Report of the

SCIENTIFIC COMMITTEE ON VETERINARY MEASURES

RELATING TO PUBLIC HEALTH (SCVPH)

on

BENEFITS AND LIMITATIONS OF

ANTIMICROBIAL TREATMENTS FOR POULTRY CARCASSES

30 October 1998

Definitions

Within the context of this report, the following definitions are used:

Antimicrobial treatment: The term "antimicrobial treatment", as applied to carcasses of meat animals or poultry, is often used as a synonym for "decontamination". It is generally understood that such a treatment is used at the slaughterhouse during or at the end of the slaughtering process. The treatment may be a physical, chemical or microbiological means of reducing the surface microbial contamination of carcass meat, with particular regard to reducing the level of pathogens.

Pathogen: The term pathogen is used to imply a human pathogen generally recognised as food-borne.

'Benefits: The benefits discussed in this paper include only the potential lowering of risk to human health from microbial hazards. This means that the influence on other management strategies and economics, such as cost-benefit considerations, are not included.'

'Limitations: The limitations discussed in this paper include only factors directly affecting the quantity and nature of the hazard and risk in question. More general adverse effects of the use of antimicrobial treatments, such as negative interactions with other risk management initiatives, i.e. good sanitation practices and hazard control throughout the production chain, are not covered.'

Caveat

The term "decontamination" may be misleading as none of the techniques proposed has the potential to eliminate high levels of pathogens nor to render the meat totally free from pathogenic microorganisms.

An expression such as "pathogen reduction treatment" is therefore preferred.

TERMS OF REFERENCE

At its first meeting on November 17, 1997, the SCVPH received a request from the EU Commission for "a scientific review concerning the use of antimicrobial treatment of poultry carcasses and the various methods of carcass rinsing including trisodium monophosphate (TSP), organic acids and hyperchlorinated water".

BACKGROUND

At its plenary session of December 2, 1996, the Scientific Veterinary Committee (Public Health Section) adopted a preliminary report on "The Decontamination of Poultry Carcasses" [Doc. VI/7785/96 Final].

The content of this document was as follows:

1- Introduction, setting the background of the discussions

2- Terminal decontamination of meat and poultry, including comments on five decontamination techniques: organic acids, irradiation, alkaline compounds, hyperchlorinated water, steam and hot water.

3- Acceptance criteria for decontamination compounds

4- Conclusions

In the conclusions, the Committee recognised that "the world-wide increase in foodborne infections and intoxications needs to be addressed in different ways". However, the Committee "expressed concern that the use of decontamination techniques during food processing would have an adverse effect on the efforts being made both at the primary production level and during the initial processing stages. In particular, the Committee pointed to the disadvantage in removing incentives for farmers to continue developing good sanitation in their flocks, and in neglecting the use of good manufacturing practice (GMP) in the whole production line". The Committee stressed also that "the good standards of husbandry and sanitation in the flocks should include the use of a HACCP approach and the application of appropriate health control methods. The evaluation, according to HACCP, of hygiene practices in processing plants and the pressure on farmers being given by the processors will necessarily lead to an improvement at the farm level". Finally, the Committee "was unable to recommend the general introduction of decontamination techniques in food processing. Nevertheless, in view of the different scientific, technological, economic and practical considerations in the member states, the Committee decided that, for the transitional period, the use of decontamination should be considered solely as a supplementary measure and must not in any way compromise the use of Good Hygienic Practice". The Committee considered also that "advice should be sought from experts as to how labelling should be sorted out".

The present report is a follow-up to the above-mentioned preliminary report. To comply with the terms of reference, the SCVPH extended its approach to an assessment of the role of specific treatments aimed at reducing microbial surface contamination (decontamination). This has been done in the general framework of the

interventions necessary to control, including where necessary to reduce, the prevalence of pathogenic microorganisms on raw poultry meat.

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SECTION 1 - THE MICROBIOLOGY OF RAW POULTRY

1.1 FOODBORNE INFECTIONS ASSOCIATED WITH RAW POULTRY MEAT

1.1.1 Foodborne diseases due to poultry

Diseases caused by foodborne pathogens constitute a worldwide public health problem. Diseases such as salmonellosis, campylobacteriosis and enterohaemorrhagic *Escherichia coli* (EHEC) infections are on the rise in many industrialised countries. Many different food categories are implicated in foodborne disease, the frequencies depending on the agent in question, the geographical area and the category of foods. Meat and poultry products are often identified as the source of foodborne outbreaks, however, egg products, milk products (especially those made from unpasteurised

milk), fish, fresh produce and, last but not least, water also play an important role. Poultry is frequently incriminated in foodborne disease caused by agents such as *Salmonella*, and *Campylobacter jejuni,* and may be contaminated with *Listeria monocytogenes.* On the other hand, poultry is not considered an important source in the epidemiology of foodborne disease caused by EHEC, *Vibrio* spp., *Yersinia enterocolitica*, *Shigella* spp., and *Clostridium botulinum*. Regarding foodborne disease caused by *Bacillus cereus*, *Clostridium perfringens*, and *Staphylococcus aureus*, several different foods may be the source, including poultry, as the occurrence of such diseases usually are a food handling and hygienic issue rather a food origin issue.

The trend reports provided for the year 1995 by the Member States to the European Commission confirm that foodborne diseases, especially salmonellosis and campylobacteriosis, represent an important public health problem.

Several Member States reported incidence rates of reported salmonellosis *cases* varying from 20.5 to 141 cases per 100 000 inhabitants with large variations between Member States. Traditionally the actual incidence rate is considered to be 10 to 20 times the rate of reported cases. A combination of studies in the Netherlands estimates the actual incidence rate within 95% confidence limits to be between 300 and 700 cases per 100.000 at risk (Hoogenboom-Verdegaal et al. 1994; Berends, 1998).

The main sources of salmonellosis were eggs, poultry, other meat and meat products and sprouts. In Denmark, it was estimated that 15-20 % of all cases of salmonellosis were attributable to poultry.

In several Member States, the incidence of campylobacteriosis is increasing; in Denmark, the incidence more than doubled form 1992 to 1995. Most cases seem to be sporadic, and are caused by *C. jejuni*. Campylobacteriosis in Europe occurs particularly as a result of consumption of poultry meat.

The US Center for Disease Control and Prevention (CDC) has summarised the surveillance for foodborne disease outbreaks for the years 1988-1992 (Bean et al, 1997). During these years the vehicle of transmission was identified as chicken or turkey in 68 out of the 1071 (6.3%) outbreaks with a confirmed vehicle. These 68 outbreaks represent 8.4% of outbreak-related cases of foodborne disease with known vehicle. Another 354 outbreaks (33%) had multiple vehicles, including poultry.

The etiologic agent was identified in 30 out of the 68 outbreaks traced specifically to chicken/turkey; *Salmonella* spp. 19, *Cl. perfringens* 6, *B. cereus* 2, *Staph. aureus* 2, *Campylobacter* spp. and *Salmonella* spp. 1. *Salmonella* spp. was the dominating etiologic agent in the outbreaks with multiple vehicles.

1.1.2   Salmonellosis and poultry

Salmonella is one of the most important causes of foodborne disease worldwide. In many industrialised countries the incidence of salmonellosis in humans and the prevalence of salmonella in many food products have increased significantly over the last twenty years. Salmonella bacteria have a broad host-spectrum, and can be isolated from a wide range of animal species, including birds and reptiles. The animals usually are healthy carriers, and contaminated feed plays an important role in the epidemiology

of salmonellosis. Salmonella can survive for a long time in the environment. Humans are usually infected through consumption of contaminated foods of animal origin. However, other food such as fresh produce, seafood and chocolate have also been implicated in outbreaks because of cross-contamination, use of contaminated water, use of manure as a fertiliser, presence of animals or birds in the production area or other factors.

Factors contributing to the increase of the salmonella problem include:

- industrialisation of animal production and the food chain

- increased international trade in foods, animals and feed

- increased international travel

- new technologies and methods for food preparation and storage

- more people belonging to high-risk populations (elderly, immunosuppressed, chronically sick people).

In developing the regulation «Pathogen Reduction; Hazard analysis and Critical Control Point (HACCP) Systems» (Federal Register July 25, 1996), the US Food Safety and Inspection Services (FSIS) conducted a baseline survey on the prevalence of salmonella in meat and poultry. The following data were obtained:

Class of product	Percent positive for *Salmonella* spp.
Steers/Heifers	1.0
Cows/Bulls	2.7
Ground beef	7.5
Hogs	8.7
Broilers	20.0
Ground chicken	49.9
Ground turkey	44.6

The above data show that US poultry is frequently contaminated with *Salmonella*. Of 272 outbreaks of salmonellosis from known vehicles, which were reported to the CDC in the period 1988-1992, 13 were traced to chicken/turkey (Bean et al, 1997). However, 132 outbreaks had more than one vehicle. There is reason to believe that poultry is implicated in many sporadic cases of salmonellosis.

The US situation is not likely to be much different from the situation in many other developed countries. It is recognised that intensive animal husbandry, which is common in the poultry industry, is often associated with a high prevalence of salmonella carriage.

The trend reports provided for the year 1995 by the Member States to the European Commission show that, in most Member States, the prevalence of salmonella in poultry is relatively high. For example, in Germany, 711 out of 3 180 (22,4 %) samples of fresh poultry meat were salmonella positive. In Denmark, 4,9 % out of 492 samples of

fresh poultry meat at retail were positive. Salmonella was detected in a total of 45,7 % of 4 099 flocks of broilers after slaughter. Before slaughter an average of 24,0 % of flocks were positive.

In Norway, Sweden and Finland, in contrast to other developed countries, the prevalence of salmonella carriage in animal husbandry is very low. For example in Norway and Sweden, surveillance has shown that salmonella is isolated from <1.0% of poultry carcasses. This low prevalence is also reflected by the incidence of salmonellosis among people. About 80 to 90% of the reported human cases of salmonellosis have acquired their infections abroad. A case-control study performed in Norway in 1993-1995 identified consumption of poultry and pork products of foreign origin as risk factors for domestically acquired salmonellosis (Kapperud G. 1996).

Antimicrobial resistance among *Salmonella* spp. is an increasing public health problem. Several outbreaks of antimicrobial resistant *Salmonella* among humans have been traced to food animals, and often associated with antimicrobial use on the farm (Cohen and Tauxe, 1986, Holmberg et al, 1984). In the 1990s the incidence of infections caused by multiresistant *S. typhimurium* DT 104 has increased significantly. In England and Wales this variant is now the second most prevalent salmonella in humans, after *S. enteritidis* PT4. Recently, quinolone-resistance has emerged among multiresistant *S. typhimurium* DT 104. Cattle is considered the most important reservoir of *S. typhimurium* DT 104, however, the bacteria can also be isolated from poultry, sheep, goats, pigs, and horses (Wall, 1997).

1.1.3 Campylobacteriosis and poultry

Campylobacter was described as a cause of enteritis in man in 1957 (King, 1957). However, the prominence of *Campylobacter* spp. in human infection remained obscure for another 12 years when a large number of cases were reported (Stern and Kazmi, 1989). Since then, campylobacter enteritis has been described from all parts of the world, and is today one of the most common causes of bacterial enteritis. It is considered to be an important food safety problem. In many industrialised countries the incidence seems to be on the rise. Data from the CDC indicate that *Campylobacter* may be a more common cause of bacterial enteritis than *Salmonella*, although *Campylobacter* is rarely associated with outbreaks (McNamara, 1997). *C. jejuni* is responsible for 80-90% of reported cases of campylobacteriosis in industrialised countries. *C. coli* is the second most important human pathogenic species within the genus *Campylobacter*. In developing countries infections with *C. coli* and *C. jejuni* are about equally common.

An important factor in the epidemiology of campylobacteriosis is the fact that the infective dose is very low. Less than 1000 bacteria might be enough to cause a clinical infection (Black, 1988). Moreover, the bacteria survive well during refrigerated storage.

Reservoirs for *Campylobacter* include cows, pigs, horses, rabbits, rodents, domestic pets, poultry and wild birds. Moreover, *Campylobacter* are frequently isolated from water. Birds are a common host for *C*ampylobacter, especially *C. jejuni*, probably because of a higher body temperature. Pigs are frequently colonised with *C. coli*.

Outbreaks of campylobacteriosis are often attributed to consumption of raw milk or contaminated water supplies. Outbreaks are also traced to poultry, and beef (Nachamkin, 1997, Stern and Kazmi , 1989). Studies have identified *C. jejuni* as an important cause of sporadic bacterial enteritis. Such cases often follow ingestion of improperly handled or cooked food, primarily poultry products (Hopkins and Scott, 1983, Stern, 1992, McNamara, 1997). Since *C. jejuni* is considered a commensal in the intestinal tract of poultry, carcasses are frequently contaminated with this organism. There is a reduction in the numbers of viable *C. jejuni* on carcasses during refrigerated storage. Freezing will significantly reduce, but not entirely eliminate viable *C. jejuni* on carcasses (Stern and Kazmi, 1989).

Serotypes of *C. jejuni* isolated from chicken carcasses include many of those most frequently isolated from human cases of campylobacteriosis, confirming that poultry is an important factor in the epidemiology of human campylobacteriosis (Stern and Kazmi, 1989).

In a study carried out by Stern et al. (1995) 9 out of 10 broiler production farms in the United States were positive for *Campylobacter*. In the study of Willis and Murray (1997) 69% (229/330) of the raw commercial broilers were positive for *C. jejuni*.

The trend reports provided for the year 1995 by the Member States to the European Commission show that also in Europe poultry are frequently contaminated with *Campylobacter*.

Sweden reported that it was possible to reduce the incidence rate of campylobacter positive broiler flocks to 10-15 % by a campylobacter surveillance programme and also the basic requirements of the salmonella control programme. During 1995, 14 % of all flocks slaughtered were found to be positive.

