



**ACIDIFIED SODIUM CHLORITE (ASC)
Chemical and Technical Assessment**

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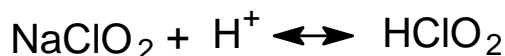
1. Summary

At the 68th meeting of JECFA, the safety of acidified sodium chlorite (ASC) for use as a surface-treatment antimicrobial agent was evaluated. This Chemical and Technical Assessment summarizes data and information on the acidified sodium chlorite as submitted to the 68th JECFA by Ecolab Inc., USA, in the dossier dated December 2006.

ASC possesses antimicrobial properties and is intended for use primarily as a spray or a dipping solution for poultry, meats, vegetables, fruits and sea foods. It is also used in poultry chilling water. ASC is produced by the addition of a food-grade acid (e.g., citric acid, phosphoric acid, hydrochloric acid, malic acid, or sodium hydrogen sulfate) to an aqueous solution of sodium chlorite (NaClO₂). Combining the acid with sodium chlorite solution results in conversion of chlorite to metastable chlorous acid (HClO₂), which can subsequently form a mixture with chlorite (ClO₂⁻), chlorine dioxide (ClO₂) and chloride (Cl⁻). The reaction, therefore, generates an oxidative solution with oxy-chlorine species with antimicrobial properties. ASC is intended for use as a part of an integrated approach designed to control microbial loads on food stuffs. ASC solution acts to reduce the number of pathogens (e.g., *Escherichia coli*, *E. coli* O157:H7, *Salmonella* spp., *Campylobacter* spp., and *Listeria monocytogenes*), as well as to a somewhat lesser extent, spoilage bacteria found on the surface of foods during processing. The solution is applied onto the surface of different types of fresh and processed foods at a concentration range of 50 to 1200 mg/l. Fresh and processed fruits and vegetables are subjected to a water rinse after ASC application followed 24-hour withholding time (for cut produce only). Treatment of whole or parts of poultry carcass, sausages or deli meats is carried out by spraying or dipping prior to or after chilling. ASC is also used to treat pre-chilling and chilling water at relatively low levels (i.e., 50 to 150 mg/l) into which poultry carcasses are submerged. Poultry and meat products are not subjected to rinse subsequent to treatment. Sodium chlorite used in the preparation of ASC has not previously been evaluated by the Committee. Among the acids mentioned above that may be used in the preparation of ASC, sodium hydrogen sulfate has not been evaluated by the Committee.

2. Description

Sodium chlorite [CAS No. 7758-19-2] is marketed in two forms, as a solid characterized by approx. 80% sodium chlorite and as an aqueous solution. Sodium chlorite is soluble in water, insoluble in non-polar solvents, and sparingly soluble in polar solvents (Merck, 2001). Aqueous solutions are colourless to greenish yellow and exhibit a slight chlorine-like odour. While the chlorite ion is stable in aqueous solution, under acidic conditions, chlorite forms a semi-stable intermediate, chlorous acid (HClO₂). Chlorous acid disintegrates to chlorine dioxide (ClO₂), which further degrades to chlorite (ClO₂⁻) and ultimately chloride (Cl⁻) is formed. The extent of each of the degradation pathways and thus the proportion of each of the oxy-chlorine species depends in part on the pH of the solution. Other factors such as temperature and alkalinity of the water also affect the composition of the oxy-chlorine constituents. At a pH in the range of 2.3-3.2, chlorous acid (5 to 35%) is the main active ingredient produced by the reaction and is in equilibrium with H⁺ and the chlorite ion (ClO₂⁻) (65-95%).



