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Opinion of the Scientific Panel on food additives, flavourings, processing aids and materials in contact with food (AFC) on a request from the Commission related to

Treatment of poultry carcasses with chlorine dioxide, acidified sodium chlorite, trisodium phosphate and peroxyacids

Question Nº EFSA Q-2005-002

Adopted on 6 December 2005

SUMMARY

The Commission has asked EFSA to update the previous opinion expressed by the Scientific Committee on Veterinary Measures Relating to Public Health (SCVPH) on 14-15 April 2003 with regard to the toxicological risks to public health from possible reaction products (e.g. semicarbazide) of chlorine dioxide, acidified sodium chlorite, trisodium phosphate and peroxyacids when applied on poultry carcasses.

When examining the possibility for reaction products, no halomethanes have been reported to be formed in treatments with chlorine dioxide in water. No chlorinated organics have been found after treatments of poultry carcasses with acidified sodium chlorite. No detectable effects on the oxidation status of fatty acids in poultry carcasses were reported following treatment with peroxyacids. Furthermore, semicarbazide was not detected (limit of detection of 1 microgram/kg) in laboratory tests on poultry carcasses after treatment by immersion with acidified sodium chlorite. The Panel notes that the initial health concerns about semicarbazide are no longer relevant. As set out in previous EFSA opinion, new data showed that semicarbazide is not genotoxic *in vivo*.
Based on conservative estimates of poultry consumption in European adults, the Panel estimated potential exposure to residues arising from these treatments.
On the basis of available data and taking into account that processing of poultry carcasses (washing, cooking) would take place before consumption, the Panel considers that treatment with trisodium phosphate, acidified sodium chlorite, chlorine dioxide, or peroxyacid solutions, under the described conditions of use, would be of no safety concern.
The Panel notes that spraying of poultry carcasses with antimicrobials, by comparison to dipping and immersion treatments, will reduce the exposure to residues and by-products that might arise.
The Panel stresses that the use of antimicrobial solutions does not replace the need for good hygienic practices during processing of poultry carcasses, particularly during handling, and also stresses the need to replace regularly the water of chiller baths.

KEY WORDS
Antimicrobials, poultry carcasses decontamination, trisodium phosphate, E 339iii, CAS No. 7601-54-9, "acidified sodium chlorite", sodium chlorite, CAS No. 7758-19-2, chlorine dioxide, CAS No. 10049-04-4, peroxyacetic acid, CAS No. 79-21-0, peroxyoctanoic acid, CAS No 33734-57-5, hydrogen peroxide, CAS No. 7722-84-1, "peroxyacids".

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BACKGROUND

Article 3(2) of Regulation (EC) No 853/2004 of the European Parliament and of the Council laying down specific hygiene rules for food of animal origin, provides a legal basis to permit the use of a substance other than potable water to remove surface contamination from products of animal origin. Such a legal basis does not exist in the current legislation for red meat (Directive 64/433/EEC) and for poultry meat (Directive 71/18/EEC), but will be available once Regulation (EC) No 853/2004 is applicable with effect from 1 January 2006.

For many decades the use of substances other than potable water, i.e. antimicrobial substances, has been resisted, because they would mask unhygienic slaughter or processing practices and would certainly not be an incentive for businesses to implement hygienic practices. If permitted for use, it was also feared that their widespread use coupled with high bacterial counts due to unhygienic practices, would induce resistance of the micro flora present on the surface of the treated products.

In an opinion prepared by the Scientific Committee on Veterinary Measures relating to Public Health (SCVPH) issued on 30 October 1998, it was stated that antimicrobial substances should only be permitted for use if a fully integrated control programme is applied throughout the entire food chain. As a first step to the authorisation of antimicrobial substances in the EU and in the framework of the veterinary Agreement between the EU and the USA, four technical dossiers were submitted by the United States of America on the use of four antimicrobial substances (chlorine dioxide, acidified sodium chlorite, tri-sodium phosphate and peroxyacids) on poultry carcasses for evaluation. The SCVPH opinion issued on 14-15 April 2003 on the evaluation of antimicrobial treatments for poultry carcasses concluded that decontamination can constitute a useful element in further reducing the number of pathogens. Both opinions stressed that antimicrobial substances shall be assessed thoroughly before their use is authorised.

With the adoption of the hygiene package and the introduction of the hazard analysis and critical control points (HACCP) principles in the entire food chain, establishments are obliged to improve their hygiene and processing procedures. Under such circumstances the use of antimicrobial substances on food of animal origin can be reconsidered. The Commission envisages the approval of certain antimicrobial substances as part of an implementing measure of the Hygiene Regulations, which will become applicable with effect from 1 January 2006.

However, approval of the antimicrobial substances will depend on a thorough evaluation of all risks to public health involved in their use. Recent research suggests the formation of reaction products (in particular semicarbazide) due to the use of active chlorine substances in food, especially on food with high protein content, such as food of animal origin (Hoenicke *et al.*, 2004). The SCVPH opinion of 2003 stated that "reactive agents like chlorine dioxide, acidified sodium chlorite and peroxyacids may induce chemical changes in poultry carcasses. However, reaction products have not been identified and consequently a toxicological evaluation is not possible". In the light of the new information on semicarbazide formation, it is necessary to complete the previous risk assessment with regard to possible reaction products of the four substances on poultry meats after treatment.

TERMS OF REFERENCE

The Commission asks EFSA to update the previous opinion expressed by the Scientific Committee on Veterinary Measures relating to Public Health on 14-15 April 2003 with regard to the toxicological risks to public health from possible reaction products (e.g. semicarbazide) of chlorine dioxide, acidified sodium chlorite, trisodium phosphate and peroxyacids when applied on poultry carcasses.

In this context EFSA is also requested to evaluate whether different ways of use of these antimicrobial substances would result in avoiding a health risk with regard to possible reaction products.

ASSESSMENT

CHEMISTRY AND COMPOSITION OF THE ANTIMICROBIAL AGENT

Trisodium phosphate

Synonym: Trisodium monophosphate
Chemical name: Trisodium orthophosphate
CAS Registry Number: 7601-54-9
Chemical formula: Na_3PO_4
Description: Colourless or white crystals

Trisodium phosphate is typically used in aqueous solutions containing 8 to 12% with a high pH value (pH 12). The solution is kept at a temperature between 7 and 13ºC and applied by dipping or spraying the carcasses for up to 15 seconds. Carcass exposure time is controlled by line speed and length of the application cabinet (USDA, 2002c). Trisodium phosphate exerts a destructive effect on pathogens and a "detergent effect" that allows the removal of bacteria by the washing process (SCVPH, 1998). The lowest effective concentration for microbial control is 8%. Trisodium phosphate is ionised in water generating Na^+ and PO_4^{3-} ions.

