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Pros and cons of carcass decontamination: The role of the European Food Safety Authority

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Abstract

Various intervention strategies to control foodborne pathogens have been identified and applied through the whole food chain. Physical, chemical, biological treatments applied alone or in combination have been studied and proved to reduce the number and the prevalence of bacterial contamination of meat surfaces such as carcasses. The various treatments have their own advantages and disadvantages. In EU, chemical decontamination was not permitted until the recent revision of European food hygiene legislation which allows the use of substances other than water for the removal of meat surface bacterial contamination. The European Commission will authorise the use of such substances after the European Food Safety Authority (EFSA) has provided a chemical and a microbiological risk assessment. For this purpose, EFSA issued a guidance document which points out the major components and prerequisites that a study/dossier should contain in order to prove that the substance intended to be used for the removal of microbial surface contamination of foods of animal origin (i) would not pose any appreciable risk to the public health (safety or chemical assessment) and (ii) would result in a significant reduction of the prevalence and the numbers of pathogenic target bacteria when compared to the control and when this reduction is at the same time of relevance to human health (efficacy or microbiological risk assessment). The current paper deals only with microbiological safety issues.

Keywords: Chemical decontamination; Physical decontamination; Biological decontamination; Foodborne pathogens; EFSA; Microbiological risk assessment

1. Introduction

Food crises occurring over the last 20 years in combination with international trade led the international community, national food safety laws and subsequent decision-making practices to be based on risk analysis – a concept consisting of risk assessment, risk management and risk communication. The ultimate scope is to provide the highest level of protection of human health and to facilitate both the domestic and the international food trade. Indeed, at international level, a risk analysis approach is fundamental for the implementation of the World Trade Organisation (WTO) Sanitary and Phytosanitary Agreement (SPS Agreement) (WTO, 1995). At European level, Regulation (EC) No. 178/2002 (OJEU, 2002) sets the general principles and requirements of food safety law including the need of a risk analysis approach, and it also establishes the European Food Safety Authority (EFSA). EFSA, as an independent scientific body in risk assessment, provides scientific and technical support to the Community institutions and Member States in order to enable them to take informed and science-based risk management decisions necessary to ensure food and feed safety. The missions assigned to EFSA are: (i) issuing scientific opinions based on risk assessment, (ii) promoting and coordinating the development of risk assessment methodologies, (iii) commissioning scientific studies, (iv) collecting and analysing scientific and technical data, (v) identifying emerging

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risks, (vi) establishing networks of relevant organisations, (vii) assisting the European Commission in crisis management, (viii) providing independent information on all matters within its mission with a high level of openness and transparency and (ix) communicate the risks. The EFSA's founding regulation establishes scientific panels and a scientific committee to deal with all areas of food and feed safety including animal health and welfare, plant health and nutrition. On the area of biological hazards, the Scientific Panel on Biological Hazards (BIOHAZ Panel), issues scientific opinions based on risk assessment of biological hazards relating to food safety and foodborne diseases including foodborne zoonoses, transmissible spongiform encephalopathies, microbiology, food hygiene and associated waste management.

The use of substances other than potable or clean water to remove microbial surface contamination from foods of animal origin is actually permitted from January 2006 when the Regulation (EC) No. 853/2004 (OJEU, 2004a) was entered into force. The authorisation of the use of these substances shall be followed after EFSA's opinion on microbiological and chemical risk assessment. The objective of this paper is to present a brief overview of various decontamination treatments of carcasses and the activities of EFSA on the microbiological risk assessment of the efficacy of chemical substances that are used to remove microbial surface contamination from foods of animal origin such as carcasses.

2. Foodborne zoonoses and control strategy in the European Union

The results on the occurrence of zoonoses, zoonotic agents, antimicrobial resistance and foodborne outbreaks that were reported from the Member States and analysed by EFSA show that in 2005 the two most commonly reported zoonotic diseases in the European Union (EU) were campylobacteriosis and salmonellosis (EFSA, 2006a). In particular, reported *Campylobacter* cases increased by 7.8% compared to the previous year rising to an incidence rate of 51.6 cases per 100,000 population and a total of 197,363 recorded cases. With regard to occurrence in the food producing line, *Campylobacter* was most frequently detected from fresh broiler meat (up to 66%), fresh pig meat (up to 7%) and fresh bovine meat (up to 2%). In animals, the prevalence in broiler flocks ranged from 0.2% to 85%, whereas in pig herds the prevalence varied from 25% to 85% and in cattle herds from 0.3% to 47%. Salmonellosis remained the second most frequent zoonosis with 176,395 reported human cases, despite a fall of 9.5% to an incidence rate of 38.2 compared to 2004. *Salmonella* was most often reported from Member States for fresh broiler and pig meat where proportions of positive samples were detected up to 18%. The reported proportions of positive findings in bovine meat were generally lower than 2%. In table eggs, findings of positive *Salmonella* samples ranged from 0% to 6%, but over the past 5 years an overall decreasing trend was observed. In animal populations, *Salmonella* was most frequently detected in poultry flocks. Particularly, the results of the mandatory control programme for *Salmonella* in breeding flocks (*Gallus gallus*) at European level indicated that 6% of the parent-breeding flocks for laying hens and 5% of parent-breeding flocks for broilers were infected with *Salmonella*.

