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Methodology for modeling the disinfection efficiency of fresh-cut leafy vegetables wash water applied on peracetic acid combined with lactic acid

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ABSTRACT

A methodology to i) assess the feasibility of water disinfection in fresh-cut leafy greens wash water and ii) to compare the disinfectant efficiency of water disinfectants was defined and applied for a combination of peracetic acid (PAA) and lactic acid (LA) and comparison with free chlorine was made. Standardized process water, a watery suspension of iceberg lettuce, was used for the experiments. First, the combination of PAA + LA was evaluated for water recycling. In this case disinfectant was added to standardized process water inoculated with *Escherichia coli* (*E. coli*) O157 (6 log CFU/mL). Regression models were constructed based on the batch inactivation data and validated in industrial process water obtained from fresh-cut leafy green processing plants. The UV_{254}(F) was the best indicator for PAA decay and as such for the *E. coli* O157 inactivation with PAA + LA. The disinfection efficiency of PAA + LA increased with decreasing pH. Furthermore, PAA + LA efficacy was assessed as a process water disinfectant to be used within the washing tank, using a dynamic washing process with continuous influx of *E. coli* O157 and organic matter in the washing tank. The process water contamination in the dynamic process was adequately estimated by the developed model that assumed that knowledge of the disinfectant residual was sufficient to estimate the microbial contamination, regardless the physicochemical load. Based on the obtained results, PAA + LA seems to be better suited than chlorine for disinfecting process wash water with a high organic load but a higher disinfectant residual is necessary due to the slower *E. coli* O157 inactivation kinetics when compared to chlorine.

1. Introduction

Pathogens have been associated with fresh produce, with leafy vegetables estimated to have the highest risk among them, and with the bacterial pathogens *Escherichia coli* (*E. coli*) O157 and *Salmonella* spp. as the most prevalent pathogens on leafy vegetables (Olaimat and Holley, 2012; Tomas-Callejas et al., 2012). Washing fresh-cut produce removes, next to soil and exudates, a part of the produce-associated microorganisms and transfers them to the water. Therefore, pathogen cross-contamination via water can occur when washing fresh produce and the risk of cross-contamination is not removed by using large quantities of water (Holvoet et al., 2012; López-Gálvez et al., 2009). Washing in disinfectant solutions can be done to enhance the removal of microorganisms from the produce, although the main motivation is to avoid cross-contamination via water. In general, chemical oxidants, including peracetic acid (PAA), are much more effective for inactivation of bacterial pathogens in wash water than for removal of these pathogens from fresh produce (Gil et al., 2009; Sapers, 2001). In addition, once cross-contamination has occurred, rewashing the newly infected lettuce in disinfectant solutions proves unable to completely remove the newly attached *E. coli* O157, even shortly after the contamination event (López-Gálvez et al., 2009, 2010; Luo et al., 2011). Therefore, the primary purpose of washing produce in disinfectant solutions seems to be avoiding cross-contamination via wash water. Furthermore, microbial contamination of produce should be avoided as much as possible by respecting good agricultural and manufacturing practices during the production and processing of fresh produce (Holvoet et al., 2012, 2013; Keskinen et al., 2009; López-Gálvez et al., 2010; Sapers, 2001).

PAA has been suggested as an alternative wash water disinfectant for chlorine. It has been intensively studied for use in wastewater disinfection due to its stability in the presence of organic matter and because it does not produce harmful disinfection by-products (DBPs) (Santoro et al., 2007; Stampi et al., 2001). These properties make it attractive for use in fruit and vegetable washing processes, it has been studied as

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disinfectant for washing a variety of fruits and vegetables (González-Aguilar et al., 2012), and it has been commercialized in combination with lactic acid (LA) for washing salads as Fresh Rinse® (Ho et al., 2011).

Water disinfection in fresh-cut industry is carried out in washing tanks (immersion washers), where fresh-cut vegetables are washed, under agitation applied by water, air, sound or mechanical devices (Pao et al., 2012). Alternatively, non-immersion washers that wash produce by spraying or rinsing can be applied but the latter is not the focus of this article. Disinfection processes in this context can be divided as: i) process wash water disinfection in the washing tank and ii) process water recycling outside the washing tank. Process water recycling is defined as inactivation of microorganisms in process water outside the processing line before reuse in the washing process. Process wash water disinfection concerns the inactivation of incoming microorganisms by keeping a disinfectant residual in the washing tank.

The performance of water disinfection in fresh(−cut) produce washing operations will depend on the disinfectant residual (and therefore the disinfectant dose, disinfectant demand and water refreshing rate), the resistance of the target microorganism and the physicochemical conditions of the wash water (organic matter, pH, T) (Van Haute et al., 2015). Mathematical models can be applied to understand the relation between the disinfection efficiency and the influential variables. This knowledge can be used to allow a more calculated approach to the decision making process of implementing a water disinfection technique in fresh(−cut) washing operations.

To study and compare the performance of water disinfectants, an experimental setup that incorporates these factors and that can be applied to study different water disinfectants under similar conditions seems to yield a greater value for industry and governmental agencies than a collection of studies, each with unique and independent experimental setups. In a previous study models were used to understand the relation between chlorine disinfection efficiency and the physicochemical quality, the disinfectant residual and the water refreshing rate in fresh(−cut) produce washing operations (Van Haute et al., 2013). In this study PAA is researched according to a similar experimental setup, both to understand the behavior of PAA in these operations, as well as to compare it with free chlorine.

2. Materials and methods

2.1. Experimental setup modeling

For the process water recycling, inactivation models were calibrated in standardized process water (SPW) with controlled physicochemical parameters and inoculated with *E. coli* O157 (Fig. 1). Both statistical and kinetic models (based on PAA decay) were considered. PAA decay is the decrease in PAA concentration in the water, primarily due to reaction with water matrix constituents. Repetition of the experiments in industrial process water (IPW) was executed for generation of validation data in order to assess the validity of the constructed models. Three replicates of each experiment were performed.

For the process wash water disinfection, a dynamic leafy vegetables washing process was simulated (Fig. 1). The microbial contamination was introduced by continuous in- and outflow of inoculated SPW. The experiment was initiated with tap water, after which a chemical oxygen demand (COD) build-up occurred due to continuous introduction of SPW. Semi-mechanistic models were constructed based on *E. coli* O157 inactivation constants and experimental operational data (water refreshing rate, contamination inflow, disinfectant residual concentration). Models were validated with measured *E. coli* O157 wash water contamination values. The experiment consisted of one trial of 60 min.