A Dutch study revealed that 29 out of 43 breeder flocks (67%) were colonised with *Campylobacter* spp. (Jacobs-Reitsma, 1995). In Norway, 10 out of 176 broiler flocks (17%) were colonised with *Campylobacter* spp. (Kapperud et al, 1993). Data from Sweden show that 522 out 3 727 flocks of poultry (14 %) were positive for *Campylobacter*.

The Danish Veterinary and Food Administration reported that 27% of retailed Danish poultry meat is contaminated with *Campylobacter* spp. (1995 data), whereas such bacteria can be isolated from 39% of poultry imported into Denmark. A recent survey revealed that *Campylobacter* could be isolated from 63% of chickens carcasses at retail outlets in New Zealand (Bongkot, 1997).

In a survey conducted in Norway in 1996-1997 *Campylobacter* were isolated from 33 out of 319 samples (10,3%) of raw poultry meat sampled at retail, and from 12 out of 364 samples (3,3%) of frozen poultry (Norwegian Food Control Authority, Anonymus, 1998).

In Finland, in 1995, 11 % of the samples of broiler meat were campylobacter positive.

None out of a total 27 outbreaks of campylobacteriosis reported to the CDC were traced to poultry in the period 1988-1992 (Bean et al, 1997). However, in 15 of these

outbreaks the vehicle was not identified, and three outbreaks had multiple vehicles. In another outbreak traced to chicken both *Salmonella* spp. and *Campylobacter* spp. were isolated. It should be emphasised that the above-mentioned statistics are based on outbreak data. It is assumed that statistics based on cases of sporadic disease would be different, as poultry is considered an important source for sporadic campylobacter infection in humans.

Usually human campylobacteriosis is self-limiting and does not require antimicrobial therapy. However, when antimicrobial therapy is indicated, fluoroquinolones are among the antimicrobials considered useful. In 1985 fluoroquinolone-resistant *Campylobacter* spp. were detected for the first time (Taylor et al, 1985). Since then fluoroquinolone-resistant strains have been reported from many countries, and the proportions are increasing. In the Netherlands, enrofloxacin was introduced in poultry production in 1987 (Endtz et al, 1991, Jacobs-Reitsma et al, 1994). By 1993, 29 % of animal isolates of *Campylobacter* were quinolone-resistant. During the same period the proportion of quinolone-resistance among human isolates had increased significantly (Piddock, 1997).

1.1.4 Listeriosis and poultry

L. monocytogenes is frequently isolated from raw poultry. Finland reports that, in 1995, 34 (40 %) of retail broiler meat samples were *Listeria monocytogenes* positive using an enrichment method. *L. monocytogenes* was detected from 20 (24 %) of these samples also by direct plating, which means the cell densities > 10 CFU/ml rinsing liquid and > 5 CFU/g sample. Although poultry is rarely associated with outbreaks, it is assumed that poultry is implicated in many sporadic cases of listeriosis (McNamara, 1997). Recent epidemiological studies conducted in the USA have implicated undercooked chicken as a risk factor for infection, particularly in immunosuppressed persons. In one case-control study, 6% of the cases of listeriosis were associated with consumption of undercooked chicken (Schuchat et al, 1992).

1.2 SOURCES OF CONTAMINATION DURING POULTRY MEAT PRODUCTION

There is an extremely large number of papers and documents on the sources of contamination of poultry meat. Comprehensive reviews may be found in particular in Mead, 1982; Smulders, 1987; Mulder, 1991; Nagy et al, 1995; van de Giessen, 1996; Mead and Mulder, 1997; White et al., 1997; ICMSF, 1998. These reviews have been extensively used for the preparation of this part.

In a typical poultry processing operation, freshly laid fertile eggs are collected and incubated. After they hatch, chicks are delivered to farms, reared until ready for slaughter and then transported to a processing plant. At the plant, the process of slaughtering includes several phases from unloading and shackling the live birds to grading and packaging the carcasses (Figure 1).

Then, carcasses are shipped and distributed chilled or frozen while a significant proportion of poultry carcasses is used for portioning and/or to produce a variety of raw or processed products. Only the primary production (breeding, hatching and rearing) and the slaughtering process will be considered in this report.

The microbiological condition of poultry carcasses is highly dependent on the manner in which animals are reared and slaughtered. The microbiological condition of live birds influences the microbiology of the products and the live animals are the principal source of microorganisms found on poultry carcasses. At the processing plant, the conditions of slaughtering will further influence the extent to which processed poultry will be contaminated.

Figure 1 : General flow diagram of poultry meat production

Primary production	Breeding
	Hatching
	Farm rearing

Transport

Slaughtering process	Unloading and shackling
	Stunning
	Bleeding
	Scalding
	Plucking
	Evisceration
	Carcass Wash
	Grading
	Packaging

Chilled storage
or freezing
Distribution
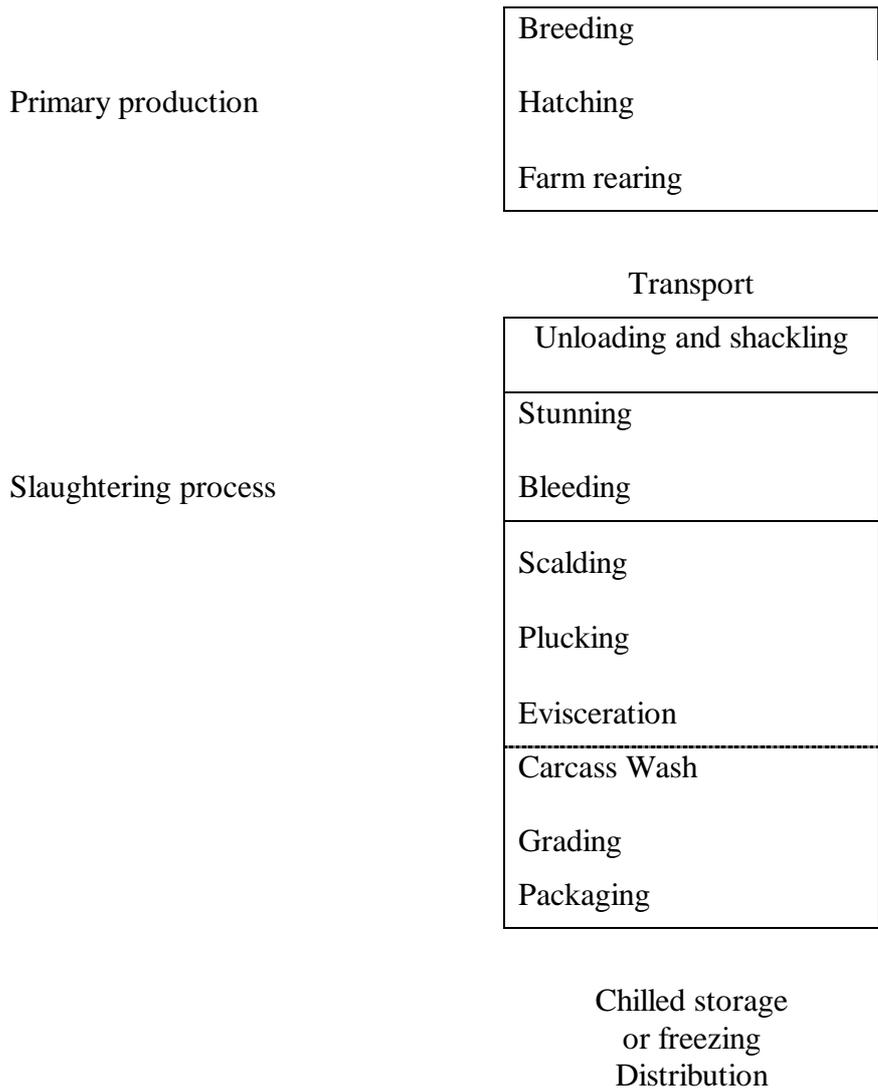
1.2.1   Sources of contamination related to animal husbandry

Poultry may be the vector of several pathogenic microorganisms (see Part 1). However, in the following section, emphasis is placed on the two principal pathogens associated with poultry, *Salmonella* and *C. jejuni*.

1.2.1.1 *Salmonella*

Poultry become infected with *Salmonella* in three main ways:

- by transmission between and within flocks;

- by the consumption of contaminated feed or water;

- through the environment.

Transmission between and within flocks

The principal sources of infection include the parent stock and the egg. Transmission of *S. enteritidis* from the ovary or the oviduct of an infected hen into the forming egg is well substantiated. *Salmonella* can be transmitted by external contamination of the egg shells, subsequent penetration of the microorganism through the pores. Survival and proliferation to hatching depend on the hatching conditions.

The hatchery can be an important point in production in which poultry become infected with *Salmonella* and this requires attention to prevent colonisation of chicken during production. At hatching, most chicken have a very limited microflora in the gut and are far more susceptible than older chicks to *Salmonella* colonisation. The infective dose of *Salmonella* for day-old chicks appears to be low whereas older birds become more resistant : it has been suggested that a bird on day 7 requires a challenge 10,000 time greater than a day-old chick to become infected with *Salmonella*. Although *Salmonella* contamination of eggs is low, 5 to 9 % of day-old chicks have been found positive when tested for *Salmonella* in commercial hatcheries in the US. Also, it has been demonstrated that the hatching of a single contaminated egg could result in the infection of all other newly hatched chicks during the hatching process. Further on, spread of salmonella among chicks occurs during transport to the farm and *Salmonella* positive chicks delivered to the growing house provide a ready source of *Salmonella* for colonisation of other chicks in the flock.

Transmission by contaminated feed and water

Various surveys have identified feed as an important source of *Salmonella* for the farm. Although raw materials used for preparation of feed may harbour the pathogen, pelleting, heating and other specific treatments are generally successful in eliminating *Salmonella*. However, the final feed may be contaminated because of an insufficient heating process or to recontamination in the feed mill, during transport or during storage at the farm. Further on, *Salmonella* may multiply in wet feeds. On the farm, water can be a source of foodborne pathogens, including *Salmonella* when it is dispensed in open troughs that can become contaminated by dust, litter, feed, feathers and faeces.

Transmission through the environment

It has been demonstrated that regardless of the time interval between the removal of one flock and the introduction of another and whether or not the litter is changed between flocks, *Salmonella* can persist in the farm environment. Moreover, in

favourable ecological niches, *Salmonella* may resist cleaning and disinfection procedures and spread to incoming flocks. Rodents, insects, birds as well as domestic pets have been suggested as potential sources of *Salmonella* in poultry flocks. Workers and visitors may also contribute as mechanical vectors of *Salmonella* contamination from the general environment.

1.2.1.2 Campylobacter jejuni

Several studies show that vertical transmission of *C. jejuni* from breeder flocks through hatcheries to farms is unlikely to occur. Broiler flocks become infected from sources in the farm environment. Potential sources are animals (other farm animals, domestic pets, rodents, wild birds, flies and vermin), contaminated litter, farm workers.

Feed has not been implicated, whereas contaminated water may be an important source. Once *C. jejuni* appears in a flock, the pathogen spreads rapidly to virtually all cohort birds.

1.2.2 Sources of contamination during processing

Commercially grown poultry flocks are collected on the farm, placed into crates, transported to the processing plant and slaughtered on the same day.

Contaminated crates can be a significant source of *Salmonella* on processed carcasses. Contamination of feathers with microorganisms of faecal origin increases as birds are confined in crates for transport to the plant and microorganisms in faeces and on feathers can be spread from bird to bird within the crates. Stress of transportation may amplify *Salmonella* levels.

In one study, faecal droppings collected in broiler houses about one week prior to slaughter were contaminated at a rate of 5.2 % while *Salmonella* was found in 33 % of the samples collected from live-haul trucks at the processing plant.

During **hanging**, as feathers, feet and bodies are contaminated with a variety of bacteria, wing flapping creates aerosols and dust, contributing to contamination of the unloading zone and transmission of pathogens at this stage.

Stunning and **killing** have few microbiological implications, although electrical water-bath stunning may lead to inhalation of contaminated water by the birds and microbial contamination of carcass tissues.

During **scalding**, soil, dust and faecal matter from the feet, feathers, skin and intestinal tract are released into the scald water and thus provide a significant opportunity for cross contamination. A large variety of bacteria, e.g. *Salmonella, Staphylococcus, Streptococcus, Clostridium spp.* have been isolated from scald water or from carcasses or air sacs immediately after scalding.

Bacterial survival in the scald water is influenced by scald temperature and time. The lethal effect of water held at 60°C (hard scald) used for carcasses intended for water chilling is measurable and greater than the lethal effect of water held at lower temperatures, e.g. 50 - 52°C (soft scald) as used for carcasses that will be air chilled.

It has also been demonstrated that scalding results in modifications to the skin : removal or damage of the epidermal layer, exposing a new surface for contamination which is smoother and less hydrophobic, exposure of microscopical channels and crevices. During and after scalding, the skin surface retains a film of scalding water which contains organic matter and large numbers of bacteria. Some of these bacteria may adhere more easily to the modified surface of the skin. Some may be retained in the channels or crevices on the skin surface as well as in the feather follicles. During the following stage of defeathering, there may be entrapment of bacteria in the channels, crevices and follicles. When entrapped, the bacteria may be difficult to remove by subsequent procedures, including mechanical and chemical decontamination treatments; they also display greater heat resistance.

Defeathering with automatic machinery may be expected to cause considerable scattering of microorganisms *in particular via aerosols.* Early findings, from work being carried out in the United Kingdom, indicate that these aerosols from defeathering can be reduced by altering the design of the equipment (Johnston, personal communication 1998). Conditions inside the machines are favourable to the establishment of a biofilm and colonisation by pathogens, in particular *S. aureus* which can survive, multiply and become indigenous to the equipment. Defeathering has been recognised as a major source of carcass contamination with *S. aureus*, *Salmonella*, *Campylobacter* spp and *E. coli*. Several studies have established that the microbial populations on poultry carcasses reflect the microbiological condition of the carcasses immediately after defeathering.

