Report of the Scientific Committee of the Spanish Agency for Food Safety and Nutrition (AESAN) on the safe use of two aqueous solutions of hydrogen peroxide, acetic acid and peracetic acid as processing aids for the bacterial disinfection of apples and peaches washing water

Reference number: AESAN-2021-009

Report approved by the Scientific Committee in its plenary session on 28 July 2021

Working group

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Abstract

The company AgroFresh Fruit Protection S.A. has requested an assessment of the safety of using two aqueous solutions of hydrogen peroxide, acetic acid and peracetic acid as processing aids. It includes 1-hydroxyethylidene-1,1-diphosphonic acid (HEDP) as stabiliser.

The proposed use is the bacterial disinfection of water used for washing apples and peaches in processing plants.

These are aqueous solutions with the same composition and manufacturer as solutions previously assessed by the Scientific Committee for the bacterial disinfection of water for washing citrus fruits and peppers. With regard to the dose proposed for use, given the reactivity of the active substances

Translated from the original published in the journal: Revista del Comité Científico de la AESAN, 34, pp: 23-37

with organic material present in the dirt covering the fruits, an initial addition of 0.1 % of FreshStart Disinfect 25-15 or 0.3 % of FreshStart Disinfect 25-5 is required. In both cases, the final concentration of peracetic acid in the washing solution will be 150 ppm. After this initial dosage of 150 ppm of peracetic acid, maintenance dosages (0.033 % of FreshStart Disinfect 25-15 or 0.1 % of FreshStart Disinfect 25-5) shall be given in order to maintain the washing solution at a peracetic acid concentration of 50 ppm.

Considering the most adverse scenario of the presence of residues in apples and peaches and their consumption, an Estimated Daily Intake (EDI) of these residues as well as a consumer risk assessment by calculating the Margin of Safety (MOS) have been made.

The Scientific Committee concludes that, based on the information provided by the applicant and taking into account the proposed composition and conditions of use, the usage of the aqueous solutions as processing aids does not involve a health risk for the consumer.

Key words

Apples, peaches, processing aid, bacterial disinfection.

Suggested citation

AESAN Scientific Committee. (Working group) Marín, S., Berrada, H., Hernando, I. and López, R. Informe del Comité Científico de la Agencia Española de Seguridad Alimentaria y Nutrición (AESAN) en relación a la seguridad del uso de dos soluciones acuosas de peróxido de hidrógeno, ácido acético y ácido peracético como coadyuvantes tecnológicos para la desinfección bacteriana del agua de lavado de manzanas y melocotones. *Revista del Comité Científico de la AESAN*, 2021, 34, pp: 23-37.

1. Introduction

The company AgroFresh Fruit Protection S.A. located in Paterna (Valencia), has requested an assessment of the safety of using two aqueous solutions of hydrogen peroxide, acetic acid and peracetic acid as processing aids for the bacterial disinfection of water used for washing apples and peaches upon arrival at the processing plant. They also include 1-hydroxyethylidene-1,1-diphosphonic acid (HEDP) as a stabiliser.

The two aqueous solutions, FreshStart Disinfect 25-15 and FreshStart Disinfect 25-5, differ in the concentrations of their active components and the stabilizer, obtaining, in all cases, the same final concentration of peracetic acid in the washing solution (150 ppm). The different presentations respond to commercial reasons, to adjust the composition to customer transport and storage regulations.

These are aqueous solutions with the same composition and manufacturer as the ones previously assessed by the Scientific Committee for the bacterial disinfection of water for washing citrus fruits and peppers (AESAN, 2018, 2020).

With regard to authorised uses in human foods, it must be pointed out that both the individual components of the processing aid and the solutions of hydrogen peroxide, acetic acid and peracetic acid have different authorised uses in several countries.

Following the "Guidelines on precise documentation required for evaluation of processing aids that are intended for use in human food" (AESAN, 2010) and given that the presence of residues in the final products (apples and peaches) after the use of these aqueous solutions cannot be disregarded, the processing aid is classified under Situation 4: substance authorised in human food whose ADI (Acceptable Daily Intake) is not established and whose use may lead to the presence of technically inevitable residue. In light of this situation, the applicant has submitted information relating to the following aspects:

- Administrative data and general presentation.
- Physicochemical characteristics.
- Technological function.
- Residue studies: analytical method and validation of method.
- Studies and data relating on safety.
- Study of consumption and assessment of anticipated intake levels by the consumer.