Acidified sodium chlorite

Definition: Acidified sodium chlorite is a combination of sodium chlorite and any acid generally approved in food
Synonym: Acidified chlorite
Chemical name: Sodium chlorite (Chlorous acid, sodium salt)
CAS Registry Number: 7758-19-2
Chemical formula: $NaClO_2$
Description: Clear, colourless, liquid

Sodium chlorite, at a concentration of 500-1200 mg/L, is activated with any acid approved for use in foods at levels sufficient to provide solutions with pH values in the range 2.3-2.9 for either a 15 second spraying or 5-8 second dipping. In the case of immersion in chilling water, the concentration is up to 150 mg/L at pH between 2.8 and 3.2. The mean residence time of poultry carcasses in the chiller is typically an hour but can be as long as 3 hours (USDA, 2002b).

The main active ingredient of acidified sodium chlorite (ACS) solution is chlorous acid which is a very strong oxidizing agent, stronger than either chlorine dioxide or chlorine. The level of chlorous acid depends on the pH of the solution. So, 31% is formed at pH 2.3, near 10% at pH 2.9 and only 6% at pH 3.2. The potential formation of chlorine dioxide is limited, not exceeding 1-3 mg/L (International registration Dossier, 2003).

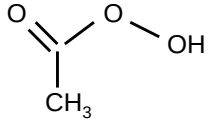
Chlorine dioxide

Synonym: Chloroperoxyl, Chlorine (IV) oxide
Chemical name: Chlorine peroxide
CAS Registry Number: 10049-04-4
Chemical formula: ClO_2
Description: Greenish yellow to orange gas with a pungent odour

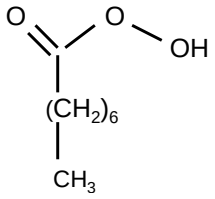
Chlorine dioxide is an oxidizing agent with a low redox potential. For use as an antimicrobial agent it is added to water in a concentration up to 50 mg/L in order to maintain a residual concentration of 2.5 mg/L (USDA, 2002a). The antimicrobial efficacy of chlorine dioxide is not affected by pH. It can be used both in on-line reprocessing (sprays or washes) or in chiller baths to limit the potential for microbial cross-contamination (SCVPH, 2003).
Chlorine dioxide is very reactive and is rapidly transformed to chlorite and chlorate ions in a ratio of 7:3. Thus, the concentrations of chlorite and chlorate would be 33 and 14 mg/L, respectively. Only 2.5 mg/L (about 5% of the initial content) remains as chlorine dioxide.

Peroxyacetic and peroxyoctanoic acids

Definition: Formulation of peroxyacetic acid (<15%), peroxyoctanoic acid (<2%) and Hydrogen Peroxide <10%)
Synonym: Peroxyacids, acetyl peroxide, acetyl hydroperoxide
Chemical name: Ethaneperoxoic acid, octaneperoxoic acid and hydrogen dioxide
CAS Registry Number: 79-21-0, 33734-57-5 and 7722-84-1, respectively
Chemical formula: $C_2H_4O_3$, $C_8H_{16}O_3$ and H_2O_2, respectively
Description: Clear, colourless, liquid



Peroxyacetic acid



Peroxyoctanoic acid

1-Hydroxyethylidene-1,1-diphosphonic acid (HEDP) is usually added to the solution as stabiliser (at <1%) because of its metal chelating activity. Acetic and octanoic acids are also present in the peroxyacids solution. Acetic acid acts as an acidifier and octanoic acid as a surfactant. Thus, the peroxyacid solution is a mixture of peroxyacetic acid, peroxyoctanoic acid, acetic acid, octanoic acid, hydrogen peroxide, and HEDP.

The solution is used at a maximum concentration of total peroxyacid, expressed as peroxyacetic acid, of 220 mg per L, a maximum concentration of hydrogen peroxide of 110 mg per L, and a maximum concentration of HEDP of 13 mg per L (USDA, 2002d). This solution may be used both in on-line reprocessing (15 second sprays or washes) or up to 60 minute immersion in chiller baths to limit the potential for microbial cross-contamination. A combined amount of peroxyacids, expressed as peroxyacetic acid, is usually given due to the difficulties in the analytical differentiation between peroxyacetic and peroxyoctanoic acids. The formula for the calculation of the concentration of the peroxyacid mixture is given in the appendix.

MECHANISMS OF ACTION OF THE ANTIMICROBIAL AGENTS

Mechanisms of action of the antimicrobial agents were recently reported by the Scientific Committee on Veterinary Measures relating to Public Health (SCVPH, 2003). Zoonotic pathogens most typically found in poultry and responsible for food borne disease are *Salmonella* spp and *Campylobacter* spp. The mechanisms of carcass contamination and distribution over a poultry carcass are rather specific. First, there is retention of bacteria in a liquid film on the skin and afterwards, bacteria are more closely associated with the skin, even untrapped in inaccessible sites. Spray rinsing at several points along the processing line is an effective means of minimising contamination but is not so effective especially in exposed areas of connective tissue that are more heavily contaminated (SCVPH, 2003). It must be emphasised that, in general, decontamination treatments are able to reduce the contamination level but do not completely eliminate pathogens. Their effectiveness depends on the initial microbial load and treatment conditions. Regarding treatment conditions, there are many factors affecting the efficacy of these antimicrobials including concentration of the substance, time of exposure, temperature, pH and hardness of water, strength of bacterial adhesion to the carcasses, biofilm formation and the presence of fat or organic material in water. The antimicrobial resistance is highly enhanced when bacteria are attached to a surface (up to 150 times) (Lechevalier *et al.*, 1988a) or forming part of a biofilm (up to 3000 times) (Lechevalier *et al.*, 1988b).

Poultry carcasses require to be cooled within defined limits before shipping. The cooling is generally accomplished by immersing the carcasses in cold water in long flow-through tanks called chillers. During immersion chilled carcasses absorb water that can represent up to 6-8 % increase in weight depending upon the size of the carcass (Schade *et al.* 1990). Since water is not regularly renewed for economic reasons, treatment with antimicrobial agents is aimed to control microbial proliferation in these chillers baths but certain by-products could be formed and therefore water treatment deserves consideration.

The proposed treatments of poultry carcasses with trisodium phosphate, acidified sodium chlorite, chlorine dioxide, and peroxyacetic and peroxyoctanoic acids have been tested for the inactivation of bacterial, viral and protozoan pathogens found on poultry and in poultry processing plants. The application in the United States can be either as spray or washes for on-line reprocessing or added to chiller baths to limit the potential for cross-contamination (USDA 2002a, b, c, d). The mechanisms of action for each specific antimicrobial agent are as follows:

Trisodium phosphate

The mechanism of action is based on its high alkalinity in solution (pH 12.1) that can disrupt cell membranes and remove fat films causing the cell to leak intracellular fluid. It can also act as a surfactant contributing to elimination of bacteria not yet strongly adhered to the surface of poultry skin (USDA, 2002c, Capita *et al.*, 2002).

Acidified sodium chlorite

Sodium chlorite is activated with acid at levels sufficient to reach pH values in the range 2.3-2.9. Its antimicrobial action is derived from chlorous acid that is determined by the pH of the solution (USDA, 2002b). Chlorous acid also oxidises cellular constituents. It also disrupts protein synthesis.