In 1980, WHO (WHO, 1980) formulated three lines of defence for the control of *Salmonella* which are still valid and may be used for other zoonotic agents. The first line focuses on the control of *Salmonella* in the food producing animal by preventing the introduction of *Salmonella* into herds and preventing the in-herd transmission and by increasing resistance to the infection. Measures to achieve the above include biosecurity, implementation of optimal hygienic and management routines, identification and removal or isolation of *Salmonella* infected animals or groups of animals, vaccination and competitive exclusion. The second line of WHO recommendations refers to the prevention or reduction of contamination of the carcass. This can be ensured by applying control measures during transport and lairage of the slaughter animals such as cleaning and disinfection procedures, separation of batches from different herds, optimization of the transport and lairage duration and hygienic procedures for the personnel and the equipment. Application of good hygiene practices (GHP) in association with Hazard Analysis Critical Control Point (HACCP) principles is essential during slaughtering and carcass dressing followed by control of rapid cooling. The third line of defence concentrates on the prevention of contamination during the final preparation of the food by the industry and consumers. Application of good hygienic practices and HACCP principles during processing, maintenance of the cold chain, application of processing techniques alone or in combination are the major examples of control measures at the industry level. At consumer level, mitigation options include hygienic handling, proper cooling, heating or cooking followed by training.

The existing European Community legislation includes a number of provisions that enhance food safety as well as control and prevent pathogenic microorganisms. Indeed, the horizontal European Regulations such as Regulation (EC) No. 852/2004 (OJEU, 2004b) on the hygiene of foodstuffs and Regulation (EC) No. 853/2004 (OJEU, 2004a) on specific hygiene rules for food of animal origin require implementation of the HACCP system in the entire food chain and the improvement of the hygiene and processing procedures of the food business including verification and validation of the safety management systems used. Moreover, Regulation (EC) No. 2160/2003 (OJEU, 2003) sets rules on the proper and effective measures that shall be taken to detect and control *Salmonella* and other specified zoonotic agents at all relevant stages of production, processing and distribution in order to reduce their prevalence and the risk they pose to public health. These specific control measures should be based on targets for the reduction of the prevalence of these agents in animal populations,

mainly at the level of primary production, and where appropriate at other stages of the food chain, including food and feed. Currently, a strategy on setting Community targets is developed for *Salmonella* spp. breeding flocks of *G. gallus*, laying hens, broilers, turkeys, slaughter and breeding pigs.

It is generally agreed that there is no universal mitigation option that can eliminate foodborne pathogens from the whole food chain. The application of good hygiene and manufacturing practices and HACCP will generally reduce the level of contamination but further means and intervention strategies of improving the safety of food products are always recommended. Moreover, as new risks continuously emerge and exposure routes are still unclear, there might be a need to apply a combination of measures which is both effective and cost efficient. For example, a recent study (Wagenaar, Mevius, & Havelaar, 2006), concluded that, the most promising intervention strategies for the control of *Campylobacter* in the poultry processing plants are the reduction of faecal leakage during scalding and defeathering and the separation of contaminated and non-contaminated flocks followed by decontamination with chemicals such as organic acids.

Decontamination could be an effective measure for reducing the microbial contamination of carcasses but should never be used as the primary measure. It should only be considered as an additional measure, to further reduce the numbers and prevalence of pathogenic microorganisms, following the application of good hygienic/manufacturing practices, and not as a substitute for those good hygienic/manufacturing practices. Moreover, most decontamination techniques result in a relative reduction, not elimination of pathogens, depending directly on the type and the extent of the initial contamination (Koutsoumanis, Geornaras, & Sofos, 2006) as well as other factors that are described in the following sections.