Evisceration can give rise to faecal contamination with enteric pathogens such as *Salmonella, Campylobacter* and *Cl. perfringens*, especially when intestines are cut and/or when automatic machines are not set properly. In addition, microorganisms may be transferred from carcass to carcass by equipment, workers, and inspectors.

Spray washing of carcasses removes visible faecal contamination and some microorganisms such as *Salmonella*. However, it does not eliminate those bacteria that have become attached to the carcass surface or entrapped in the inaccessible sites of the skin surface. It has been demonstrated that continuous carcass washing or applying a series of sprays at the various stages of evisceration removes bacteria before they are retained, and this is much more effective than a single wash after evisceration. There is a danger that use of water sprays, in particular those used in carcass washing, may create aerosols that can spread microbiological contamination.

Three types of **chilling** processes may be used : air blast, water immersion and a combination of air and water chilling. All three methods may lead to some degree of cross contamination. With regard to the final microbiological load on the carcass, it has been demonstrated that properly controlled water immersion chilling can reduce overall levels of carcass contamination. However, high levels of contamination of carcasses before chilling and insufficient water used per carcass (amount of fresh water replacement; number of carcasses in relation to the volume of chilled water) may result in an increase in the level of microbial contamination on carcasses rather than a decrease.

There have been numerous studies to determine the relative effect of each processing step on carcass contamination. Generally, the results show that aerobic plate counts or count of Enterobacteriaceae decrease during processing.

The data on the prevalence of salmonella contaminated carcasses are highly variable. The proportion of contaminated carcasses appears to be influenced mainly by the condition of incoming birds and also by processing. Although the prevalence of salmonella contaminated carcasses can be high, the number of *Salmonella* per carcass is usually quite low. For example, Todd and Harwig, 1996, found that salmonella positive poultry carcasses usually carried not more than 30 salmonella cells per carcass. However, those results would depend on the methods used particularly for carcass sampling.

In comparison with salmonella, campylobacters are generally carried in high numbers by poultry. Therefore carcasses are more readily contaminated during processing and the numbers present are correspondingly higher.

SECTION 2: PATHOGEN REDUCTION

It is widely recognised that successful control of foodborne human pathogens requires a "farm to fork" approach, involving producers, processors, distributors, retailers, caterers and consumers. In the case of poultry meat production, the aim must be to establish a fully integrated control programme throughout the poultry supply chain and beginning on the farm.

2.1 The test/control strategy

A logical strategy for salmonella reduction in poultry would be to ensure the lowest possible prevalence of salmonella in the production pyramid, starting from the top, moving downwards, i.e. from grandparent to breeding to production flocks. This strategy could be based upon flock testing at relevant intervals. For positive flocks appropriate control measures are necessary. Another key element of such a strategy would be hygiene measures which should be applied on the farm and in the slaughtering process (see below). A third element would be the control of salmonella in feed. This control should include process control (HACCP) and the appropriate corrective action when salmonella contamination occurs in processed feed.

At present this type of strategy has been fully introduced for instance in Finland and Sweden, whereas it has been introduced at least in part in several other countries. Elements of this approach have been formally introduced in European poultry production via regulations laid down in the "Zoonosis" Directive 92/117/EEC[1]. Here specific rules for monitoring and control of presence of, especially, *S. typhimurium* and *S. enteritidis* have been laid down for breeding flocks

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[1] Concerning measures for protection against specified zoonoses and specified zoonotic agents in animals and products of animal origin in order to prevent outbreaks of food-borne infections and intoxications, as amended by Directive 97/22/EC

The test/control strategy has not been applied directly to the campylobacter problem. However, in Sweden a campylobacter surveillance programme and the implementation of the basic requirements of the salmonella control programme seem to have resulted in a decrease in the campylobacter flock prevalence from 30 - 40% to 10 - 15% (report on trends and sources of zoonoses 1995).

2.2 Hygienic measures at farm level

Because of the relatively large numbers of birds that are kept together, conditions of intensive rearing tend to favour the spread of any pathogens that gain access to the flocks; however, the use of controlled-environment housing for this purpose does provide an opportunity to exclude undesirable micro-organisms by maintaining an appropriate level of biosecurity.

For salmonella, high standards of personal hygiene are essential and must include proper use of protective clothing, disinfectant footbaths etc.

It is also necessary for each farm to exclude biological vectors as far as possible and to implement rodent-baiting programmes. Ideally, all birds should be taken for slaughter at the same time - the "all-in, all-out" rearing principle. When the houses are empty, spent litter and faeces must be removed from the farm after which the houses, their equipment and the immediate environment must be thoroughly cleaned and disinfected before re-use. It is also advisable to allow the houses to remain empty for as long as possible to allow a natural die-off of any pathogens present.

Successful control of salmonellas on the farm is dependent on high standards of hatchery hygiene and a consistent supply of salmonella-free feed. Measures taken at feed mills to safeguard the final product include the use of a heating process, sometimes combined with chemical treatment of the feed, and care to prevent recontamination during cooling. Short-chain fatty acids, such as formic and propionic acids, may be incorporated in feed and have the advantage of protecting it against recontamination during distribution and storage. The acids can reduce the incidence of salmonella infections in poultry but are active only when the feed is moistened following consumption by the birds. Acids have no beneficial effect once the birds have become infected.

From the moment of arrival on the farm, the young chick is highly susceptible to salmonella infection because it is placed in a strictly sanitised environment, has no contact with parent birds and is slow to develop an intestinal microflora that could successfully compete with any ingested pathogens. It has been found, however, that an adult-type microflora can be established in chicks by oral dosing with suspensions or anaerobic cultures of gut contents from mature, salmonella-free birds (Nurmi and Rantala, 1973). In this way, chicks become more resistant to an oral challenge with salmonellas. The protective effect, which is usually termed "competitive exclusion" (CE, reviewed by e.g. Pivnick and Nurmi, 1982; Schleifer, 1985), is now available in the form of several commercial preparations, any of which can be administered in the hatchery to protect chicks at the earliest possible opportunity.

A particular advantage of CE treatment is that it is not specific and protection has been demonstrated against a variety of salmonella serotypes, including both invasive and non-invasive strains. In contrast, vaccines are generally more specific but have an important role in preventing vertical transmission of invasive serotypes. Research has centred on live,

attenuated and dead vaccines and some are available commercially. An inactivated vaccine is becoming widely used and aims to reduce vertical and horizontal transmission of *S. enteritidis.*

When broiler parent stock are vaccinated with the preparation, it is claimed that chicks show passive immunity for at least 21 days. As with CE treatment, effective vaccination depends upon the simultaneous use of other control measures, especially a high standard of biosecurity (Methner et al. 1997).

Although attention to husbandry hygiene also helps to reduce flock infection with *Campylobacter*, control of the organism is hampered by lack of knowledge on the sources of flock infection, modes of transmission to poultry flocks and availability of suitable preventive measures. It is clear that animal vectors can play a part, as can farm personnel, if hygiene precautions are inadequate. Vertical transmission seems unlikely because campylobacters show poor survival in egg contents and newly hatched chicks are invariably free from overt infection. Nevertheless, some evidence suggests that vertical transmission could occur.

Feed, on the other hand, is too dry to favour survival and is not regarded as a source of campylobacter infection. There is a possibility that untreated water-supplies can transmit the organisms and, if mains water is not available, the supply to the growing houses should be chlorinated. Since campylobacters survive well in biofilms, thorough cleaning and disinfection of the water-supply system in each house is essential between different crops of birds.

2.3   Hygienic measures in the processing plant

In relation to slaughter and processing of the birds, the main problem is control of cross-contamination, which begins at the transportation stage. If live-bird crates and lorries are not adequately cleaned and disinfected after delivering one batch of birds and before transporting another, then there is a strong possibility of transmitting any pathogens present from the first flock to the second (Bolder and Mulder, 1987). At the processing plant, contamination of carcasses can occur via contact with soiled surfaces, equipment or the hands of operatives. Microorganisms can also be spread in airborne dust particles and droplets and through any rupture of the intestines during evisceration. Certain of the requirements in Directive 71/118/EEC aim to limit opportunities for cross-contamination, for example physical separation of the hanging-on bay and the scalding and plucking area. There is also stringent control of conditions used in water immersion chilling. Other precautions include avoidance of unnecessary contact between carcasses and soiled surfaces and the use of chlorinated water-sprays to sanitise equipment and surfaces during the processing period (Mead et al., 1980). It is also beneficial to wash the carcasses at any point where they may be exposed to faecal contamination. In this way, microbial contaminants are removed before they can become irreversibly attached to the carcass surface (Notermans et al., 1980). Such measures, along with e.g. proper use of protective clothing and hand-washing facilities by operatives, are regarded as Good Manufacturing Practices and are part of the overall HACCP system in processing. In this situation, a terminal decontamination step would provide a much-needed Critical Control Point that does not exist at present, but it would need to supplement existing measures and cannot be allowed to mask bad practices.

Poultry meat production is now highly automated, although the technology involved was not developed with hygiene in mind. Instead, the main considerations are cost-effective operation of the machinery and maintenance of product yield (Hupkes, 1996). More recently, however, hygiene control has been given a higher priority and several new developments have been made. These include multi-stage scalding, a new type of evisceration system, better equipment for cleaning and disinfecting live-bird delivery crates and cleaning-in-place systems for eviscerators and conveyor belts.

Although they are not yet used universally within the poultry industry, most of the newer machines are gradually gaining acceptance. However, there is no clear evidence that they significantly affect the microbiological condition of the finished carcass.

SECTION 3: ROLE OF POULTRY CARCASS DECONTAMINATION IN PREVENTION OF FOODBORNE DISEASES - RISK ASSESSMENT

3.1 The assessment of management options to reach acceptable risk levels for poultry consumption

An acceptable level of microbial safety of poultry can be achieved by various means. It is generally agreed that the determination of actual and acceptable risk should be based on scientific data. However, the pathogen situation in poultry production and the production systems can differ significantly between countries or regions, as can the quality and amount of data available. The Sanitary and Phytosanitary (SPS) agreement recognises this and states that the definition of an acceptable level of risk is a national decision. It should also be recognised that the determination of safe, realistic and achievable risk levels depend not only upon science, but also upon a number of social, economic and technological factors.

The risk management strategies for zoonotic pathogens vary among regions and nations. Modern concepts of dealing with zoonotic pathogens 'from farm to fork' works best in an infrastructure where a relatively high level of control can be achieved throughout the food chain. Such levels of control should be achievable in industrialised production systems, such as poultry production.

The discussion of risk management options to reach a 'target level' of acceptable risk from the consumption of poultry is ongoing. The use of the word 'target level' reflects the dynamic nature of foodborne disease risk and the fact that this risk will never reach zero. In principle, targets should be set primarily in relation to the incidence of human disease, since the risk concept inherently relates to this. However, in a number of cases risk management initiatives will only relate indirectly to human disease and the primary initiatives will centre around the definition of tolerable hazard levels or prevalence in the food. Possible management initiatives could encompass control at source, action plans at the production level, criteria for the final product, treatment strategies for the final product or a combination of these.

The use of antimicrobial treatment for poultry carcasses represents (part of) a management option. When evaluating this option using scientific data, it is important to realise that the process can be considered both from a risk assessment and a risk management angle. When using only the risk assessment angle one option can be

evaluated separately, but in risk management the option and its alternatives should be evaluated and compared, for example using cost-benefit analysis based on scientific data and risk-benefit evaluation. Therefore an interaction between risk assessment and risk management is necessary here. The use of such antimicrobial treatments should at some stage of the evaluation process be related to the alternative, i.e. the production of poultry without, or with a very low level of, specific pathogens. These considerations are not straight forward, especially in relation to the magnitude and the diversity of the microbiological problems in poultry production. Nevertheless for the purpose of this report a partial risk assessment is sufficient to indicate the likely benefit of carcass decontamination.

3.2   Partical risk assessment

Potential of Antimicrobial Treatments to reduce the Risk of Foodborne Disease

In the case of pathogens such as *Salmonella*, which can grow on chicken carcasses if the temperature is right, the initial level of contamination is of minor importance. If temperature abuse occurs the organisms will reach infectious levels in about the same time.

However, it makes a difference whether there are no salmonellas or some salmonellas and the production goal should be to have as large a proportion of chicken carcasses as possible free of pathogens. If a carcass is free of pathogens there will be no risk to the consumer, even if temperature abuse occurs.

In experimental studies of pathogen reduction the results are expressed as per cent reduction or log reduction, while regulatory agencies (Code of Federal Regulations, 1996) and consumers will focus on the proportion of the positive carcasses. This creates a need to translate proportionate reduction into the proportion of positive carcasses.

If the assumption is made that pathogens on poultry carcasses follow a Poisson distribution and that poultry belong to the same general population, the mean number, u, of pathogens per carcass can be calculated as in equation **1**

$$u = \ln 100 - \ln \text{(per cent negative carcasses)}^{(*)} \quad [\mathbf{1}]$$

A reduced mean number, u, resulting from a pathogen reduction process can be translated into the probability, P, of <u>no</u> pathogens on a carcass using the first element of the Poisson distribution as in equation **2**

$$P = (e^{-u} \times u^{0})/0! = e^{-u \,(*)} \quad [\mathbf{2}]$$

(*)where: ln = natural logarithm

(*)where: e =base of natural logarithm = 2.7182; u= the mean number and 0!=1

The probability of a consumer getting at least <u>one</u> contaminated carcass per year, if buying one chicken per week, can then be calculated using a binomial sampling distribution. The use of one year as a unit may seem arbitrary, but was selected because most foodborne diseases are reported on a yearly basis.

Example:

Assume 70% of poultry carcasses are positive for *Salmonella* if not subjected to a pathogen reduction process. The probability of getting at least one contaminated chicken per year is calculated as in equation **3**

$$1 - (0.30)^{52} \sim 100\% \quad \textbf{[3]}$$

and the mean number us shown in equation **4**

$$u = \ln 100 - \ln 30 = 1.204 \quad \textbf{[4]}$$

This represents the most probable number of salmonellas per carcass.