Chlorine dioxide

Its main action consists in the oxidation of cellular constituents. Chlorine dioxide has a direct action on cell membranes, either altering (at high concentrations) or disrupting their permeability (at low concentrations) (USDA, 2002a) and then penetrating into the cell and disrupting the protein synthesis. At a pH of 8.5, chlorine dioxide was reported as 20 times more effective than chlorine at killing *E. coli* (Benarde *et al.*, 1965).

Peroxyacetic and peroxyoctanoic acids

Peroxyacids consist of a mixture of peroxyacetic acid, octanoic acid, acetic acid, peroxyoctanoic acid, hydrogen peroxide, and HEDP (1-hydroxy-1,1-diphosphonic acid). Microorganisms are killed by oxidation of the outer cellular membrane (USDA, 2002d). A secondary mechanism could be the acidification of the carcass surface (SCVPH, 2003).

FORMATION OF DISINFECTION BY-PRODUCTS AND FURTHER REACTION PRODUCTS

Trisodium phosphate

On dissolution in water, the ionisation products of trisodium phosphate are Na^+ and PO_4^{3-}. These ions can be absorbed into the carcass but no further reactions are likely. The poultry carcass can be affected when exposed to the high alkalinity of the solutions. However, the possible consequences of this is not part of this evaluation. For instance, the action of endogenous poultry muscle enzymes or the water retention capacity could be altered during the post-treatment period of time. However, a study on broiler products reported no detectable effects of treatment on taste, texture or appearance (Hollender *et al.*, 1993). There would be no possibility of the formation of semicarbazide after treatment with trisodium phosphate.

Acidified sodium chlorite

The use of acidified sodium chlorite generates chlorous acid as well as other species like chlorite, chlorate and chlorine dioxide. The proportion depends on the pH of the mixture. The extent of formation of chlorous acid from chlorite is about 31% at pH 2.3, 10% at pH 2.9 and 6% at pH 3.2, and the amount of chlorine dioxide does not exceed 1-3 mg/L (USDA, 2002b). The initial sodium chlorite concentration is in the range 500-1200 mg/L for spray and dip solutions (pH 2.3-2.9) and 50-150 mg/L for chilling water (pH 2.8-3.2).

The formation of semicarbazide in nitrogen-containing products after hypochlorite treatment has been recently reported (Hoenicke *et al.*, 2004). Therefore, the possibility that this substance could also be formed after treatment of chicken meat with other active chlorine substances, like acidified sodium chlorite, has been examined. Three concentration levels (0.012, 0.12 and 1.2% equivalent to 120, 1200 and 12000 mg/L, respectively) of sodium chlorite were used in the application solutions and they were kept in contact with chicken legs overnight. In all 3 cases, semicarbazide was not detected (<1μg/kg) in the treated samples even though the chlorite concentration was 10 times the maximum use level and time of exposure was overnight instead of 1 hour.

Acidified sodium chlorite may interact with either organic matter in solution or protein and fat compounds in the carcasses giving rise to different reaction products. The potential reactions are described below.

According to a manufacturer (International Registration Dossier, 2003), amino acid profiles in poultry carcasses were analysed after treatment under exaggerated conditions of immersion in 2525 mg of acidified sodium chlorite per L at pH 2.78 for 5 min. The distribution of amino acids obtained by hydrolysis of the proteins of the control poultry carcasses was identical to the distribution in the disinfected carcasses. The concentration of amino acids like cysteine, tyrosine, threonine and tryptophan, with easily oxidisable functional groups, was basically the same in the treated carcasses and the control carcasses. However, potential reaction products were not analysed.

Reactions of acidified sodium chlorite with lipids in poultry carcasses

Additional chlorine to unsaturated free fatty acids and their methyl esters may occur after treatment with ASC. The potential formation of chlorinated organic compounds has been analysed by a manufacturer in poultry carcasses under different conditions. The treatment consisted of immersion in 2525 mg acidified sodium chlorite per L, pH 2.78, for 5 min. No chlorinated organics could be detected. The detection limit for single-chlorinated molecules was about 0.05 mg per kg.

In further studies, a manufacturer (International Registration Dossier, 2003) treated carcasses by spray for 15 seconds with 1200 mg ASC per L, pH 2.5, followed by 2-hour air chilling. No apparent increases of organically bound chlorine were observed in the carcasses at the same detection limit (0.05 mg/kg).

The manufacturer also analysed the poultry carcasses to detect oxidation or changes in the fatty acids profiles under different treatment conditions. The treatments consisted of:

- immersion for 5 seconds in 1200 mg ASC per L, 5 min drip and 1 hour of immersion in water (pre-chill study)
- immersion for 1 hour in 150 mg ASC per L and 5 minutes of drip (chiller study).
- 15 or 30 seconds dip in 1200 mg ASC per L, with no rinsing and dwell times of 1, 2, 4 and 8 hours (post-chill study).
- 15 or 30 seconds dip in 1200 mg ASC per L, followed by 5 seconds of water rinsing and 30 seconds dwell time (post-chill study).
- 15 or 30 seconds dip in 1200 mg ASC per L, with no rinsing and 30 seconds dwell time (post-chill study).

In all cases, samples and controls were cooked before analysed. No chlorinated organics were found at a detection limit of 0.05 mg/kg.

The fatty acid profiles determined in the lipid fractions of the carcasses after the treatments with acidified sodium chlorite, as described above, were similar to those of the controls. No detectable changes were observed in the fatty acid profiles even in polyunsaturated fatty acids, which are more sensitive to oxidation. When performing the thiobarbituric acid (TBA) assay, which measures the oxidation of lipids, an increase in TBA reactive substances (TBARS) values was observed in the skin after the treatments but not in the muscle that remained unaffected regardless of the treatment. The use of ASC in spray gave lower TBARS values in the skin than the chill treatment. At 1200 mg ASC per L, a mild transitory whitening of the skin has been reported (Kemp *et al.*, 2000).

Chlorine dioxide

Chlorite and chlorate are the primary by-products resulting from the use of chlorine dioxide. Chlorite and chlorate formation increase (in a ratio of 7:3) with increasing concentration of chlorine dioxide and increased treatment time. Chlorine dioxide decreases rapidly. Generally, around 5% of an initial concentration of 50 mg/L, remains as chlorine dioxide (Tsai *et al.*, 1995; USDA, 2002a).

The organic by-products produced after treatment of drinking water by either liquid or gaseous chlorine dioxide have been determined by Richardson *et al.* (1994). In contrast to chlorine treatment, no halomethanes were detected in treated drinking water (Richardson *et al.*, 1994, 2003). However, other disinfection by-products were present (Richardson, 2003). Thus, a large number of fatty acids and other substances were found. Substances containing chlorine were found; for instance, 1-chloroethyldimethylbenzene and tetrachloropropanone were detected. The approximate concentrations reported by the authors for these by-products were within the range 1-10 ng per L for semi volatile compounds and around 0.05 mg/L for total organic halide compounds (Richardson *et al.*, 1994).