3. Decontamination treatments of carcasses

Numerous decontamination technologies have been found to reduce the microbial contamination of carcasses. These can be divided into three major types: (i) physical (e.g. hot water, steam, steam vacuuming), (ii) chemical (e.g. organic acids, chlorine, acidified sodium chlorite, polyphosphates) and (iii) biological (bacteriophages, bacteriocins). Combinations of the above technologies could also be used. Some of the decontamination technologies can be considered as traditional methods (e.g. water treatments) or emerging techniques (e.g. bacteriophages or lysins from phages) which are under development for application on meat. (The latter are presented in detail in another part of this special issue.)

3.1. Physical decontamination treatments of carcasses

Many physical technologies have been used or are being developed to reduce and/or control the microbial contamination on meat surfaces such as carcasses. They include mainly, trimming, water treatments, steam vacuum, steam pasteurizing, electromagnetic, ionising radiation and freezing.

3.1.1. Water treatments

Washing with water is routinely used in meat processing plants and it has been shown to be effective in removing physical/visible contaminants such as soil, feathers and other debris. Effectiveness in the reduction in microbial numbers increases by increasing the temperature of the water (Sofos & Smith, 1998). For heat treatments (80–85 °C) of carcasses, hot water by spray or immersion, steam pasteurizing or pressurizing have been described as potential applications (Huffman, 2002).

In general, different studies conducted on various meat types under different conditions reported a 1–3 $\log_{10}$ reduction of bacterial counts and a reduction of pathogen prevalence (Sofos & Smith, 1998). For example, a commercial hot water carcass wash cabinet applying 74 °C for 5.5 s reduced both aerobic plate counts and *Enterobacteriaceae* counts by 2.7 $\log_{10}$ on pre-evisceration beef carcasses, and *Escherichia coli* O157:H7 prevalence by 81% (Bosilevac, Nou, Barkocy-Gallagher, Arthur, & Koohmaraie, 2006). This effect is strongly affected by the temperature of the water and the type of meat tissue. Eggenberger-Solorzano, Niebuhr, Acuff, and Dickson (2002) spray-washed pork skin and muscle tissue surfaces with water ranging from 25 to 80 °C. They reported that the reductions of *Enterobacteriaceae* on pork muscle tissue were greater following washing at 65 and 80 °C than at lower water temperatures; however, no effect of water temperature on population reductions of *Enterobacteriaceae* was observed on pork skin. Spray treatment of poultry carcasses with hot water (55 and 60 °C) in an inside–outside bird washer after evisceration and before chilling reduced the *Campylobacter* contamination by more than 0.78 $\log_{10}$ cfu/carcass compared with a 20 °C water spray treatment (Li, Yang, & Swem, 2002). Application of a second scald after defeathering (60 °C for 28 s or 73 °C for 20 s), did not further lower the *Campylobacter* counts on the carcasses compared to the first scald (Berrang, Dickens, & Musgrove, 2000). The technique, by which the heat is applied to the carcasses, or, alternatively, the point in processing, (e.g. after defeathering, after evisceration or before chilling) seems to have great impact on the effectiveness of the treatment.

In general, the use of spray treatments may cause some disadvantages if appropriate validation and verification of the methodology applied are not followed. For example, high pressures could result in the penetration of bacteria into the tissue (Sofos & Smith, 1998), and spray treatments easily loses heat by evaporation. Moreover, elevated tissue surface moisture could stimulate the growth of microorganisms. Concerns have also been expressed about the organoleptic properties of the spray treated carcass (e.g. appearance and colour) and the risk of redistributing or spreading of a localized microbiological population to adjacent tissue surfaces or equipments (Sofos & Smith,

1998). Subsequent inhibitory effects after heat treatments during aerobic storage of the treated meat are essentially absent; however the treatments could result in an increased lag phase in the population among the spoilage microorganisms (Koutsoumanis et al., 2004).

An alternative to treatment with hot water is the application of steam. The main advantage of using steam is that steam at 100 °C has a greater heat capacity than the same amount of water at that temperature and therefore is able to penetrate cavities, crevices and feather follicles (James et al., 2007). Steam treatments for up to 20 s at atmospheric pressure has been found to reduce numbers of *Campylobacter jejuni* and *E. coli*, but damaged the appearance of the carcasses (James et al., 2007).