2. General presentation and physicochemical characteristics

2.1 Detailed composition and formulation

The products proposed as processing aids and bearing the commercial names FreshStart Disinfect 25-15 and FreshStart Disinfect 25-5, are aqueous solutions of peracetic acid, hydrogen peroxide, acetic acid and 1-hydroxyethylidene-1,1-diphosphonic acid (HEDP) as stabiliser. The two compositions seek to maintain the chemical equilibrium of the components.

The two aqueous solutions differ in the concentrations of their active components (hydrogen peroxide, acetic acid and peracetic acid) and the stabilizer (HEDP), obtaining, in all cases, the same final concentration of peracetic acid (150 ppm) in the washing solution (Table 1).

Table 1. Composition of the processing aids

Substance	Function	CAS No.	Concentrations (%)	
			FreshStart Disinfect 25-15	FreshStart Disinfect 25-5
Peracetic acid	Active substance	79-21-0	15	5
Hydrogen peroxide	Active substance	7722-84-1	25	25
Acetic acid	Active substance	64-19-7	16	8
1-hydroxyethylidene-1,1-diphosphonic acid (HEDP)	Stabilizer	2809-21-4	0.6	0.5

The pH of the solution at 100 % is <1, at a temperature of 20 ºC.

2.2 Product specifications

Tables 2 and 3 list the specifications and results of the analysis of several batches of the processing aids.

Table 2. Specifications and results of the analysis of FreshStart Disinfect 25-15

Substance	Specifications (% w/w)	Certificates of analysis (% w/w)			
Peracetic acid	15 ± 1.0	15.2	15.8	15.6	15.6
Hydrogen peroxide	25 ± 2.0	24.8	24.6	24.6	24.3
Acetic acid	16 ± 2.0	16.2	15.7	16.6	15.9
1-hydroxyethylidene--1,1-diphosphonic acid (HEDP)	0.6	-	-	-	-

Table 3. Specifications and results of the analysis of FreshStart Disinfect 25-5

Substance	Specifications (% w/w)	Analysis certificates (% w/w)					
Peracetic acid	4.5-5	4.5	4.5	4.8	5.1	4.8	4.6
Hydrogen peroxide	25 ± 2.0	25.4	25.7	25.4	25.5	25.5	25.9
Acetic acid	8 ± 2.0	7.7	7.8	7.6	7.4	7.6	7.8
1-hydroxyethylidene--1,1-diphosphonic acid (HEDP)	0.5	-	-	-	-	-	-

The applicant has not provided data on compliance with the HEDP specifications.

2.2.1 Product stability

The applicant has submitted a study on the evolution of the concentration of peracetic acid by means of a model based on calorimetric analysis conducted with solutions that have a similar composition.

Based on the results obtained, the applicant indicates that FreshStart Disinfect 25-15 has a stability of 9 months and FreshStart Disinfect 25-5 has a stability of 1 year.

2.2.2 Reactivity

As mentioned in previous assessments of solutions with a similar composition, the reactions that take place in water are the decomposition of the compounds with peroxide groups to create acetic acid and water (EFSA, 2005, 2014).

The Joint FAO/WHO Expert Committee on Food Additives (JECFA) indicates that, in contact with food, the active ingredients in these kinds of disinfectant solutions (with hydrogen peroxide, peracetic acid, octanoic acid, peroxyoctanoic acid and HEDP) decompose rapidly in non-toxic substances and that the quantities of acetic acid and octanoic acid that may remain as a result of decomposition of peracetic acid and peroxyoctanoic acid do not constitute a safety issue. It also states that hydrogen peroxide decomposes rapidly in contact with foods, obtaining water and oxygen (JECFA, 2004, 2005).

Furthermore, the use of these types of solutions does not appear to negatively affect the nutrient content (vitamin C and β-carotene) of fruits based on the results of a study conducted by the JECFA (2006) using washing solutions with 80 ppm of peracetic acid and 50 ppm of hydrogen peroxide for 5 minutes.

2.3 Authorised use in human food

As mentioned, these are aqueous solutions with the same composition and manufacturer as the ones previously assessed by the Scientific Committee for the bacterial disinfection of water for washing citrus fruits and peppers (AESAN, 2018, 2020). Table 4 contains examples of authorised and assessed uses of these substances.