Chlorine dioxide may interact with either organic matter in solution or protein and fat compounds in the carcasses giving different reaction products. The potential reactions are described below.

Reactions of chlorine dioxide with proteins, peptides and amino acids

Proteins, peptides and some amino acids, especially tyrosine, tryptophan and cysteine can undergo oxidation and/or substitution when exposed to chlorine dioxide (Fukayama *et al.*, 1986). A study was conducted on the reaction of chlorine dioxide with 21 amino acids but only 6 of the amino acids reacted. Amino acids that showed positive reaction with chlorine dioxide contain sulphur or an aromatic ring in their structures. Amino acids at low pH are expected to be more inert towards oxidation because of the presence of an electron-deficient centre on the amino-nitrogen atom (Tan *et al.*, 1987a). Tyrosine, tryptophan and cysteine reacted very rapidly at all assayed pH values (3, 6 and 9); methionine reacted only at pH 9 while hydroxyproline, histidine and proline mainly reacted at pH 6 and 9 (Tan *et al.*, 1987a). Chlorine dioxide is reduced to chlorite ion and the amino acids are oxidized as follows: cysteine produces cysteic acid, tryptophan forms indoxyl, isatine and indigo red, methionine is oxidised to sulphoxide and finally, to the corresponding sulphone, and tyrosine forms dopaquinone (Tan *et al.*, 1987a).

Studies of 2 proteins (bovine serum albumin and casein) and 3 peptides (L-aspartyl-L-phenylalanine, L-glycyl-L-tryptophan and L-tryptophylglycine) have shown a rapid

reaction with chlorine dioxide at pH 6 except for L-aspartyl-L-phenylalanine, which was not reactive under these conditions (Tan *et al.*, 1987a). The proteins reacted very rapidly and the other two dipeptides also reacted rapidly with the heterocyclic ring of tryptophan being the major reaction site (Tan *et al.*, 1987a). Proteins represent the main constituent in poultry but some peptides are also present. Main dipeptides are carnosine (β-alanyl-L-histidine), anserine (β-alanyl-L-1-methylhistidine) and balenine (β-alanyl-L-3-methylhistidine); their concentrations vary depending on the muscle type. The concentrations of these dipeptides in poultry meat are within the following ranges: 60-180 mg/100g for carnosine, 200-780 mg/100g for anserine and 2-10 mg/100g for balenine (Aristoy and Toldrá, 2004). Other natural peptides are glutathione (L-γ-glutamyl-L-cysteinglycine) which is in the range of 14-30 mg/100g (Jahan *et al.*, 2004) and carnitine (β-hydroxy γ-N-trimethylysine) within the range 12-24 mg/100g muscle (Shimada *et al.*, 2004). The amount of free amino acids in meat, before any ageing, is very low; usual values in meat are below 30 mg/100g (Aristoy and Toldrá, 1991; Aliani and Farmer, 2005).

The Panel has received no data on potential semicarbazide formation following treatment of poultry with chlorine dioxide. However, the Panel notes that chlorine dioxide is a less aggressive oxidant than acidified sodium chlorite and also it is used in lower concentrations. Therefore, bearing in mind that the worst-case laboratory experiments using acidified sodium chlorite did not form any detectable semicarbazide, it seems unlikely that chlorine dioxide has the potential to form semicarbazide either.

Reactions of chlorine dioxide with lipids

Chlorine compounds can readily react with lipids. The extent of incorporation of chlorine into free fatty acids and their methyl esters was studied by Ghanbari *et al.* (1982) using radio labelled chlorine dioxide solutions. The main results are shown in table 1.

Table 1. Incorporation of ^{36}Cl into free fatty acids and methyl esters after treatment with $^{36}ClO_2$ solutions, at pH 6.0 for 60 min,. From Ghanbari et al. (1982)

Lipids	Formula	Double bonds	^{36}Cl [a]
Oleic acid	C 18:1	1	0.006
Linoleic acid	C18:2	2	0.013
Linolenic acid	C18:3	3	0.021
Arachidonic acid	C22:4	4	0.023
Methyl oleate			0.0039
Methyl linoleate			0.0075
Methyl linolenate			0.0094
Methyl arachidonate			0.0080
Triolein			0.0031

[a] Chlorine incorporated as moles/mole lipid. Values were calculated using the following formula: Percent chlorine incorporated/100 x molar concentration of available chlorine/5 x concentration of lipids

As can be observed in table 1, the extent of incorporation of chlorine into lipids is very low when exposed to chlorine dioxide. Chlorine dioxide is by and large less reactive with lipids than hypochlorous acid (Ghanbari *et al.*, 1982). The double bonds in the fatty acid moieties can undergo oxidation and addition in the presence of electrophiles

such as chlorine dioxide. The major reaction of chlorine dioxide is oxidation, rather than chlorination.

The amount of fat in poultry varies depending on the location. The skin contains up to 30g/100g, mostly triacylglycerols. Breast contains around 1g fat/100g with similar amounts of triacylglycerols and phospholipids and thigh contains around 2-3g fat/100g, most of them triacylglycerols. Poultry is rich in polyunsaturated fatty acids (PUFA). Linoleic acid is the major PUFA present in poultry fat as corn, wheat and/or barley are main cereals used for poultry feeds.

Reactions of chlorine dioxide with carbohydrates

Chlorine dioxide can react with carbohydrates through two types of reactions: Oxidation of the glycosidic bond and oxidative cleavage of the C2 and C3 carbon bonds to form carboxylic acids. The reactions of chlorine dioxide with carbohydrates generally result in oxidation products (Fukayama *et al.*, 1986). However, the amount of carbohydrate in poultry carcasses is extremely low so that any significant reaction of antimicrobial agents or production of disinfection by-products with carbohydrates would be unlikely.

Peroxyacetic and peroxyoctanoic acids

The peroxyacids solution used consists of a mixture of peroxyacetic acid, peroxyoctanoic acid, hydrogen peroxide and HEDP (1-hydroxy-1,1-diphosphonic acid). Upon application to the carcasses, acetic acid, octanoic acid, water and oxygen are generated as natural breakdown products.

Several products have been identified after disinfection treatment of surface water with peroxyacetic acid. These compounds are 1-methoxy-4-methylbenzene, nonanal and decanal (Monarca *et al.*, 2003; 2004).