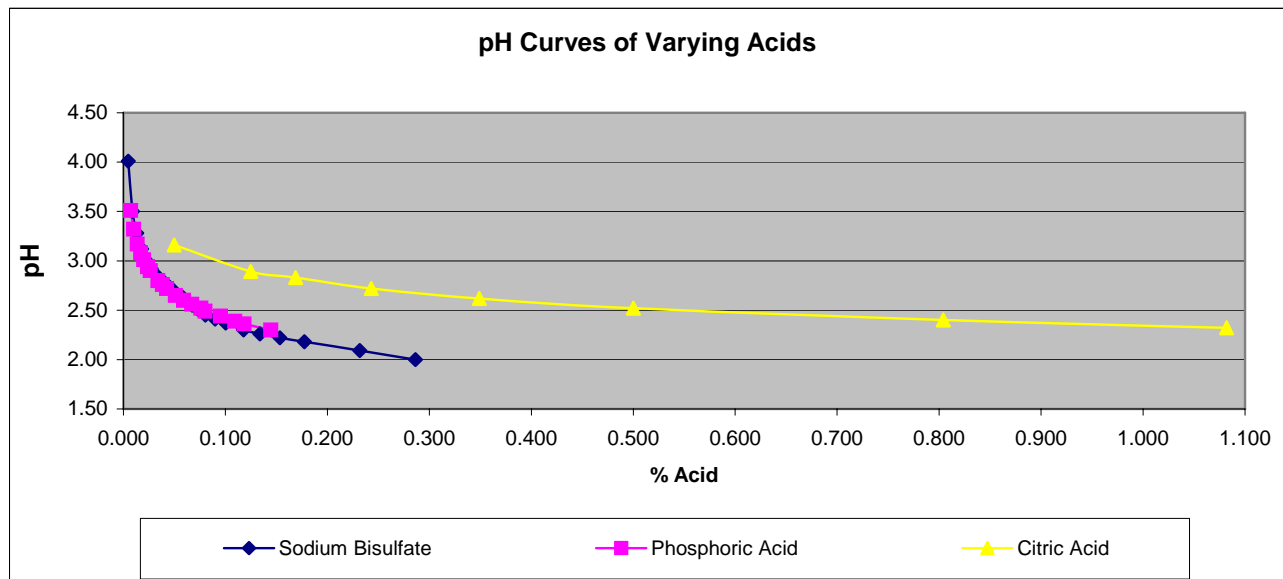
Concentrations of chlorous acid and its degradation products are determined by the pH, temperature, and acid/base concentrations of the solution. Sufficient food-grade acid is used to maintain a pH between 2.3 and 2.9 and sufficient technical-grade sodium chlorite is used to produce a 0.050 to 0.120% solution (500-1200 ppm). Among the different applications, the highest concentration of chlorous acid is attained at a pH of 2.3 in 1200 ppm ASC solution. At a pH of 2.3, approximately 31% of chlorite (from sodium chlorite) is converted to chlorous acid and 10 and 6% at pH 2.9 and 3.2, respectively. Concentrations of chlorous acid found in ASC solutions with different pH levels are summarized in Table 1.

Table 1. Concentration of chlorous acid (ppm) in solutions of Acidified Sodium Chlorite (ASC) with different chlorite concentrations and pH levels

pH	Concentration of Sodium chlorite			
	50 ppm	150 ppm	500 ppm	1200 ppm
2.3	16	-	157	376
2.8	6	19	-	-
2.9	5	-	51	123
3.2	3	8	-	-

Since different acids have varying buffering capacities, the acidity of the solution is dependent on the particular acidulant used to acidify sodium chlorite. The pH values at different concentrations of citric acid, phosphoric acid, and sodium hydrogen sulfate to prepare a 1000 mg/l sodium chlorite solution are presented in Figure 1.

Figure 1. pH curves for citric acid, phosphoric acid and sodium hydrogen sulfate (sodium bisulfate) to prepare a 1000 mg/l sodium chlorite solution



Sodium chlorite is largely used in the disinfection of water. It is also used, in substantial amounts, in the production of chlorine dioxide for applications such as bleaching of textiles, in the processing of pulp and paper (IARC, 1991). Accordingly, drinking water and occupational exposure are primary sources of sodium chlorite in the environment. In the United States, 7700 tonnes of sodium chlorite (80% purity)

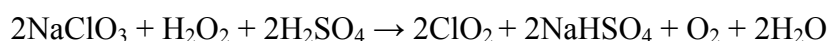
was manufactured in 1991 (Kaczur & Cawfield, 2001). In European countries, annual production volumes in the range of 1500 to 10000 tonnes were reported.

3. Method of Manufacture

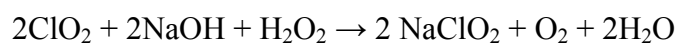
3.1. Sodium Chlorite

Commercially, sodium chlorite is manufactured *via* chemical or electrochemical reduction of sodium chlorate to chlorine dioxide gas (IARC, 1991; ATSDR, 2004). Chlorine dioxide dissolved in aqueous sodium hydroxide is subsequently reduced with hydrogen peroxide to form sodium chlorite. The resulting solution contains sodium chlorite at concentrations ranging from 30 to 50% and dried to a solid characterized by a sodium chlorite content of ca. 80% or diluted to an aqueous solution. More specifically, sodium chlorate, which is obtained by electrolytic oxidation of sodium chloride, is reduced electrochemically in the presence of hydrochloric acid to chlorine dioxide and chlorine, with sodium chloride formed as a by-product (sodium chloride is recycled to form sodium chlorate).

