin has an antibacterial effect only when in its iron-free state and the iron-saturated lactoferrin has a limited antimicrobial activity. The Lactoperoxidase oxidizes the sulfhydryl groups of the proteins present in the bacterial membrane, which could be injured by the efflux of the potassium ions, the amino acids, the peptides, and the enzymes (177,178,179,180,181 and 182).

### The Lysozyme

The Lysozyme (the muramidase or the N-acetylmuramichydrolase) is mainly extracted from the hen egg whites and is known as an antimicrobial enzyme. The Lysozyme is a glycoside hydrolase that hydrolyses the linkages in the peptidoglycan at the Gram-positive bacterial cell wall. The Lysozyme is composed of 129 amino acids, which contain the disulfide bonds and the tryptophan, the tyrosine, and the phenylalanine residues. The Lysozyme has been used commercially, named the Inovapure, against the spoilage microorganisms and the food-borne pathogens to prolong the shelf-life of the raw and the processed meat. The Modified lysozyme, the high hydrophobicity, and the low hydrolytic activity compared to the lysozyme monomer, at the concentrations of 5%, exhibited low microbial growth rates (the total viable count 4.59 log CFU/cm<sup>2</sup>; the molds and the yeasts 2.17 log CFU/cm<sup>2</sup>) in the pork meat surface with the modified atmosphere packaging with composites of 50% O<sub>2</sub>, 40% CO<sub>2</sub>, and 10% N<sub>2</sub>. The mixed antimicrobials consisting of the lysozyme (250 ppm), the nisin (250 ppm), and the disodium

ethylenediaminetetraacetic acid (the EDTA) (20 mM) had antibacterial effects against the *L. monocytogenes*, the total viable counts, the *Enterobacteriaceae*, the *Pseudomonas* spp., and the lactic acid microorganisms in the ostrich meat patties with the air and the vacuum packaging. The mixed lysozyme preparations reduced the *L. monocytogenes* below the official detection limit of the EU (<2 log CFU/ gram) in the ostrich meat patties. The treated samples showed a decrease in the total viable counts by 1 log CFU/ gram after 2 days of the storage and tended to increase thereafter. The *Enterobacteriaceae* and the *Pseudomonas* spp. were not affected by the mixed antimicrobials in either the packaging atmosphere, and the reduction in the lactic acid microorganisms was detected at 2 log CFU/ gram. The combination of the lysozyme with the chitooligosaccharide presented a more effective antibacterial effect against the Gram-negative microorganisms than the lysozyme alone. In the minced lamb meat, the mixture of the lysozyme and the chitooligosaccharide led to complete removal of 3–4 log CFU/ gram of the inoculated *E. coli*, *Pseudomonas fluorescens*, and *B. cereus* during 4 hours at the ambient temperature. The *S. aureus* was not completely eliminated, but was reduced up to 2 log CFU/gram (183,184,185,186 and 187)

### The Ovotransferrin

The Egg white contains 13% ovotransferrin (the conalbumin), which is a monomeric 77.9 kDa glycoprotein comprised of 686 amino acid residues. The ovotransferrin contains N- and C- globular parts, each of which can reversibly Fe<sup>3+</sup> and CO<sub>3</sub><sup>2-</sup>. The ovotransferrin is the main constituent of the egg's defense system for the microorganisms, as it renders the iron unusable for the microbial growth within the albumen. The antimicrobial effects of the ovotransferrin against the *E. coli* in the fresh chicken breast involved in  $\kappa$ -carrageenan film. The growth of the *E. coli* in the fresh chicken breast wrapped with the active film was 2.7 log CFU/ gram by the addition of 25 mg of the ovotransferrin in combination with 5 mM EDTA. The ham models, 25 mg/mL of ovotransferrin with 100 mM sodium bicarbonate (NaHCO<sub>3</sub>) did not show any antibacterial effects against the *E. coli* O157:H7 and the *L. monocytogenes* in commercial hams, whereas 25 mg/mL ovotransferrin with 0.5% citric acid had bacteriostatic effects against *L. monocytogenes* (188,189,190,191,192 and 193).