After pathogen reduction of 90 % the mean number is 0.1204, and the probability, P, of <u>no</u> *Salmonella* on a carcass is shown in equation **5**

$$P = (e^{-0.1204} \times 0.1204^{0})/0! = e^{-0.1204} = 0.8866 \quad \textbf{[5]}$$

The risk of getting at least one contaminated chicken per year is shown in equation **6**

$$1 - 0.8866^{52} = 0.9981 \text{ or } 99.81\% \quad \textbf{[6]}$$

The following table shows the probability of getting at least one contaminated carcass per year in relation to different initial contamination levels and different efficiencies of pathogen reduction

<u>Probability of getting at least one contaminated carcass per year, when buying one per week</u>

% reduction	Per cent carcasses positive before pathogen reduction			
	70	**50**	**20**	**5**
60	100%	100%	99.04%	65.69%
70	100 -	100 -	96.92 -	55.07 -
80	100 -	99.93 -	90.18 -	42.34 -
90	99.81 -	97.28 -	68.66 -	23.41 -
99	46.53 -	30.26 -	10.96 -	2.63 -
99.9	6.07 -	1.84 -	1.15 -	0.27 -

These data show that the protective effect of a hypothetical antimicrobial treatment depends strongly on the level of contamination. For example if the goal is, with a *99%* probability, to reduce consumer risk to less than one contaminated chicken per year per family, the proportion of contaminated carcasses must be below 5% and the efficiency of pathogen reduction above 99.9%.

SECTION 4: PRINCIPLES FOR THE ASSESSMENT OF ANTIMICROBIAL TREATMENTS (DECONTAMINATION TREATMENTS)

From a public health point of view, a given decontamination treatment can be sustainable if, and only if, the **efficacy**, the **safety** and the **controllability** of that treatment can be demonstrated, using the best available information and data. Other considerations may refer to the impact of the technique on product quality, worker safety, protection of the environment and consumer acceptance.

The aim of this section is to identify the aspects / characteristics of a decontamination treatment to be addressed. The discussion is to serve as guidance for the assessment of any technique already in use or proposed for use, for decontamination of poultry carcasses.

It has to be recognised that, when assessing a given decontamination treatment, not all of the following considerations apply equally to every kind of treatment. Also, where some essential information is missing, additional research work may be necessary before an informed judgement can be made.

When assessing a decontamination treatment, the following aspects should be considered:

- Overall efficacy
- Microflora changes and implications
- Potential for introducing other food safety hazards
- Potential for occupational hazard (worker safety)
- Impact on the environment
- Effects on sensory properties and quality of the product
- Feasibility and effectiveness of control under commercial conditions
- Consumer perception

These aspects will be considered in turn in the following.

4.1 Overall efficacy

- The *sine qua non* for a decontamination treatment is that it effectively reduces the level of target pathogens. Therefore, the effectiveness of the treatment on pathogen reduction should always be appropriately demonstrated.

- Demonstration of effectiveness should include not only laboratory tests but also in-plant investigations.

- Demonstration of efficacy basically involves measuring the reduction in pathogens of concern by determining the levels present before and after treatment.

- The results should also be expressed in terms of the proportion of positive carcasses before and after treatment.

- Because a given decontamination technique may impact differently on different groups of bacteria, investigations should include different pathogens of concern.

- When assessing the overall efficacy of a decontamination treatment, it has to be borne in mind that the efficacy may be affected by a number of factors. Depending on the technique at hand such factors may include:

  - efficacy in the presence of organic material
  - characteristics of the skin surface e.g. channels, crevices, follicles that facilitate entrapment of bacteria
  - processing stage at which treatment is applied (effectiveness of treatment is expected to be greater when the treatment is applied before the contaminants have attached firmly to the carcass).
  - physical parameters such as concentration of chemicals, pH, temperature of a of treatment.

- Finally, in assessing test results and when comparing them, due consideration should be given to the experimental conditions and to the microbiological technique used in the investigation because not all techniques will adequately recover attached bacteria.

4.2 Microflora changes and implications

- The effects on both pathogenic and spoilage microorganisms should always be investigated.

- It has to be realised that the treatment may in turn influence:

  - the equilibrium and development of residual or resistant spoilage microorganisms with possible impact on spoilage characteristics and duration of shelf life
  - the development of residual levels of pathogens, or of pathogens which are less sensitive to the treatment, or of pathogens that may be added by re-contamination after treatment. Reducing or eliminating the competitive microflora may allow uninhibited growth of pathogens. This is of particular concern when the overall reduction of spoilage microorganisms results in an extended shelf life.

Therefore, consideration of changes in microflora and their implications is an essential part of any decontamination treatment assessment.

This may involve:

  - effect on spoilage microorganisms and changes on spoilage microflora
  - conditions under which a modified new spoilage microflora may develop impact on shelf life
  - implications of such changes on the potential development of pathogens

- assessment of such potential development of pathogens vs potential shelf life extension

- It has to be recognised that:
  - strains of a given bacterial species may vary widely, especially with regard to their attachment capability and sensitivity to antimicrobial treatments;
  - repeated or continuous use of a given antimicrobial treatment may select strains resistant to factors usually used to control the microbial development (e.g. resistance to acid conditions, resistance to higher temperatures, resistance to antibiotics etc.)
  - decontamination treatments may impact on the physiological status of surviving pathogens (e.g. transformation to viable but non cultivable forms)

At present, these aspects have been poorly investigated, if ever. Such considerations may probably not contradict the immediate utility of antimicrobial treatments to reduce pathogen levels in foods. However, additional research in these fields is necessary to fully assess the potential long term consequences of otherwise accepted treatments.

4.3 Potential for introducing other food safety hazards

- Antimicrobial treatments used for pathogen reduction should not increase the risk of other health hazards.

- In particular, a toxicological assessment should always be performed. For chemical compounds it should be determined whether the decontamination treatment leaves residues on/in the meat and / or whether the treatment leads to the formation of secondary toxic compounds.

4.4 Potential for occupational hazards

- The implementation of a given decontamination treatment should not introduce an occupational hazard for the workers, such as potential for causing skin irritation, hypersensitivity, carcinogenic effect, etc.

4.5 Impact on the environment

- The use of a decontamination treatment should not negatively impact the environment.

- In particular, where a chemical compound is used, assessment may involve determining:

  - the frequency and level of chemical compound released
  - the degradability of the compound
  - the activity of the compound
  - the environmental impact of releasing the compound

- opportunities for recycling

4.6 Effect on the sensory properties and quality of the product

- Decontamination treatments should not significantly alter the sensory properties of the food nor its characteristics with particular regard to adulteration and possible change in nutritional value.

- Assessing a decontamination technique may include investigations on possible changes in

  · organoleptic properties e.g. colour, odour, taste

  · composition e.g. water retention, drip loss

  · cooking or technological properties e.g. melting of fat resulting in partial cooking nutritional properties

4.7   Feasibility and effectiveness of control under commercial conditions

- Investigations are needed to determine the feasibility and effectiveness under commercial conditions.

- Appropriate assurance should be provided on effective control and monitoring of treatment conditions as well as factors affecting efficacy and safety of the technique.

This may require in particular:
· the establishment of the required conditions for application and their documentation (e.g. written procedures) as appropriate. Such documentation should also include procedures aimed at avoiding re-contamination after treatment;
· the establishment of limits or values of parameters critical to efficacy and safe use (e.g. dose, concentration, temperature, duration)
· the establishment and implementation of appropriate monitoring systems.

- Evidence should be provided on how the technique can fit within the HACCP plan of the users and whether the decontamination process can be operated as a Critical Control Point.

The Committee recommends that the competent authorities develop appropriate control systems for the residues of a substance in the poultry meat.

4.8   Consumer perception

- Notwithstanding safety considerations, consumers may have some reservation regarding the decontamination treatment used , e.g. possible changes in the sensory properties of the product, the use of irradiation or for religious reasons (e.g. kosher process)

- An important part of any decontamination treatment assessment is a detailed investigation of consumer perception and acceptance. It may, therefore, be necessary for products receiving a specific decontamination process to be labelled accordingly e.g. if irradiation has been used. The information given should be clear and concise and without ambiguity.

Introduction

The poultry production allows many opportunities for microbial contamination and cross-contamination. Numerous attempts have been made to find an appropriate means of eliminating or at least reducing such contamination by the use of an end-product treatment. The ideal method would have no adverse effect on the appearance, smell, taste or nutritional properties of the meat, leave no undesirable residues, pose no threat to the environment and raise no objections from consumers or legislators. It would also be low in cost and easy to apply effectively. The treatment should be used as an adjunct to good hygienic practices and not as a substitute for them.

Does such an ideal treatment exist? Table 1 shows in summary form the many and varied options that have been investigated. These have involved a variety of antimicrobial chemicals, some of which would appear to be unsuitable for use in a food processing environment or carry a risk of tainting the product. For example, dipping chicken carcasses in a solution of hot succinic acid was found to be highly effective in destroying salmonellas, but had an adverse effect on meat colour. Physical treatments, too, have been studied and new methods are under development. These have the advantage of avoiding any chemical residues or problems of waste disposal. However, they may still affect the appearance of the product unless a high degree of control can be exercised. Combinations of different treatment are also possible, but have yet to be fully investigated.

Important remark

This report will be confined to certain treatments involving the use of one single physical, chemical or biological agents that appear feasible under commercial conditions.

As other techniques may be proposed, these are covered as examples only, to enlighten some of the factors which may be included in the assessment. Furthermore, the following examples should not be perceived as complete assessments, as some important elements, such as toxicology or consumer perception, have not been addressed.

The techniques covered as examples in this report and other techniques proposed for use, in particular those which involve a combination of treatments or agents, still need to be fully assessed with regard to all elements listed in Section 4. Related investigations and demonstrations should be carried out by the person / company proposing such techniques and the results should be presented for review to the competent authorities.

Table 1: Possible treatments for reducing microbial contamination of poultry carcasses, either during or after processing

Physical	Chemicals	Microbial

Cold and hot water	Organic acids	Lactic acid bacteria
Steam	Inorganic acids	Bacteriocins
Steam-vacuum	Alkalis	Microbial parasites
Ultrasound	Sorbates	
Electromagnetic radiation	Chlorine and related compounds	
Ionising radiation	Phosphates	
Electrical methods	Ozone	
High pressure	Hydrogen peroxide	
Air ions	Salts (other than above)	
Freeze-thaw cycling	Lysozyme	
	Disinfectants	
	Antibiotics	
	Lactoperoxidase system	
	EDTA	
	Ethanol	

5.1   TREATMENT WITH WATER, HOT WATER, STEAM

5.1.1   Safety of procedures

Washing with potable water is safe but care should be taken to avoid excessive generation of aerosols. Treatment with hot water or steam must also be considered safe provided that carcasses are cooled promptly after treatment.

5.1.2   Efficacy of treatments

The efficacy of treatments has been evaluated in laboratory experiments using artificially contaminated carcasses and in some cases in slaughter establishments using naturally contaminated carcasses. The results are generally expressed as per cent reduction or log unit reduction. In this review, data - when necessary - have been translated to per cent reduction.

<u>Washing with water</u>

Washing with water is routinely used in poultry processing plants and the carcasses are washed after different operations; this may result in an overall reduction of surface contamination by 90- 99%. This is not achieved by a single wash but through the combined effect of a number of washes.

<u>Hot water</u>

Most of the published studies have been done with beef, relatively few with poultry. Hot water (74-95°C) was shown to reduce *E. coli* by 99-99.9%. Hot water sprays were most effective when they raised the carcass surface temperature to 82°C for 10 seconds; 20 seconds resulted in permanent colour change of the beef surface (Federal Register, 1995).

Another study (Federal Register, 1995) found that water at 70°C resulted in a 99% or higher reduction in *E. coli* inoculated on beef. Dipping sheep carcasses for 10 seconds in 80°C water reduced *E. coli* by 99% and total counts by 98%. Pouring 77°C water on beef slices for 10 seconds reduced inoculated *E. coli* and *Salmonella* by more than 99%. Other studies with hot water applied to beef slices (Federal Register, 1995) found that time of exposure was not a factor, but a progressive reduction was achieved with increasing temperature from 90% reduction at 60°C to more than 99.99% at 90°C. One study (Federal Register, 1995) compared the effect of cold water (16°C), hot water (76-80°C) and steam (95°C) on total counts on previously frozen beef. Cold water reduced the counts by 90%, steam alone by 13% and hot water by 99%. During subsequent holding at 3.3°C bacteria grew most rapidly on cold water and steam treated samples; the rate of growth on the hot water treated samples was the same as on the controls.

In a study using broiler carcass wings, Rodriguez de Ledesma et al., (1996) compared the effect of water at 95°C for 10 seconds to the effect of dipping in 10% trisodium phosphate or 10% sodium carbonate for 10 seconds and to the effect of combination treatments, where treatment with hot water was applied after the treatment with alkali. Trisodium phosphate was more effective than sodium carbonate. Trisodium phosphate alone caused reductions of 84.33%, 65.33% and 60.19% of *S. typhimurium*, *Staph. aureus* and *L. monocytogenes,* respectively. Hot water treatment alone caused 64.52%, 91.29% and 91.27% reductions of these organisms. Combination of trisodium phosphate and hot water resulted in 98.59%, 99.54% and 99.73% reductions.

A combination of hot water and sodium carbonate had a similar effect. The hot water treatment, alone or in combination with alkali, changed the appearance of the skin, making it less translucent. The study did not reveal any presence of viable but injured bacteria after treatments. Castillo et al. (1998) observed that hot water (95°C) reduced experimentally inoculated *S. typhimurium* and *E. coli* O157:H7 on beef carcasses by 99.98%, and naturally occurring coliforms by 99.95%. Validation of the efficacy of hot water treatment as a critical control point in HACCP may be possible using coliform counts.