Reactions of peroxyacids compounds with proteins, peptides and amino acids

Sulphur amino acids of proteins are susceptible to oxidation by peroxide reagents, like hydrogen peroxide, present in the peroxyacids solution. For instance, cystine is oxidised only partly to cysteic acid while methionine is oxidised to methionine sulphoxide and also produce a minor amount of methionine sulphone (Slump and Schreuder, 1973; Strange, 1984). Lanthionine generates lanthionine sulphoxide, lanthinine sulphone and some unidentified products. The oxidation of homocystine generates homolanthionine sulfoxide as main product and homolanthionine sulphone and homocysteic acid (Lipton *et al.*, 1977). Reduced glutathione can be oxidised by hydrogen peroxide. The oxidation rates increase with the pH and most of the cysteine in the glutathione is oxidised to the monoxide or dioxide. Sulphinic acid and cysteic acid are also produced by direct oxidation of cysteine (Finley *et al.*, 1981). Also tryptophan is easily oxidised. Main degradation products, when treating 5 mM tryptophan with 0.2 M H_2O_2 within the pH range 4.0 to 8.5 and heated for 60 min at 25, 60 and 100ºC, included other amino acids like alanine, glycine or serine as well as other products like kynurenic acid and 3-OH-kyrunenine. Xanthurenic acid and indolacetic acid were formed only at alkaline pH values (Kell and Steinhart, 1990) which are far from those of the applied solution.

Dipeptides containing tryptophan, ala-trp and phe-trp, were also oxidised by hydrogen peroxide. The observed degradation at pH 7.0 and 8.0 was due to the oxidation of

tryptophan, most important in ala-trp than in phe-trp (Kell and Steinhart, 1990). The formation of oxidation products for ala-trp was of the same order as with free tryptophan at pH 7.0. In the case of phe-trp, the formation of oxidation products was lower indicating that the phenyl ring of phenylalanine exerted a negative induction effect (Kell and Steinhart, 1990).

Reactions of peroxyacids compounds with lipids in poultry carcasses

The application of peroxyacids solution could cause oxidation of lipids, especially through the action of peroxyacids and hydrogen peroxide, which are strong oxidizing agents, on fatty acids with one or more double bonds (Rhee *et al.*, 1989). A manufacturer (Ecolab, 2004) analysed the potential oxidation of unsaturated fatty acids, measured as TBARS, and the alteration in the fatty acid profiles. Poultry carcasses were treated by spray with 200 mg total peroxyacetic acid per L for 15 seconds (spray treatment) or immersion for 60 minutes (chiller treatment). In both cases samples and controls were cooked at 90-95ºC for 45 minutes and also analysed. The results showed no significant alteration in the TBARS values or the fatty acids profiles when comparing treated samples, either raw or cooked, with respective controls.

ASSESSMENT OF EXPOSURE FROM ANTIMICROBIAL USE

The consumption of poultry can be estimated from the draft EU concise food consumption database, which is currently being developed by EFSA. This database is compiling mean and high percentiles of consumption for about 16 broad food categories from 3 European countries. Mean and high consumption of meat and meat products (including offals) by adults were extracted from the 3 national food consumption surveys currently considered, namely Italy (Turrini *et al.*, 2001), France (Volatier *et al.*, 2000) and Sweden (Becker *et al.*, 2002) which are based on 7 days records for individuals. Average mean daily consumption of meat (edible portion) varies from 120 g/day to 151 g/day, reaching 240 to 260 g/day at the 95th percentile and 320 to 350 g/day at the 99th percentile (see table 2). By using these figures on meat consumption, the consumption values provide a conservative estimate of mean and high consumption of poultry in Europe.

Potential dietary exposure to all substances was estimated based on the conservative hypothesis that the concentration in the edible part of meat is identical to the concentration in the carcass.

Table 2: Consumption of meat and meat products (including offal) in the adult population of Sweden, France and Italy

			Average daily consumption in consumers only (g/day)						
	Number of subjects	Number of consumers	mean	SD	50th	90th	95th	97.5th	99th
France	1875	1861	120	66	110	206	243	274	321
Sweden	1214	1204	151	68	141	233	263	297	346
Italy	1425	1419	137	67	127	224	264	292	351

Trisodium phosphate

According to previous estimations by the SCVPH (2003), the treatment of poultry carcasses with trisodium phosphate (TSP) would incorporate 480 mg TSP per kg carcass. Based on meat consumption data in European adults, as reported above, potential daily exposure to TSP for a 60 kg individual would be up to 1.21 mg/kg bw at the mean and up to 2.08 and 2.80 mg/kg bw at the 95th and 99th percentile of meat consumption, respectively.

Acidified sodium chlorite

The levels of chlorite and chlorate ions were determined by a manufacturer, under maximised treatment conditions (International registration dossier 2003). This was a pre-chill study consisting of immersion for 5 seconds in 1200 mg ASC per L, pH 2.5, on the wet carcasses after removal from a chiller tank and after 5 minutes post-removal drip period. The levels of chlorite and chlorate in the carcasses were 9 and 11 μg per kg carcass, respectively. When carcasses were submitted to 15 or 30 seconds dip in 1,200 mg ASC per L, with or without post-treatment water rinsing, the levels of chlorate and chlorite ions remained below 0.2 mg per kg carcass.

The fate of any residual chlorite or chlorate ions on poultry carcasses upon exiting from the commercial chiller water process (chiller study) consisting of 1 hour of immersion in 150 mg ASC per L, pH 2.8, was also determined by the manufacturer. The levels of chlorite and chlorate in the carcasses were 0.54 mg and 19 μg per kg carcass, respectively. The levels of chlorite and chlorate were also determined in post-treated carcasses up to 20 hours after the treatment. The residual chlorite and chlorate levels in the poultry carcasses were 16 and 19 μg per kg carcass, respectively. This leads to a potential dietary exposure to chlorite and chlorate of up to 0.04 and 0.05 μg/kg bw/day, respectively, at the mean for a 60 kg individual. Potential dietary exposure to chlorite would reach 0.07 and 0.09 μg/kg bw at the 95th and 99th percentile of meat consumption, respectively. Potential dietary exposure to chlorate would reach 0.08 and 0.11 μg/kg bw at the 95th and 99th percentile of meat consumption, respectively.

Chlorine dioxide

According to previous estimations by the SCVPH (2003), poultry carcasses would incorporate, after decontamination with chlorine dioxide for 1 hour, 0.13 mg chlorite and 0.06 mg chlorate per kg carcass. In addition, 0.01 mg chlorine dioxide, in the form of chlorite, per kg carcass would also be incorporated. Based on meat consumption data in European adults, as reported above, potential dietary exposure to chlorine dioxide for a 60 kg individual would be up to 0.02 μg/kg bw at the mean and up to 0.04 and 0.06 μg/kg bw at the 95th and 99th percentile of meat consumption, respectively. In the case of chlorite and chlorate, potential dietary exposure for a 60 kg individual would be up to 0.33 and 0.15 μg/kg bw/day, respectively, at the mean. High dietary exposure to chlorite and chlorate would be 0.56 and 0.26 μg/kg bw/day, respectively, at the 95th percentile of meat consumption and 0.76 and 0.35 μg/kg bw/day, respectively, at the 99th percentile of meat consumption.

