Efficacy of 2000 ppm PAA (Perasan MP-2®) On *Salmonella* Heidelberg-Inoculated Chicken Halves

Background

The contamination of food products by pathogenic organisms such as *Salmonella spp.* is an on-going problem that is addressed within the processing plant using antimicrobial products. *Salmonella enterica enterica* is a subspecies of *Salmonella enterica.* *Salmonella* Heidelberg is an antibiotic resistant, pathogenic serovar belonging to this subspecies and has recently been implicated in a recent outbreak of Salmonellosis traced to poultry products.

The use of PAA is becoming popular for the secondary processing of poultry products, as it applies a much higher dose to the whole birds, or parts and pieces, skin on or off and organs to aid in pathogenic and microbial kill where parts and pieces are often co-mingled from various birds. This study is designed to determine the efficacy of 2,000 ppm peroxyacetic acid (PAA) from Perasan MP-2® against *Salmonella* Heidelberg inoculated chicken halves at a short contact time. The level of planktonic *Salmonella* Heidelberg will also be measured.

Experiment Conditions

Part 1- *Salmonella* Heidelberg (Planktonic):

a) Control, 5 minutes: 100 ml inoculum in 20 L city water

b) PAA, 5 minutes: 100 ml inoculum in 20 L of 2000 ppm PAA (from Perasan MP-2)

Part 2- *Salmonella* Heidelberg (Sessile):

a) Control, 5 minutes: Ten chicken halves- city water

b) PAA, 5 minutes: Ten chicken halves-2000 ppm (from Perasan MP-2)

Note- Both Part 1 and Part 2 of this study were conducted at the same time, i.e., 10 of the *Salmonella* Heidelberg inoculated chicken halves were placed in each of the 20 L inoculated bins prior to treatment.

The Food Contact Substances

The MP-2 used to prepare the test solutions containing PAA was made from Perasan MP-2. Perasan MP-2 is a product that contains 15% peroxyacetic acid, 5.5% hydrogen peroxide, and 0.7% HEDP (hydroxyethylidene diphosphonic acid). The FCS has been issued prior approvals in the form of FCN's #887, #908 and #1132 for uses on meat and poultry.

Methods

Test System:
Salmonella Heidelberg bacteria (ATCC 8326)

Salmonella Heidelberg bacteria were cultured in nutrient broth (Sigma, St. Louis, MS, lot number 095K0035) by incubation for two days at 35^{o} C. The bacteria were separated from the nutrient broth by centrifugation, and carefully re-suspended in approximately 800 mL of sterile phosphate buffer, which was later used to inoculate the test materials. The concentration of the *Salmonella* Heidelberg was measured in the inoculum by plating using 3M Petrifilms Enterobacteriaceae Plates which were then incubated at 35°C for 24 hours, upon which the duplicate plates were enumerated.

Ten whole, uncooked chickens were purchased from a local grocer. Each chicken was cut evenly into two halves after the removal of internal organs. This resulted in a total of 20 chicken halves.

100 mL of the *Salmonella* Heidelberg inoculum was added to each of two 30 gallon plastic storage bins containing 20 liters of chilled water.

The twenty chicken halves were patted dry with a towel, randomly split into groups of ten, and sprayed with the remaining *Salmonella* inoculum.

Each set of ten chicken halves was placed in 30 gallon plastic storage bins containing 20 liters of the chilled water and *Salmonella* inoculum. Immediately thereafter, a calculated amount of PAA was added to the appropriate bin. The actual concentration of PAA was measured using the Modified DPD method. Separately, one bin containing 20 liters of the chilled water and *Salmonella* inoculum was designated as the control (no antimicrobial added) and also had a 5 minute contact time. All sets of control and challenge chicken halves were agitated using gloved hands to simulate the movement through a typical chiller during the 5 minute contact time.

After the challenge testing of *Salmonella*, at the time allotted, the plastic bin containing the PAA solution was treated with 90.6g of erythorbic acid to neutralize any remaining PAA and H_2O_2 oxidant. After neutralization, a sample of the water was removed and plated for planktonic *Salmonella* using 3M Petrifilm Enterobacteriaceae Plates. The chicken halves were then removed and gently shaken three times to remove excess liquid and returned to a new, sterile plastic bag. 200 g of city water was introduced to each bag and subsequently tumbled gently for one minute to dislodge remaining sessile *Salmonella* bacteria. The water remaining at the bottom of the bag was plated in duplicate using 3M Petrifilm

Enterobacteriaceae Plates. All plates were incubated at 35°C for 24 hours, upon which the plates were enumerated.