Steam pasteurisation (Phebus et al., 1997) resulting in a carcass surface temperature of 90-100°C for approximately 15 seconds was found to be more efficient than other decontamination procedures, and resulted in 99.68% to 99.98% reduction of *L. monocytogenes, S. typhimurium* and *E. coli* O157:H7 on the surface of beef carcasses. The process was evaluated in a commercial beef processing facility; it reduced the number of carcasses positive for *Enterobacteriaceae* from 46.4% to 2.9% and for generic *E. coli* from 16.4% to 0% (Nutch et al., 1997). A pasteurisation process using steam at 140°C for 50 milliseconds was found to reduce *L. inocua* on poultry carcasses by 99.99% (Morgan et al., 1995).

The results of pathogen reduction studies using hot water or steam are quite variable, probably depending on the way in which the carcasses were contaminated. Steam pasteurisation seems the most promising. However, there is a paucity of studies using naturally contaminated chicken carcasses and the potential for regrowth has not been investigated in most studies.

5.2   IONISING RADIATION

5.2.1   Effects on microorganisms

The biological effects of ionising radiation on cells can be due both to direct interactions with critical cell components and to indirect actions on these critical targets by molecular entities such as free radicals formed from water. The response of a microbial cell and hence its resistance to ionising radiation depends on the nature and amount of direct damage, the amount of reactive chemical entities and the ability of the cell to repair damage. Ionising radiation is capable of causing a variety of chemical changes in microorganisms. It is generally assumed that the DNA is the most critical target of ionising radiation and the inactivation of microorganisms by ionising radiation is a result of damage to the DNA. Radiation resistance varies widely among different microorganisms.   There can be differences in inherent resistance from species to species, and even among different strains of the same species.

The major environmental factors that influence the survival of irradiated cells are: temperature, gaseous atmosphere, water activity, pH and chemical composition of the food (Grecz et al., 1983). Bacterial spores appear to be less susceptible to modifying factors than are vegetative cells. Since part of the effect of ionising radiation on a microorganism is due to indirect action mediated through free radicals, the nature of the medium or menstruum in which the microorganisms are suspended obviously plays an important role in determining the dose required for a given microbicidal effect. The more complex the medium, the greater is the competition by its components for the free radicals formed by irradiation within the cell, thus protecting the microorganisms by absorbing the free radicals.

Therefore care should be taken in extrapolating resistance data from use of in laboratory media to foodstuffs. Generally microorganisms in food are more resistant to irradiation than those tested in vitro.

Microorganisms that survive irradiation treatment seem to be more sensitive to environmental conditions (temperature, pH, nutrients, inhibitors, etc.) than untreated cells (Welch and Maxcy, 1979).

The dose required to decontaminate a food depends on the initial level of the contaminating microorganisms. Thus, it requires a larger dose to inactivate a large number of microorganisms than to inactivate a small number. This corresponds to the fact that the time necessary for temperature inactivation of microbial populations also depends on the initial concentration of microorganisms.

Highly radiation resistant vegetative bacteria do exist. Some strains, notably belonging to the Moraxella-Acinetobacter group have a radiation resistance greater than that of bacterial spores. Such bacteria appear to be part of the normal flora of meats (Welch and Maxcy, 1975).

5.2.2   Effect on pathogens commonly occurring in poultry

Certain bacterial species and genera are more resistant to irradiation than others. Sporeforming genera (*Bacillus* and *Clostridium*) are resistant but also other Gram positive families (i.e. Micrococcaceae and Lactobacillaceae) show high resistance.

Some of the most important poultry related pathogens are described in relation to irradiation efficiency.

Bacillus and Clostridium

Thayer and Boyd (1994) investigated the gamma-irradiation resistance of vegetative cells and endospores of enterotoxigenic and emetic toxin producing strains of *B. cereus* in chicken meat and other meat types. In this study the D_{10} values *for B. cereus* were 0.18, 0.43, and 2.6 kGy for logarithmic-phase cells, stationary-phase cells, and endospores at 5 °C. A dose of 7.5 kGy at 5°C was required to eliminate a challenge of 4.6 x 10^3 *B. cereus* from temperature-abused chicken meat.

This indicates that irradiation of meat or poultry can provide significant protection from vegetative cells but not from endospores of *B. cereus*. Other studies with spore-forming bacteria in chicken meat relate primarily to *Cl. botulinum* and temperature abuse of food later in the production chain. Such studies are not considered further here.

Listeria

Patterson et al. (1993) inoculated raw and cooked minced poultry meat with *L. monocytogenes* at levels allowing detectable numbers to survive irradiation (1.0 or 2.5 kGy). Lag phases of *L. monocytogenes* were 1 and 18 days in unirradiated and irradiated (2.5 kGy) cooked poultry meat, respectively. The authors conclude that the significant recovery time required for *Listeria* suggests that growth of the bacteria would not pose a food safety risk during shelf-life of the poultry meat. It should be noted also, that long lag phases of surviving populations pose a special problem in relation to the bacteriological monitoring of irradiation efficiency.

Mead et al. (1990) surface-inoculated poultry carcasses with *L. monocytogenes* at counts of approx. 10^2 or 10^4 cfu/cm-2. The carcasses were then gamma-irradiated at 2.5 kGy and stored at 5 and 10°C. Immediately after irradiation, only 1 of 12 carcasses was positive for *L. monocytogenes*, and no further positive sample was detected until day 14 at 5°C or day 5 at 10°C. Twenty two percent of the samples contained viable *L. monocytogenes* at the end of storage, and growth of surviving *L. monocytogenes* in irradiated samples was slow. In control samples, counts increased by a factor of 10^2 in 7 days at 5 °C, 3 days at 10 °C.

Patterson (1989) found D_{10} values on poultry meat of 0.42-0.55 kGy depending on strain and plating medium used. D_{10} values for *L. monocytogenes* were similar to those reported for *Salmonella* spp. irradiated under similar conditions. Therefore irradiation doses suggested to eliminate salmonella[e] from poultry would according to the author also be sufficient to remove *L. monocytogenes*. Huhtanen et al. (1989) found that *L. monocytogenes* surviving an irradiation dose of 1.5 kGy were no more radiation resistant than those which had had no previous exposure to irradiation. These studies indicated that a dose of 2 kGy was sufficient to destroy 1 x 10^4 cells of *L. monocytogenes*.

Kamat & Nair (1995) found some radiation protection of *L. monocytogenes* in chicken homogenates in comparison to irradiation in phosphate buffer at pH 7.00. The D_{10}

values in chicken was 0.5 kGy. The results were reproduced with 4 different serotypes of *L. monocytogenes* and it is suggested that a dose of 3 kGy is needed for elimination of 10^3 *L. monocytogenes*/g in chicken meat.

Campylobacter

Patterson (1995) investigated the sensitivity of *C. jejuni, C. coli, C. fetus* and to irradiation in poultry meat. The D^{10} values ranged from 0.12 to 0.25 kGy and there was a significant difference in the radiation sensitivity between different *Campylobacter* spp. and within strains of the same species. The values indicated that *Campylobacter* spp. are more sensitive than *Salmonella* and *L. monocytogenes* irradiated under similar conditions, and the authors concluded that irradiation treatments suggested to eliminate *Salmonella* and *L. monocytogenes* from poultry would also be sufficient to inactivate *Campylobacter*.

E. coli

The effect of electron beam radiation on survival of *E. coli* on chicken meat was studied by Banati et al. (1993). Radiation dose of 5 kGy gave a 3-4 log reduction when the bacteria was present on meat, but there was a pronounced tailing effect with increasing doses of ionising radiations.

Salmonella

Thayer et al. (1991) examined the effects of gamma-radiation preceded or followed by heating at 60°C for 3 min on the survival of *S. typhimurium* in chicken meat. Treating inoculated chicken meat with gamma-radiation made much more sensitive to the effects of heat. A radiation dose of 0.90 kGy followed by above-mentioned heat decreased the number of survivors by 8 to 9 log units.

Kamat et al. (1991) treated fresh and frozen chicken meat with a dose of 2 kGy at -40°C, and thereby eliminated the natural *Salmonella* contamination of poultry (all untreated samples positive for *Salmonella*). Chicken samples artificially inoculated with 10^8 cells/g of *S. seftenberg* and *S. typhimurium* required gamma radiation doses of up to 4-5 kGy. This would suggest that a dose of 2 kGy is adequate for normally contaminated chicken samples, whereas heavily contaminated chicken requires 4-5 kGy, depending upon the predominant *Salmonella* serotype.

Heath et al. (1990) investigated the effect of radiation at 100-700 krad upon counts of salmonellae in naturally contaminated poultry. Radiation at 100 krad almost totally eliminated *Salmonella* in chicken meat, whereas a majority of non-irradiated samples were *Salmonella*-positive.

5.2.3   Influence on microbial ecology of food

Any treatment (including irradiation) which partially eliminates the indigenous flora of carcasses or food may allow a surviving population of pathogenic organisms to grow faster than in untreated food. In the absence of the spoilage organisms, the food could then appear fit for consumption, based on typical organoleptic properties, yet contain pathogens. It has been stated that it is necessary to conduct studies on the effect of

irradiation on the numbers and types of microorganisms in food immediately following treatment and under the conditions that might prevail in commerce and in the home (Anon., 1994). The potential growth of pathogens in different types of meat packaged under modified atmosphere is particularly important. Likewise, the 'Sous-vide' process has a potential to affect the balance between the natural microbial flora of meat and any pathogens that may be present, resulting in a situation conducive to pathogen growth. These are few scientific data on these aspects.

The FDA has approved a proposal to irradiate chicken at a maximum dose of 3kGy to control foodborne pathogens such as *Salmonella* and *Campylobacter* (Anon., 1990). At such doses, the pathogens likely to survive will be spore-forming microorganisms. Thus *Cl. botulinum* poses a real problem in regard to microbial safety because its spores are more resistant than vegetative forms to radiation (Anon., 1990). The possibility that *Cl. botulinum* could produce toxins without any sign of spoilage obvious to the consumer, has been investigated. A series of studies with *Cl. botulinum* type E revealed that both irradiated and nonirradiated chicken stored at 30°C developed toxin, but chicken irradiated at 3 kGy did not develop toxin under any storage conditions before off-odours characteristic of spoilage became apparent (Firstenberg-Eden et al., 1983, Rowley et al., 1983). In another study (Dezfulian & Bartlett, 1987) examined the effect of irradiation (3 kGy) on growth and toxin production of *Cl. botulinum* types A and B on chicken skin. These clostridia are more commonly found on chicken than type E. At the abuse temperature of 30 degrees *Cl. botulinum* toxin was formed in both irradiated and nonirradiated chicken but toxin formation was delayed in irradiated chicken. No toxin was found before off-odours, signalling spoilage of the food, were produced.

5.2.4   Mutations and other changes

It has been known for many years that ionising radiation increases the rate of mutation in living organisms. It has been shown that irradiation can result in loss of virulence and infectivity (Farkas, 1988). In theory, mutations could also result in an increase in virulence, but there are few, if any, examples of this.

The use of ionising radiation might select for radiation resistance strains. It is extremely difficult to induce such mutations, especially by a single treatment. The difficulties in isolating radiation-resistant mutants from single radiation treatment, suggests that wild types of natural strains of bacteria have already evolved an adequate DNA repair capacity (Anon., 1994). It is possible to develop radiation-resistant populations by subjecting bacteria to many sequential radiation treatments.

Some data suggest that irradiation could increase mycotoxin production, whereas other studies indicate just the opposite (Anon., 1994).

5.2.5   Outstanding general questions

The microbial safety of irradiated food was addressed by the International Committee on Food Microbiology and Hygiene. The Committee concluded that food irradiation could be an important addition to existing methods of control of foodborne pathogens and did not present any additional hazards to health (FAO, 1983).

Nevertheless, consideration should be given to the following :

- The influence of surviving pathogens relative to other surviving microorganisms.

- The problem of potential radiation resistance.

- Long lag phases of surviving pathogen populations could pose a special problem in relation to bacteriological monitoring.

- The varied D_{10} values could give cause for concern, e.g. *L. monocytogenes* with a D_{10} value reported for some organisms of 0,4 to 3 kGy in poultry carcasses or meat.

- Some studies show a tailing effect on the survivor curve with increasing doses of radiation.

- There seem to be no reported investigations relating radiation to the formation of viable but not cultivable forms.

- Consumer perception should be taken into account.

5.3   ORGANIC ACIDS

The effort to decontaminate poultry meat with organic acids and their salts to extend shelf life and reduce pathogens, especially *Salmonella*, started in the late 1950's. Efforts have been concentrated on short-chain organic acids and mainly those that are generally recognised as safe (GRAS). Acids which have been evaluated include acetic, lactic, citric, propionic, fumaric, tartaric and succinic. Acceptable daily intakes for humans are given in table 1.

5.3.1   The acids

Acetic acid

Acetic acid is the principal component of vinegar and as such is primarily used for flavouring but also for its antimicrobial action. It is a GRAS substance (21CFR 184.1005) with no upper limit of daily intake for humans (Directive 95/2, FAO 1965).

Lactic acid

Lactic acid is one of the most widely distributed acids in nature. It is produced as the principal acid during all food fermentations involving lactic acid bacteria (LAB). As a commercial product it is used in the manufacture of jams, jellies, sherbets, confectionery products, etc. It is also used to improve quality and to control microbial growth in a variety of foods. Lactic acid is approved as a GRAS substance (21 CFR 184.1067) with no upper limit of daily intake for humans (Directive 95/2, FAO 1965).

Citric acid

Citric acid occurs in nature in all citrus juices and it is used in citrus-based foods for flavouring and preservation purposes. It is approved as a GRAS substance (21 CFR 182.1033) with no upper limit of daily intake for humans (Directive 95/2, FAO 1966).

Propionic acid

Propionic acid is normally produced during fermentation by propionic acid bacteria of some foods mainly cheeses. Certain types of cheese, e.g."Swiss cheese" contain up to 1% propionic acid. Proprionic acid exerts antibacterial and anti-mold action and it is approved as a GRAS substance (21 CFR 184.1081) with no upper limit of daily intake for humans (FAO 1965).