Alternatively, chlorine dioxide is obtained by reacting together sodium chlorate, hydrogen peroxide, and sulphuric acid according to the following:



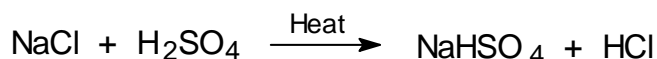
Subsequently, chlorine dioxide is absorbed in a solution of sodium hydroxide and hydrogen peroxide to form a sodium chlorite solution according to the following:



Sulphuric acid can be used to neutralize any excess alkali and the resulting sodium chlorite solution can be drum-dried to form solid sodium chlorite. The dried product is adjusted to contain at least 79% of sodium chlorite by addition of sodium chloride, sodium sulphate, or sodium carbonate. Instead of producing a solid product, the resultant sodium chlorite solution also can be diluted to a 25% solution.

3.2. Sodium Hydrogen Sulfate

Sodium chloride and sulfuric acid are the starting materials in the manufacture of sodium hydrogen sulfate. Natural salt is first dissolved in water and then re-crystallized to produce a refined salt. Sulfuric acid is obtained from the roasting of natural sulfur-containing ores, resulting in the production of sulfur oxides that are dissolved in water to produce sulfuric acid. The sodium chloride and sulfuric acid are combined at elevated temperatures to produce molten sodium hydrogen sulfate according to the following reaction:



Once the reaction is complete, the molten sodium hydrogen sulfate is sprayed and cooled to form a solid product of consistent particle size.

3.3 Other Acids (Citric acid, phosphoric acid, hydrochloric acid and malic acid)

Manufacturing of the above acids are not included in this CTA as they have been previously evaluated by the JECFA. Appropriate JECFA specifications may be consulted for their manufacturing processes.

3.4 Acidified Sodium Chlorite

ASC solutions are prepared just prior to use. Sodium chlorite and acids used in the preparation of ASC are stored either in bulk as concentrates or as formulated solutions. When stored as bulk concentrates, the individual components are diluted with water to a double-strength solution before mixing. The acid and base solutions, whether stored as such or diluted from concentrates at the site of mixing, are combined in a 1:1 ratio. Once mixed, the solution is applied within one to two minutes. The amount of food-grade acid that needs to be added to sodium chlorite in order to achieve a solution with a specific pH and level of chlorous acid depends on the strength of the acid and the buffering capacity of the solution. The alkalinity of the water, although not a primary determinant, also needs to be considered. Thus, weaker acids are added at concentrations of 0.2 to 1.2%, whereas stronger acids (*e.g.*, phosphoric acid) are used at lower concentrations (approximately 0.04 to 0.1%). A strong acid such as sulfuric acid would be added at a concentration of less than 50 µg/ml.

4. Characterization

4.1 Composition of sodium chlorite

The technical-grade of sodium chlorite used to prepare ASC is comprised of 80% of sodium chlorite, with sodium chloride, sodium carbonate, sodium hydroxide, sodium sulfate, and sodium chlorate making up the remainder of the composition.

4.2 Possible impurities (including degradation products)

4.2.1 Sodium chlorite

The manufacturing process employed in the production of sodium chlorite, does not include any specific purification steps. Impurities occurring in sodium chlorite comprises of unreacted starting materials (*e.g.*, sodium chlorate) as well as reaction by-products (*e.g.*, sodium chloride). Lead may occur in the final product as a result of their occurrence in the starting materials which are obtained from natural sources. Lead is limited by the specifications indicating maximum levels of 5 mg/kg.

4.2.2 Sodium hydrogen sulfate

Lead and selenium may occur in the final product as a result of their occurrence in the starting materials which are obtained from natural sources. Lead and selenium are limited by the specifications indicating maximum levels of 2 and 5 mg/kg, respectively.

4.2.3 Acidified sodium chlorite

Since ASC is a mixture of sodium chlorite and a food-grade acid, any impurities in the resulting ASC solution are expected to be those of the individual acid and base components. As such, there are no set purity criteria for ASC. Rather, the acid used to acidify sodium chlorite (*i.e.*, citric acid, phosphoric acid, hydrochloric acid, malic acid, or sodium hydrogen sulfate) must meet JECFA specifications its identity and purity.

Mixing of sodium chlorite with the food-grade acid results in the formation of chlorite, chlorate, and chlorine dioxide. Although levels of chlorine dioxide do not exceed 3 ppm in the solution initially, chlorine dioxide levels increase during aging of the solution. Chlorine dioxide is very volatile and is therefore lost by evaporation or reduction to chlorite by oxidation of organic matter (*e.g.*, bacteria). Consequently, it is not present as a residue of the treated food-product. Moreover, if ASC is used

according to good manufacturing practice (GMP), no residues of any of the oxy-chlorine species should remain on the sprayed or dipped food product (for further details see Section 6.1).