2.2. Preparation of standardized process water (SPW)

Two types of standardized process water were produced. For the water recycling experiments, iceberg lettuce (*Lactuca sativa* L.) was purchased from a local wholesale market in Ghent (Belgium) and transported within 15 min to the laboratory, where it was kept at 4 °C before use. Water with high COD was prepared following the procedure described in López-Gálvez et al. (2012), and then was filtered through the filter of a stomacher bag (Seward, UK), in order to separate big solid particles. Afterwards, samples were taken to measure COD and water was kept at 4 °C before use (always the same day as the preparation). Finally, SPW with different levels of COD (500, 800, or 1500 mg/L) was prepared by mixing the adequate volume of high COD water with tap water. In the case of the process wash water disinfection experiments, SPW from spinach (*Spinacia olearacea* L.) was made as described by Gómez-López et al. (2014).

2.3. Collection of industrial process water (IPW)

Wash water from two fresh-cut produce companies was collected into sterile recipient containers and transported under refrigerated conditions to the laboratory, where it was stored at 4 °C for a maximum of 24 h. At company 1, tap water was used as the water source during washing of sugarloaf (*Cichorium intybus*), iceberg lettuce, endive and radicchio. Company 2 utilized borehole water for processing butterhead lettuce, iceberg lettuce, endive, and radicchio.

2.4. Bacterial inoculation

Two attenuated (non-verotoxin producing) nalidixic acid resistant *E. coli* O157 strains (LFMFP 662 and 679) were used. The strains were grown at 37 °C for 24 h in Brain Heart Infusion (Oxoid, United Kingdom) containing 50 μg/mL nalidixic acid (Sigma-Aldrich, Belgium). LFMFP 662 is a nalidixic acid-resistant version of the strain CECT 5947 provided by the Hibro Group from the University of Cordoba (Spain), while LFMFP 679 is a nalidixic acid-resistant version of the strain MB3885 provided by the Technology and Food Science Unit from ILVO (Belgium). A cocktail was made by combining volumes of individual strains. Cocktails were centrifuged at 4 °C, 1800 *g* for 10 min. The pellets were washed twice in phosphate buffer (pH 7), with intermittent centrifugation, and subsequently resuspended in phosphate buffer.

2.5. Disinfection treatments

Disinfectant solutions consisted of a combination of PAA (Chriox 5, Christeyns NV, Belgium) and LA (Purac Biochem, The Netherlands). PAA + LA solutions were used in a mass ratio of 1:40 in all experiments (Ho et al., 2011).

2.5.1. Process water recycling

The two different types of SPW and the IPW from two fresh-cut produce companies were inoculated with the *E. coli* O157 cocktail to a level of approximately 6 log CFU/mL just before the beginning of the treatment. The SPW was continuously stirred during the experiment. Disinfectant solution was added to obtain the desired PAA and LA concentrations, and samples for microbiological analysis were taken periodically. All water recycling experiments were performed in triplicate at 5 °C. To assess the influence of pH on *E. coli* O157 inactivation, inactivation in oxidant demand free buffer was executed in the same way as described for the water recycling experiments. The acid dissociation constant of PAA is 8.2 (Kitis, 2004). Buffer solutions at pH 6, pH 8.2 (consisting both of phosphate buffer 0.07 M) and pH 10.2 (carbonate buffer 0.1 M) were used to manipulate the acid dissociation of PAA to 1%, 50% and 99% respectively.

2.5.2. Process wash water disinfection

Disinfection experiments were performed using a pilot plant system that has been used as a standard dynamic system in previous studies (Gómez-López et al., 2014). Process wash water disinfection treatments were performed starting with clean potable water and applying a

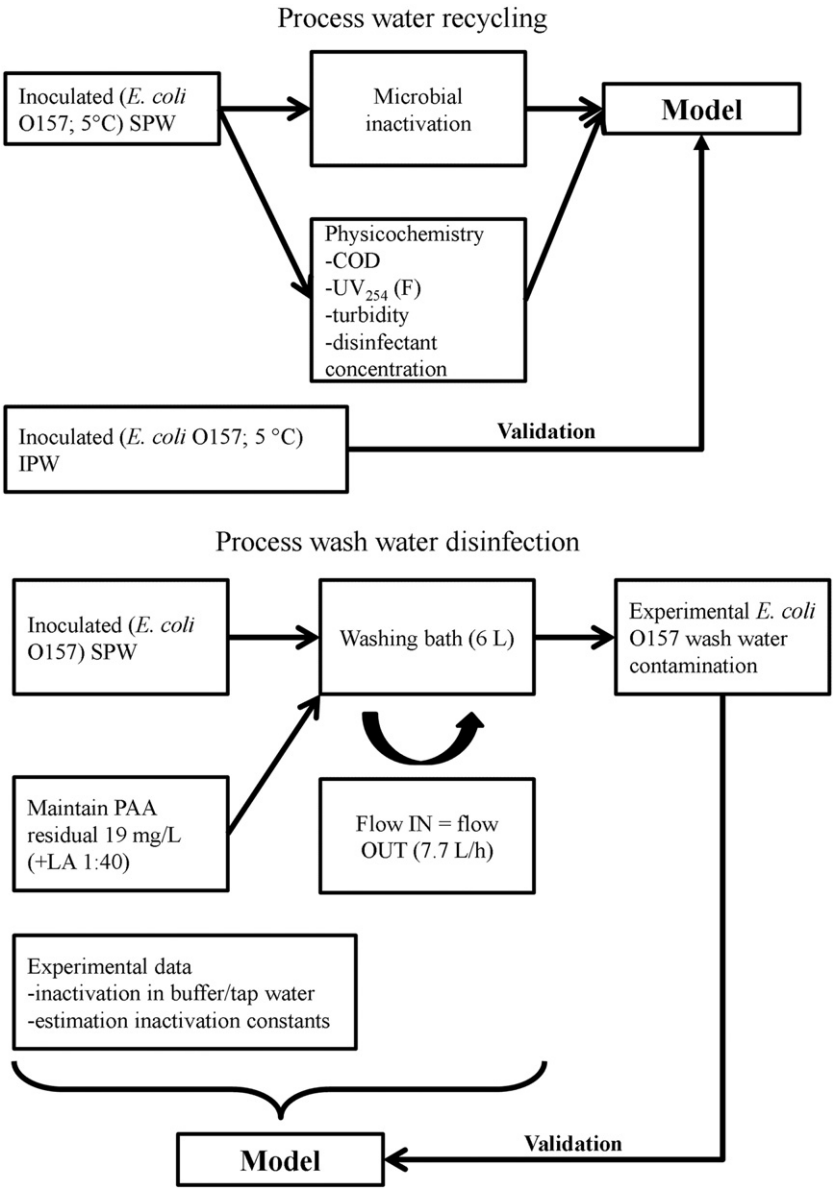


Fig. 1. Overview of experimental setup, SPW = standardized process water, IPW = industrial process water, $UV_{254}(F)$ = UV absorbance at 254 nm after filtration.