Table 4. Examples of authorised uses and assessments

Substance	Authorised use/assessment	Country/Reference
Hydrogen peroxide	Regulation (EC) No. 853/2004 establishes a limit of 10 ppm hydrogen peroxide residue for gelatin and collagen	European Union (EU, 2004)
	Favourable toxicological assessment as processing aid in the processing of blood derivatives and cephalopods	Spain (AESAN, 2011)
	Authorised for use in the production of beer as a clarifying agent (maximum quantity 135 mg/kg), in buttermilk to discolour and maintain pH (100 mg/kg) and in oat hulls as a bleaching agent (GMP)	Canada (DJC, 2021)
	Generally recognised as safe (GRAS) (21 CFR 184.1366), used in milk (0.05 %), whey (0.04 %), whey cheese coloured with annatto (0.05 %), starch (0.15 %), corn syrup (0.15 %), emulsifiers (1.25 %), dehydrated eggs, stomachs, beef trotters, herring, wine, tea and wine vinegar	United States (ECFR, 2021a)
	Authorised in combination with acetic acid for the process of washing or peeling fruit and vegetables that are not unprocessed raw material and not exceeding 59 mg/kg in the washing solution	United States ECFR, 2021b)
	Authorised for use as processing aid (bleaching, washing and peeling agent, pH stabilizer and inhibitor) in several foods and water (5 mg/kg)	Australia (ANZFSC, 2021)
Acetic acid	Authorised as food additive (E 260) in accordance with Regulation (EC) No. 1333/2008, with maximum specific dose as *quantum satis*	European Union (EU, 2008)
Peracetic acid	Authorised for use as processing aid of peracetic acid in solution with hydrogen peroxide and acetic acid, in egg shells intended for the manufacture of *ille flotant* (solution at 2.5 % with 4.5 % of peracetic acid); in peas and green beans for sterilization (500 mg/l of peracetic acid); in starch, potato starch and derivatives (1 kg/tonne); in raw ready-to-eat salads (4th range); in blanched spinach, aromatic herbs and unbleached leeks for freezing (75 mg/l of peracetic acid) and wheat prior to milling (3 l of a solution based on 15 % peracetic acid and 23 % hydrogen peroxide per tonne of wheat) and dehydrated legumes (500 mg/l of peracetic acid)	France (Arrêté, 2006)
	Authorised for the process of washing or peeling fruits and vegetables that are not unprocessed raw material and not exceeding 80 mg/kg in the washing solution	United States (ECFR, 2021b)
	Authorised as a food additive (starch modifying agent)	Canada (DJC, 2021)
	Authorised as processing aid as bleaching agent, for washing and peeling foods, and as a catalyst with maximum permitted level of 0.7 mg/kg	Canada (DJC, 2021)

Table 4. Examples of authorised uses and assessments

Substance	Authorised use/assessment	Country/Reference
1-hydroxyethylidene-1,1-diphosphonic acid (HEDP)	Toxicological assessment favourable for acetic acid, peracetic acid, hydrogen peroxide and HEDP solutions (may also include octanoic and peroxyoctanoic acid) for use in poultry and meat carcasses	(EFSA, 2014)
	Authorised together with peracetic acid for the process of washing or peeling fruits and vegetables that are not unprocessed raw material and not exceeding 4.8 mg/kg in the washed solution	United States (ECFR, 2021b)
	Mixed peracetic acid, acetic acid, hydrogen peroxide, peroxyoctanoic acid and HEDP additive authorised as disinfectant for poultry carcasses, parts, tripe and organs with a maximum peroxyacid concentration of 220 mg/kg as peracetic acid, 110 mg/kg of hydrogen peroxide and 13 mg/kg of HEDP	United States (ECFR, 2021c)
	Authorised for use as a processing aid in water and as a chelating agent in disinfectants for meat, fruit and vegetables, and as a processing aid in water	Australia (ANZFSC, 2021)

2.4 Acceptable Daily Intake

No ADI has been established for hydrogen peroxide, peracetic acid and HEDP as individual components (EFSA, 2021a) (JECFA, 2021a). With regard to acetic acid, it is authorised as a food additive (E 260) with a maximum specific dose as *quantum satis* (EU, 2008).