Fumaric acid

Fumaric acid is widely distributed in nature and occurs in many foods as a result of fermentation of glucose and molasses. It is used in fruit drinks, gelatine desserts, biscuits and wines. It is naturally present in fresh meat in concentration of 0.14 to 0.20% (Stoll 1970). For this reason fumaric acid is considered as a (GRAS) substance (21 CFR 172.350) but an upper limit of daily intake for humans of 6 mg/Kg of body weight is set by FAO (1974) or from 1 to 4 g/Kg of foods (Directive 95/2).

Succinic acid

Succinic acid is a GRAS substance (21 CFR 184.1091). It is incorporated into gelatine desserts and in cakes as flavouring agent. No limit has been set on the acceptable daily intake for humans (Doores 1993).

Tartaric acid

Tartaric acid is a GRAS substance (21 CFR 184.1099). It is used in marmalades, jellies, fruit jams and grape flavoured beverages. An upper limit of 30 mg/Kg of body weight for humans has been set by FAO (1973).

Table 1.   Acceptable Daily Intake for Humans

Acid	Limitations (mg/kg body weight)	Reference	
Acetic	Not limited	Directive 95/2/EC	FAO 1965
Lactic	Not limited	Directive 95/2/EC	FAO 1965
Citric	Not limited	Directive 95/2/EC	FAO 1966
Propionic	Not limited	-	FAO 1965
Succinic	Not limited	-	(Doores 1993)
Fumaric	0-6	Directive 95/2/EC	FAO 1974
Tartaric	0-30	Directive 95/2/EC	FAO 1973

5.3.2 Antibacterial activity

The effectiveness of short-chain organic acids as antimicrobials is influenced by many factors such as carbon chain length and variant susceptibility of different microorganisms. However, the most important factor is the pH of the environment.

The magnitude of the antimicrobial activity depends on the nature of the acid and pH. This is understandable since the bactericidal and bacteriostatic effects of acids in general have been shown to depend on the degree of acid dissociation at a particular pH and the concentration of undissociated acid remaining in the solution. The antimicrobial effect increases with increasing concentration of undissociated acid (Doores, 1993). The antimicrobial activity is also affected by the nature of the acid molecule, the type of bacteria, the temperature of the acid solution and the exposure time (Mountney and O' Malley, 1965; Juven et al., 1974; Thomson et al., 1976). In relation to meat surfaces (carcasses) additional factors are the type of tissue being treated and the stage of application. Antimicrobial effects are much more marked on fat tissue surfaces than on lean, because lean meat has greater buffering (neutralising) capacity. On the other hand the earlier the application, before bacteria are firmly attached to fresh meat surfaces, the better the antibacterial action (Smulders, 1987).

Decontamination of poultry meat

Thomson et al., (1976) compared the effect of acid, heat treatment and chlorine on *Salmonella* contamination of broiler carcasses and observed that succinic acid alone (1% at 55 ^{o}C) reduced *Salmonella* prevalence to 50%.

Lillard et al., (1987) reported that 0.2-0.5% acetic acid in the scalding water reduced Total Plate Count (TPC) and Enterobacteriaceae. No significant reductions occurred in the prevalence of *Salmonella* or on levels of TPC and Enterobacteriaceae on picked carcasses that were sprayed with 0.5% acetic acid. On the contrary Okrend et al., (1986) added 0.1% acetic acid to scald water and observed a reduction in levels of *S. typhimurium and C. jejuni* by 0.5 to 1.5 log CFU.

Van der Marel et al., (1988) investigated the effect of immersing broiler carcasses in 1-2% lactic acid (pH 2.2 at 19 ^{o}C) for 15 seconds at various stages of processing on surface TPC, total psychrophiles, Enterobacteriaceae and *Staph. aureus*, as well as their growth at 0 ^{o}C for 23 days. They reported that immediately after treatment colonisation per gram of skin was generally reduced by 1 log and the pH by 3.2 to 4 units. Treatment with 2% lactic acid suppressed post-decontamination growth of microorganisms more effectively than 1%. The treatment was most effective when applied shortly before chilling.

Bautista et al., (1995) studied the effect of lactic acid, chlorine (50 ppm) and trisodiumphosphate (TSP) sprays under various pressures, on turkey carcasses. They observed that 1.25% and 4.25% lactic acid caused a 2.4 and 4.4 $\log_{10}$ reduction in APC respectively. The acid spray had even greater impact on the reduction of coliforms and also reduced *Salmonella* contamination.

Hwang and Beuchat (1995) inoculated chicken skin samples with a mixture of *Salmonella, L. monocytogenes, C. jejuni* and *Staph. aureus* and then washed the samples for 30 min, with water or solutions of 0.3 to 0.5 lactic acid / 0.05% sodium benzoate. They observed a decrease in pathogens on the skin treated with antimicrobials, with *C. jejuni* being more sensitive.

Decontamination of large animal carcasses

Many investigators have used organic acids to decontaminate large animal carcasses (Dickson and Anderson 1992). Ockerman et al. (1974) used sprays of lactic and acetic acid in concentrations from 6% up to 24% on lamb carcasses and concluded that 18% acetic acid was the most effective but concentrations of acid in excess of 12% produced bleaching of the carcasses. Reynolds and Carpenter (1974) sprayed pork carcasses with a 60:40 mixture of acetic and propionic acids and reported a 2 log reduction in total microbial population, while only 1 out of 16 *Salmonella* positive carcasses remained positive after treatment.

Osthold et al. (1984) showed that a mixture of acids (acetic, lactic, citric and ascorbic) had a selective inhibitory effect on Enterobacteriaceae.

Woolthuis and Smulders (1985) sprayed calf carcasses with lactic acid solutions containing 0.75% to 2.5% of the acid and found that 1.25% acid resulted in a substantial reduction in total aerobic counts with minimal carcass discoloration. This concentration reduced total aerobic plate count by 1 log, with similar reductions in Enterobacteriaceae.

Snijders et al. (1985) concluded that the use of lactic acid sprays as a terminal process in carcass processing could provide significant microbiological advantages, while Smulders et al. (1986) reviewing the literature on the use of lactic acid in sanitising meat, recommend that public health authorities allow the use of lactic acid as a decontaminating agent.

Bell et al. (1986) demonstrated the effectiveness of 1.2% acetic acid as well as a mixture of 0.6% acetic and 0.046% formic acid, on a mixture of pathogenic bacteria pre-inoculated onto beef meat cubes and observed a 65% reduction in *S. typhimurium, Y. enterocolitica, Shigella sonnei* and *Ps. aeruginosa* and a 46% reduction in *E. coli*.

The interaction between the type of acid and treatment temperature has been studied on lamb carcasses and beef tissue (Anderson and Marshall 1989, 1990). These researchers evaluated a mixture of 2% acetic, 1% lactic, 0.25% citric and 0.1% ascorbic acids and found that acetic and lactic acids alone were as effective as the mixture, reducing *S. typhimurium* and APC by one $\log_{10}$ and *E. coli* by 0.5 log. Typically the bactericidal effect increased with increasing acid concentration and temperature of the acid solution.

Beef and pork carcasses were sprayed at various stages of processing with hot (55 ^{o}C) dilute (1%) lactic acid and a reduction of about one log was observed in APC (Prasai et al. 1991, 1992). The same authors also tested for *Listeria* before and after treatment. From 3 carcasses that were positive for *Listeria* before treatment, one remained positive after treatment.

Simulated water spray chilling included acetic acid (0.5, 1.0, and 2.0%) were tested for effectiveness in reducing *S. typhimurium, L. monocytogenes* and *E. coli* 0157:H7 on beef fat and lean tissue (Dickson 1991). The author observed a reduction of up to 3 log for all three pathogens on fat tissue and a smaller reduction on lean tissue which was significant when compared to controls.

Siragusa and Dickson (1992) observed an increase in the activity of organic acids against *L. monocytogenes* on beef tissue when the acids were immobilised in calcium alginate gel.

Cutter and Siragusa (1994) used a pilot-scale carcass washer to test the efficacy of organic acids against *E. coli* 0_{157}:H$_7$ and *Ps. fluorecens*, attached to beef carcass tissue. Tissues where sprayed either with water or 1, 3 or 5% lactic, acetic or citric acids at 24 ^oC. A reduction was observed of 1 to 2 log cfu/cm^2 but pathogens were not eliminated. However it was concluded that organic acid spraying of carcasses may be beneficial as part of an overall HACCP approach.

Dickson and Anderson (1992) in a review of washing and sanitising systems for microbial decontamination of carcasses concluded that a decontamination step, in the form of washing and sanitising during the slaughter process, can improve the microbial safety and shelf life of the meat and should be considered an integral part of the production process.

The decontaminating effect of lactic, acetic and fumaric acids, either alone or in a mixture was tested by Podolak et al. (1996) using lean beef tissue at 55 ^oC, against *L. monocytogenes* and *E. coli* 0_{157}:H$_7$. It was found that fumaric acid at a concentration of 1% was the most effective in reducing the populations of pathogens tested by 1 to 1.3 log units when compared with acetic or lactic acids.

5.4 CHLORINE

5.4.1 Mode of action and range of antimicrobial activity

Super-chlorination of water used in poultry processing usually involves the addition of either chlorine gas or a solution of sodium hypochlorite. In both cases, the chlorine is most active in the form of undissociated hypochlorous acid (HOCl), but, in the presence of organic matter, its antimicrobial activity is either reduced through the formation of chloramines or eliminated.

Other factors that influence the effectiveness of chlorination are the concentration of residual chlorine, temperature and pH of the solution and the contact time.

Chlorine is active against a wide range of microorganisms and is also sporicidal, but susceptibility varies. For example, at pH 6 or 8 and 4 or 25°C, *C. jejuni* was more susceptible than *E. coli* to 0.1 mg/l of free available chlorine and 1.0 mg/l of monochloramine (Blaser *et al.*, 1986). For a 99% kill, the necessary contact time varied between 5 and 15 min.

5.4.2 Use in poultry processing

Addition of chlorine to water used in processing eliminates any spoilage bacteria in the water supply. It also helps to control the spread of bacterial pathogens and the build-up of microorganisms on working surfaces and equipment and in chiller water. For example, in some countries, chlorine is used in strategically placed water sprays to minimise cross-contamination from equipment to carcasses. Contamination of processing equipment is progressively reduced by increasing the chlorine concentration to 70 mg/l (Bailey et al., 1986).

In the U.S.A., chlorine has been used in poultry processing for more than 40 years and was originally introduced to reduce microbial contamination of carcasses, extend shelf-life and shorten plant clean-up time.

These objectives were supported by several studies, generally involving in-plant chlorination at up to 20 mg/l, with higher levels in the water chilling system to compensate for the organic loading of the chill water (Goresline et al., 1951; Drewniak et al.; 1954; Gunderson et al.; 1954, Ziegler & Stadelman, 1955; Dawson et al., 1956; McVicker et al., 1958; Mallman et al. 1959; Ranken et al., 1965). It is now recognised, however, that chlorine is principally an aid to hygienic processing rather than a true decontamination treatment.

This is because, under conditions of commercial processing, not all studies have shown a reduction in carcass contamination. In one case, neither levels of contamination nor the occurrence of cross-contamination were reduced by spray-washing in chlorinated water after evisceration (Mead et al., 1975). Another study showed little effect of chlorine in the final wash unless at least 40 mg/l were used (Sanders & Blackshear, 1971). Washing carcasses post-chill with water containing up to 50 mg/l of chlorine did not reduce the proportion of salmonella-positive samples (Kotula et al., 1967). These studies serve to emphasise the importance of an adequate contact time, which may not be achieved while carcasses remain on a rapidly moving processing line. Larger reductions in carcass contamination have been observed in some laboratory trials; for example, elimination of artificially added salmonellas was achieved by holding carcasses in 200 mg/l chlorine for 10 min. (Dixon & Pooley, 1961). Salmonellas were virtually eliminated when carcasses were held for one hour in water containing 20 mg/l of chlorine at 6°C (Nilsson & Regner, 1963). However, the conditions of these experiments were not comparable with those of commercial processing.

Results have also been variable in relation to chlorination of chill water, with some studies showing no effect on either carcass counts or the prevalence of salmonella-positive carcasses (Mead & Thomas, 1973b; James et al., 1992), while others have found that addition of chlorine reduced carcass contamination by 0.5 - 1.0 log unit (Patterson, 1968a; May, 1974). In this situation, chlorine is more effective in controlling microbial levels in the chill water.

 Maintaining a total chlorine residual of 45 - 50 mg/l can keep the water essentially sterile (Mead & Thomas, 1973a), while the use of 200 mg/l in static slush-ice tanks was shown to minimise both cross-contamination and the growth of psychotrophic spoilage bacteria (Barnes, 1965). The chlorine had no effect on the numbers or types of bacteria on the carcasses and, in another study (Patterson, 1968b), had no influence on the spoilage flora of chicken carcasses held at 1°C.

5.4.3   Effects on organoleptic properties and composition of poultry meat

In most studies where effects of chlorine on sensory properties of the meat have been considered, concentrations up to 200 mg/l have had no adverse effect on the appearance, taste or odour of the meat. Although chlorine enhances water uptake during processing, there is evidence (Mast *et al.*, 1977) that chlorine reaction products are mainly confined to the skin, unless contact is prolonged, and most of the chlorine is likely to be present in the form of an inorganic salt.

5.4.4   Safety aspects

Possible human health hazards associated with the use of chlorine have been recognised (Dickson and Andersen, 1992).