continuous increase of COD. The COD of the SPW was analyzed in triplicate before use in the experiment. Concentrated process water was diluted to a COD of 500 mg O_2/L with tap water and inoculated with the *E. coli* O157 cocktail in order to obtain ca. 6 log CFU/mL. The volume of water in the washing tank was 6 L. An opening at the appropriate height served as a water exit to keep the volume constant against the continuous income of process water and PAA + LA solution. Before starting, the washing tank was filled with clean tap water. PAA and LA concentration in the washing tank was adjusted to the desired level by addition of PAA and LA from stock solutions. Inoculated SPW was continuously dosed to the treatment tank at a flow rate of 7.7 L/h; simultaneously, a PAA + LA solution (stock concentration 100 mg/L and 4000 mg/L respectively) was dosed to the treatment tank at a flow rate necessary to maintain a constant PAA residual (target value 20 mg/L). Water samples were collected every 3 min in order to monitor PAA and re-adjust PAA + LA solution flow, and at pre-set time intervals in order to determine the microbial contamination during the 60 min trial. Sampling for measuring PAA and microbial load was done at 2 locations in the washing tank. The cumulative PAA dose to maintain the desired residual was monitored.

2.6. Physicochemical measurements

Turbidity was measured with a turbidimeter (HI98703; HANNA Instruments; Belgium), COD according to the small-scale sealed-tube method (ISO, 15705:2002; LCI 400; Hach Lange; Belgium), absorbance at UV 254 nm with a UV–visible (UV–vis) spectrophotometer (UV1601, Shimadzu, Belgium) and quartz cuvettes with a 1-cm path length (Hellma, Belgium) after filtration through a 0.45 μm polytetrafluorethylene filter (Macherey-Nagel, Belgium) ($UV_{254}(F)$). PAA concentration during water treatments was assessed by means of spectrophotometric measurements as described by Cavallini et al. (2012) with some modifications. Briefly, samples of 10 mL were taken from beakers containing 100 mL of SPW + PAA + LA at different time intervals and were disposed in test tubes containing 1 mL of catalase (320 mg/L) (Sigma-Aldrich, Belgium). Catalase was used to eliminate

interference by H_2O_2 in the PAA measurement. From these tubes, 10 mL were taken and analyzed according to the DPD method for total chlorine (Eaton et al., 2005) with absorbance measurement at 530 nm.

2.7. Microbiological analyses

Changes in levels of *E. coli* O157 in water were measured at different time intervals. For the water recycling experiments samples of 1 mL were taken from treated water and transferred into tubes containing 8 mL of a neutralizing solution. Neutralizing solution contained phosphate buffer saline (Na_2HPO_4 1.2 g/L, NaH_2PO_4 0.22 g/L, NaCl 8.5 g/L) supplemented with sodium thiosulphate (1 g/L) (all reagents from Sigma-Aldrich, Belgium). After mixing by means of a vortex, 1 mL of catalase solution (500 mg/L) was added. Phosphate buffer saline was used to keep samples at neutral pH, sodium thiosulphate neutralized PAA, and catalase neutralized residual H_2O_2. Then, samples were diluted when needed, using neutralizing solution, and plated in Chromocult coliform agar + nalidixic acid (Merck, Germany) (50 μg/mL) for *E. coli* O157. Plates were incubated at 37 °C for 24 h before counting.

2.8. Statistics used for assessing the models

SPSS statistics 22 and Microsoft Excel 2010 were used for statistical analysis. The Kolmogorov–Smirnov test and Levene's test were used to assess normality and equality of variance ($P \geq 0.05$) respectively. Differences between treatments, i.e. influence of physicochemical parameters, disinfectant concentration or bacterial species, were determined with ANOVA. If normality or equality of variance could not be assumed, the Kruskal–Wallis or Brown–Forsythe tests were used respectively as alternative to ANOVA. If significant differences were found, the Tukey HSD or Games-Howell post-hoc tests were used at a significance level of $P \leq 0.05$ for further analysis if the group variances were equal or unequal respectively. Non-linear regression for fitting the Hom model (Haas and Joffe, 1994) was executed with SPSS statistics 20 (sequential quadratic programming algorithm). The Geeraerd model was fitted by using the Excel add-on GinaFit v1.6 (generalized reduced gradient algorithm) (Geeraerd et al., 2000). To investigate for possible local minima, the values of the parameters at which the search algorithm initiates were altered to observe whether different solutions were obtained (depending on the initial parameter values), which was not the case. Noted deviations on measurements represent standard deviations unless otherwise stated.

2.8.1. Process water recycling

Multilinear regression was executed in JMP 10 (SAS Institute Inc.). Model selection was done based on variable significance level and ranked according to the Akaike Information Criterion. For assessing the overall quantitative quality of the models, both the squared correlation coefficient (r^2) for predicted values versus measured values and the ratio of prediction to deviation (RPD) were used. The RPD is the ratio of the standard deviation of the measured log reduction values to the root mean square error (RMSE) of the predicted values. The RPD expresses the increase of prediction accuracy compared to using the mean log reduction value to predict all disinfection trials. A ratio larger than 2 is necessary for a decent calibration, whereas a ratio below 1.5 indicates insufficient prediction potential of the model (Karoui et al., 2007). In order to provide additional qualitative information on the origin of the error, the Theil's decomposition of the mean square error (MSE) was used (Van Haute et al., 2013). Decomposition of the MSE can be done as following:

$$MSE = (\bar{y}-\bar{y}_m)^2 + \left(s'_{\bar{y}} - r s'_{\bar{y}_m}\right)^2 + (1-r^2)s'^2_{\bar{y}_m} \quad (1)$$

where

$\bar{y}_m$	mean of measured data points
$\bar{y}$	mean of modeled data points
r	the Pearson's coefficient of correlation

$$s'_{\bar{y}_m} = \sqrt{\frac{\sum_i (y_{m,i}-\bar{y}_m)^2}{N}} \quad (2)$$

$$s'_{\bar{y}} = \sqrt{\frac{\sum_i (y_i-\bar{y})^2}{N}}. \quad (3)$$

Dividing Eq. (1) by the MSE allows a proportional representation of the three decomposed factors of the MSE (Eqs. (4) and (5)).