Peroxyacetic and peroxyoctanoic acids

The residues in poultry carcasses were analysed after treatment with peroxyacids (peroxyacetic and peroxyoctanoic acids) solutions (Ecolab, 2004). The results were as follows:

Peroxyacids and hydrogen peroxide: Chicken carcasses were treated with 200 mg total peroxyacids per L for 15 s spray at ambient temperature followed by 60 min immersion chill at <4ºC. The concentrations were determined at 2, 5 and 10 min after completion of the chill treatment. The concentrations of both, peroxyacids and hydrogen peroxide, were below the detection limit of 1 mg/L. Based on these results, the SCVPH (2003) estimated that the residues of peroxyacids and hydrogen peroxide, 2 min after the decontamination treatment, would be equivalent to <0.25 mg per kg carcass. Based on meat consumption data in European adults, as reported above, potential dietary exposure to peroxyacetic acid and hydrogen peroxide for a 60 kg individual would be up to 0.63 μg/kg bw/day at the mean reaching up to 1.08 and 1.46 μg/kg bw at the 95th and 99th percentile of meat consumption, respectively.

HEDP (1-hydroxyethylidene-1, 1-diphosphonic acid): Six chicken carcasses were treated with two different solutions. Solution 1 containing 200 mg peroxyacids (as peroxyacetic acid) per L and 10 mg HEDP per L, and solution 2 containing 30 mg peroxyacids per L with 1.5 mg HEDP per L. All carcasses were sprayed 15 s with solution 1 at ambient temperature. Then, 3 carcasses were treated with immersion for 60 min in a chiller bath at 3ºC with solution 1 and the other 3 carcasses were immersed for 60 min in a bath at 2ºC with solution 2. Carcasses treated in the chiller bath with solution 1 gave a residual amount of 120-170 μg HEDP per kg carcass. In the case of solution 2, the residual amount was of 40-50 μg HEDP per kg carcass (reported by the manufacturer as approximate value because it was near the detection limit of the method). The manufacturer estimated a potential 10% variability in the final solution composition. Potential dietary exposure to HEDP for a 60 kg individual would be up to 0.43 μg/kg bw/day at the mean, with high potential exposure of up to 0.74 and 0.99 μg/kg bw/day at the 95th and 99th percentile of meat consumption, respectively.

The residue levels used in the above estimates of exposure were obtained under the treatment conditions. It is evident that any washing and cooking treatment of poultry before consumption could affect the presence of residues and concentration of certain disinfection-by-products. So, the final real dietary exposure by consumers is likely to be lower than the levels given in this section.

TOXICOLOGICAL EVALUATION

The present evaluation focus on the safety of residues of the active antimicrobial substances in poultry carcasses and of potential by-products arising from these treatments.

Trisodium phosphate

Background information

Trisodium phosphate is a permitted food additive in Europe identified as E 339 (iii) and authorised in several processed foods, including meat products (EC, 1995).

In the USA, sodium phosphates (mono-, di-, and tri-) are considered GRAS as multipurpose ingredients in food (21 CFR 182.1778). This GRAS status recognition was issued through experience based on common use in food and considering that the substance was used in food prior to January 1, 1958.

A maximum tolerable daily intake (MTDI) of 70 mg/kg body weight for phosphates was established by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) (WHO, 1982).
The SCF (1991) confirmed the MTDI value estimated by the JECFA for phosphates used as food additives. Both evaluations concluded that the main risk related to the ingestion of these additives was their potential effect on the calcium-phosphorus-magnesium balance of the body.

Residues evaluation
Based on meat consumption data in European adults, as reported above, as an estimate of poultry consumption, potential dietary exposure to trisodium phosphate for a 60 kg individual would be up to 1.2 mg/kg bw/day at the mean, reaching up to 2.1 and 2.8 mg/kg bw at the 95^{th} and 99^{th} percentiles of meat consumption, respectively.
Treated poultry carcasses are only consumed after processing (cooking, frying, etc) and final concentrations of phosphate residues to which the consumer would actually be exposed are likely less than what has been estimated above. Dietary exposures would thus only be a fraction of MTDI value (up to 4 %, 99^{th} percentiles) and the Panel considers that this exposure is of no safety concern.

By-products evaluation
The rapid dissociation of trisodium phosphate into its constituent ions and the relatively low chemical reactivity of Na^+ and PO_4^{3-} makes it very unlikely that significant levels of by-products would be produced after treatment of poultry carcasses.

Acidified sodium chlorite

Background information
Several national and international committees and agencies have evaluated acidified sodium chlorite. The International Programme on Chemical Safety (IPCS) derived a tolerable daily intake (TDI) of 0.03 mg/kg bw for chlorite while the US Environmental Protection Agency (EPA) designated the same level (0.03 mg/kg) as a reference dose (RfD) (SCVPH, 2003). The WHO guidelines set a guideline value of 0.7 mg/L for chlorite for drinking-water, based on a TDI of 0.03 mg/kg bw for chlorite (WHO, 2004).

Residues evaluation
Based on meat consumption data in European adults, as reported above, as an estimate of poultry consumption the potential dietary exposure to chlorite and chlorate for a 60 kg individual would be up to 0.04 and 0.05 µg/kg bw/day, respectively, at the mean. The potential dietary exposures to chlorite would reach 0.07 and 0.1 µg/kg bw at the 95^{th} and 99^{th} percentile of meat consumption, respectively. The exposures to chlorate would reach 0.08 and 0.11 µg/kg bw at the 95^{th} and 99^{th} percentile of meat consumption, respectively.
The estimated intakes are between 1000 and 300 times less the TDI value of 0.03 mg/kg bw for chlorite set by IPCS, EPA and WHO. Considering that the poultry carcass is to be consumed only after processing, the actual levels would be less and accordingly the final exposure likely to be lower. Therefore the Panel considers that the exposure to chlorite residues arising from treated poultry carcasses would be of no safety concern.

By-products evaluation
The results available from recent studies (International Registration Dossier, 2003) showed no apparent increase in organically bound chlorine or formation of chlorinated organics in poultry carcasses treated with acidified sodium chlorite. Fatty acids profiles including polyunsaturated fatty acids determined in the lipid fractions of carcasses treated with acidified sodium chlorite did not differ from those of untreated controls. Furthermore, oxidative changes in poultry carcasses, as followed by TBA measurements, remained essentially unchanged regardless of the treatment.
Comparative analytical results also showed that the amino acid profiles from untreated poultry carcasses and poultry carcasses treated with acidified sodium chlorite under stringent conditions are identical, In particular, the concentrations of the amino acids having reactive functional groups (cys, tyr, thr, trp) were the same in treated and control carcasses (International Registration Dossier, 2003).

Chlorine dioxide

Background information
Several national and international committees and agencies have evaluated chlorine dioxide. The International Programme on Chemical Safety (IPCS) derived a tolerable daily intake (TDI) value expressed as chlorine of 0.03 mg/kg bw, while the US Environmental Protection Agency (EPA) designated the same level (0.03 mg chlorite /kg bw) as a reference dose (RfD) (SCVPH, 2003). The WHO guidelines for drinking-water quality set a guideline value of 5 mg/L for chlorine based on a TDI of 0.150 mg/kg bw, allocating 100 % of this TDI to water and assuming a 60 kg bw individual consumes 2 litres of water per day (WHO, 2004).