Results and Discussion

The *Salmonella* inoculum used to inoculate the 20 chicken halves yielded a *Salmonella* Heidelberg population of 8.82×10^8 or $\log_{10}$ 8.82.

The actual concentrations of the test solutions were measured by DPD method immediately after adding the test chemicals. The concentration was 2157ppm PAA from Perasan MP-2®.

Part 1- *Salmonella* Heidelberg (Planktonic)
Table 1 demonstrates the average number of planktonic *Salmonella* bacteria present in the test solutions containing the 10 chicken halves after the five minute contact time compared to the control. The $\log_{10}$ reduction in *Salmonella* Heidelberg compared to the control was >7.54 (>99.9999%).

Table 1: Planktonic *Salmonella* bacteria 5 minute microbiological results

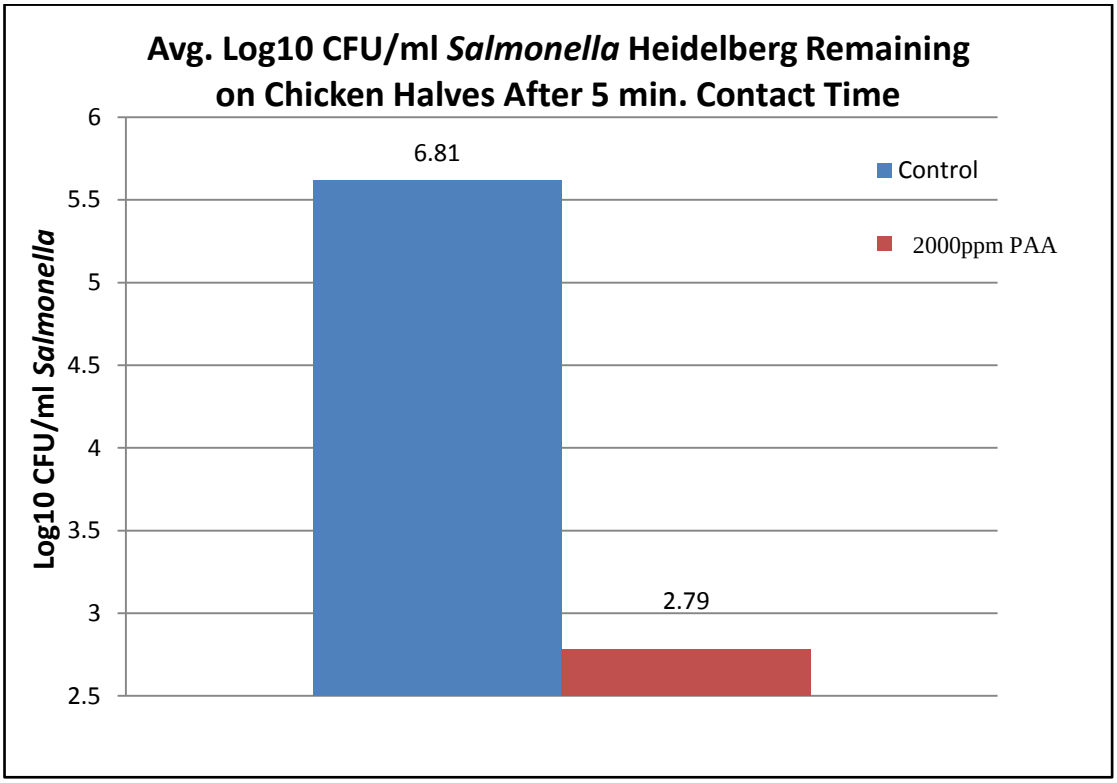
Description	Log10 CFU/ml Salmonella (average)	Log 10 Reduction	Percent Reduction
Control	7.54	N/A	N/A
PAA (MP-2)	0	>7.54	>99.9999%

Part 2- *Salmonella* Heidelberg (Sessile)
Table 2 and Figure 1 demonstrate the average number of bacteria left on the chicken after being submerged for five minutes in chilled city water (control) or PAA-challenge solution. It can be seen that the control averaged a $\log_{10}$ of 6.81 CFU/ml. The $\log_{10}$ reduction in *Salmonella* Heidelberg bacteria on chicken halves submerged in a ~2000 ppm PAA solution compared to the control was 4.02.