$$1 = \frac{(\bar{y}-\bar{y}_m)^2}{MSE} + \frac{\left(s'_{\bar{y}} - r s'_{\bar{y}_m}\right)^2}{MSE} + \frac{(1-r^2)s'^2_{\bar{y}_m}}{MSE} \quad (4)$$

$$1 = U_m + U_r + U_d. \quad (5)$$

U_m measures the proportion of the MSE related to bias in the prediction model. U_r represents the proportion of the MSE that is caused by deviation of the regression line between measured and predicted data points from the 45° perfect fit line. U_d represents random prediction errors that cannot be reduced. Ideally, U_m and U_r would be zero, while U_d would equal the MSE.

To assess for nested models whether adding parameters to the model (i.e. increasing complexity) leads to a significantly better fitted model (with consideration of the difference in prediction error of both models, the number of data points and the number of parameters of both models), the partial F-test was used (Eq. (6)):

$$F = \frac{(SSE_r - SSE_f)/SSE_f}{(f-r)/(N-f)} \quad (6)$$

where

SSE_r	sum of square errors of prediction of the reduced model
SSEf	sum of square errors of prediction of the full model
N	number of data points
r	number of parameters in reduced model
f	number of parameters in full model.

The resulting F-value is compared with the critical F-value for a given α (α = 0.05), which is given by $F_{f-r,N-f,1-\alpha}$ (Glatting et al., 2007).

2.8.2. Process wash water disinfection in the washing tank

The models were constructed in @RISK (add-on Excel). Distributions were fitted to the measured parameters that were used for constructing the model. Monte Carlo simulation was used to select random samples from the input distributions as input for the model. As such, a set of output samples (*E. coli* O157 wash water contamination) were obtained, and distributions were fitted to these output samples. For assessing the overall quality of the time series models in this experiment, the Theil's inequality coefficient (TIC) was used (Eq. (7)). TIC values range from 0 to 1 and values below 0.3 indicate a decent agreement of the model with the experimental data (Audenaert et al., 2010).

$$TIC = \frac{\sqrt{\sum_i (y_i - y_{m,i})^2}}{\sqrt{\sum_i y^2_i} + \sqrt{\sum_i y^2_{i,m}}}. \quad (7)$$

3. Results

3.1. Peracetic acid as water recycling agent

When considering both the SPW and the IPW, the PAA decay rate correlated better with the UV_{254}(F) than with the COD or the turbidity

(Fig. 2). Usually, when oxidizing agents are used as disinfectants, an initial decay in disinfectant concentration is often observed. This initial decay means a very rapid disinfectant depletion (within the first min) that is not explained by the further (in time) observed disinfectant decay rate. It is determined as the difference between the added disinfectant concentration and the concentration at which the fitted curve cuts the y-axis. ANOVA analysis showed no difference among initial decay in function of the variables $UV_{254}(F)$ and PAA concentration, unless for $UV_{254}(F) \geq 0.29$ and PAA ≥ 1.4 mg/L, where there was a significant initial decay, which correlated with $UV_{254}(F)$ (Table 1).

To estimate the reaction order of PAA decay, linear regression of PAA (0th), ln(PAA) (1st), and PAA^{-1} (2nd) in function of time was executed for all PAA concentrations, and physicochemical loads (n = 122 time points). Within the studied range of PAA concentration (0.7–2.1 mg/L) and physicochemical load ($UV_{254}(F) = 0.02 - 0.54$), the prediction quality of PAA decay was as followed: 0 order ($r^2 = 0.97$; RPD = 6.33) > 1st order ($r^2 = 0.95$; RPD = 3.15) >> 2nd order ($r^2 = 0.78$; RPD = 1.41). Note however that these parameter relations are only valid for the considered range of $UV_{254}(F)$ and PAA concentration. At higher $UV_{254}(F)$, i.e. in the IPW with $UV_{254}(F)$ of 1.23, initial decay was dependent on both the $UV_{254}(F)$ and the PAA concentration, and the time dependent decay approximated 1st order kinetics: 1st order ($r^2 = 0.97$; RPD = 6.29) >> 0 order ($r^2 = 0.81$; RPD = 2.33) > 2nd order ($r^2 = 0.79$; RPD = 2.20).

The PAA decay was estimated in function of $UV_{254}(F)$ and PAA concentration with multilinear regression. For the PAA decay in function of time, the three models with the highest RPD value for the calibration also yielded the highest RPD values for the validation, and this for both 0 and 1st order approximations (Table 1). In addition, the models with lower complexity (lower amount of parameters) showed the highest prediction quality for both 0th (Model 1) and 1st order (Model 4) (Table 1).

Comparison of the microbial inactivation with 1.4 mg/L of PAA and 1.4 mg/L PAA + LA (1:40) showed that the addition of LA improved the disinfection efficiency (Fig. 3). The inactivation of *E. coli* O157 in function of time occurred in two phases, a phase of slow microbial inactivation (shoulder) followed by a phase of more rapid inactivation. Both the PAA concentration and physicochemical quality ($UV_{254}(F)$) of the SPW influenced the slope of the microbial kill-off (increasing and decreasing influence respectively). Two models were assessed to predict the microbial inactivation: the shoulder + log-linear model by Geeraerd et al. (2000), and the modified Hom model with disinfectant decay by Haas and Joffe (1994). The modified Hom model can be used to model the observed shoulder effect and incorporate the first order disinfectant decay (Eqs. (8) and (9)):

$$\ln\left(\frac{N(t)}{N0}\right) = -\left(\frac{m}{n \cdot kPAA}\right)^m \left(k(PAA(0) - InDecay)^n\right)\left(1 - e^{\frac{-n \cdot kPAA \cdot t}{m}}\right)^m \quad (8)$$

$$PAA = PAA(0) \cdot e^{-kPAA \cdot t} \quad (9)$$

where

N(t)	*E. coli* O157 concentration (CFU/mL) at time t N(0) initial *E. coli* O157 concentration (CFU/mL)
m, n, and k	the Hom model parameters
kPAA	the first order PAA decay rate constant
InDecay	initial decay
PAA(0)	added PAA dose.