Residues evaluation
Based on meat consumption data in European adults, as reported above, as an estimate of poultry consumption the potential dietary exposure to chlorite and chlorate for a 60 kg individual would be up to 0.3 µg/kg bw/day at the mean. The potential dietary exposure to chlorite and chlorate would reach 0.6 and 0.7 µg/kg bw at the 95^{th} and 99^{th} percentiles of meat consumption, respectively
These estimated exposure levels are between 1000 and 40 times lower than the TDI value of 0.03 mg/kg bw for chlorine and chlorite set by IPCS and EPA. Furthermore, considering that poultry carcass is to be consumed only after processing the exposure levels would diminish further. The Panel therefore concluded that after processing (washing, cooking) the actual exposure to chlorine dioxide residues arising from treated poultry carcasses would be of no safety concern.

By-products evaluation
Experimental results have shown that chlorine dioxide can readily react with amino acids, peptides, proteins and lipids. Chlorine dioxide reacts with free amino acids and dipeptides in solution giving rise to by-products. Reaction of chlorine dioxide with 21 amino acids and 3 peptides under laboratory conditions showed that only 2 amino acids (tryptophan and hydroxyproline) and 1 dipeptide (L-glycyl-L-tryptophan) produced by-products with mutagenic potential in the Ames *Salmonella* assay using strains TA100 and TA98 with and without metabolic activation. With metabolic activation, the mutagenic activity was lower. Chemical by-product species responsible for this activity were not identified (Tan *et al.*, 1987b). The mutagenic potential of products arising

from the reaction of chlorine dioxide with L-tryptophan were also confirmed in another study under the same test conditions (Owusu-Yaw *et al.*, 1990).
However, chlorine dioxide treatment of chiller water samples have been found not to induce significant levels of revertants in the Ames *Salmonella* assay using TA100 bacteria without S9 metabolic activation (not tested with metabolic activation) (Tsai *et al.* 1997). Furthermore, organic extracts of salmon and red grouper fillets did not showed mutagenicity activity in the Ames *Salmonella* assay using TA98 and TA100 bacteria, with and without S-9 activation, after treatment with 20 and 200 mg/kg aqueous chlorine dioxide (Kim *et al.*, 1999). The reaction products in the treated aqueous solutions processed similarly did not show mutagenic activity either (Kim *et al.*, 1999). Chlorine can also be incorporated into free fatty acids as shown by model experiments using radiolabelled aqueous solutions of chlorine dioxide (Table 1). Total susceptible fatty acids represent up 50 % of the total lipid content of poultry muscles and radiolabelled analytical results show that the most susceptible are polyunsaturated fatty acids. However the extent of incorporation of chlorine into lipids was shown to be very low. Furthermore, no effects on protein or lipid contents were reported after treatment of salmon and red grouper fillets with 20, 40 100 and 200 mg/kg chlorine dioxide in brine (3.5 % NaCl solution) (Kim *et al.*, 1998). Such treatments did not cause any change in the fatty acid compositions of treated fishes, according to the authors, and only thiamine and riboflavin contents were lowered after treatment with 40 mg/kg chlorine dioxide and higher concentrations.
No specific data on chlorine dioxide by-products formation from poultry proteins or lipids were available to the Panel. Chlorine dioxide is a less potent oxidizing agent than acidified sodium chlorite and results showing that treatment with the latter has no effect on the amino acid profiles of poultry carcasses and on organically bound chlorine levels, in fatty acids profiles or oxidative status of meat lipids (International Registration Dossier, 2003), strongly suggest that chlorine dioxide will not show any effect either.
Reactions of chlorine dioxide with aldehydes and ketones as well as carbohydrates have also been reported under laboratory conditions, giving rise to the formation of carbonyl compounds and oxidation reaction products, respectively. However, it appears that the amounts of carbohydrates and volatile aldehydes and ketones in poultry carcasses are too low to result in formation of significant levels of by-products of toxicological relevance.

Peroxyacetic and peroxyoctanoic acids

Background information
Both national and international committees and agencies have evaluated peroxyacid solutions for antimicrobial treatment of food (SCVPH, 2003). The most recent evaluation has been performed by JECFA (WHO, 2005). Whereas 1-hydroxyethylidene-1, 1-diphosphonic acid (HEDP) is stable in the solutions, the peroxyacids rapidly breaks down to acetic acid, hydrogen peroxide, octanoic acid, water and oxygen upon contact with organic matter.
Food containing residues of acetic acid and octanoic acid arising from the use of peroxyacid antimicrobial solutions has previously been considered as safe for human consumption (SCVPH, 2003; WHO, 2005).
For the peroxyacids (as peroxyacetic acid), SCVPH (2003) cites a LOAEL of 0.13 mg peroxyacetic acid/kg bw/day based on increased spleen weight and increased

hemosiderin in spleen red mater in rats receiving the compound via drinking water for four weeks.
For hydrogen peroxide a NOAEL of 26 mg/kg bw/day for males and 37 mg/kg bw/day for females was identified in a 90-day oral study using catalase-deficient mice. The NOAEL from a rat gavage study was 30 mg/kg bw per day (SCVPH, 2003).
For HEDP a NOAEL of 500 mg/kg bw/day was identified from two 90-days feeding studies in rats (WHO, 2005). Notably, histopathological lesions including gastrointestinal erosion were observed upon HEDP treatment at the higher doses tested (SCVPH, 2003, WHO, 2005). When tested in a 90-day study in dogs at oral dose levels up to 250 mg HEDP/kg bw/day no adverse effects were reported (WHO, 2005), whereas a NOAEL of 50 mg HEDP/kg bw/day via the diet was found in a combined two-generation study of reproductive toxicity and teratogenicity in rats. No evidence of teratogenicity was found, but HEDP was embryotoxic at 250 mg/kg bw/day. HEDP was not teratogenic in rabbits but a similar NOAEL of 50 mg HEDP/kg bw/day was found for embryotoxicity (WHO, 2005). In humans, an oral starting dose of 5 mg HEDP/kg bw/day, for not longer than 6 months, is used to treat Paget disease (WHO, 2005).

Residues evaluation
In its evaluation, JECFA considered that due to the high reactivity of the peroxyacids and hydrogen peroxide towards organic matter they would break down into acetic acid, octanoic acid, and water, respectively and therefore be of no safety concern.
However, in their report the SCVPH assumed that there would be residual peroxyacids in the poultry carcass. Based on meat consumption data in European adults, as reported above, as an estimate of poultry consumption the potential dietary exposure to peroxyacids and hydrogen peroxide for a 60 kg individual would be up to 0.6 µg/kg bw/day at the mean. The potential dietary exposure to peroxyacids and hydrogen peroxide would reach 1.1 and 1.5 µg/kg bw at the 95^{th} and 99^{th} percentiles of meat consumption, respectively. Potential dietary exposure to HEDP for a 60 kg individual would reach up to 0.4 µg/kg bw/day at the mean. The potential dietary exposure to HEDP would reach up to 0.7 and 1.0 µg/kg bw at the 95^{th} and 99^{th} percentiles of meat consumption, respectively.
From a comparison with the toxicological reference values outlined above the Panel concluded that, the estimated intakes of residues of peroxyacetic acid, hydrogen peroxide, acetic acid, octanoic acid and HEDP arising from the treatment of poultry carcasses would be of no safety concern. As the poultry carcass is only consumed after processing (i.e. washing and cooking) these intake levels are likely to be less.