Generally, the effectiveness of hot water treatment as decontamination technique depends on both operational factors and factors related to the product itself. Operational factors may include water temperature, pressure, flow rate and target surface distance, method of application, the time or stage of application in the slaughtering sequence and plant variation (e.g. rate of slaughtering, size and design of the plant) (Gill & Badoni, 2004; Kiermeier, Bobbitt, Vanderlinde, Higgs, Pointon, & Sumner, 2006; Sofos & Smith, 1998). Factors related to the product itself (intrinsic and implicit) such as animal lots, type of meat tissue, initial microbial load, the type of the microbial ecology of the product, and the time of exposure to contamination which can affect bacterial attachment and biofilm formation could influence the effectiveness of water decontamination treatments.

3.1.2. Ionising radiation

The effectiveness of irradiation on foodborne pathogens depends on the type of the target microorganism, the type of the food, the irradiation temperature, the presence of oxygen and water content (Farkas, 1998). Vegetative bacterial cells are generally more sensitive to radiation than bacterial spores. *Campylobacter* spp., *Yersinia* spp., *Vibrio* spp., *Listeria monocytogenes* and *E. coli* seem to have low resistance to ionising radiation while *Salmonella* serotypes vary in their radiation sensitivity (Farkas, 1998). The inactivation effect of ionising radiation on microorganisms can be due to direct inactivation of the genetic material of a cell or indirect due to reactions between the generic materials of the cell with the free radicals resulting from the ionisation of molecules of other substances, usually water.

Radiation readily eliminates *Campylobacter* from poultry meat at doses of 3–5 kGy for frozen poultry and 1.5–2.5 kGy for chilled poultry (Farkas, 1998). In most cases, the microbial reduction rates achieved are within the range of 2–3 $\log_{10}$ for vegetative forms of the main foodborne pathogens (e.g. *Salmonella*). However, this does not account for viruses and microbial toxins, which are more persistent to radiation.

In the EU, irradiated foods are regulated by Directive 1999/2 (OJEU, 1999) which covers general and technical aspects for carrying out the processes, labelling and conditions of authorisation. So far the irradiation of dried aromatic herbs, spices and vegetable seasonings is authorised in the EU. In addition, some Member States maintain national authorisations for certain foods such as shrimps, frog legs, eggs, frozen vegetables, dried fruits and meat. The latter as well as with the EU enlargement has developed an increased interest at Community level to harmonise the legislation on food irradiation and the EC is considering different options for drawing up a proposal to complete the list of food and food ingredients authorised for treatment with ionising radiation at European level. EFSA is currently updating a former risk assessment on the safety of irradiation for certain food products including foods of animal origin.

3.1.3. Freezing

Freezing and further storage at freezing temperatures will reduce the prevalence and numbers of contamination on meat products. For example, freezing at −20 °C reduced the numbers of *Campylobacter* on inoculated broiler carcasses by approximately 1 $\log_{10}$ (Georgsson, orkelsson, Geirdsdóttir, Reiersen, & Stern, 2006). Zhao, Ezeike, Doyle, Hung, and Howell (2003) found that the numbers of *Campylobacter* on inoculated chicken wings declined by 1.3 and 1.8 $\log_{10}$ after storage at −20 °C and −30 °C for 72 h, respectively, with further reductions of 4 $\log_{10}$ after storage at −20 °C for 52 weeks. In 2003, freezing of meat from *Campylobacter* positive flocks was implemented as an intervention in Denmark in combination with other intervention strategies such as biosecurity and education of consumers. It was observed a 13% decrease in the broiler flock prevalence and a 20% decrease in the prevalence of *Campylobacter* in Danish fresh chilled chicken meat (Rosenquit, Helwigh, & Boysen, 2007).

The BIOHAZ Panel, in its Opinion on *Campylobacter* in animals and foodstuffs (EFSA, 2005a) concluded that reducing numbers of *Campylobacter* on poultry carcasses can be achieved by reducing faecal spread during slaughter and further processing and by using appropriate physical such as freezing or chemical decontamination techniques were permitted. Moreover, it was identified that there is a need to better understand the effects of freezing on the occurrence and survival of *Campylobacter* during retail shelf-life and to determine the optimum rate of freezing, freezing temperature and frozen storage time for reduction of *Campylobacter* in poultry.

3.2. Chemical decontamination treatments of carcasses

Chemical decontamination treatment involves the application of a chemical substance at a given step during the slaughter process. The most common substances which have been used for chemical decontamination of carcasses and have been extensively studied, are low-molecule organic acids (e.g. lactic, acetic, citric, fumaric) but also other chemical substances such as chlorine, acidified sodium chlorite, peroxyacids and trisodium phosphate.