### The Lactoferrin

The Lactoferrin, a glycoprotein that belongs to the transferrin protein family in milk and milk products as well as neutrophil granules and exocrine secretions in mammals,

was able to bind iron within the cells. The ability of this 80 kDa protein to control free iron levels contributes to its bacteriostatic and health-beneficial characteristics, such as stimulating bone growth, protecting the intestinal epithelium, and promoting the immune system in animals. In the ground beef, application of the active lactoferrin, the immobilized lactoferrin with the glycosaminoglycans, and solubilized in citrate/bicarbonate buffer systems at concentrations of 3% and 5% resulted in 2 log CFU/ gram reductions of *E. coli* O157:H7 at 10 °C for 9 days. The reduction of the *S. Enteritidis* growth was 0.8 log CFU/ gram when the active lactoferrin concentration was increased to 2.5%. A single application of 0.5% active lactoferrin reduced *L. monocytogenes* in the beef, resulting in 2 log CFU/ gram. The Bovine lactoferrin (0.5 mg) was tested against the *E. coli* O157:H7 and *P. fluorescens* inoculated on the chicken with HPP treatments between 200 and 500 MPa for 10 min at 10 °C. As a result, the *P. fluorescens* was decreased when the lactoferrin was combined with the HPP treatment at 300 MPa for 2.3 log CFU/ gram additional reduction compared to only 300 MPa treatment on day 9. Additional reductions in the *E. coli* O157:H7 counts obtained by combined treatments remained below 0.5 log CFU/ gram. (194,195,196,197 and 198)

### The Lactoperoxidase

The Lactoperoxidase is a member of the peroxidase family. It is a ubiquitous active enzyme in bovine milk, which has antimicrobial effects. Bovine lactoperoxidase is a glycoprotein that contains a peptide chain of 78.4 kDa and catalyzes the oxidation of thiocyanate ions (SCN-) in lactoperoxidase, producing oxidizing products, such as hypothiocyanite and hypothiocyanous acid. The lactoperoxidase coated with alginate at concentrations of 2, 4, and 6% on the shelf-life of the chicken breast filets. The chicken samples with active coating of alginate and 6% lactoperoxidase showed a reduction of *Enterobacteriaceae*, *P. aeruginosa*, and aerobic mesophilic microorganisms by approximately 5 log CFU/ gram, 4 log CFU/ gram, and 2.5 log CFU/ gram at 16 days of refrigerated storage, respectively. The antimicrobial effects of lactoperoxidase were also assessed against *L. monocytogenes* and *S. Enteritidis* in sliced dry-cured-ham for 60 d at 8 °C treated with HPP at 450 MPa. The synergistic effect of lactoperoxidase and pressure was confirmed as *S. Enteritidis* decreased below the detection limit (1 log CFU/ gram). For *L. monocytogenes*, the synergistic effect reduced cell viability by 0.86 log CFU/ gram compared with untreated samples at the end of storage. In the beef, the effect of the lactoperoxidase on the growth of the inoculated pathogens (4 log CFU/ gram)

composed of *S. aureus*, *L. monocytogenes*, *E. coli* O157:H7, *S. Typhimurium*, *P. aeruginosa*, *Yersinia enterocolitica*, and indigenous microbiota was investigated. All pathogens used in the experiment were reduced compared to the control at a chilling regime (−1 to 12 °C) for 42 days. The total aerobe and *Pseudomonas* spp. increased less in the lactoperoxidase treated group than in the control group, but the antibacterial effect was not exhibited for anaerobes and lactic acid microorganisms (199,200,201,202 and 203).