JECFA has established non-specified ADI for antimicrobial solutions of peroxyacids which may include hydrogen peroxide, acetic acid and peracetic acid, also including HEDP as a stabilizer (JECFA, 2021b). JECFA also considers that in the intended conditions of use for these solutions, the quantities of residue in treated foods are not in any way concerning from a food safety perspective (JECFA, 2004, 2005).

3. Technological function

3.1 Technological use claimed

The applicant claims that the technological use is that of bacterial disinfectant of water used for washing apples and peaches.

3.2 Level of use requested

As mentioned by the applicant, given the reactivity of the active substances with the organic material present in the dirt covering the fruits, an initial addition of 0.1 % of FreshStart Disinfect 25-15 or 0.3 % of FreshStart Disinfect 25-5 is required. In both cases, the final concentration of peracetic acid in the washing solution shall be 150 ppm. After this initial dosage of 150 ppm of peracetic acid, maintenance dosages (0.033 % of FreshStart Disinfect 25-15 or 0.1 % of FreshStart Disinfect 25-5) shall be given in order to maintain the washing solution at a peracetic acid concentration of 50 ppm.

The washing solution shall be reused for days or weeks and, in the case of washing by immersion (drums), it will be left to circulate for at least 45 seconds before washing the apples and peaches, with a contact time of 90 seconds. With regard to washing by spraying (showers), the contact time shall be 30 seconds. After washing, a final rinsing of the apples and peaches shall be performed using potable water.

3.3 Justification of use, interest and efficacy

As indicated in the assessments of similar products, the first post-harvest treatment of plant products is the washing, it being essential to maintain the hygiene of the washing solution, as it recirculates, and therefore, dirt from the harvesting passes into the solution, as well as microorganisms deposited on plant material. This leads to the accumulation of contamination, which increases on each recirculation. To prevent the washing solution from becoming a vector of propagation of microorganisms due to cross-contamination, it must be ensured that the microbiological quality is retained, using disinfectant products for that purpose while ensuring that the degradation products and residue of the antimicrobial agent used do not constitute a risk to the health of the consumer or to the environment (AESAN, 2020).

3.3.1 Efficacy trials

The applicant has submitted the details of two efficacy trials conducted on apples and peaches, taking into account the microbiological parameters established in Royal Decree 140/2003 establishing the health criteria of water quality for human consumption (*Escherichia coli*, *Enterococcus* and *Clostridium perfringens*) (BOE, 2003). The trials were conducted with the replenishment dose of FreshStart Disinfect 25-15, as it is the processing aid with the same final concentration of peracetic acid (50 ppm) in the washing solutions and the lowest concentration of hydrogen peroxide. Likewise, different modes of application were used emulating both application by immersion and by spraying.

The results obtained showed reductions of 100 % of the added inoculates (at levels of 104 cfu/ml) of *Escherichia coli, Enterococcus faecalis* and *Clostridium perfringens* in the washing solutions, after submerging apples for 90 seconds or after spraying peaches (150 ml of solution/kg of fruit).

3.4 Description of the process

3.4.1 Forms of incorporating the processing aid

The incorporation of the processing aid takes place during the washing of apples and peaches upon arrival at the processing plants, using the spray or the drum washing systems. The processing aid is added by an automatic dispenser, with an initial dose added so that the concentration of peracetic acid in the washing solution is 150 ppm, and subsequently replenishing the amount is required to maintain a peracetic acid concentration of 50 ppm. Additionally, eventual controls of the concentration of peracetic acid are conducted using a spectrometer or reactive strips.

The contact time of the washing solution for apples and peaches is 90 seconds in the case of washing in drums and 30 seconds for spraying.

3.4.2 Identification of phases of elimination of the processing aid

In the case of active substances, it is expected that their presence in fruits is negligible given that these substances decompose rapidly, giving rise to acetic acid, water and oxygen.

As the applicant indicates, both the hydrogen peroxide and the peracetic acid in the solution are unstable, especially in the presence of oxidable organic material. Hydrogen peroxide decomposes

into water and oxygen and peracetic acid decomposes into acetic acid and water.

The applicant also states that the fruit is subjected to a final rinsing with drinking water for the purpose of removing any potential water-soluble residue from its surface.

The results of peracetic acid and HEDP trials conducted on the washing and rinsing solutions for apples and peaches are displayed here.

4. Residue studies

The results of two studies conducted by an independent laboratory to determine the residues of peracetic acid and HEDP were submitted. The trials were conducted with FreshStart Disinfect 25-5, as it is the processing aid with the same final concentration of peracetic acid (150 ppm) as in the washing solutions, and with the highest concentration of HEDP (15 ppm).