Generally, the efficacy of the above mentioned substances depends on their ability to destroy the cellular membranes and other cellular constituents and pathways of the bacteria. This action results in a decrease in numbers of the various types of bacterial flora. The result will be an improvement of the microbiological quality and safety of products by a reduction in the number of pathogenic bacteria and, an improved shelf-life of products in the refrigerated state (0–4 °C) because of destruction of a part of the spoilage bacteria (Koutsoumanis et al., 2006).

3.2.1. Organic acids

Decontamination with organic acid solutions will reduce the number and prevalence of foodborne pathogens and the microbial load of meat carcasses (Huffman, 2002; Smulders & Greer, 1998). For example, a commercial lactic acid spray cabinet applying 2% lactic acid at approximately, 42 °C to beef carcasses (pre-evisceration) has been shown to reduce aerobic plate counts by 1.6$\log_{10}$, *Enterobacteriaceae* counts by 1.0$\log_{10}$ and *E. coli* O157:H7 prevalence by 35% (Bosilevac et al., 2006). A stepwise increase in pH from 2.6 to 3.5 and 4.0 in an *in vitro* model resulted in a decrease of the bactericidal effect of lactic acid decontamination on meat-borne pathogens (Van Netten, Huis in't Veld, & Mossel, 1994). Moreover, these authors reported that a 2% solution of lactic acid at 37 °C for 30–90 s would eliminate *Salmonella* but not *L. monocytogenes* in pork skin suspensions. This is because Gram-positive bacteria are generally more resistant to lactic acid than Gram-negative bacteria. Combination of cold or hot water followed by lactic acid treatment resulted in lower populations of aerobic bacteria, psychotrophic bacteria, coliforms, *E. coli* and lactic acid bacteria on fat-covered pork trim tissue than on lean pork trim tissue (Castelo, Kang, Siragusa, Koohmaraie, & Berry, 2001). It is obvious that the efficacy of organic acid decontamination will depend on the type of the meat tissue, the type and the load of initial microbial contamination, as well as the pH, the concentration and the temperature of the organic acid solution (Koutsoumanis et al., 2006; Sofos & Smith, 1998).

Mesophilic enterobacteria are usually more sensitive to organic acid decontamination than other pathogens (Smulders & Greer, 1998). However, concerns have been raised whether changes in the natural microflora of meat caused by decontamination technologies will emerge risks such as higher acid tolerance of pathogens and increase of their growth due to possible reduction of microbial competition. For example, it is reported that the aerobic storage of fresh beef meat treated with 2% lactic acid at 55 °C shifted the predominant spoilage association from Gram-negative to Gram-positive bacteria and yeasts (Koutsoumanis et al., 2004). The above spoilage microbial profile could be beneficial from the safety perspective since growth of *L. monocytogenes* was limited compared to the untreated samples. However, Nissen, Maugesten, and Lea (2001) reported that growth of *E. coli* O157:H7 was 2–3$\log_{10}$ greater on beef treated with a combination of steam vacuum and 0.2 M lactic acid than on untreated samples stored at 10 °C in air and vacuum packaging. Thus, the growth inhibition of *E. coli* O157:H7 on untreated samples could also be explained by the competitive high numbers of the background flora.

Concern has been raised that decontamination treatments may lead to increased tolerance after adaptation to stressful environments and the microorganisms may become highly resistant to further stresses and resulting in an increased survival of pathogens on meat. However, there are conflicting results in the literature. For example, the number of Gram-negative spoilage organisms of skin surface of pork belly after treatment with hot 2–5% lactic acid at 55 °C (120 s) were drastically reduced and intrinsic parameters (lactic acid content, pH) conditions were created that might benefit the survival and growth of acid-adapted pathogens, and therefore possess a health hazard (Van Netten, Valentijn, Mossel, & Huis in't Veld, 1997, 1998). However, growth of acid-adapted mesophilic pathogens, *L. monocytogens* and *Yersinia enterocolitica* did not cause a health hazard during aerobic storage at 4 °C. On the contrary, survival of acid-adapted *E. coli* O157:H7 was observed on vacuum packaged beef treated with 2% acetic acid and stored at 4 °C for 14 days (Berry & Cutter, 2000). The natural acid resistance of each species or strain and the method used for acid adaptation (e.g. pH adjustment with lactic acid or addition of fermented glucose in the culture broth) could account for these inconsistencies (Samelis, 2005).

Finally, studies in meat acid decontamination dripping wastes that were used as a model system to simulate conditions existing in meat plant environments, have also indicated the potential to cause acid stressing and selection of acid-resistant survivors. Particularly, bacterial pathogens may survive for several days, and possibly proliferate in acidic water washings of meat serving as potential cross-contamination sources of subsequent batches of fresh meat, if pathogen niches are established in the plant (Samelis, 2005).