### The Livestock Animal-Derived AMPs

The Livestock animal-derived products have been used as a source of AMPs. Among these by-products of livestock, blood, bones, collagen, gelatin, liver, lungs, placenta, skin, and visceral mass are important sources of AMPs, as well as muscle parts. The bovine cruor, a slaughterhouse byproduct containing mainly hemoglobin, broadly described as a rich source of fibrin proteins, was investigated for the extraction of AMPs. The fraction named  $\alpha$ 137–141 (polypeptide with five components, Thr-Ser-Lys-Tyr-Arg), a small (0.65 kDa), and hydrophilic AMPs deviated from hemoglobin. The  $\alpha$ 137–141 preservative (0.5%, w/w) had bacteriostatic effects on the total microbial population, coliform microorganisms, yeasts, and molds at 4 °C for 14 d on beef. The AMPs isolated from porcine leukocytes had antibacterial effects on the proliferation of *S. aureus* and *E. coli* inoculated in the ground meat (the boneless ham) and the sausage minces. The 20  $\mu$ g/ gram AMPs decreased by 1.3 log CFU/ gram of *S. aureus* and 1.5 log CFU/ gram of *E. coli* in ground meat. It was also achieved that 160  $\mu$ g/ gram of AMPs had the best inhibition and decreased in 3.9 log CFU/ gram of *S. aureus* and 3.3 log CFU/ gram of *E. coli* at 6 hours in the ground meats. In sausage mince, the AMPs at concentrations of 160  $\mu$ g/ gram could decrease by 3 log CFU/g of *S. aureus* and 2.7 log CFU/ gram of *E. coli* at 12 hours. After 24 hours of storage, no visible colonies of *S. aureus* or *E. coli* were detected in the sausage mince (203,204,205,206,207 and 208).

### The Natural Preservatives from the Microorganism and Their Application for the Meat and the Meat Products

The Lactic acid microorganisms (the LAB) strains secrete several bacterial growth inhibitory substances (the organic acids, the diacetyl, the phenyl-lactate, the hydroxyphenyl-lactate, the cyclic dipeptides, the hydroxy fatty acid, the propionate, and the hydrogen peroxide), the bacteriocins (the nisin, the acidophilin, the bulgaricin, the helveticin, the lactacin, the pediocin, the plantarim, the diplococcin, and the bifidocin), and the bacteriocin-like inhibitory substances (the BLIS), which exhibit antibacterial activity and can control the spoilage microorganisms and the food-borne



pathogens. Among various bacteriocins, commercial bacteriocin preparations have been applied using nisin and pediocin. Bacteriocins are peptides or proteins with antibacterial and antifungal effects that produce microorganisms, mainly lactic acid microorganisms. These compounds are considered potential natural preservatives because of their inhibitory effects on the food spoilage microorganisms or pathogens. LAB bacteriocins vary in accordance with molecular size, the chemical structure, modifications during biosynthesis, presence of modified amino acid residues, and antimicrobial mechanisms. The LAB bacteriocins can be categorized into two major classes: class I (lanthionine-containing antibiotics) with three subclasses (Ia, Ib, and Ic) and the class II with four subclasses (IIa, IIb, IIc, and IId). The Class I bacteriocins generally include 19–50 amino acid residues (<5 kDa) and are largely post-translationally modified, ensuring the non-standard amino acids, such as the lanthionine, the  $\beta$ -methyllanthionine, and the labyrinthine. The class I bacteriocins are further subdivided into the class Ia (the lantibiotics), the class Ib (the labyrinthopeptins), and the class Ic (the sanctibiotics). The Class II bacteriocins comprise the small, heat-stable, the non-modified peptides (<10 kDa). It can be further subdivided into the class IIa (the pediocin-like bacteriocins), the class IIb (the non-modified bacteriocins with two or more peptides), the class IIc (the circular bacteriocins), and the class IId (the non-pediocin-like bacteriocins). The Pediocin-like bacteriocins (the class IIa) can be regarded as the main subgroup among all classified the LAB bacteriocins. The Class III bacteriocins are classified as the high molecular weight (>30 kDa) and the thermally unstable peptides. The Class IV bacteriocins are large peptides complexed with the lipids or the carbohydrates. The bacterial cell surface exhibits a negative charge because the anionic characteristics of the cell membrane consist of the phosphatidylethanolamine, the phosphatidylglycerol, the lipopolysaccharide, the lipoteichoic acid, and the cardiolipin, and is generally captured by the positively charged bacteriocins. The cationic charged groups of the bacteriocins electrostatically interact with the anionic bacterial cell surface, while the hydrophobic surfaces are attached to the membrane and traverse the lipid bilayer. The bacteriocins self-associate or polymerize to develop complexes after passing through the lipid bilayer. The bacteriocins induce the cell death by increasing the permeability of the bacterial membrane, forming pores that cause dissipation of the proton motive force, exhaustion of ATP, and leakage of intracellular substrates. Gram-positive microorganisms -derived bacteriocins only perform for Gram-positive microorganisms and are not effective against Gram-negative microorganisms because of their different

membrane compositions and selective membrane permeability. These disadvantages could be compensated by mixing processing with other preservatives and the application of further preservation methods (209,210,211,212 and 213).