Table 2: Sessile *Salmonella* bacteria microbiological results

Description	Log10 CFU/ml Salmonella (average)	Log 10 Reduction	Percent Reduction
Control	6.81	N/A	N/A
PAA (MP-2)	2.79	4.02	99.99

Figure 1:



Statistical analysis was completed to evaluate the data associated with the Control and PAA treated Salmonella inoculated chicken halves. The statistical analysis was run with an alpha of 0.1%, meaning that there is 99.9% confidence in difference between the treatments to make a conclusion. The F-value necessary to conclude a difference in these treatments at 99.9% confidence is 15.38 ("F-crit"), and the value calculated from the ANOVA is 4,098 ("F"). The p-value is almost zero within the precision of the calculation such that there is 99.99999+% confidence in difference between treatments.

Table 3: Statistical analysis comparing the Control and PAA treated *Salmonella* inoculated chicken halves.

Summary

Groups	*Sample size*	*Sum*	*Mean*	*Variance*
Control	10	68.26302	6.82630	0.01392
PAA	10	27.94110	2.79411	0.02575

ANOVA

Source of Variation	*SS*	*df*	*MS*	*F*	*p-level*	*F crit*
Between Groups	81.29287	1	81.29287	4,098.43682	0.00000E+0	15.37931
Within Groups	0.35703	18	0.01984			
Total	81.64990	19				

Conclusions:

- Because of the recent outbreak of the antibiotic resistant *Salmonella Heidelberg* linked to poultry, it was decided to perform efficacy testing against this particular strain. This study was designed to give the reader a reasonably accurate idea of the relative efficacy of peroxyacetic acid (from Perasan MP-2®) against *Salmonella Heidelberg* bacteria in solution and on chicken when submerged for 5 minutes.

- The planktonic *Salmonella Heidelberg* present in solution was eradicated using 2157 ppm PAA after the five minute contact time compared to the control.

- This study also indicates that PAA at approximately 2157ppm provided a 4 log reduction of sessile *Salmonella Heidelberg.*

- The PAA challenge bleached the chicken carcass during the 5 minute test. However, about 10-15 minutes after the conclusion of the PAA challenge test about half the normal yellow color of the carcass returned to normal. It is suggested that concentrations of the PAA approaching 2000 ppm be limited to shorter contact times than used herein to avoid bleaching of the poultry carcass.

12-16-2013
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Attachment #11
Environmental Assessment

1. **Date:** December 20, 2013

2. **Submitter:** Enviro Tech Chemical Services, Inc.

3. **Address:** 500 Winmoore Way, Modesto, CA. 95358

4. **Description of Proposed Action:**

   a. The FCS proposed in the Food Contact Notification is composed of peroxyacetic acid, hydrogen peroxide, acetic acid, HEDP, and (optionally) sulfuric acid for microbiological control in process water during the production and preparation of whole birds, parts and pieces, skin on or off and organs. Maximum concentration of the FCS is 2,000 ppm as peroxyacetic acid, 770 ppm as H_2O_2, and 100 ppm as HEDP.

   b. The FCS is requested for use in pre-air chiller poultry processing dip tanks and in secondary commercial processing of whole birds, parts and pieces, skin on or off and organs using spray and dip applications. Secondary commercial processing is defined as post-main chiller (air or water) spray or dip applications until packaging the poultry products.

5. **Identification of Substance:**

   The FCS is a liquid equilibrium mixture of peroxyacetic acid, hydrogen peroxide and acetic acid. It is made by blending acetic acid, hydrogen peroxide, RO water, and HEDP as a transition metal stabilizer. Sulfuric acid is optionally added in winter time to aid in the speed of the reaction process.

   Ingredients: (note pg. 4 of FDA Form 3480):

Acetic acid	CAS # 64-19-7
Hydrogen Peroxide	CAS # 7722-84-1
HEDP	CAS # 2809-21-4
Sulfuric acid	CAS # 7664-93-9
Purified water	CAS # 7732-18-5

   The basic reaction by the above combination is as follows:

$CH_3CO_2H + H_2O_2 = CH_3CO_3H + H_2O$ (molecular weight is 76 gms/mole)

6. Introduction of Substance into the Environment:

a. The FCS is currently manufactured in an EPA approved facility (EPA Establishment Number 63838-CA-01) at the address listed above, and no unusual or factual threat to the environment exists. No extraordinary environmental circumstances would apply to the continued on-going manufacture of the FCS.

b. The FCS is proposed for use in water as an antimicrobial agent during the commercial processing and storage of poultry products. In these types of facilities the predominant type of discharge would be to an on-site pretreatment facility that would discharge to a local sewer treatment facility.