3.2.2. Other chemical substances

Other chemicals used for meat decontamination include chlorine, trisodium phosphate, acidified sodium chlorite and peroxyacids. Generally, the use of these substances leads to 1–1.5$\log_{10}$ reductions of foodborne pathogens such as *Salmonella* and *E. coli* O157 (Smulders & Greer, 1998). Most of these substances have been suggested or authorised as intervention strategies in the US but are not currently permitted in the EU.

Chlorine is the most traditional chemical decontamination treatment for poultry and beef carcasses and can be used and applied with various forms. Chlorine has proved to reduce total counts and pathogenic bacteria such as *Salmonella* and *E. coli* O157:H7 (Sofos & Smith, 1998). Park, Hung, and Brackett (2002) observed an increase in the reduction during washing of poultry carcasses of two logs when the washing water contained 50 ppm of residual

chlorine. A recent study showed that spray of 20–50 ppm chlorinated water on poultry carcasses applied individually at different processing points (after defeathering, after evisceration and following neck removal) reduced the aerobic counts, total coliforms and *E. coli* but not more than 0.4 $\log_{10}$ while *Salmonella* incidence was reduced by 20–25% (Stopforth, O'Connor, Lopes, Kottapalli, Hill, & Samadroup, 2007).

Reductions following immersion dipping of pre-washed broiler carcasses in a solution containing acidified sodium chlorite with citric or phosphoric acid for 5 s before chilling were demonstrated at different concentrations (500, 850 and 1200 ppm) for total aerobes (0.76–1.03 log cfu/g), for *E. coli* (2.29–2.31 log cfu/g) and for total coliforms (0.85–1.96 log cfu/g) (Kemp, Aldrich, & Waldroup, 2000). Treatment of chilled beef carcasses with acidified sodium chlorite (0.16% w/v with citric acid) was less effective in reducing total aerobic bacteria (<0.5 log cfu/g) or *E. coli* (none) compared to the treatments with water (Gill & Badoni, 2004).

Dipping fresh beef trim pieces in a peroxyacetic acid solution (200 ppm; 43 °C; 15 s) resulted in a 0.6 and 1.0 $\log_{10}$ cfu/g reduction in *E. coli* O157:H7 and *Salmonella typhimurium*, respectively. The application of different concentrations (500 and 1000 ppm) did not significantly increase the $\log_{10}$ reductions of both pathogens (Ellebracht et al., 2005). Lower reduction levels of these pathogens were observed on chilled beef carcass surfaces when they were sprayed (200 ppm; 43 °C; 15 s) with peroxyacetic acid (King, Lucia, Castillo, Acuff, Harris, & Savell, 2005). However, peroxyacetic acid was found to be more effective resulting to greatest microbial reductions when applied to hot beef carcasses, hot-boned bobby calf and hot-boned beef flaps (King et al., 2005; Penney et al., 2007).

Immersion of carcasses in 10% trisodium phosphate solution (TSP) has shown microbial decontamination capabilities. Whyte, Collins, McGill, Monahan, and ÓMahony (2001) found a reduction of 1.71 $\log_{10}$ by dipping broiler carcasses for 15 s in TSP (pH 12) compared to a reduction of 0.55 $\log_{10}$ obtained by dipping the carcasses in water. Spray application of TSP (8–12%) immediately before chilling of poultry carcasses resulted in a 0.5–0.8 $\log_{10}$ reduction of bacterial populations (aerobic counts, total coliforms and *E. coli*) and 70% reduction of *Salmonella* incidence (Stopforth et al., 2007).

3.3. Combined physical and/or chemical treatments

The most widely accepted approach in food preservation is based on the theory of the multiple hurdles (Leistner, 2000). The application of combined hurdles techniques can have a synergistic effect on the inhibition or inactivation of the prevalence and the numbers of microbial contaminants in the food included carcasses; two or more hurdle methods at suboptimal levels are more effective than one at optimal level. For example, reductions of numbers of *Enterobacteriaceae*, total coliforms, thermotolerant coliforms, and *E. coli* obtained by steam vacuuming were significantly lower than those obtained by a combination of steam vacuuming with any other sanitizing treatment, e.g. treatments of hot water (95 °C) or 2% lactic acid (Castillo, Lucia, Goodson, Savell, & Acuff, 1999). The hurdle technology may involve the simultaneous application (e.g. acid solutions) or the sequential application of treatments (e.g. hot water treatments and organic acids) (Geornaras & Sofos, 2005). The most important factor that has to be considered during the multiple hurdle technology is the sequence in which the treatments are applied. For example, *L. monocytogenes* was reduced by 2.68 $\log_{10}$ cfu/g on beef tissue treated with hot water (75 °C) followed by lactic acid (2%, 55 °C) or treated with lactic acid followed by hot water (Koutsoumanis et al., 2004). However, only the latter treatment allowed the growth of *L. monocytogenes* at the same growth rate as in untreated samples during storage at 10 °C.