Duplicate samples were taken of the washing solution for apples and peaches, corresponding to three phases of the process:

- Pre-treatment solution: sample taken once the processing aid was added and prior to the processing.
- Post-treatment solution: sample taken after washing by immersion.
- Post-rinsing water: sample taken after final rinsing with drinking water.

The analyses of peracetic acid were conducted by nuclear magnetic resonance (1HNMR) with limit of detection and limit of quantification of 8 mg/l and 25 mg/l, respectively (Table 5).

Table 5. Contents of peracetic acid (mg/l) in the washing solutions and post-rinsing water

Samples	Pre-treatment solution	Post-treatment solution	Water post-rinsing
Apples	82.6	<8	<8
	81.7	<8	<8
Peaches	53.9	<8	<8
	52.8	<8	<8

The peracetic acid residues present in the post-rinsing water were below the limit of detection, proving the rapid decomposition of peracetic acid.

In relation to the possible presence of HEDP residues, the analyses were conducted via Nuclear Magnetic Resonance (^{31}PNMR) with limit of detection and limit of quantification of 1.5 mg/l and 4 mg/l, respectively.

No residues of HEDP were detected after final rinsing with drinking water (post-rinsing water). Nevertheless, it is noted that HEDP was not detected in the initial (pre-treatment) solutions either, prior to washing the apples and peaches, where the concentration should be 15 ppm. The applicant indicates that this may be due to the degradation of HEDP in contact with peracetic acid and acetic acid. In this regard, it must be pointed out that in other assessments of similar solutions conducted by the Scientific Committee, HEDP was detected and quantified.

Given that it is considered that the HEDP analysis results presented are not adequate, as it was not detected in the pre-treatment solutions, a theoretical estimate was made for the maximum quantities of HEDP in apples and peaches, considering the worst case scenario, that is assuming that the post-treatment solution contains the same concentration of HEDP as the pre-treatment solution (15 ppm) (with no degradation, evaporation, etc.) and the rinsing water applied subsequently does not eliminate the HEDP that could remain on the surface of the apples and peaches.

This situation implies an overestimation of the possible residue, and to calculate it, it is considered that the immersed fruit retains 0.007 l of solution/kg while spraying increases it to 0.15 l/kg of fruit. Assuming an HEDP concentration of 15 ppm in the post-treatment solution and the use of 0.007 l solution/kg for immersion and 0.15 l solution/kg for spraying, the estimated HEDP residues would be 0.105 mg HEDP/kg of fruit and 2.25 mg HEDP/kg of fruit, respectively.

5. Study and data relating to the innocuousness of HEDP

Given that there is no ADI established for HEDP, the risk assessment is based on the determination of the Margin of Safety (MOS), considering that where the MOS >100, there is no risk to the consumer. The MOS is calculated taking into account the No Observed Adverse Effect Level (NOAEL) and the Estimated Daily Intake (EDI).

In the case of HEDP, several studies have been carried out on the toxicity establishing different NOAELs (EFSA, 2014). Following the same criteria as EFSA, for the calculation of the MOS, a NOAEL of 50 mg/kg b.w./day will be used, as established based on studies conducted on rats and rabbits.

6. Study of consumption and assessment of the anticipated intake level of HEDP by the consumer

To estimate exposure, the data for the European Union Member State with the highest chronic consumption (mean and 95th percentile consumers only) of apples (sum of apples and apple juice) and peaches (sum of peaches and peach juice) for adults, infants (below the age of 1) and toddlers (12 to 35 months), was taken into account, in accordance with the Comprehensive European Food Consumption Database of EFSA (2021b) (data updated as of July 2021).

In the case of adults, the highest consumption of apples (resulting from the sum of the consumption of apples and apple juice, data from Germany) is 8.05 and 22.97 g/kg b.w./day for the mean and 95th percentile consumers respectively, whereas for peaches (the sum of the consumption of peaches and peach juice, data from Estonia) the highest consumptions are 4.45 and 9.20 g/kg b.w./day for the mean and 95th percentile consumers, respectively. With reference to children below the age of 1, the highest consumptions of apples are 13.58 and 40.53 g/kg b.w./day (mean and 95th percentile, for the sum of apples and apple juice, data from Estonia), while the highest consumptions of peaches correspond to 11.76 and 29.30 g/kg b.w./day (mean and 95th, for the sum of peaches and peach juice, data from Bulgaria). Following the same criteria, for children between 12 and 35 months, consumptions of 16.16 and 50.24 g/kg b.w./day are obtained for apples (data from Estonia) and 16.19 and 34.79 g/kg b.w./day in the case of peaches (data from Spain).