c. For land-based operations the balance of the process water including the FCS would be discharged to the local municipal waste treatment or on-site pretreatment facility, whereas the peroxygen components of the FCS would have a very short half-life (less than hours[1, 10]). The FCS substance, if accidentally discharged or released as over-flow from the process area, would be directed to the food plant wastewater discharge system. Treatment of the FCS in this method would represent a 99.4% degradation of the peroxyacetic acid, hydrogen peroxide and acetic acid into their degradation products carbon dioxide, water, oxygen, sulfate and acetic acid[2, 3]. The active components and HEDP stabilizer in the formulation would subsequently be diluted proportional to the combined wastewater discharge, which would not present an environmental concern.

Estimates for uses:

A. The proposed use of the FCS is intended as an antimicrobial water treatment in water that is used to treat poultry in pre-air chiller dip tanks and during secondary processing of whole birds, parts and pieces, skin on or off and organs prior to packaging. This applicant has asked for a limit of 2000 ppm of peroxyacetic

acid (PAA) as the upper limit. We know of some cases where the FCS will be used successfully at up to the maximum concentration utilizing short contact times[23]. Undoubtedly the FCS may be used at somewhat lower concentrations during secondary processing of poultry and in pre-air chiller dip tanks, but the focus on this new use shall be at the highest limits requested in this application.

i) The FCS will be used in both spray and dip water treatment applications of whole birds, parts and pieces, skin on or off and organs. The processing of whole birds, parts and pieces, skin on or off and organs following the exit of the poultry from the main chiller (air or water) until packaging may be described as the secondary poultry treatment process. Secondary processing may occur within the same poultry processing plant or the parts may be transported to a separate poultry processing plant for processing. The use of this FCS is becoming popular for the secondary processing of poultry products, as it applies a much higher dose to the whole birds, parts and pieces, skin on or off and organs to aid in pathogenic and microbial kill where parts and pieces are often co-mingled from various birds using short exposure contact times.

ii) In the first described application of the FCS there up to nine individual poultry sprays that are applied during secondary processing of whole birds, poultry parts and pieces. The FCS sprays are often used to remove microbial contamination prior to and after the mechanical processing systems that debone, desinew or defat poultry parts and pieces. Mechanical systems such as a POSS system are often used for deboning, desinewing and/or defatting poultry parts and pieces. FCS treated water flows from each of the possible nine spray nozzles at approximately 1 gallon per minute (gpm) and the mechanical sprayers are used for up to 16 hrs/day in many facilities. Secondary poultry processing operations may run continuously up to 16 hrs/day which is similar to primary poultry processing operations for whole birds where a finishing chiller operates for up to 16 hrs/day. The total FCS spray water treatment usage rate per day is as follows:

9 spray nozzles x 1gal/min FCS x 60min/hr = 540 gal/hr FCS
540 gal/hr FCS x 16 hrs/day = 8640 gal/day FCS

iii) The second described application of the FCS involves use in pre-air chiller dip tanks and in secondary processing dip tanks for whole birds and organs (such as giblets), respectively. The FCS treated water used in pre-air chiller dip tanks contains approximately 400 gallons of the FCS. The 400 gallon dip tank requires make up water at the rate of up to 10% per hour for a total of 40 gallons per hour for this FCS application. In the secondary processing of poultry organs such as giblets, the giblets are mechanically removed from the whole bird and dipped in a separate 400 gallon dip tank filled with the FCS to reduce microbial contamination. Similar to the pre-air chiller dip tank, the secondary processing dip tank requires make up water at a rate of up to 10% per hour and operates for up to 16 hrs/day. The total FCS dip tank water treatment usage rate per day is as follows:

(40 gal/hr FCS (make up water) x 2 tanks) x 16 hrs/day = 1280 gal/day FCS
(400 gal FCS (dip tank volume) x 2 tanks) + 1280 gal/day = 2080 gal/day FCS

iv) The FDA has examined processing water dilution factors (DF) at poultry processing plants and found that 71% of facilities had DF's >100, and 96% had a DF of 20 or greater[(11)]. In a letter dated October 27, 2010, page 4 (g), the Agency noted "…the Agency generally allows a dilution factor (DF) for the EIC to be 10 to obtain the EEC in receiving water[(24)]…"

This applicant will use the figures of 10,720 gal per day water used (8,640 gallons for sprays plus 2,080 gallons for dip tank application) for secondary processing of whole birds, or parts and pieces, skin on or off and organs in an average 200,000 bird/day processing plant. As previously stated in this applicant's FCN 1132 application to the Agency, the water used for poultry spray and chiller operations was calculated to be 125,000 gal per day for a 200,000 bird/day processing plant