### The Nisin

The Nisin is the most representative class I bacteriocin. The Nisin is produced by several strains of the *Lactococcus lactis*, a species that is widely used for the dairy production. The Nisin was first approved as a food preservative in the United Kingdom in the 1950s and is now widely used worldwide and is permitted in over 50 countries. The structure of the nisin consists of a polypeptide with 34 amino acids, a 3.5 kDa molecular mass, and contains the methyllanthionine and the lanthionine groups. The Nisin has antimicrobial activities against a wide range of the Gram-positive microorganisms, including the *Staphylococcus* spp., the *Bacillus* spp., the *Listeria* spp., and the *Enterococcus* spp. The Nisaplin is a typical commercial nisin formulation. The Nisin could provide long-lasting bacteriostatic effects on the pathogenic microorganisms in the beef jerky at room temperature. The shelf-life extensive effect of the nisin in the *B. cereus* inoculated with the beef jerky. In the beef jerky without the nisin, the counts of the mesophilic microorganisms and the *B. cereus* increasing is unlikely for the beef jerky treated with the nisin at 25 °C for 60 days. The *B. cereus* grew after 3 days in the 100 IU/ gram. The treated groups and after 21 days in the 500 IU/ gram nisin-treated groups. The nisin-containing fermentate from the *L. lactis* 537 strain was evaluated for the inhibition of the *L. monocytogenes* in ready-to-eat (RTE) sliced ham. The addition of the fermentate to the RTE sliced ham led to an immediate decrease in the *L. monocytogenes* counts from 3 log CFU/ gram to below the detection limit stored at 4 °C (20 CFU/ gram). The Nisin with the cinnamaldehyde and the grapefruit seed extract presented synergistic antibacterial effects. It reduced the counts of the *L. monocytogenes* by 3 log CFU/ gram in the raw pork loin at 4 °C for 12 hours. The minimum inhibitory concentration of the nisin against *L. monocytogenes* was 250 ppm in laboratory media, but it was possible to reduce the concentration of 5–6 ppm against the growth of *L. monocytogenes* by mixing with the natural antibacterial substances in the pork (214,215,216,217,218 and 219).

### The Pediocin

The *Pediococcus* spp., *Pediococcus acidilactici*, and *Pediococcus pentosaceus* are the main pediocin-producing strains. Pediocin was classified into the bacteriocin group class IIa, characterized as small non-

modified peptides (<5 kDa) comprising less than 50 amino acids.. Remarkably, pediocin showed antimicrobial activity even at nanomolar concentrations. The Food grade pediocin-containing formulations are commercially available and marketed as ALTA 2341 and MicroGARD. The Pediocin has been studied for the inhibition of the *Listeria* spp. for the meat preservation. The antibacterial activities of pediocin PA-1 in the frankfurters and the *P. acidilactici* MCH14, the pediocin PA-1 producing strain, in the Spanish dry-fermented sausages were assessed against the *L. monocytogenes* and the *C. perfringens*. In frankfurters treated with 5000 bacteriocin units (BU)/mL of the pediocin PA-1 produced by the *P. acidilactici* the MCH14, the *L. monocytogenes* was reduced by 2 and 0.6 log CFU/ gram after the storage at 4 °C for 60 days and at 15 °C for 30 days, respectively. The *C. perfringens* decreased with 5000 BU/mL of the pediocin PA-1 by 2 and 0.8 log CFU/ gram after the storage at 10 °C for 60 d and at 15 °C for 30 days, respectively. The growth of the *L. monocytogenes* was inhibited by the pediocin-producing strain, the *P. acidilactici* MCH14, in the Spanish dry-fermented sausages at 2 log CFU/ gram compared to the control. The bacHA-6111-2, the pediocin from the *P. acidilactici* HA-6111-2, was applied to the Portuguese fermented meat sausage (the Alheira) with the HPP treatment (300 MPa, 5 min, 10 °C) to inhibit the *Listeria innocua*. The bacteriostatic effect was verified for high inoculation counts of the *L. innocua* at 4 °C for 60 days. For lower the inoculated *L. innocua*, antibacterial effect was observed below 2 log CFU/ gram from day 3 of storage until the end of storage. The antibacterial activities of a mixed preparation containing pediocin from the *P. pentosaceus* and *Murraya koenigii* (curry tree) berries in a raw goat meat emulsion at 4 °C for 9 days. The *L. innocua* was reduced for 4.1 log CFU/ gram in the treated samples concentrations at 8.3 mL pediocin/1000 grams of the meat emulsion with 10% (v/w) *Murraya koenigii* berries extract at the end of the storage. The total viable count and the psychrophilic count were also observed lower in the treated samples, 2.2 log CFU/gram and 1.6 log CFU/ gram, respectively (220,221,222,223,224 and 225).