4.3 *Rationale for proposed specifications*

4.3.1 Sodium chlorite

Sodium chlorite is obtained by *via* chemical or electrochemical reduction of sodium chlorate to chlorine dioxide gas, which is further reduced to sodium chlorite in sodium hydroxide solution with hydrogen peroxide as the reducing agent. Chlorine dioxide can be obtained by reduction of sodium chlorate in the presence of hydrochloric acid (HCl), with sodium chloride formed as a by-product. In an alternative production process, chlorine dioxide is obtained by reacting together sodium chlorate, hydrogen peroxide, and sulfuric acid. Next, chlorine dioxide is absorbed in a solution of sodium hydroxide and hydrogen peroxide to form a sodium chlorite solution. Sulfuric acid can be used to neutralize any excess alkali. Sodium chloride, sodium sulfate, or sodium carbonate can be added to adjust the concentration of the solid form of sodium chlorite.

Purity of the final product is determined by assaying the sodium chlorite content and its identity is confirmed by positive tests for chlorite and sodium. Since the final product is not subject to any specific purification steps, purity parameters were based on residual levels of starting materials, as well as quantities of compounds added to adjust the content of sodium chlorite in the final product. As such, specifications for sodium chlorite include parameters for sodium chlorate, sodium chloride, and sodium hydroxide, as well as sodium carbonate and sodium sulfate. Heavy metal content is limited by a specification parameter for lead.

4.3.2 Acidified sodium chlorite

Since no residues of the oxy-chlorine species are expected on ASC-treated food products, an assay for available free chlorine levels in a solution of ASC is not considered to be appropriate. Instead, the ASC solution is monitored for sodium chlorite concentration and pH, which are known to result in acceptable levels of chlorate, chlorite, and chlorine dioxide. Depending on the food application, the solution is characterized by a sodium chlorite concentration in the range of 50-150 mg/l and a pH of 2.8-3.2 or 500-1200 mg/l and a pH of 2.5-2.9.

4.4 *Analytical methods*

4.4.1 Sodium chlorite

The test methods for the determination of lead and loss on drying are standard methods, published in the Combined Compendium of Food Additive Specifications [FAO JECFA Monographs, 1(4), 2006]. Sodium chlorate and sodium sulfate are determined by ion chromatography method based on the reference methods developed by the U.S. Environmental Protection Agency (U.S. EPA, 1993). Determination of sodium carbonate, sodium chloride, and sodium hydroxide, as well as the assay are conducted based on the validated test methods by the American Water Works Association (AWWA, 2005).

4.4.2 Acidified sodium chlorite

Chlorite and chlorine dioxide levels (in the range of 50 -1500 mg/l) in the ASC solution are determined together by an iodometric method. Essentially, the amount of chlorite in the solution is measured by determining consumption of iodine in the acidified solution. The analytical method used to determine the

pH of the ASC solution is an adaptation of a standard method for the determination of pH published in the U.S. Pharmacopoeia (USP, 2005a). The total acidity of the solution is measured using an adaptation of the USP assay method for citric acid (USP, 2005b).

Absence of potential chlorite and chlorate residues on the final food product is assessed using a potentiometric titration method (Tang & Gorden, 1980) and ion chromatography based on a validated method (Waters Innovative Methods for Ion analysis, unpublished) or a method used by the U.S. EPA (1993).

5. Functional uses

5.1 Technological functions

ASC is intended for use as an antimicrobial agent as part of integrated, “multiple-hurdle” approach designed to control growth of microbial species. The ASC solution acts to reduce the number of pathogenic, as well as to a somewhat lesser extent, spoilage bacterial species found on the surface of foods during processing. *Escherichia coli*, *E. coli* O157:H7, *Salmonella* spp., *Campylobacter* spp., and *Listeria monocytogenes* are among some of the bacteria that are effectively killed by interaction with the oxy-chlorine species in the solution. Use of ASC, therefore, results in a reduce risk for the occurrence of food borne illnesses, as well as extension of a food’s shelf-life. The solution is applied onto the surface of different types of fresh and processed foods (*i.e.*, poultry meats, fresh and processed meats, fruits and vegetables, and freshwater fish and seafood) at relatively low levels (*i.e.*, 50 to 1200 mg/l). The solution can be applied as a spray or dip prior to or after chilling (*e.g.*, whole or parts of poultry carcass) or cooking (*e.g.*, sausages, deli meats). It can also be used as a component of pre-chilling and chilling water into which poultry carcasses are submerged (*i.e.*, 50 to 150 mg/l). Depending on the food, a post-treatment water rinse may or may not follow application of ASC. Specifically, while poultry and meat products are not rinsed subsequent to treatment with ASC, both fresh and processed fruits and vegetables are subjected to a water rinse and a 24-hour withholding time (processed produce *i.e.*, cut only) after application of ASC.