(per Attachment #14). It should be understood that every plant uses a considerable amount of water for other non-processing purposes, such as cleaning, spraying, lubricating, bathroom uses and boiler operations, to name a few. It is estimated that the average poultry plant consumes a total of 26.0 L/bird[(9)]. This total consumption is: 26L/3.784 *(number of liters per gal)* = 6.871036 gal/bird x 200,000 birds/day = 1,374,207 gal/day. Therefore, the Dilution Factor (DF) for an average poultry plant becomes 1,374,207 gal/day ÷ (125,000 gal + 10,720 gal) = 10.13. This agrees quite well with the estimate given in A (iii) above.

Using the FCS at its target dilution rate (PAA 2000ppm, 770 ppm H_2O_2 and 100 ppm as HEDP), an environmental introduction concentration (EIC) can be calculated by multiplying the starting concentration of the compound by the estimated percentage of degradation associated with use of the compound in a secondary poultry processing dip or spray application to yield an estimate of the EIC. In commercial practice, water containing PAA, H_2O_2 and HEDP (up to 2000ppm, 770ppm and 100ppm respectively), is not redirected into any other application or re-used anywhere within the poultry processing plant so it flows only to a primary wastewater treatment facility. To determine the estimated percentage degradation associated with PAA and H_2O_2 in a secondary poultry processing application, a study on Perasan MP-2 was conducted (see Attachment #7). In the Perasan MP-2 study, a 1000ppm PAA and 250ppm H_2O_2 solution is introduced to chicken halves in a simulated bath where over 99% of the PAA degraded on contact with chicken halves over a period of 3 hours and all of the detectable H_2O_2 degraded after just 10 minutes. This study serves as a reference point for the decay percentage for both PAA and H_2O_2 in a dip or spray application of the FCS. No HEDP degradation was assumed for this calculation. The EIC is calculated for 1 hour to be conservative as follows:

2000ppm PAA x 10% compound remaining (ppm) = 200ppm PAA EIC
770ppm H_2O_2 x 0% compound remaining (ppm) = 0ppm H_2O_2 EIC
100ppm HEDP x 100% compound remaining (ppm) = 100ppm HEDP EIC

The effective environmental concentration (EEC) for PAA at 2000ppm, H_2O_2 at 770ppm and HEDP at 100ppm can be calculated by starting with the EIC for each compound, then dividing by a DF of 10 per A (iii). This calculation does not take into account additional time that it takes for movement of water containing PAA, H_2O_2 and HEDP to flow to a primary wastewater treatment facility which would increase the rate of decay. PAA and H_2O_2 are not expected to survive treatment at a primary wastewater treatment facility due to their reactivity and pH sensitivity [(1)]. However, when considering just the DF of 10, the EEC for PAA may be calculated as follows:

200ppm PAA ÷ 10 (DF) = 20ppm PAA EEC
The EEC for H_2O_2 will be 0ppm per the EIC.
100ppm HEDP ÷ 10 (DF) = 10ppm HEDP EEC

On January 27, 2012, the USDA Food Safety Inspection Service (FSIS) published a proposed rule entitled "Modernization of Poultry Slaughter Inspection."[(25)] This rule would provide a new, voluntary inspection system for poultry slaughter facilities that would replace current systems, and allow for higher line speeds and more birds processed per minute (max. 175 birds/min). Currently, slaughter establishments process approximately 70-140 birds/min [(25)]. The higher processing rate would be expected to increase chemical use and effluent amounts, potentially altering the environmental impacts related to the use of the chemical. This respondent assumes that the dip and spray application during secondary processing of poultry products will continue to operate 16 hrs/day under the proposed rule similar to primary poultry processing operations. Based on the new maximum processing speed of 175 birds/min., an estimated 250,000 carcasses will be processed per day resulting in a 25% increase in the number of poultry carcasses processed under the FSIS proposed rule. The estimated water use following implementation of this new rule is as follows:

Spray Application: 1.25 (25% increase in number of carcasses processed) x 8640 gal of spray/day = 10,800 gallons/day.

<u>Dip Application:</u> 400 gallons initially in each tank…Make up water is required at the rate of 1280 gallons/day for both tanks combined, and we assume 25% more carcasses and organs are processed per day, which increases the amount of make up water needed by 25%. Thus, 1600 gallons of water are added to the dip tanks combined during the course of the day. This brings the total dip tank water consumption to 2,400 gal for a 250,000 bird/day processing plant.