### The Sakacin

The Sakacins, a class II bacteriocin, are mainly produced by the *Lactobacillus sakei* or the *Lactobacillus curvatus* strains. The Commercial sakacin products are currently not presented. Compared to the nisin and the pediocin, the sakacins have a relatively narrow antimicrobial spectrum, especially with effective inhibition against the *Listeria* species. The antibacterial effect of the sakacin-producing strain, the *L. sakei* CWBI-B1365, and the *L. curvatus* CWBI-B28, on the fate of the *L.*

*monocytogenes* in the raw beef and poultry. In the refrigerated (5 °C) the raw beef, the *L. sakei* induced a decrease in the *L. monocytogenes* concentration by 1.5 log CFU/ gram after 7 days to 2 log CFU/ gram after 14 days, and below the detection limit at 21 days. The addition of the *L. curvatus* reduced the *L. monocytogenes* to below the detection limit after 7 days. However, in the poultry, the bacteriocin-producing strain did not affect the inhibition of the *L. monocytogenes*. It was assumed that the type of the meat may have influenced the bacteriocin production by the LAB. The antibacterial activity of different bacteriocin preparations using the sakacin Q produced by the *L. curvatus* ACU-1 on the meat surface was evaluated against *L. innocua*. The freeze-dried reconstituted cell-free supernatant (3200 AU/mL) was effective for the inhibition of *L. innocua* on the meat surface, decreasing its bacterial cell number to the detection limit (<2 log CFU/ gram) after 2 weeks of storage at 4–5 °C. The adsorption of the sakacin Q to the meat products, the main ingredients, the meat proteins, and the fat tissues did not affect its antibacterial activity (226,227,228,229,230 and 231).

### The Bacteriocin-Like Inhibitory Substance (BLIS)

The BLIS are among the antimicrobial substances produced by microorganisms and are not completely categorized in terms of amino acid composition, molecular size, and nucleotide sequence. In RTE pork ham, the antibacterial effects of BLIS produced by *P. pentosaceus* American Type Culture Collection (ATCC) 43200 were assessed and compared with those of commercially available nisin preparations (the Nisaplin). BLIS showed effective antibacterial activity against the *Listeria seeligeri* by 0.74 log CFU/ gram in the RTE ham stored at 4 °C after 2 days. However, a slight increase in the *L. seeligeri* counts was detected in the BLIS-treated samples from 6 days to the end of storage. Nisaplin did not present any antibacterial effect for up to 2 days. After 2 days, Nisaplin started to induce a decrease in the *L. seeligeri* counts throughout the refrigerated storage. This might have been due to the higher sensitivity of BLIS to residual proteases compared to the nisin, thus weakening its antibacterial effect. The BLIS-producing the LAB strains, the *P. acidilactici* KTU05-7, *P. pentosaceus* KTU05-9, and the *L. sakei* KTU05-6, were used to ferment the plant (the Jerusalem artichoke, the *Helianthus tuberosus* L.), and 5% of the fermented products were tested to inhibit the food-borne pathogen at 18 °C for 12 hours in the ready-to-cook (the RTC) minced pork. The *P. acidilactici* fermented product presented the highest antimicrobial activity compared to the other strains. The counts of the *E. coli*, the *Enterococcus faecalis*, the *S. aureus*, and the *Streptococcus* spp. were reduced by 5.53, 4.37, 4.86, and 3.84 log CFU/ gram, respectively, compared