The antimicrobial activity of the acidified sodium chlorite system results from the oxidative interaction between the oxy-chlorine breakdown products of the acidification process and organic matter. When solutions of the acid and sodium chlorite are combined, metastable chlorous acid is formed in an equilibrium reaction. Chlorous acid is a precursor to a series of strong oxidants (*i.e.*, chlorate, chlorite, chlorine dioxide), able to kill bacteria, fungi, viruses and algae. As chlorous acid is depleted from solution, more sodium chlorite becomes acidified in the solution to maintain chemical equilibrium. More specifically, the oxy-chlorine species found in the acidified solution, oxidize sulphide (S-H) bonds of amino acids and the disulfide (S-S) bonds of enzymes and, ultimately, disrupts cellular function. Chlorous acid is the primary antimicrobial oxy-chlorine species in ASC. It exerts its antimicrobial activity by direct disruption of the cellular membrane, as well as by oxidation of cellular constituents. The antimicrobial properties of ASC also are related to the presence of chlorine dioxide in the solution, which forms at low levels as one of the degradation products.

5.2 Food categories and use levels

ASC is intended to be applied at use-levels of (i) 50 to 150 mg/l ASC in chilling solution for immersion of whole carcass of poultry (pH 2.8 to 3.2); (ii) 500 to 1200 mg/l for whole carcass or parts of poultry and meats, and formed meats (*e.g.*, sausages, luncheon meats, pressed hams) (pH 2.5 to 2.9); and (iii) 500 to 1200 mg/l ASC for fruits and vegetables (whole and sliced), freshwater fish and seafood, and eggs (pH 2.5 to 2.9). The intended food-uses and use-levels for ASC are presented in Table 2.

Table 2. Summary of the individual proposed food uses and use-levels for Acidified Sodium Chlorite (ASC)

FOOD CATEGORY ¹	FOOD-USE ¹		USE-LEVELS (MGL)
04.0 Fruits and vegetables (including mushrooms and fungi, roots and tubers, pulses and legumes, and aloe vera), seaweeds, nuts and seeds	0.4.1.1.1	Untreated fresh fruit	500 to 1200
	0.4.1.1.2	Surface-treated fresh fruit	
	0.4.1.1.3	Peeled or cut fresh fruit	
	0.4.2.1.1	Untreated fresh vegetables (including mushrooms and fungi, roots and tubers, pulses and legumes (including soybeans), and aloe vera), seaweeds, nuts and seeds	
	0.4.2.1.2	Surface-treated fresh vegetables (including mushrooms and fungi, roots and tubers, pulses and legumes, and aloe vera), seaweeds, nuts and seeds	
	0.4.2.1.3	Peeled, cut or shredded fresh vegetables (including mushrooms and fungi, roots and tubers, pulses and legumes, and aloe vera), seaweeds, nuts and seeds	
08.0 Meat and meat products, including poultry and game	0.8.1.1	Fresh meat, poultry and game, whole pieces or cuts	500 to 1200 ²
	0.8.1.2	Fresh meat, poultry and game, comminuted	500 to 1200
09.0 Fish and fish products, including molluscs, crustaceans, and echinoderms	0.9.1.1	Fresh fish and fish products	500 to 1200
	0.9.1.2	Fresh molluscs, crustaceans and echinoderms	500 to 1200
10.0 Eggs and egg products	10.1	Fresh eggs	500 to 1200

¹ Food category system (Annex B) of the GSFA of the Codex Alimentarius Commission [CODEX STAN 192-1995 (Rev. 6-2005)] (CODEX, 2005).

² 50 to 150 ppm for whole poultry carcasses

6. Reactions and fate in foods

6.1 Potential residues of oxychlorine species

When ASC is sprayed onto raw food, chlorous acid (HClO₂) is consumed by reacting with microorganisms and other organic matter found on the surface of the treated food. As chlorous acid is consumed, the equilibrium shifts resulting in the generation of more chlorous acid. Chlorous acid may degrade to chlorite *via* transient intermediates including hypochlorous acid (HClO), dichlorine dioxide (Cl₂O₂) and dichlorine tetraoxide (Cl₂O₄). Chlorite that does not form chlorous acid may remain as chlorite ion in solution, or may react with water to produce chlorate. Thus, the predominant chemical residues from the acidified sodium chlorite system are chlorine dioxide, chlorite, and chlorate. Within the acidified sodium chlorite system, chlorine dioxide forms as an extremely low proportion (<3 mg/l) of the final solution.

6.1.1 Chlorite (ClO₂⁻) and Chlorate (ClO₃⁻)

The metastable chlorous acid is degraded to chlorite under the acidic conditions of the ASC solution. Chlorite is further oxidized to chlorate and chlorate is subsequently reduced to chloride, which is the

major residue component of ASC solutions. A number of studies were conducted to determine residual levels of chlorite and chlorate following treatment with ASC solutions applied under different conditions. Results of these assays are summarized below in Table 3.