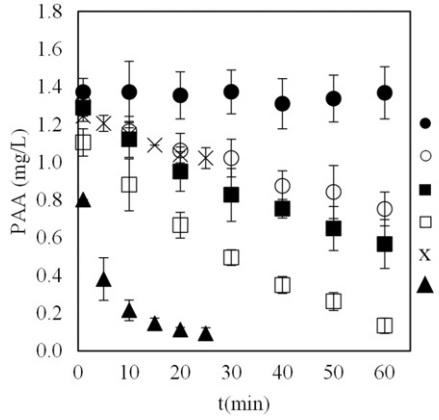
The 1st order PAA decay model (initial decay, kPAA) with the highest RPD validation value (Model 4, Table 1), was used for the modified Hom model (Table 2). With m > 1, a shoulder effect was described. With an RPD value of 2.76 and 99% of the error due to random prediction errors, the model does not suffer from systematic deviations between the measured and predicted inactivation data.

The Geeraerd equation has the following form:

$$\log\frac{N(t)}{N0} = \frac{-kmax \cdot t}{\ln(10)} + \log\left(\frac{e^{kmax \cdot SL}}{1 + \left(e^{kmax \cdot SL} - 1\right) \cdot e^{-kmax \cdot t}}\right) \quad (10)$$

For the Geeraerd model, the data from addition of 0.7 mg/L PAA to SPW with $UV_{254}(F)$ absorption of 0.54 were discarded due to lack of significant microbial inactivation which interfered with the estimation of the maximum inactivation constant (kmax) and the shoulder length (SL). The Geeraerd model was fitted to the calibration set (n = 33; 171 time points) and for each setting (PAA conc, $UV_{254}(F)$ value) and repeat (n = 3) the parameters (kmax and SL) were determined. The relation between the kmax/SL values and the physicochemical parameters was assessed with multilinear regression. The three microbial inactivation models (kmax, SL combinations) that yielded the best RPD values are shown in Table 2. Both the Hom (Table 2) and Geeraerd models (Models 3–5, Table 2) contained an equal amount of parameters (i.e. 6 parameters). The RPD and r^2 values were slightly higher for the Geeraerd model (Models 3–5, Table 2) than for the Hom model (Model 0, Table 2). On the other hand, a larger percentage (11% vs 1%) of the error was systematic. According to the model, SL is dependent on PAA concentration but not on the physicochemical water quality, whereas kmax is influenced by both PAA concentration and $UV_{254}(F)$. However, these dependencies can only be suggested within the experimental limits (PAA concentration, $UV_{254}(F)$) of the experiment. Comparing the reduced Geeraerd models 1 or 2 (Table 2) with the more complex model 3 (Table 2) resulted in p values $<10^{-5}$ (partial F-test) for both comparisons. Therefore, lower model complexity for the Geeraerd models could not be justified.

Contrary to the *E. coli* O157 inactivation in SPW, the inactivation in IPW could not be predicted solely based on the PAA exposure (in turn



	Turbidity (NTU)	COD (mg O_2/L)	UV_{254} (F)
●	<5	30	0.022 ± 0.004
○	25.3 ± 4.3	516 ± 4	0.18 ± 0.02
■	37.5 ± 6.1	835 ± 6	0.29 ± 0.03
□	68.9 ± 10.6	1551 ± 33	0.54 ± 0.04
x	11.3 ± 0.1	177 ± 1	0.26 ± 0.03
▲	32.1 ± 2.9	518 ± 10	1.23 ± 0.02

Fig. 2. PAA decay (1.4 mg/L at t = 0 min; PAA + LA 1:40) in function of time in SPW/IPW, ● ○ ■ □ = SPW, ▲ = IPW Company 1, × = IPW Company 2 (n = 3).

Table 1
Prediction quality of the PAA concentration during water disinfection in SPW/IPW wash waters with PAA + LA.

Model number	InDecay[a]	kPAA[b]	Calibration SPW					Validation IPW				
			r^2	U_m	U_r	U_d	RPD	r^2	U_m	U_r	U_d	RPD
	Zero order											
1	$(0.49 \pm 0.06$[c]$) \cdot UV_{254}(F)$	$(0.003 \pm 0.001) \cdot PAA + (0.020 \pm 0.003) \cdot UV_{254}(F)$	0.964	0.051	0.016	0.933	5.07	0.997	0.004	0.009	0.987	12.2
2	$(0.49 \pm 0.06) \cdot UV_{254}(F)$	$(-7 \pm 11).10^{-4} + (0.001 \pm 0.0007) \cdot PAA + (0.034 \pm 0.006) \cdot UV_{254}(F) - (0.054 \pm 0.008) \cdot UV_{254}(F)^2 + (0.015 \pm 0.002) \cdot PAA \cdot UV_{254}(F)$	0.966	0.121	10^{-4}	0.879	5.13	0.993	0.028	0.010	0.962	11.5
3	$(0.49 \pm 0.06) \cdot UV_{254}(F)$	$-0.003 \pm 0.001 + (0.0046 \pm 0.0009) \cdot PAA + (0.023 \pm 0.003) \cdot UV_{254}(F)$	0.969	0.119	0.002	0.879	5.30	0.994	0.075	0.166	0.759	11.3
	First order											
4	$(0.49 \pm 0.06) \cdot UV_{254}(F)$	$(0.090 \pm 0.007) \cdot UV_{254}(F) - (0.028 \pm 0.005) \cdot PAA \cdot UV_{254}(F)$	0.978	0.001	0.082	0.917	6.47	0.990	0.348	0.005	0.647	8.25
5	$(0.49 \pm 0.06) \cdot UV_{254}(F)$	$(6 \pm 10) \cdot 10^{-4} \cdot PAA + (0.090 \pm 0.007) \cdot UV_{254}(F) - (0.029 \pm 0.005) \cdot PAA \cdot UV_{254}(F)$	0.981	0.015	0.037	0.948	7.08	0.990	0.362	0.003	0.635	7.98
6	$-0.042 \pm 0.026 + (0.58 \pm 0.23) \cdot UV_{254}(F)$	$(6 \pm 10) \cdot 10^{-4} \cdot PAA + (0.090 \pm 0.007) \cdot UV_{254}(F) - (0.029 \pm 0.005) \cdot PAA \cdot UV_{254}(F)$	0.981	0.014	0.039	0.947	7.12	0.990	0.362	0.003	0.635	7.98