Under the proposed rule, this applicant will use the figures of 13,200 gal per day water used for secondary processing of poultry products in an average 250,000 bird/day processing plant. This is in comparison to 10,720 gal per day water used for secondary processing of poultry products in an average 200,000 bird/day processing plant. As previously stated in this applicant's FCN 1132 application to the Agency, under the proposed rule, the water used for poultry spray and chiller operations was calculated to be 150,000 gal per day for a 250,000 bird/day processing plant (per Attachment #14). The average water consumption per bird will remain at 26.0L/bird as previously stated. This total consumption is: 26L/3.784 (number of liters per gal) = 6.871036 gal/bird x 250,000 birds/day = 1,717,759 gal/day. Therefore, the Dilution Factor (DF) for an average poultry plant becomes 1,717,759 gal/day ÷ (150,000 gal + 13,200 gal) = 10.53. This DF estimate is in relative proximity to the estimate given in A (iii) above.

When considering the impact of the proposed rule for increasing the poultry processing speed to 175 birds/minute maximum, we do not expect any change to the EIC because the concentration of PAA will be maintained in the current concentration range of 2000ppm. The EIC under the proposed rule would continue to be 200ppm PAA, 0ppm H_2O_2 and 100ppm HEDP. The EEC under the proposed rule can be calculated using the new DF of 10.60 as follows:

200ppm PAA ÷ 10.60 (DF) = 18.9ppm PAA EEC
0ppm H_2O_2 ÷ 10.60 (DF) = 0ppm H_2O_2 EEC
100ppm HEDP ÷ 10.60 (DF) = 9.4ppm HEDP EEC

B. Assuming the FCS is used at its target diluted concentration for the above uses, a maximum anticipated ingredient discharge of the FCS would thus be: (assumptions are the FCS contains 15.5% PAA, 5.5% H_2O_2 and 0.75% HEDP by weight of the as-is FCS):

i) **Poultry:**

10,720 gal x 8.34 lbs (wt of water) = 89,405 lbs
89,405 lbs water requires 44.70 lbs active PAA @ 2000 ppm active
44.70 lbs active / 0.155% = 288 lbs of the FCS
288 lbs FCS x 0.0075 (HEDP activity by wt) = 2.16 lbs HEDP

Under the USDA Food Safety Inspection Service (FSIS) proposed rule entitled "Modernization of Poultry Slaughter Inspection,"

13,200 gal x 8.34 lbs (wt of water) = 110,088 lbs
110,088 lbs water requires 55.04 lbs active PAA @ 2000 ppm active
55.04 lbs active / 0.155% = 355 lbs of the FCS
355 lbs FCS x 0.0075 (HEDP activity by wt) = 2.66 lbs HEDP

At 10,720 gal per day process discharge and a DF (dilution factor) of 10.13, this equals 108,594 gal/day total discharge. The wt of water is 8.34 lbs/gal, so the total discharge per day would be approximately 905,674 lbs. 905,674 lbs ÷ 2.16 lbs HEDP equals a 419,294:1 dilution. The net result is 2.38 ppm of HEDP (1 mil ÷ 419,294). Assuming the HEDP was released to the wastewater treatment facility, and also assuming that all wastewater is treated, and that 80% of the HEDP is removed from the water via adsorption[9, 12], the expected environmental concentration (EEC) in surface waters is then 0.238 ppm, depending on one's DF. Additionally, resultant wastewater sludge may be land applied. However, due to the FCS's projected low end-use level, compared to concentrations where terrestrial toxicity is expected (1000 mg/kg soil dry weight), no environmental toxicity would be expected to occur[8].

7. Fate of the Substance in the Environment:

It is well documented and accepted in the scientific community that PAA and HP are short lived in the environment, do not bioaccumulate, have innocuous degradation byproducts, and are of no toxicological or ecotoxicity concern[1, 2, 3].

The HEDP biodegrades into carbon dioxide, water, and simple orthophosphate[8].

Peroxyacetic acid and hydrogen peroxide are not expected to survive treatment at the primary wastewater treatment facility due to their reactivity and pH sensitivity[1]. Both compounds are rapidly degraded on contact with organic matter, transition metals, and upon exposure to sunlight[2, 3]. The half-life of PAA in buffered solution solutions was 63 hrs at pH 7 for a 748 ppm solution, and 48 hrs for a 95 ppm solution, also at pH 7[2].