Table 3. Residual chlorite and chlorate concentrations for Acidified Sodium Chlorite (ASC) treated foods

FOOD	TYPE OF APPLICATION		CONCENTRATION (MG/L)*			
	Treatment 1	Treatment 2	Immediately after Treatment Completion		After a Specified Dwelling/Holding Time	
			ClO ₂ ⁻	ClO ₃ ⁻	ClO ₂ ⁻	ClO ₃ ⁻
Poultry						
Chicken carcass (pre-chill or chilled)	5-sec immersion; 1200 ppm; pH = 2.5	5-min drip; 1-hour immersion in chilled water	-	-	<0.009	<0.011
	1-hr immersion; 150 ppm; pH = 2.8	5-min drip	0.54	<0.019	-	-
		5-min drip; 10 min, 1-, 2-, 4-, or 20-hr dwell time	-	-	<0.016 (t ≥ 2 h)	-
	15-sec spray; 1200 ppm; pH = 2.5	45-min hydrocooling; deboning and breast removal for analysis (24 hrs)	-	-	<0.1 ¹	<0.1 ¹
15-sec spray or dip; 1200 ppm; pH = 2.5	2-hr air-chilling (2-3°C)	-	-	<0.006 ¹	<0.006 ¹	
Chicken breasts (post-chilled) ²	15- or 30-sec immersion; 1200 ppm; pH = 2.5	With or without 5-sec water rinse; 30-sec dwell time	<0.1	<0.1	-	-
		No water rinse; 1-, 2-, 4-, or 8-hr dwell time	-	-	<0.1	<0.1
Red Meat						
Steak (pre-chilled)	5-sec spray; 500, 850, or 1200 ppm; pH = 2.5	30-sec drip; 1- to 2-sec water rinse	<0.1 ¹	<0.1 ¹	-	-
	5- or 10-sec spray; 1200 ppm; pH = 2.5	30 sec drip; 1- to 2-sec water rinse or air dry	<0.1	<0.16 – <0.22 ³	-	-
	15-sec spray; 1200 ppm; pH = 2.5	Air dry	<0.06	<0.06		
Steak (double-treated)	15-sec spray; 1200 ppm; pH = 2.5; air dry	10-sec dip; 1200 ppm; pH = 2.5	<0.06	<0.06		
	15-sec spray; 1200 ppm; pH = 2.5; air dry and chill	15-sec spray; 1200 ppm; pH = 2.5; air dry	<0.06	<0.06	-	-
Processed, comminuted or formed meats ⁴	30-sec spray or dip; 1200 ppm; pH = 2.5	Drip and air dry or water wash and air dry; surface rinse	<0.045	<0.045		
		Same as above; 24 hr dwell time	-	-	<0.045 ¹	<0.045 ¹
Seafood and Freshwater Fish						
Fresh	30-sec immersion; 1200	30-sec drip; no water	1.70-	<0.1	--	-

salmon, grouper, catfish, scallops, and shrimps	ppm; pH = 2.3	rinse	38.14			
		30-sec drip; water rinse	<0.01-2.76	<0.1	-	-
		30-sec drip; with or without a water rinse; 24- or 48-hr dwell time	-	-	<0.01	<0.1
Produce						
Pre-processed ⁵ , 4	10-sec spray; 2,400 ppm; pH = 2.5	30-sec drip; high-volume tap water rinse	<0.1 ⁶	<0.1 ⁶	-	-
Post-processed (cut up/sliced)	5- or 30-sec spray; 1200 ppm; pH = 2.5	30-sec drip; air dry ⁷	≤17.58	≤0.64	-	-
		30-sec drip duplicate water rinse ⁷	≤7.12	<0.1		
		30-sec drip duplicate water rinse; 6-hr dwell time ⁸	-	-	<0.1	<0.1
	30-sec spray; 1200 ppm; pH = 2.5 ⁹	Drip and air dry for 24 hours	-	-	<0.01-1.49	<0.01
		Water wash and air dry for 24 hours			<0.01	<0.01
	30-sec dip; 1200 ppm; pH = 2.5 ⁹	Drip and air dry for 24 hours	-	-	<0.01-16.82	<0.01
Water wash and air dry for 24 hours				<0.01 (0.23 ¹⁰)	<0.01	

* Values of “less than” indicate the limit of detection.

¹ Cooked and uncooked.

² 2nd ASC treatment; 1st ASC treatment applied “pre-chill” (1200 ppm, 15-sec spray).

³ Levels detected in both test and control samples.

⁴ Hot dogs (4 x 55.5 g).

⁵ One whole apple, orange, strawberries, carrot, and cucumber.