3.4. Biological treatments or biocontrol

Some bacteria may produce bacteriocins which are antimicrobial proteinaceous compounds that have a lethal or bacteriostatic effect on other related microorganisms. The most widely studied bacteriocin is nisin which is produced by *Lactobacillus lactis* susp. *lactis* and is effective against Gram-positive bacteria. Nisin is authorised for food preservation in the European Union and is permitted in ripened cheese and processed cheese, certain puddings, clotted cream and mascarpone. With respect to carcass decontamination, the use of nisin has not been very successful because of the deficient inhibitory effect on Gram-negative bacteria (Bolder, 1997). In addition, other limitations that generally encounter the use of bacteriocins on carcasses may be the low level of production *in vivo*, the likely inactivation of their effect due to interactions with other food components and the potential development of resistance of the target microorganisms (Bolder, 1997; Hugas, 1998). However, a combination of bacteriocin (e.g. nisin) with other decontamination treatments such as lactic acid through spray washing reduced the levels of microbial contamination on beef carcasses (Barboza de Marinez, Ferrer, & Salas, 2002).

Bacteriophages, which are ubiquitously and naturally viruses occurring in the environment with the characteristic to infect and kill bacteria, have also been proved experimentally to reduce *C. jejuni* and *Salmonella enteritidis* on chicken skin (Atterbury, Connerton, Dodd, Rees, & Connerton, 2003; Goode, Allen, & Barrow, 2003). The main advantage of using of bacteriophages as an alternative biocontrol measure is that due to their highly strain specificity to control a target species, no interactions with the background microbial flora are expected (Greer, 2005). Moreover, they are easy to prepare and apply, do not cause any organoleptic changes of the food and are able to survive under commercial processing procedures. However, the interactions between bacteriophages and their host taking also into consideration the possible influence of environmental factors and barriers present in food (such as structure and pH of the food, physiological state of bacte-

rial strains, immune responsiveness) are still to be explored. Other limitations of using bacteriophages as a measure to control bacterial meat surface contamination may include the transfer of undesirable characteristics (e.g. virulence genes), the occurrence of phage-resistant bacterial mutants and the ability to lyse only the homologous host due to narrow host range (Greer, 2005).

4. Evaluation of the microbial efficacy of substances to be used to remove the microbial contamination of carcasses in EU

For many years, European legislation limited the use of substances for the removal of microbial surface contamination of foods of animal origin, highlighting the significance and application of good manufacturing practices and/or other safety management systems throughout the whole production line. Currently, Article 3(2) of the Regulation (EC) No. 853/2004 (OJEU, 2004a) provides a legal basis for the use of substances other than potable water or clean water to remove microbial surface contamination from foods of animal origin. A draft Regulation proposal which sets the specific conditions for such decontamination treatments is under discussion with the Member States and stakeholders. Some of the provisions proposed in this draft Regulation are limitation to use only one substance at a time and to apply only for poultry carcasses, the need of rinsing after the application of the substance and consumer information by labelling.

Previous opinions of the Scientific Committee on Veterinary Measures relating to Public Health (SCVPH, 1998, 2003) concluded that decontamination treatments should be considered as supplementary means of reducing the microbial load of foods of animal origin and should be a part of an integrated control programme throughout the whole food chain. Both opinions stressed that the authorisation of substances for decontamination treatment of foods of animal origin should be based on risk assessment considering various factors such as (i) the safety and the efficacy of the substances from a public health view (chemical and microbiological risk assessment, respectively), (ii) the occupational risk assessment, (iii) the impact on the environment, (iv) the product acceptability and quality (e.g. appearance, taste), (v) the consumer perception and (vi) the industrial interest and cost-benefit analysis.