[a] Presence of an initial decay (InDecay) was only significant for UV254(F) ≥ 0.29 and PAA ≥ 1.4 mg/L.
[b] PAA decay rate.
[c] Standard error of mean.

dependent on added PAA concentration and $UV_{254}(F)$ absorbance). Despite similar PAA exposure, the inactivation in IPW was preceded by a longer lag phase (Fig. 4). As the temperature was kept constant (5 °C) in both wash waters, the pH seems the most obvious factor of influence. The initial pH in the wash water was higher in the IPW than in the SPW. Due to the added LA, the pH dropped from 8.2 to 7.8–7.3 (72–89% PAA in acid form) in the IPW, and from 7.4 to 6.7–6.5 (96–98% PAA in acid form) in the SPW. The influence of dissociation of the PAA was assessed in buffered solutions. Lowering the pH increased the reduction of *E. coli* O157 (Fig. 5).

3.2. Peracetic acid as process wash water disinfectant

A model to assess the *E. coli* O157 contamination in the washing tank during continuous influx of SPW (COD 504 mg O_2/L; $UV_{254}(F)$ 0.945; 6 log CFU/mL *E. coli* O157) was constructed based on the following assumption: PAA is free to inactivate bacteria, or otherwise stated, knowledge of the residual PAA in the washing tank can be used to estimate the microbial kill-off regardless the physicochemical load.

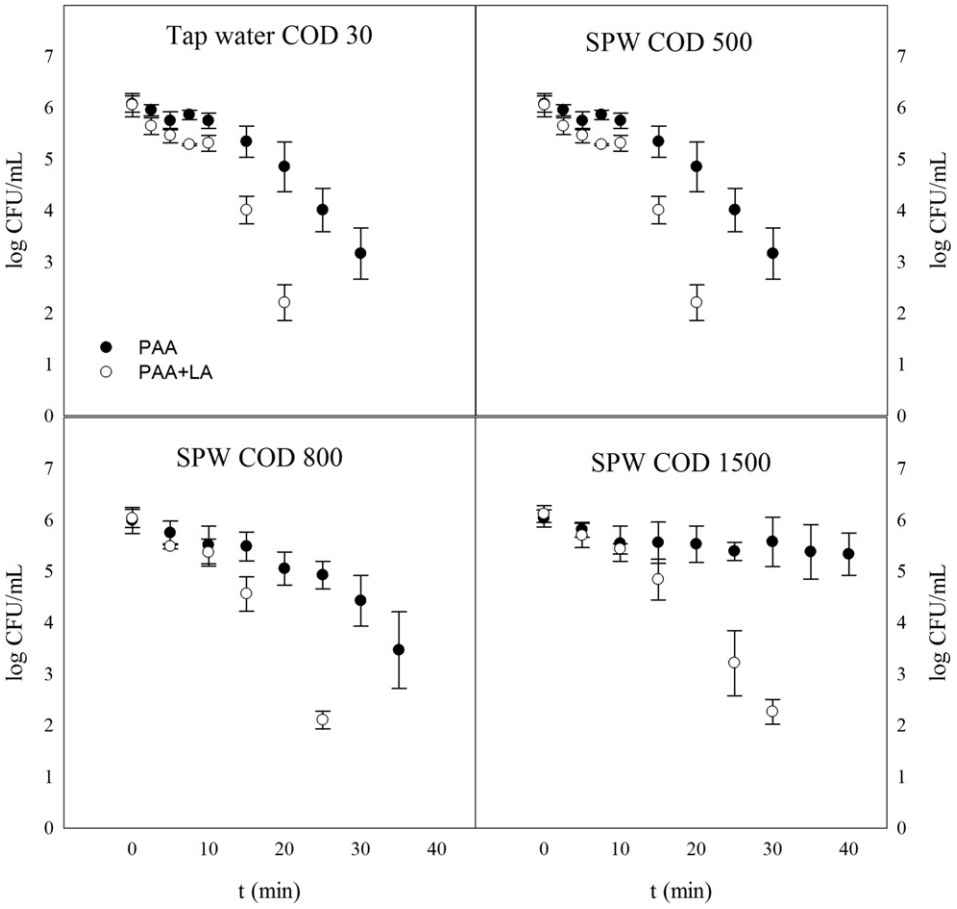


Fig. 3. Comparison of *E. coli* O157 inactivation with 1.4 mg/L PAA compared to 1.4 mg/L PAA + LA(1:40) (n = 3).

To estimate the Geeraerd inactivation constants, *E. coli* O157 was inactivated in tap water, i.e. in water with virtually no PAA demand (Table 3). Within the range of 15 to 24 mg/L PAA the kmax and SL did not significantly change. During the dynamic PAA + LA experiment, the PAA concentration was 18.5 ± 1.1 mg/L. Therefore, the parameter estimates of kmax and SL for PAA = 19 mg/L (Table 3) were used in the model. The dynamic model was constructed by inserting a water dilution factor in the derivative of the Geeraerd equation, in order to quantify the change in *E. coli* O157 load with respect to time (Eq. (11)):

$$\frac{dNs(t)}{dt} = \left(-kmax \cdot \frac{-kmax}{1 + e^{-kmax \cdot t} \cdot \left(e^{kmax \cdot SL} - 1\right)} - D\right) \cdot Ns(t) \quad (11)$$

where

Ns(t) *E. coli* O157 wash water contamination (CFU/mL) of a 'single' influx event of microbial contamination
D dilution factor (s^{-1}), i.e. the fraction of the water volume in the washing tank that is exiting the washing tank in function of time.