By-products evaluation
As mentioned before lipid peroxidation of polyunsaturated fatty acids, particularly membrane phospholipids, could take place upon peroxyacetic acid treatment of poultry carcasses. Aldehydes such as 4-hydroxynonenal, malonaldehyde, propionaldehyde, methylglyoxal, and hexanal can be formed by lipid peroxidation of unsaturated fatty acids (Esterbauer *et al.*, 1991).
The results provided show that peroxyacetic acid treatment, monitored as TBARS, had no detectable effect on the oxidation status of poultry fatty acids. Furthermore, fatty acid profiles of treated poultry carcasses were not altered by any of the tested treatments.
Additionally, it is expected that no significant levels of amino acids by-products will be produced after treatment with peroxyacids since free amino acids levels in poultry meat, just before ageing, are very low.

No data on by-products formation from poultry proteins was available to the Panel.

CONCLUSIONS AND RECOMMENDATIONS

The Panel emphasises that its up-date of the previous opinion of the Scientific Committee on Veterinary Measures Relating to Public Health (SCVPH) with regard to toxicological risks to public health of residues and possible reaction products arising from the use of the antimicrobial substances only concerns the described conditions of use.
The Panel also took into consideration that processing of poultry carcasses (washing, cooking) would take place before consumption.

Trisodium phosphate:
On the basis of the available data, the Panel considers that treatment of poultry carcasses with trisodium phosphate as described is of no safety concern. The Panel considers that the rapid dissociation of trisodium phosphate into its constituent ions (Na^+ and PO_4^{3-}) and their relatively low chemical reactivity make it very unlikely that by-products of toxicological relevance are formed after this treatment.
There is no possibility of formation of semicarbazide from the use of trisodium phosphate.

Acidified sodium chlorite:
On the basis of available data, the Panel considers that treatment of poultry carcasses with acidified sodium chlorite as described is of no safety concern.
No chlorinated organics have been found upon treatment of poultry carcasses with acidified chlorite. Furthermore, potential semicarbazide levels from this treatment were below the limit of quantification of the analytical method ($\leq$ 1 μg/kg) and would therefore be of no safety concern.

Chlorine dioxide:
In contrast to the situation with acidified sodium chlorite, no specific data on chlorine dioxide by-products formation from poultry proteins or lipids were available to the Panel. Nevertheless, the Panel notes that chlorine dioxide is a less aggressive oxidant than acidified sodium chlorite and that it is used in lower concentration. Therefore, the Panel assumes chlorine dioxide will not significantly affect poultry lipids. In the case of potential chlorination of amino acids, aromatic amino acids constitute the preferential target but these amino acids are absent in identified peptides in poultry. Furthermore, the concentration of free aromatic amino acids in poultry is very low.
The Panel considers that the available data on the treatment of poultry carcasses with chlorine dioxide does not indicate a safety concern. Further data might be needed to confirm that chlorinated compounds are not generated to a significant extent.

Peroxyacids:
On the basis of available data, the Panel considers that treatment of poultry carcasses with peroxyacids as described is of no safety concern.
No detectable effects on the oxidation status of fatty acids or fatty acid profiles in poultry carcasses were reported following treatment with peroxyacids.
There is no possibility of formation of semicarbazide from the use of peroxyacids.

General:
The Panel notes that the initial health concerns about semicarbazide are no longer relevant. As set out in the EFSA opinion on semicarbazide (EFSA, 2005), new data showed that semicarbazide is not genotoxic *in vivo*.

Overall the Panel notes that since poultry carcasses absorb water, by comparison to dipping and immersion in repeatedly used water of chiller baths, spraying will reduce the exposure to residues and by-products that might arise from these treatments.
The Panel stresses that the use of antimicrobial solutions does not replace the need for good hygienic practices during processing of poultry carcasses, particularly during handling, and also stresses the need to replace regularly the water of chiller baths.

DOCUMENTATION PROVIDED TO EFSA

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ANNEX I

Acidified sodium chlorite

The addition of acid to sodium chlorite generates chlorous acid through the following reaction:

$NaClO_2 \rightarrow ClO_2^- + Na^+$

$ClO_2^- + H^+ \rightarrow HClO_2$

Other compounds like chlorine dioxide, chlorite ion and chlorate ion are generated, their proportion depending on the pH of the mixture. Below pH 4.0, chlorite ion reacts with extra acid to give chlorous acid but it is unstable and dissociates back to chlorite ion, reaching an equilibrium:

$ClO_2^- + H^+ \leftrightarrow HClO_2$

Chloride ion may be formed from the oxidation/reduction of chlorous acid and chlorite ion via the following reactions:

$HClO_2 + 3H^+ + 4e^- \rightarrow Cl^- + 2H_2O$

$ClO_2^- + 4H^+ + 4e^- \rightarrow Cl^- + 2H_2O$

Chlorine dioxide

Chlorine dioxide is reduced in water generating the chlorite ion:

$ClO_2 + e^- \rightarrow ClO_2^-$

Chlorite is reduced to chloride ion:

$ClO_2^- + 4H^+ + 4e^- \rightarrow Cl^- + 2H_2O$

In the absence of oxidisable substances in water and presence of alkali, chlorine dioxide gives chlorite and chlorate ions:

$2ClO_2 + H_2O \rightarrow ClO_2^- + ClO_3^- + 2H^+$

Peroxyacetic and peroxyoctanoic acids

Total peroxyacids as peroxyacetic acid is calculated as follows (USDA, 2002d):
PA = weight percent of peroxyacetic acid + [(weight percent of peroxyoctanoic acid /160)x76]
where 160 is the molecular weight of peroxyoctanoic acid and 76 is the molecular weight of peroxyacetic acid.

Oxidation/reduction reactions of peroxyacids take place in water generating water, acetic acid and octanoic acid as follows:

$H_2O_2 + 2H^+ + 2e^- \rightarrow 2H_2O$

$CH_3COOOH + 2H^+ + 2e^- \rightarrow CH_3COOH + H_2O$

$CH_3(CH_2)_6COOOH + 2H^+ + 2e^- \rightarrow CH_3(CH_2)_6COOH + H_2O$

When peroxyacids contact the poultry carcasses, they decompose as follows:

$2H_2O_2 \rightarrow 2H_2O + O_2$

$CH_3COOOH + H_2O \rightarrow CH_3COOH + H_2O_2$

$CH_3(CH_2)_6COOOH + H_2O \rightarrow CH_3(CH_2)_6COOH + H_2O_2$

$2CH_3COOOH \rightarrow 2CH_3COOH + O_2$

$2\ CH_3(CH_2)_6COOOH \rightarrow 2\ CH_3(CH_2)_6COOH + O_2$