Additionally, considering the estimated HEDP residues (2.25 mg HEDP/kg fruit being the worst case scenario), we obtain the estimated daily intake (EDI). The Margin of Safety (MOS) (Table 6) is calculated on the basis of the estimated daily intake and the NOAEL (50 mg HEDP/kg b.w./day).

Table 6. Estimate of HEDP exposure and MOS calculation considering residues of 2.25 HEDP/kg of fruit

Population	Fruit	Consumption (g/kg b.w./day)		EDI (mg HEDP/kg b.w./day)	MOS
Adults	Apples	Mean	8.05	0.018	2761
		P95	22.97	0.052	967
	Peaches	Mean	4.45	0.010	4994
		P95	9.20	0.021	2415
Infants (<1 year)	Apples	Mean	13.58	0.031	1636
		P95	40.53	0.091	548
	Peaches	Mean	11.76	0.026	1890
		P95	29.30	0.066	758
Toddlers (12-35 months)	Apples	Mean	16.16	0.036	1375
		P95	50.24	0.113	442
	Peaches	Mean	16.19	0.036	1373
		P95	34.79	0.078	639

An alternative scenario proposed is the possibility of considering that the process of rinsing with drinking water practically eliminates HEDP residue present in fruit, given its high solubility, such that the concentration of estimated residues in fruits would be 0.225 mg/kg, by considering the limit of detection (1.5 mg/l) as the HEDP concentration present in the post-treatment solution. Taking into consideration this premise and the previously cited consumption data, the exposure estimate and MOS calculation is performed (Table 7).

Table 7. Estimate of HEDP exposure and MOS calculation considering residues of 0.225 mg HEDP/kg of fruit

Population	Fruit	Consumption (g/kg b.w./day)		EDI (mg HEDP/kg b.w./day)	MOS
Adults	Apples	Mean	8.05	0.002	27 605
		P95	22.97	0.005	9674
	Peaches	Mean	4.45	0.001	49 938
		P95	9.20	0.002	24 155
Infants (<1 year)	Apples	Mean	13.58	0.003	16 364
		P95	40.53	0.009	5483
	Peaches	Mean	11.76	0.003	18 896
		P95	29.30	0.007	7584
Toddlers (12-35 months)	Apples	Mean	16.16	0.004	13 751
		P95	50.24	0.011	4423
	Peaches	Mean	16.19	0.004	13 726
		P95	34.79	0.008	6388

Although, initially, we may consider the scenario that is the closest to reality to be the result of estimating that the post-rinsing water would eliminate most of the HEDP residue in fruit (Table 7), we cannot discard other scenarios (Table 6), given the absence of corroborating analysis data.

Conclusions of the Scientific Committee

The Scientific Committee, having assessed the request for use of the aqueous solutions with the commercial names FreshStart Disinfect 25-15 and FreshStart Disinfect 25-5 as processing aids in the process of bacterial disinfection of water used for washing of apples and peaches upon arrival at processing plants, based on the information provided by the applicant and taking into consideration the composition and conditions of use proposed, deems that the use of the processing aid does not pose any risk to consumer health.

The conclusions of this report refer exclusively to the solutions subject to assessment as processing aids under the conditions of use proposed and their current composition, both in terms of their active components and their stabilizers and cannot be extrapolated to any formulations or conditions other than those assessed herein. One must bear in mind that the kg of fruit processed, the climate conditions or dirt may influence the concentrations of the components of the processing aids in the washing solutions and therefore, their eventual residues.

This assessment does not constitute an authorisation for use nor does it affect uses other than its use as a processing aid for the bacterial disinfection of water used to wash apple and peaches upon arrival at the processing plants. This use involves a final rinsing with drinking water, subsequent to the application of rinsing water with the processing aid, in order to eliminate any possible residues on apples and peaches.

The products thus processed, must comply with all applicable food safety legislation and, once on the market, the operator must ensure the absence of undesirable contaminants, residues or micro-organisms or ensure that their presence remains under the established maximum limits.

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