⁶ When fruits/vegetables were only air-dried or immersed in water (*i.e.*, light-water wash) following treatment with a 5 or 10-sec spray of 1200 ppm ASC, up to 803 µg of chlorite and 495 µg of chlorate recovered per piece of produce.

⁷ 100 g of French fries; cubed cantaloupe; diced onions; or baby carrots.

⁸ 100 g of cubed cantaloupe.

⁹ 100 g of baby carrots; cubed cantaloupe; lettuce; oranges; strawberries; diced onions; or French fries.

¹⁰ Lettuce only.

In assays in which levels of residual oxy-chlorine species were monitored over a period of time (up to 48 hours), time-dependent reductions were observed in the levels oxy-chlorine residues in cases where residues were detected immediately following treatment application. In most instances, residue levels declined to levels below the limits of detection with time. For example, chlorite levels of 0.54 ppm detected on poultry carcasses immediately after ASC application, were reduced to 0.021 ppm within the first hour following treatment and were not detectable thereafter (<0.016 ppm). Likewise, chlorite residues on fish and seafood were detected only immediately following treatment, but not following a 24-hour holding period. Moreover, considering the highly labile nature of chlorite, it is expected that in the unlikely event that residues remain on the treated food product, they will be degraded during cooking.

While poultry, fresh meat and meat products, seafood, and fish are generally not rinsed following application of ASC, a water rinse follows ASC treatment in the cases of fruits and vegetables. In general, in comparison to unwashed produce, detectable levels of residues were considerably lower following rinsing (see Table 3; produce). While chlorite residues were identified on pre-processed produce that had only been immersed in water following treatment with ASC, residues fell below the level of detection when a high-volume wash was applied. In another study conducted with various types of processed (cut-up) fruits and vegetables (*i.e.*, carrots, melons, lettuce, organs, strawberries, onions, and French fries) dipped in or sprayed with a 1200 mg/l ASC solution (pH = 2.5) for 30 seconds, rinsed, and held for 24 hours, measurable chlorite levels were identified on many of the treated samples when they were not rinsed. However, a post-treatment wash reduced chlorite levels to below detection in all cases except for ASC-dipped (but not sprayed) lettuce (0.23 ppm). It was suggested that the natural crenulations and folds of the lettuce leaves are conducive to the pooling of the ASC solution and the rinsing water, resulting in some residual material persisting on the leaves at 24 hours after the solution had been applied. This was more apparent in cases where the lettuce was dipped in the ASC solution as dipping results in a greater volume of ASC solution coming in contact with the produce surface. In an attempt to identify additional processing steps that would further reduce levels of chlorite residues on lettuce, samples of lettuce were washed prior to treatment application. No detectable levels of chlorite residues were identified on lettuce that had been pre-treated with water, prior to ASC application (spray or dip).

6.1.2 Chlorine dioxide (ClO₂)

Following acidification, a fresh ASC solution will typically contain less than 3 ppm of chlorine dioxide. Chlorine dioxide is relatively soluble and will therefore remain in solution; however, because chlorine dioxide also is extremely volatile, it will be off-gassed, with the off-gassing increasing as particle size decreases, from the solution when it is sprayed onto food. Likewise, for foods that are treated *via* immersion into the ASC solution, chlorine dioxide remaining in the solution is expected to evaporate from the food's surface as the food dries. However, any chlorine dioxide that does come into contact with the food surface also may exert antimicrobial activity. Reaction of chlorine dioxide with microbial contaminants results in the formation of chlorite, which is further degraded to chlorate and, ultimately, chloride.

6.1.3 Chloride (Cl⁻)

Although, chloride is the major by-product of the antimicrobial activity of the ASC solution, because of background sodium chloride levels in food, the chloride generated as a result of treatment with ASC solution is not quantifiable. However, given the ubiquitous nature of chloride ions in food, the amount of chloride produced as a by-product of the interaction of the oxy-chlorine species with microbial matter, is not expected to be a toxicological concern. Moreover, chloride occurs endogenously under normal physiological conditions in human body water.

6.2 Potential reaction with organic compounds

Chlorous acid that does not react with microbial substrates may potentially react with organic substrates on the outer surface of the ASC-treated food itself. Since disinfection with chlorine-based systems is known to result in the formation of chlorinated organic compounds, the potential for the occurrence of chlorinated organic products on different types of ASC-treated foods was evaluated under standard and exaggerated treatment conditions.