For the initial condition Ns(t) = N0 with N0 is influx of contamination (CFU/s/mL), solving the differential equation yields (Eq. (12)):

$$Ns(t) = \frac{N0e^{kmax \cdot SL} \cdot \left(e^{-kmax \cdot t}\right)^{\frac{D}{kmax}}}{e^{kmax \cdot SL} - 1 + e^{kmax \cdot t}}. \quad (12)$$

This equation describes a 'single' contamination event in function of time. As for all intents and purposes D << kmax, the factor D/kmax approaches 0 and the equation is reduced to the Geeraerd equation (Eq. (13)):

$$Ns(t) = \frac{N0e^{kmax \cdot SL}}{e^{kmax \cdot SL} - 1 + e^{kmax \cdot t}}. \quad (13)$$

As the inactivation rate is time dependent, the total microbial contamination at a certain time point is described as the sum of all the 'single' contamination events up to that point. Therefore, the total microbial wash water contamination equals the integration of the Geeraerd function (Eq. (13)) in function of time (Eq. (14)):

$$N(t)_{mL} = \int_0^t Ns(t)dt = \left[-\frac{N0 \cdot e^{kmax \cdot SL} \cdot \left(\ln\left(e^{kmax \cdot SL} - 1 + e^{kmax \cdot t}\right) - kmax \cdot t\right)}{kmax \cdot \left(e^{kmax \cdot SL} - 1\right)}\right]_0^t \quad (14)$$

where $N(t)_{mL}$ = the *E. coli* O157 wash water concentration at time t (CFU/mL).

The *E. coli* O157 wash water contamination in the absence of PAA + LA was estimated by measuring the *E. coli* O157 contamination in the inoculated SPW (n = 6) and entering these contamination data in Eq. (16).

$$\frac{dN(t)}{dt} = a \cdot inflow - \frac{inflow}{V} \cdot N(t) \quad (15)$$

Where

N(t) the number of *E. coli* O157 cells in the wash water at time t (CFU)
a the *E. coli* O157 concentration in the influent (CFU/mL)
inflow outflow = the rate of inflow of SPW in the washing tank (mL/s)
V volume of the washing tank (mL).

Eq. (15) can be solved for N(0) = 0:

$$N(t)_{mL} = a \cdot \left(1 - e^{\left(-\frac{inflow \cdot t}{V}\right)}\right) \quad (16)$$

where $N(t)_{mL}$ = the *E. coli* O157 wash water concentration at time t (CFU/mL).

When wash water disinfection was applied, the *E. coli* O157 wash water contamination converged rapidly (Fig. 6, panel PAA 18.5 mg/L) and so did the model output as can be seen when zooming in on the first 30 s of the washing process (Fig. 7). The model corresponded well to the measured *E. coli* O157 contamination values (TIC = 0.020).

3.3. Peracetic acid dose and residual in function of COD and water refreshing during process wash water disinfection

The total COD that entered the washing tank via SPW during the wash water disinfection experiment can be calculated as following:

$$COD_T = COD_{SPW} \cdot inflow \cdot t \quad (17)$$

where

COD_T total COD that entered the washing tank during the experiment (mg)
COD_{SPW} COD content of SPW (mg O_2/L)
inflow water refreshing rate (L/h)
t time of experiment (h).

By keeping track of the disinfectant dose, both the total demand of PAA and the demand in function of COD could be calculated as following:

$$PAAdemand_T = Dose - Residual \cdot (V + inflow \cdot t) \quad (18)$$

Table 2
Prediction quality of the modified Hom model with disinfectant decay (Model 0) and of the Geeraerd models (Model 1–5) for *E. coli* O157 inactivation in SPW with PAA + LA.

Model number	InDecay	kPAA	k	n	m	r^2	U_m	U_r	U_d	RPD
0	(0.49 ± 0.06[a]) · UV_{254}(F)	(0.090 ± 0.007) · UV_{254}(F) − (0.028 ± 0.005) · PAA · UV_{254}(F)	0.015 ± 0.004	2.12 ± 0.09	1.92 ± 0.08	0.867	0.001	0.001	0.998	2.76

Model number	SL	kmax	r^2	U_m	U_r	U_d	RPD
1	19.7 ± 1.4[a] − (7.27 ± 0.87) · PAA	−0.22 ± 0.09 + (0.63 ± 0.05) · PAA − (0.40 ± 0.12) · UV_{254}(F)	0.826	0.001	0.691	0.307	2.00
2	31.0 ± 3.7 − (26.1 ± 5.6) · PAA + (6.6 ± 1.9) · PAA^2	(0.59 ± 0.05) · PAA − (0.78 ± 0.25) · UV_{254}(F)	0.878	0.004	0.352	0.640	2.30
3	31.0 ± 3.7 − (26.1 ± 5.6) · PAA + (6.6 ± 1.9) · PAA^2	−0.22 ± 0.09 + (0.63 ± 0.05) · PAA − 0.40 ± 0.12) · UV_{254}(F)	0.910	0.072	0.040	0.888	3.15
4	31.0 ± 3.7 − (26.1 ± 5.6) · PAA + (6.6 ± 1.9) · PAA^2	−0.25 ± 0.15 + (0.635 ± 0.091) · PAA − (0.81 ± 0.33) · $UV_{254}(F)^2$	0.918	0.080	0.028	0.892	3.31
5	31.0 ± 3.7 − (26.1 ± 5.6) · PAA + (6.6 ± 1.9) · PAA^2	(0.26 ± 0.11) · PAA + (0.13 ± 0.08) · PAA^2 − (0.38 ± 0.17) · UV_{254}(F)	0.915	0.082	0.022	0.896	3.25

[a] Standard error of mean.