These include the formation of trihalomethanes, notably chloroform (Robinson *et al.*, 1981; Schade *et al.*, 1990), chloramines and cyclic imides, which were shown to be mutagens in the Ames test (Haddon *et al.*, 1996). Some countries consider that the risk to human health is low (e.g. Anon, 1995), while others do not allow any use of chlorine in processing. In N. America, there is a restriction on the chlorine concentration that can be used. Chlorine itself, whether in gaseous form or as hypochlorite, is corrosive and a powerful irritant, but at working concentrations normally used (20 or 50 ppm) does not affect those that come into contact with it, provided that plant ventilation is adequate.

5.4.5   Chlorine dioxide

An alternative to chlorine, which is sometimes used in poultry processing, is chlorine dioxide. This acts as a gas in solution and does not form hypochlorous acid or react with ammonia. It has a wide spectrum of antimicrobial activity, but much less potential than chlorine for trihalomethane formation and no mutagenic activity when used to treat poultry chiller water (Anon, 1995b). It reduces microbial contamination of carcasses to much the same extent as chlorine, but is up to seven times more active (Lillard, 1979) and therefore can be used at lower concentrations, e.g. 3 - 5 mg/l in chiller water, which are less corrosive.   Working concentrations have no effect on meat flavour, but tend to result in a slightly lighter skin colour (Thiessen *et al.*, 1984). High levels of chlorine dioxide in the processing plant atmosphere can have adverse effects on workers , and are to be avoided.

5.5   TRISODIUM MONOPHOSPHATE (TSP)

Trisodium monophosphate (TSP) is a trisodium orthophosphate (formula $Na_3 PO_4$), food grade (E 339 iii), currently used for several food applications such as emulsifier for cream cheeses. TSP has been proposed as a means of reducing microbial surface contamination of poultry carcasses (as well as for beef or lamb carcasses and vegetables such as tomatoes). The process involves dipping or spraying the carcass with a solution of food grade TSP. Solutions of 8 to 12 % are used. The contact time is usually 15 seconds and the temperature of application 20 to 30°C.

5.5.1 Overall efficacy on pathogens

The bactericidal effect of TSP is well documented in the scientific literature and confirmed in several industrial trials. TSP is more active on Gram negative pathogens

(e.g. *Salmonella, Campylobacter, E. coli*) than against Gram positive ones (e.g. *L.. monocytogenes*).

Though the mechanism of action is still uncertain, it seems to result from a combination of several factors:

- a destructive effect of the high pH (pH 12)
- removal of bacteria that are not yet firmly adherent to the skin surface
- removal of some of the surface fat ("detergent effect") which facilitates the removal of bacteria by the washing process
- activity on bacterial walls which may account for the greater sensitivity of Gram negative bacteria due to the structure of the cell membrane. This effect has been demonstrated by electronic microscopy.

The efficacy of the TSP treatment depends on several factors including the following:

The nature of the pathogen(s)

The variable sensitivity of different groups of pathogens has been demonstrated in several experiments.

- The overall activity of TSP against **Salmonella** has been well documented (Bender, 1992; Gudmunsdottir *et al*. 1993; Li *et al*. 1994; Lillard, 1995; Hwang and Beuchat, 1995). When using commercial concentrations (12-15%) of TSP on beef tissues, Dickson *et al* (1994) found a reduction rate of 1 to 1.5 log on lean tissues and a maximum reduction rate of 2 to 2.5 log on adipose tissue. Salvat *et al.* (1997) and Coppen *et al.* (1998) investigated TSP treatment of poultry carcasses and found 2 log reduction of Salmonella. Pre-chill spraying of chicken with a TSP solution (10%) resulted in a 2.1 to 2.2 log reduction of *S. typhimurium* (Xiong et al, 1998).

- In France, Coppen et al. (1998) found that in industrial trials involving an air-chill process, TSP reduced the prevalence of *Salmonella* positive birds from 57.5 to 0.5 %.

- The effect of TSP on **C. jejuni/coli** has been demonstrated, although marked differences in effect have been observed *in-vitro* and in industrial tests. Using artificial biofilms or suspension of cells, Somers *et al* (1994) reported a reduction rate of 5 log. However, in industrial trials, the reduction observed was 1.2 to 1.5 log (Slavik *et al*, 1994; Federighi *et al*, 1995).

- Salvat *et al* (1997) found that the proportion of positive carcasses was reduced from 100% to 0%. Slavik *et al* (1994) found that *Campylobacter* was present on 96 and 100% of control carcasses and only on 24 and 28% of treated carcasses respectively.

- TSP has also shown to be effective against **E. coli**. On broiler carcasses, Salvat et al (1994) found a 2 log reduction rate on thermotolerant coliforms. Dickson *et al* (1994) showed that on beef tissues population reduction was comparable for *E.*

coli ATCC 25922 and E. coli O157:H7. Reductions of 1 to 1.5 log were obtained on lean tissues and reduction of 2 to 2.5 log on adipose tissue.

Kim and Slavik (1994) reported that on beef surfaces, compared with controls, the level of *E.coli* O157:H7 was 1.35 and 0.92 logs lower on TSP treated fat and fascia surfaces respectively and that, in that experiment, TSP was more effective in removing *E. coli* O157:H7 than *S. typhimurium*.

- A maximum reduction of 1 log has been reported for **L. monocytogenes** (Somers et al, 1994; Dickson et al 1994.; Hwang and Beuchat, 1995). Salvat et al (1995) found 3 positive samples out of 15 on controls and 2/15 on treated samples (statistically not significant). However, after 7 days at 2°C only 7 out of 20 samples were found to be Listeria positive for treated products against 14 out of 20 for controls (P<0.03).

Other factors

- The above mentioned results were obtained using commercial application (10%ww+/-2% TSP; 11°C+/-3°C; 15 seconds). However, the **temperature** of application, the **concentration** of the solution and the **duration** of application may influence the efficacy of the TSP treatment. The higher the temperature, the greater the activity (Dickson *et al*, 1994; Corry and Mead, 1996). Varying the concentration of TSP in the range of 8 to 12% was not a significant factor in reducing the population of bacteria on carcasses under commercial conditions (Dickson *et al*, 1994). Gudmunsdottir *et al* (1993) investigated the effect of TSP on 4 serotypes of *Salmonella* at different concentrations (10, 20, 40, 60, 80 g/l) and for different duration of application (5, 15, 30 and 60 sec.). All samples containing 60 and 80 g/l TSP were negative after 5 sec. exposure or more. At lower concentrations 5 to 15 sec. exposure was not efficient to kill all *Salmonella.* Following exposure at 30 to 60 sec. no *Salmonella* were recovered from 63 of 64 solutions containing 20g/l TSP.

- The **stage of application** may influence the effectiveness of the process in two ways. As one of the mechanisms of action of TSP is a removal of bacteria that are not yet firmly attached to carcass surfaces, the process has been shown to be more effective when applied immediately after evisceration than when applied later in the slaughtering process. It has been demonstrated (Salvat *et al*, 1995) that when treated post evisceration and pre air chill, opportunities for recontamination after treatment may exist (air-borne contamination, contamination by equipment) that should be appropriately overcome.

5.5.2   Microflora changes and implications

Action on spoilage microorganisms - conditions of development of the new spoilage microflora - impact on shelf life

The composition and evolution of the spoilage microflora of broiler carcasses is widely documented (ICMSF, 1998).

Initially Enterobacteriaceae form the dominant microflora (ca 70%) and *Pseudomonas spp* represent ca 20 %. At 2°C, psychrotrophic bacteria develop and *Pseudomonas spp* become progressively dominant (up to 93 - 96%) and constitute the main cause of spoilage on chicken carcasses. The TSP treatment results in a complete change of the dominant microflora. The activity of TSP on Enterobacteriaceae and *Pseudomonas spp* is well substantiated (Gudmunsdottir, 1993; Dickson *et al*, 1994; Somers *et al*, 1994; Gorman *et al*, 1995). Coppen *et al* (1998) found an average of 2.5 log reduction in Enterobacteriaceae and 1.5 log reduction in *Pseudomonas spp*.

The destruction, or sub-lethal injury in *Pseudomonas spp.* induced by TSP encourages the development of *Brochothrix thermosphacta* which is not markedly affected by the treatment and becomes the new cause of spoilage on treated carcasses. Coppen *et al* (1998) observed that after 10 days of cold storage, *Br. thermosphacta* represented ca 96% of the spoilage microflora. After 21 days, the small proportion of *Pseudomonas* that has not been eliminated by the treatment develop again and constitute ca 16% of the total psychrotrophic microflora at this stage.

These changes account for a longer shelf life. Salvat *et al* (1995) mention that the development of the spoilage psychrotrophic microflora is noticeable after 10 days only on treated samples. In their experiment control samples presented high numbers of psychrotrophic microorganisms (6.81 log cfu/ml) after 10 days of cold storage and were in advanced spoilage (8.61 log cfu/ml) after 15 days. Treated samples were of good microbiological quality after 10 days (3.96 log cfu/ml) and presented 6.0 log cfu/ml after 15 days and 9.09 log cfu/ml after 21 days.

Implication of changes on the potential development of pathogens

Concerns have been raised that the severe reduction (or increase of the lag phase) of Gram negative competing spoilage microflora may favour the emergence of Gram positive pathogens and *L. monocytogenes* in particular. Such a relative development has been poorly investigated to date. Salvat et al (1995) showed that, at day 1 no significant difference was observed in the number of samples tested positive for *L. monocytogenes* before and after a TSP treatment. However, at day 7 a statistically significant reduction of *L. monocytogenes* positive carcasses was observed (7/20 treated vs 14/20 controls).

5.5.3   Potential for introducing other food safety hazards

The question of whether the treatment leaves residues (Phosphorus, expressed as P_2O_5) on/in the skin/meat should be assessed.

- TSP is a food phosphate naturally present in foods.
  In Europe, it is an authorised food additive (E 339iii) which can be used alone or in combination with other authorised orthophosphates. Authorised uses include sterilised and UHT milks (1 g/l P_2O_5 w/w); partly dehydrated milk with less than 28% solids (1 g/Kg); partly dehydrated milk with more than 28% solids (1,5 g/Kg); dried milk and dried skimmed milk (2.5 g/Kg); sterilised and UHT creams (5 g/Kg); whipped cream and vegetable fat analogues (5 g/Kg); unripened cheeses (2 g/Kg); processed cheeses (20 g/Kg); meat products (5 g/kg); sport drinks and prepared table waters (0.5 g/Kg); cereals (1 g/Kg). In the US, TSP is a GRAS

substance (21 CFR 182.1778) and no limitation of use is imposed when TSP is utilised according to good manufacturing practice.

5.5.4 Impact on the environment

The use of TSP result in release of orthophosphate (PO_4^{---}). It can be found at high concentrations in the dipping / spraying solution and at low concentrations in the drainage from the carcasses after treatment.

The impact on the environment is highly dependent on the level of compound release and the treatment of these effluents at the slaughterhouse.

The impact on the environment of the use of TSP treatment in slaughtering premises has to be assessed in relation with the conditions of use and of effluent treatment.

5.5.5 Effects on sensory properties and quality of the product

- As an humectant effect is recognised for some classes of phosphates (e.g. polyphosphates), concerns have been raised about water absorption and retention in TSP treated birds.
  In the US, experiments carried out by USDA-FSIS on water chilled carcasses showed that the water absorption and retention were below acceptable levels as in CFR 381.66 (FSIS Backgrounder, October 1992).

- Any decontamination treatment should not cause undesirable sensory change. Consumer evaluation of TSP treated chicken has been investigated by Hollender et al (1993). This was conducted to measure the effect of the treatment on taste, texture and appearance of the meat. The flavour and texture hedonic scores for fried thigh and breast meat were the same for treated and control samples. The results were similar for baked thigh and breast samples. The visual appearance of fresh whole broiler carcasses was not significantly different for treated and control samples, either on day 1 or on day 8. However, whereas at day 1 there was not a significant purchase preference for either treated or control samples, on day 8 the purchase preference for treated samples was significant.

5.5.6 Other classes of phosphates

Several other classes of phosphates have been tested with regard to their antimicrobial activity on broiler carcasses. The effectiveness of tripotassium phosphate has been demonstrated, but is less than that of TSP (Gudmunsdottir *et al*, 1993). Long chain polyphosphates have been shown to be effective against *S. aureus* by Lee *et al* (1994) and against *L. monocytogenes* by Zaika and Kim (1993). However, the effect of polyphosphates on *L. monocytogenes*, *E. coli* and *S. typhimurium* has been refuted by Flores *et al* (1996). Pyrophosphates in chiller water are only mildly effective against *Enterobacteriaceae* on broiler carcasses with a reduction of more than 1 log (Rathberger and Waldroup, 1995). Sodium hypophosphite is effective against Enterobacteriaceae at concentrations of 1000-3000 µg/ml but ineffective against *C. jejuni* or *Ps. fluorescens* (Rhodehamel and Pierson, 1990). Therefore, when used alone, these other classes of phosphates do not represent an effective alternative for antimicrobial treatment of poultry carcasses.

5.6 ANTIBIOTICS AND BIOPRESERVATION

5.6.1 Antibiotics

Antibiotics are secondary metabolites produced by some microorganisms that inhibit or kill other microorganisms. At present, only one antibiotic, natamycin, is approved for use in food. In several countries, natamycin can be applied in the dairy industry for production of cheese. Other antibiotics such as tetracyclines, tylosin and subtilin have also been found effective for various food applications, but are for various reasons not used in foods today.

Generally, antibiotics should not be used as food additives or processing aids because of the risk of selecting strains that are resistant to agents that are used in human or veterinary medicine. If an antibiotic is considered for antimicrobial treatment of foods, the following should be addressed:

- Possible toxicological effects

- Biodegradability

- Possible medical applications (human and veterinary)

- Development of resistance

- Possible cross-resistance to clinically useful antibiotics

Natamycin (also known as pimarcin, tennectin and myprozin), which was accepted by JECFA in 1976, is only effective against yeasts and moulds. Thus, it is not considered an option for antimicrobial treatment of poultry carcasses.

5.6.2 Biopreservation

Biopreservation has been defined as the use of LAB (lactic acid bacteria), their metabolic products, or both to improve or assure the safety and quality of foods that are not generally considered fermented (Montville TJ, Winskowski K, 1997).