The half-life of hydrogen peroxide in natural river water ranged from 2.5 days when initial concentrations were 10,000 ppm, and increased to 15.2 days when the concentration decreased to 250 ppm[3]. In filtered lake water the half-life of H_2O_2 (initial concentration 3.4 ug/l) was 8.6 hrs-31 hrs. (page 21 reference #3).

Since PAA and HP rapidly degrade, they will not be introduced into the natural environment in wastewater at toxic levels. Therefore toxicity and fate data should not be required for these compounds. In biodegradation studies of acetic acid, 99% degraded in 7 days under anaerobic conditions[5].

The optional ingredient, sulfuric acid, degrades into sulfate (SO4), which is not a toxicological or environmental concern at the proposed use levels.

Degradation of HEDP phosphonate occurs slowly in sunlight-illuminated river water as shown by loss of chelant titer and the production of orthophosphate. Some species of algae can slowly utilize the phosphorous present in HEDP as a nutrient, and thus degrading the active molecule [6].

In addition, literature reports indicate that HEDP is removed from water and wastewater by classical precipitation treatment with aluminum sulfate or lime[7, 8].

According to HERA[8], HEDP has a very high adsorption rate coefficient in wastewater activated sludge operations, and this rate of removal has been estimated at >90% for secondary-treated wastewater (page 20, HERA), and further proportionate reductions for tertiary treatment[8].

For sea-based wastewater discharges of this FCS, the peroxygen ingredients would decay rapidly[1, 2, 3, 10]. Since a significant amount of species in ocean-based sea life is dependent upon the food-chain, beginning with plankton-like subspecies, bioaccumulation of the HEDP may be a consideration. However, page 27 of HERA states: "the low K_{ow} values (octanol/water partition coefficient) are extremely low and range from –3.4 to –4.4 depending on the type of (phosphonate) product. Tests on …HEDP (EG&G bionomics, 1976c; Sterber and Wierich, 1986) gave BCF values of …<2-18 (Chemstar PAC, 2003)". This confirms that there is no risk of bioaccumulation in the organism and subsequently in the food chain[8] " This applicant cannot find any references citing the half-life of HEDP in seawater, so we will assume it is no less than reported for fresh water. The half-life for HEDP in water was estimated in another risk assessment to be 395 days based on reported average data of 10% degradation over 60 days[8].

8. Environmental Effects of Released Substances:

In the current FCN, the FCS is proposed for use in water used to commercially process poultry. The concentrations proposed are quite diluted, and once the FCS contacts the balance of the site's wastewater, and subsequently further downstream with the main body of discharge/waste water, the pH would be such that the peroxygens PAA and H_2O_2 would degrade rapidly[1, 2, 3]. HEDP would be the most probable candidate for any potential for environmental toxicity.

a. Aquatic Environment

HEDP is a strong chelating agent and can result in adverse effects on environmental organisms by complexation of essential nutrients[8]. For strong chelating agents, it is suggested that two types of NOEC's be determined: an intrinsic NOEC (NOECi) measured with excess nutrients available and an NOEC measured to protect from the chelating effects in natural waters (NOECc)[12]. A realistic NOECc should be determined by testing in natural waters, by predicting metal speciation and algal trace element requirements, and/or using metal speciation modeling programs[12]. However, excess nutrients are expected to be present in industrial wastewater as eutrophication is a well known phenomenon seen in industrial wastewaters from food processing facilities[13, 14, 15].

However, it should be noted that derivations of NOEC's and their intrinsic uncertainties has recently come into light, and their conclusions make the usefulness of the parameter debatable[19], even to the point that it has been recommended that NOECs be abandoned as a consideration[17].

Table 1. Environmental toxicity data for HEDP.[a]

Species	Endpoint[b]	mg/L
Lepomis macrochirus	96 hour LC50	868
Oncorhynchus mykiss	96 hour LC50	360
Cyprinodon variegatus	96 hour LC50	2180
Ictalurus punctatus	96 hour LC50	695
Leuciscus idus melanatus	48 hour LC50	207-350
Daphnia magna	24-48 hour EC50	165-500
Palaemonetes pugio	96 hour EC50	1770
Crassostrea virginica	96 hour EC50	89
Selenastrum capricornutum	96 hour EC50	3
Selenastrum capricornutum	96 hour NOEC	1.3
Algae	96 hour NOEC	0.74
Chlorella vulgaris	48 hour NOEC	≥100
Pseudomonas putida	30 minute NOEC	1000
Oncorhychus mykiss	14 day NOEC	60-180
Daphnia magna	28 day NOEC	10-<12.5
Algae	14 day NOEC	13

[a] All data from Jaworska et al. (2002) and the HERA risk assessment, references 12 and 9.