to the control groups, suggesting that the fermented product of the BLIS-producing strains showed an enhanced antibacterial effect. The BLIS obtained from *Enterococcus faecium* DB1 inhibited the growth and formation of biofilms of *C. perfringens* in the chicken meat. The 2.5 mg/mL of DB1 BLIS suppressed the growth of the *C. perfringens* by approximately 30%. The *C. perfringens* growth was inhibited by 50% at 5 mg/mL The DB1 BLIS. Biofilm formation by the *C. perfringens* treated with 5 mg/mL DB1 BLIS was radically reduced by approximately 90% at 4 °C for 72 hours compared to the control groups. The 2.5 mg/mL of the DB1 BLIS also inhibited biofilm formation by the *C. perfringens* under the same conditions. The BLIS could inhibit the formation of the *C. perfringens* biofilms on the chicken surfaces due to its antibacterial effect (232,233,234,235,236 and 237).

### Other Microorganism Sources

The mytichitin-CB peptide, which was isolated from the blood lymphocytes of the *Mytilus coruscus*, showed antibacterial effects against the Gram-positive microorganisms and fungi. The mytichitin-CB peptide expressed by *Pichia pastoris* and applied it to the pork preservation. The total viable counts of the treated group with 6 mg/L of mytichitin-CB derived from the *P. pastoris* was reduced by 33% (1–2 log CFU/ gram) compared to the control group after the storage at 4 °C for 5 days. The Mytichitin-CB effectively inhibited the total bacterial growth during the storage compared to the groups treated with 50 mg/L of the nisin. The Mytichitin-CB at 6 and 12 mg/L suppressed *Staphylococcus* spp. and *Escherichia* spp., respectively, with a reduction of 1–2 log CFU/ gram, respectively. The *Listeria* spp. and the *Pseudomonas* spp. were not detected during the storage, unlike the control and nisin-treated groups. The Hispidalin is a unique AMP derived from the seeds of the *Benincasa hispida* and has been shown to exhibit the antimicrobial effects against the various microorganisms. The hispidalin expressed by the *P. pastoris* was used as a preservative for the pork meat. The Pork meat treated with 100 µg/mL hispidalin showed bacteriostatic effects during the entire refrigerated storage period. The total viable count of the pork with 100 µg/mL hispidalin was 1 log CFU/gram lower than that of the control group at 4 °C for 7 days (238,239,240, 241, 242, and 243).

### Conclusions

The Meat and the meat products are excellent nutrient sources due to their abundant protein content, the essential amino acids, the vitamins, and the minerals. The meat and meat products are susceptible to the contamination by the food-borne pathogens and the various spoilage

microorganisms because of their high water activity and the nutrient content. The application of the preservatives is an indispensable element in the livestock food industry to prevent the food poisoning, delay the spoilage, and extend their shelf life. The Industrial preservatives, commonly made up of the synthetic chemicals, are not demanded by the food customers because of their negative health concerns. The natural preservatives derived from the plants (the rosemary, the sage, the chestnut, the GSE, and the tumeric), the animals (the lysozyme, the lactoferrin, the lactoferoxidase, the ovotransferrin, and others), and the microorganisms (the organic acids, the bacteriocins, and the BLIS) have been explored as alternatives to the synthetic chemical preservatives. The versatility of the natural preservatives compared to the synthetic preservatives is limited due to the production cost, the standardization, the insufficient toxicity studies, and the negative sensory effects on the food. To compensate for these disadvantages, various applications have been studied for their synergistic effect with the other natural preservatives with reduced the application concentrations compared to single use, the application of the physical treatment (the gamma irradiation, the high pressure processing, and the drying), the encapsulation, and the possibility of the packaging materials. The various natural preservatives and the application methods to inhibit the growth of the food-borne pathogens and the spoilage microorganisms in the livestock foods. The Natural preservatives are expected to be in high demand due to the consumer and the industrial requests. Therefore, it is necessary to explore various applications of the existing natural preservatives, while continuously searching for the novel ones.

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