*Biopreservation by controlled **acidification***

LAB can produce lactic acid in situ and thereby inhibit microbial growth. Many factors determine the effectiveness of in situ acidification including the product's initial pH, its buffering capacity, the type and level of challenge organisms, the nature and concentration of the fermentable carbohydrate, ingredients that might influence the viability and growth rate of LAB, and the growth rates of the LAB and target pathogen at refrigerated and abused temperatures (Montville TJ, Winskowski K, 1997). It has been shown that lactic acid at 1 to 2% reduces Enterobacteriaceae and aerobic mesophilic microorganisms on beef, veal, pork and poultry, and delays growth of spoilage microflora during long-term storage of products. Sodium lactate (2.5 to 5.0%) inhibits *Cl. botulinum*, *Cl. sporogenes*, *L. monocytogenes*, and spoilage bacteria in various meat products (Montville TJ, Winskowski K, 1997).

There are no significant risks associated with the consumption of LAB per see. However, LAB that are to be used as starter cultures or as probiotics should not contain acquired genes conferring resistance to therapeutic antibiotics as such genes may promote an increase in antibiotic resistance among intestinal bacteria.

Because the major pathogens associated with fresh meat and poultry are Gram-negative bacteria, and LAB only have limited effect on such bacteria, the use of LAB does not seem to be a reliable method for decontamination of poultry carcasses.

Bacteriocins

Bacteriocins are secondary metabolites produced by microorganisms that inhibit or kill other microorganisms, but unlike antibiotics, they inhibit or kill only closely related species or different strains of the same species. Many, if not all, are peptides, and they are typically bactericidal in action. LAB bacteriocins act at the cell membrane and disrupt the integrity of the cytoplasmic membrane, thus increasing its permeability to small compounds. Ultimately, this leads to cell inhibition and possibly death. The LAB associated with culture antagonism include the lactococci, enterococci, enterococci, lactobacilli, carnobacteria and pediococci. A wide variety of foodborne pathogens are either inhibited or killed, and many spoilage organisms are affected in similar ways, especially Gram-negative psychrotrophs (Montville TJ, Winskowski K, 1997). Some bacteriocins, e.g. nisin, act against spores.

It is renewed interest in LAB bacteriocins as an alternative to the use of chemicals to control undesirable organisms in foods. The use of bacteriocins, the organism which produce them, or both is attractive because of the increasing consumer demand for natural products and increasing concern about foodborne disease.

Nisin is the first and best known and studied of the bacteriocins produced by LAB.

In the scientific community bacteriocins are generally considered safe for human consumption as bacteriocinogenic bacteria are isolated from a variety of foods all over the world. Nevertheless, the approval of a bacteriocin as a food additive should be based upon a toxicological assessment. In the US, nisin is the only bacteriocin that has GRAS affirmation, and its affirmation by the FDA was supported by toxicological data (Montville TJ, Winskowski K, 1997).

Nisin

Nisin (a polypeptide produced by some strains of *Lactococcus lactis)* is the only bacteriocin approved internationally for use in foods, with around 46 countries permitting its use in food to varying degrees (Delves-Broughton J, 1990). The Joint FAO/WHO Committee accepted nisin as a food additive in 1969 and set maximum intake levels as 33000 IU/kg body weight.

Nisin has a narrow spectrum of antimicrobial activity being effective against Gram-positive bacteria, primarily spore-formers.

Typical usable levels are in the range of about 2.5 to 100 ppm. By adding nisin, the heat process for low-acid canned foods can be reduced. In addition to its use in certain

canned foods, nisin is most often employed in dairy products, e.g., processed cheeses, condensed milk, and pasteurised milk, as well as mayonnaise and baby foods.

Because the major pathogens associated with fresh meat and poultry are Gram-negative bacteria and nisin only has effect on Gram-positive bacteria, nisin is not considered an option for antimicrobial treatment of poultry carcasses. However, in combination with other antimicrobial treatments, e.g., chelators such as EDTA or citric acid, lactic acid, or nitrite, nisin may inhibit growth or attachment of Gram-negative bacteria such as *Salmonella* spp. and *E. coli* (Bolder NM, 1997). More research is needed in this regard to evaluate the usefulness of such approaches and to perform risk assessments.

Pediocin

Pediocins inhibit *L. monocytogenes*, but are inactive against spores. Pediocin PA-1 is used to extend the shelf-life of salads and salad dressing and as an antilisterial agent in food such as cream, cottage cheese, meats and salads (Montville TJ, Winskowski K, 1997). Pediocins are more effective than nisin in meat and are even more effective in dairy products. Dipping meat in 5000 AU of crude pediocin PA-1/ml decreases the viability of attached *L. monocytogenes* 100- to 1000-fold. Pretreating meat with pediocin reduces subsequent *L. monocytogenes* attachment. Pediocin AcH at 1350 AU/ml reduces *Listeria* in ground beef, sausage and other products by between 1 and 7 log cycles. A recent study showed that applying pediocin onto food packaging film is an effective approach to reduce *L. monocytogenes* contamination in fresh and processed meats and poultry (Ming X et al., 1997)

Because the major pathogens associated with fresh meat and poultry are Gram-negative bacteria and pediocins **merely** have effect on Gram-positive bacteria, pediocins are not considered an alternative for antimicrobial treatment of poultry carcasses.

Addition of bacteriocin-producing bacteria to non-fermented foods

The addition of a bacteriocinogenic culture rather than the pure bacteriocin has in many instances proved effective as an antitilisterial method (Montville TJ, Winskowski K, 1997).

Bacteriocin resistance

While nisin-resistant starter cultures can be used to great advantage, the appearance of nisin-resistant bacteria can undermine the use of nisin as an inhibitor. The study of bacteriocin resistance is in its infancy.

5.7   SUMMARY OF PRINCIPAL FINDINGS

Decontamination should not be used as the primary pathogen reduction measure nor as a substitute for appropriate preventive measures at the production level or at the slaughterhouse. This is partly because most decontamination techniques have only a limited effect in reducing pathogen contamination, the result of which is directly related to the initial level of contamination. As an example a treatment resulting in a 3 log

reduction will reduce a population of 10^8 to 10^5, a population of 10^3 to 1 and will eradicate a population of 10^2. This means that the efficacy of a pathogen reduction treatment should generally be expressed as the fraction killed (per time unit), expressed either as % or as log units. Thus, if the initial pathogen load is high, decontamination may not reduce the proportion of positive carcasses. Ultimately, if the goal is to reduce consumer exposure to less than one contaminated chicken per year, the proportion of contaminated carcasses prior to decontamination must be below 5% and the efficiency of pathogen reduction no less than 99.9% (see Section 3).

Furthermore, in a final evaluation it is important to consider, that the use of decontamination techniques can compromise traditional monitoring of food quality and safety through changes in the frequency or level of indicator parameters, such as *E. coli* and total viable count.

Finally, the process of monitoring the pathogen situation, which can have implications in relation to other management options, can be significantly influenced by the use of antimicrobial treatments. Since it has been shown (see below) that these treatments will generally only reduce and not eliminate the relevant pathogens, the pathogen concentrations will in some cases only intermittently be below the detection limit. This will seriously hamper the potential to monitor the pathogen situation throughout the food production chain, and thereby some of the incentives to adhering to Good Hygienic Practices can be lost.'

In this report, some specific decontamination compounds/techniques have been considered . These are only to be considered as examples because not all of the elements listed in Section4 have been addressed:

    a) cold and hot water, steam, steam vacuum;
    b) ionising radiations;
    c) organic acids;
    d) chlorine and related compounds;
    e) trisodium monophosphate;
    f) antibiotics and biopreservation.

    a) <u>Cold and hot water, steam, steam vacuum</u>

Washing with potable water is safe but care should be taken to avoid excessive generation of aerosols. Treatment with hot water or steam must also be considered safe, provided that carcasses are cooled promptly after treatment.

Washing with water may result in an overall reduction of surface contamination by 90-99 %. This is not achieved by a single wash but through the combined effect of a number of washes.

The results of pathogen reduction studies using hot water or steam are quite variable, depending on the way in which the carcasses were contaminated. High temperature short time steam vacuum pasteurisation seems the most promising.

    b) <u>Ionising radiations</u>

If a pathogen reduction treatment has to be used, irradiation is clearly the most efficient technique at present. A treatment of 3 kGy will, in most cases, give more than a 3 log reduction in the important pathogens in poultry. Certain other bacteria and spore-forming organisms can have a significantly higher resistance. Questions of tailing in survivor curves and the importance of changes in microflora and regrowth have not been adequately elucidated. The technology needs special equipment, also for worker protection.

c) Organic acids

Short-chain organic acids (acetic, lactic, citric, propionic, fumaric, tartaric and succinic) exert antibacterial activity and some of them (sorbic, propionic) also posess anti-fungal activity. Traditionally, they have been used as food preservatives as well as flavouring agents. They are GRAS substances approved as food additives by E.C., FAO/WHO, and FDA.

The use of organic acids in poultry carcasses decontamination result in 90-99 % reduction of bacterial load and 30 to 90 % reduction in pathogens. Thus it can be considered a useful treatment for improving the microbial safety of meat but only in the frame of an overall HACCP approach.

d) Chlorine

Super-chlorination of process water has been advocated as an aid to hygienic processing to prevent a build-up of microorganisms on equipment, surfaces and chilled water during the working period. It has also been used as a means of limiting the spread of pathogens among the carcasses being processed.

Chlorine is rapidly inactivated in contact with carcasses and therefore is not consistently effective as a direct carcass decontamination. Moreover, its use is associated with the formation of small amounts of certain mutagens. The public health significance of these substances needs to be clarified. The use of chlorine as antimicrobial treatment for carcasses cannot be recommended. Chlorine dioxide is more active and can be used at lower concentrations.

e) Trisodium phosphate (TSP)

TSP has been shown to achieve 1 to 2 log reduction in Gram-negative pathogens (e.g. Salmonella, Campylobacter) in poultry carcasses. It is less effective against Gram-positive pathogens (e.g. L. monocytogenes).

The significance of changes in microflora and regrowth still need to be fully evaluated.

TSP is naturally present in tissues and bones and may be considered as harmless. However, appropriate conditions of application should be designed and controlled so as to minimise water intake and retention in carcasses and to avoid increasing the polluting effluents from poultry slaughterhouses.

f) Antibiotics and Biopreservation

Antibiotics:

In poultry processing, antibiotics should not be used as food additives or processing aids because of possible adverse effects on consumer health such as possible development of antimicrobial resistance.

At present, only one antibiotic, natamycin, is approved for use in food processing, as an antifungal agent in cheese production. (cf. 5.6.1)

Antibiotics are not an option for antimicrobial treatment of poultry carcasses.

Biopreservation:

Lactic acid bacteria (LAB) and bacteriocins can be used in the food industry for inhibition of certain microorganisms. Nisin is the only bacteriocin approved internationally for use in foods. It is effective against spore-formers and Listeria and is used in canned foods and dairy products.

Because the major pathogens associated with fresh meat and poultry are Gram-negative bacteria and LAB and bacteriocins only have limited effect on such bacteria, the use of biopreservation does not seem to be an option for decontamination of poultry carcasses.

SECTION 6: CONCLUSIONS AND RECOMMENDATIONS

CONCLUSIONS

- There is a need to reduce the burden of foodborne disease by reducing the prevalence of zoonotic pathogens in poultry.

- Successful control of foodborne human pathogens requires a "farm to fork" approach involving input from producers, processors, distributors, retailers, caterers and consumers. In the case of poultry meat production, the aim must be to establish a fully integrated control programme throughout the poultry supply chain.

- A logical strategy for pathogen reduction in poultry would be to ensure the lowest possible prevalence of pathogens in the production pyramid, starting from the top and moving downwards, i.e. from primary breeders throughout the chain to production flocks

- Another key element of such a strategy are hygienic measures which should be applied on the farm, during transport and in the processing plant.

- Decontamination should not be used as the primary pathogen reduction measure nor as a substitute for appropriate preventive measures at the production level or at the slaughterhouse. This is partly because most decontamination techniques have only a limited effect in reducing pathogen contamination, this effect depending directly on the initial level of contamination. .

- This means that the efficacy of a pathogen reduction treatment should generally be expressed as the fraction killed (per time unit), expressed either as % or as log units. Thus, if the initial pathogen load is high, decontamination may not reduce the proportion of positive carcasses.

- If decontamination treatments are used in the wrong way as a substitute for good hygienic practice, they may even be counterproductive to food safety.

- The use of antimicrobial treatment of poultry carcasses is part of a management option based on an assessment of available scientific data. The evaluation should be an interactive process between risk management and risk assessment. The use of such antimicrobial treatments should at some stage of the evaluation process be related to other alternatives such as the production of poultry without, or with a very low level of, specific pathogens. In general, the final selection of antimicrobial treatment as an option should be based on thorough analysis of risks, benefits and alternative options.

- When decontamination of poultry carcasses is being considered, it should be demonstrated that the decontamination procedures are effective and safe and that their use is controllable under practical conditions. In particular the occurrence of persistence levels of zoonotic pathogens, recontaminations, and residual levels of potentially hazardous compounds should be considered

- Consumer information and labelling requirements for the endproduct must be considered

RECOMMENDATIONS

The Committee recommends that:

- antimicrobial treatment should only be used as part of an overall strategy for pathogen control throughout the whole production chain;

- before any decontamination compound or decontamination technique is authorised for use it should be fully assessed;

- the person/company proposing such a decontamination compound or decontamination technique must demonstrate that all aspects considered in the 4th section of this report are covered;

- the person/organisation using a decontamination compound or decontamination technique must demonstrate that effective control of parameters critical for efficacy and safe use are in place and that good practice and appropriate HACCP plans are implemented;

- based on the conclusions of this report, a framework is established for the assessment of decontamination compounds or decontamination techniques proposed.

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