While authorisation is under the remit of the European Commission according to the existing European legislative procedures, EFSA in its role as the risk assessment body in food safety in the EU will be responsible for the risk assessment after an official request from the EC to evaluate the safety and efficacy of substances to be used to remove microbial surface contamination of foods of animal origin. A substance is considered to be safe when it does not pose any appreciable risk to the public health. With regard to the efficacy of substances to be used to remove the microbial surface contamination of carcasses, it is essential to have a common understanding of the term 'efficacy' when BIOHAZ Panel will evaluate dossiers and thus define it. Therefore, a substance will be regarded efficacious when its application on the surface of foods of animal origin results in any statistically significant reduction of the percentage of contaminated samples/carcasses and concentration of pathogenic target bacteria when compared to the control, e.g. application of water instead of the substance. In addition, this reduction is of relevance to human health, e.g. would result in a decrease of foodborne infections.

Table 1
Some factors for consideration in the evaluation of the safety and the efficacy of chemical decontamination treatments

Technical data
Identity of the substance and specifications
Manufacturing process
Reactions and fate in the treated product (residual levels, degradation and reaction by-products)
Methods of analysis
The process and its purpose
Exposure assessment
Estimate of potential daily exposure of the consumer to residues, degradation products and any relevant reaction by-products
Toxicological data
Available toxicological data on each substance, including its potential degradation products and any identified reaction by-products
Data to evaluate efficacy
All experimental conditions to be performed with the product formulation for authorisation
Comparison of the prevalence and/or numbers of the pathogenic microorganisms between the treated food and the control food
Measurement of the prevalence and/or numbers of the target pathogenic microorganisms before and after application of the product formulation
Measurement of the prevalence and/or numbers of the target pathogenic microorganisms at the end of the shelf-life of both the treated and control food
Behaviour of non-pathogenic microorganisms, such as indicator microorganisms and total viable counts
Available scientific information on adaptation as well as on natural or acquired resistance of bacteria to the active substance
Tests on naturally contaminated foods
Proof that the concentration of the product formulation proposed is justified
Description of the methods to control and monitor the concentration of the active substance in the processing plant during operational time
Identification of factors that may influence the efficacy of the active substance

The extent of this reduction remains to the decision of the risk managers, e.g. European Commission or national authorities.

To assess the impact of decontamination on public health on microbial risk aspects the following need to be considered: the prevalence of pathogens on carcasses, distributions of numbers of pathogens on each carcass, the detection limit for the microbiological procedure which may be a fixed number or a probability distribution, the infectious dose which may also be a fixed number of a probability distribution, the conditions along the food chain determining growth and survival/death of bacteria after decontamination, the type of pathogen.

EFSA has so far provided several scientific opinions on the efficacy and safety of substances to be used to remove the microbial surface contamination of foods of animal origin (EFSA, 2005b, 2006b, 2006c). The evaluation of the safety of these substances falls within the remit of the Scientific Panel on food additives, flavourings, processing aids and materials in contact with food (AFC) while the evaluation of the efficacy is under the remit of the BIOHAZ Panel. A joint AFC/BIOHAZ guidance document for the submission of data for the evaluation of the safety and the efficacy of substances intended to be used to remove microbial surface contamination of food of animal origin has been developed (EFSA, 2006d). The scope of this document is to provide guidance on the major factors and data that a dossier under evaluation should contain, such as technical information, toxicological data, and data on exposure assessment and on microbial efficacy of the substance to be used for decontamination (Table 1).

5. Conclusions

Accurate evaluation of overall effects of meat decontamination treatments is difficult, as most efficacy data may result from laboratory studies while extrapolation to commercial practice is not warranted (Smulders & Greer, 1998). In general, the efficacy of decontamination treatments will depend on the validation and the verification of the methodology applied, on the type of meat tissue, the microbial ecology of the product and the initial microbial load, the ability of bacteria to attach to the product and to produce biofilm and the operational factors such as temperature, pH of the solution, time and stage of application in the process. Advantages include improved meat safety through reduction of the occurrence and the numbers of microbial hazards on meat, provision for inclusion of a hazard-reducing CCP into HACCP, and reduction of overall pathogens populations being passed into the meat processing/distribution stages. Disadvantages include potential problems with disproportionate reliance on the decontamination step and consequent reduction of the process hygiene, limited reduction rates achievable enabling positive selection for surviving resistant strains, increase of virulence of the surviving strains and subsequent enhanced growth of surviving pathogens due to elimination of background meat microflora. Except of the above microbiological safety issues, the application of any chemical decontamination method on meat carcasses should take into consideration other issues such as the product quality, e.g. discolouration and defects on meat flavour and odour, as well as environmental and health occupational matters.

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