No evidence of either oxidized or chlorinated organic residues, including lipids or amino acids and proteins, was found. Specifically, a study was performed in which chicken wings were immersed for 5 minutes in a 2525 mg/l ASC solution (*i.e.*, 2 times the permitted concentration) and subsequently rinsed with deionised water and left to soak overnight in hexane. Analysis of the hexane extract did not reveal

any detectable levels of chlorinated lipids at a detection limit of 0.05 mg/l. Thus, assuming that a poultry carcass has a 5% lipid content, levels of chlorinated organic compounds, if any present, would occur at levels of less than 0.0025 mg/l. Likewise, no differences were identified in the amino acid and fatty acid distribution of ASC-treated poultry carcasses in comparison to untreated samples. The fatty acid profiles of ASC-treated red meat, seafood, and fish, also were comparable to untreated controls. Additionally, pre- and post-chilled red meat samples, with the latter treated twice with ASC, as well as seafood and fish were tested for total organic halides. No differences were identified in the total organic halide content between treated and untreated food products.

In addition to the potential for chlorination of organics, potential oxidative changes also were evaluated in poultry, pre- and post-chilled red meat, and seafood and fresh water fish. ASC-treated food products were assessed for malondialdehyde (MAD) oxidation products using the thiobarbituric acid (TBA) assay. Although TBA reactive substances in treated samples were generally elevated, the increases were marginal in comparison to changes that occur as a result of normal storage and cooking of both ASC-treated and untreated samples. In red meats and seafood/fish, peroxide values were measured as an indicator of potential lipid peroxidation. No changes were identified in the peroxide value of food samples following application of ASC.

7. References

ATSDR, 2004. *Toxicological Profile for Chlorine Dioxide and Chlorite*. Atlanta (GA): U.S. Department of Health and Human Services, Public Health Service (PHS), Agency for Toxic Substances and Disease Registry (ATSDR). <http://www.atsdr.cdc.gov/toxprofiles/tp160.pdf>

AWWA, 2005. Test Method B303-5: Section 5: Verification: Sodium chlorite. Denver (CO): American Water Works Association (AWWA), pp. 4-12.

CODEX (2005) Food category system: Annex B. In: *General Standards for Food Additives CODEX STAN 192-1995 (Rev. 6-2005)*. Rome: Food and Agriculture Organization of the United Nations (FAO), World Health Organization (WHO), Codex Alimentarius Commission, pp. 9-53. (http://www.codexalimentarius.net/download/standards/4/CXS_192e.pdf).

Combined Compendium of Food Additive Specifications, FAO JECFA Monographs 1, Volume 4, 2006. Food and Agriculture Organization of United Nations (FAO) <http://www.fao.org/docrep/009/a0691e/a0691e00.htm>

IARC, 1991. Sodium chlorite. In: *Chlorinated Drinking-Water; Chlorination By-Products; Some Other Compounds; Cobalt and Cobalt Compounds*. IARC Working Group, June 12-19, 1990, Lyon, France. Lyon (France): World Health Organization (WHO), International Agency for Research on Cancer (IARC), IARC Monographs on the Evaluation of Carcinogen Risks to Humans, Vol. 52, pp. 145-158.

Kaczur, J.J., and Cawfield, D.W., 2001. Chlorine oxygen acids and salts, chlorous acid, chlorites, and chlorine dioxide. In: *Kirk-Othmer Encyclopedia of Chemical Technology*, 5th Ed. Vol 6. New York (NY), John Wiley and Sons Inc.

Merck, 2001. Sodium chlorite. In: *The Merck Index: An Encyclopedia of Chemicals, Drugs, and Biologicals*, 13th Ed. Whitehouse Station (NJ), Merck & Co., Inc., p. 1538 [Abstract No. 8672].

Tang, T.-F., and Gordon, G., 1980. Quantitative determination of chloride, chlorite, and chlorate ions in a mixture by successive potentiometric titrations. *Anal. Chem.* 52, 1430-1433.

U.S. EPA, 1993. *Determination of Inorganic Anions in Drinking Water by Ion Chromatography. Revision 1.0.* Cincinnati (OH): National Exposure Research Laboratory, Office of Research and Development (ORD), U.S. Environmental Protection Agency (U.S. EPA), Method 300.1 (<http://www.epa.gov/safewater/methods/met300.pdf>, updated to 1997 with 1999 Errata).

USP, 2005a. Method: <791> pH. In: *United States Pharmacopeia, 28th Ed. & National Formulary, 23rd Ed.* Rockville (MD): U.S. Pharmacopeia (USP) Convention Inc., pp. 2455-2456.

USP, 2005b. Anhydrous citric acid. In: *United States Pharmacopeia, 28th Ed. & National Formulary, 23rd Ed.* Rockville (MD): U.S. Pharmacopeia (USP) Convention Inc., pp. 483-484.

Waters Innovative Methods for Ion analysis (Unpublished) Method No. A119. In: *Oxyhalide Analysis Using IC-Pak A HC Column.* Millipore Corporation, Waters Chromatography Division. Manual Number 22340, Revision 1.