[b] The median lethal concentration (LC50) is a statistically derived concentration of a substance that can be expected to cause death in 50% of test animals.
The median effects concentration (EC50) is a statistically derived concentration of a substance that can be expected to cause a specified effect in 50% of test animals.

The lowest toxicity endpoints published for algae, *Selenastrum capricornutm*, *Daphnia magna*, and *Crassostrea virginica* are the result of the chelation effect and not the intrinsic toxicity of HEDP [12]. These values are not relevant when excess nutrients are present as expected in food processing wastewaters[14]. This leaves the lowest aquatic toxicity endpoint published by Jaworska et al. at 3 mg/L (96 hr) EC50. The values calculated herein for the current use request of the FCS fall far below these limits.

Eutrophication is a process whereby water bodies, such as lakes, rivers, and streams, receive excess nutrients that stimulate excessive growth of algae and other plant material. This enhanced plant growth can result in low dissolved oxygen, fish kills, and a depletion of desirable flora and fauna. The relevance of this environmental issue is reflected in reports from the Environmental Protection Agency (EPA) stating that, "As much as half of the nation's waters surveyed by states and tribes do not support aquatic life because of excess nutrients"[14]. The main cause of eutrophication in lakes and streams are high levels of nitrogen and phosphorus and phosphates which usually originate from municipal or industrial effluents [13, 14]. Primary industrial point source contributions of phosphorus include dairy, meat, and vegetable processing facilities, indicating that excess phosphates in food processing effluent is a relevant environmental issue[16]. HEDP contains phosphorus and has the potential to contribute to eutrophication.

In 1998, permissible discharge levels for industries ranged from 0.1 – 0.5 mg/L total phosphorus and a goal of 1 mg/L total phosphorus was set in a phosphorus management plan for POTWs in the Upper Mississippi River Basin[13, 16, 17]. Since HEDP is only 30% phosphorus by weight[6], this applicant expects the proposed use of the FCS to contribute only a small percentage of total phosphorus load in wastewater[18]. On the other hand however, food processing effluent released to POTWs and surface waters are typically treated to reduce total phosphorus prior to discharge[15].

b. Terrestrial Environment

HEDP in effluent discharged to land is not expected to have any adverse environmental impact. The process effluent concentration DF of 2.0 mg/L (an EEC of 0.4 ppm) is expected to result in soil concentrations lower than terrestrial toxicity endpoints available for plants, earthworms, and birds[8]. The NOEC for soil-dwelling organisms was 1000 mg/kg soil dry weight, and this includes plants and earthworms[8]. The 14 day median lethal dose (LD50) for birds was greater than 284 mg/kg body weight[8]. Application of the wastewater to land will result in phosphorus concentrations in soil that are a small fraction of total phosphorus concentrations currently found in the environment and used in fertilizers[17, 19].

Runoff of phosphorus into groundwater or surface waters depends on the management practices and site-specific factors. When best management practices (BMP's) developed by the EPA are followed, this applicant believes that land application of wastewater will reduce use of water by recycling water for irrigation and the overall cost of treatment of wastewater.

9. Use of Resources and Energy:

The proposed FCS would not pose any additional burden on existing resources or energy in the manufacture, transport, use or disposal of the FCS above and beyond those already existing, and the proposed use will not create any additional burden on resources or energy.

10. Mitigation Measures:

The proposed FCS is not reasonably expected to result in any new or extraordinary environmental problems that would require mitigation measures of any kind. The FCS is a relatively benign compound that may replace other more toxic compounds in use presently at the use sites. In addition, discharge permits are mandated by the National Pollutant Discharge Elimination System (NPDES), in which all pollutants or components of discharges are reported by the discharger, and each location's discharge permit is then monitored and controlled by each state and region within a state.

11. Alternatives to Proposed Action:

There are no known alternatives to this proposed FCN.

12. List of Preparers:

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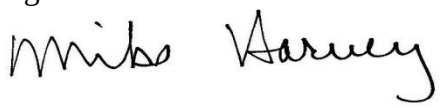
13. Certification:

The undersigned official certifies that the information presented is true, accurate, and complete to the best of the knowledge of Enviro Tech Chemical Services, Inc.

Date: January 6, 2013
Signature: Name and Title: Michael S. Harvey, President



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