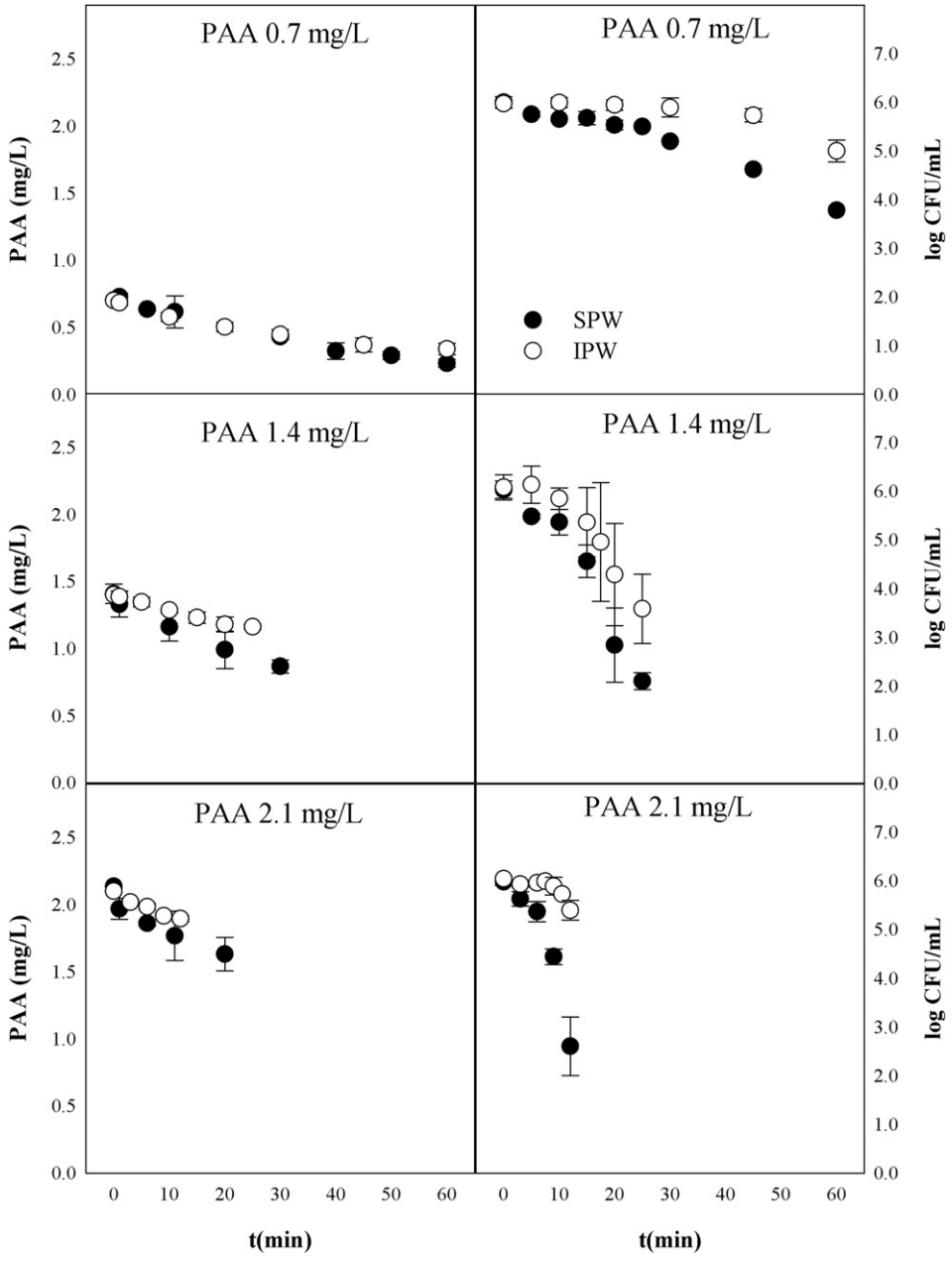


Fig. 4. Comparison of PAA decay and *E. coli* O157 inactivation in SPW (with $UV_{254}(F) = 0.29$) and IPW (with $UV_{254}(F) = 0.26$) for added PAA concentrations of 0.7, 1.4 and 2.1 mg/L (n = 3).

$$\text{PAAdemand} = \frac{\text{PAAdemand}_T}{\text{COD}_T} \quad (19)$$

where:

Dose PAA dose added during the experiment (mg)
PAAdemand_T total PAA demand (mg)
PAAdemand PAA demand per mg O_2 COD
Residual PAA residual per L (mg/L)
V volume of washing tank (L).

Knowing the added COD and PAA + LA during the washing simulation in SPW from spinach (COD = 504 mg O_2/L; $UV_{254}(F) = 0.944$), the PAA demand could be calculated. To maintain 18.5 mg/L residual of PAA (to achieve about 2.3 log reduction at 60 min of operation), 0.003 mg PAA per mg COD was consumed. The pH did not change significantly in function of time (pH 3.8 ± 0.1). The same experimental setup was used by Gómez-López et al. (2014) but for studying free chlorine. To maintain 1 mg/L of free chlorine (to achieve >3 log reduction at 60 min of operation) in the same conditions 0.124 mg chlorine per mg COD was consumed, i.e. about 41.3 times more free chlorine was needed. 50 ± 4 mg/L of PAA (+LA) was needed to achieve >3 log reduction in an identical dynamic setup (Fig. 6, panel PAA 50 mg/L). These data were not modeled because of microbial values below the limit of detection (1 log CFU/mL). The pH did not change significantly in function of time (pH 2.9 ± 0.1).

Present-day postharvest washing operations refresh the water to some degree i) to maintain a positive transfer of matter from the lettuce to the water, ii) because of the misconception that it can effectively be used to control the microbial load, iii) to control the accumulation of pesticides, mycotoxins etc., iv) to control the concentration of DBPs. Refreshing also has impact on the necessary disinfectant dose to maintain an opted residual. As free chlorine and PAA show vastly different behaviors in both inactivation kinetics and disinfectant stability, comparing

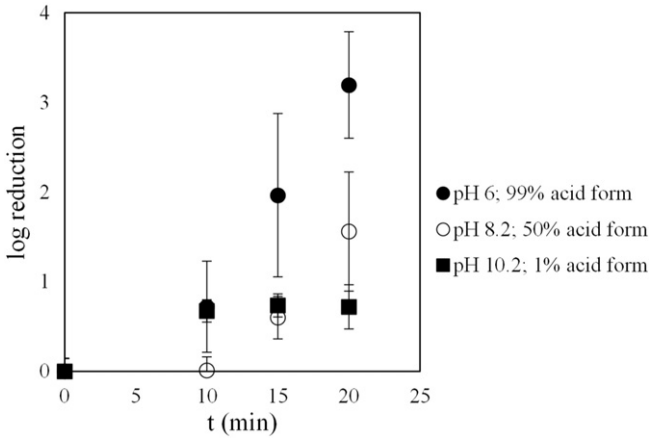


Fig. 5. Inactivation of *E. coli* O157 in oxidant demand free buffer with PAA + LA (1:40; 1.4 mg/L PAA) (n = 3).

these disinfectants is an interesting setup to assess how the COD load and refreshing rate influence the disinfectant consumption.

Suppose a fresh-cut spinach washing process with the following parameters:

- influx of organic matter (COD) from washing spinach (range 0 to 1500 mg O_2/h),
- assume a disinfectant demand of 0.003 mg PAA/mg COD and 0.124 mg free chlorine/mg COD (the data for the latter not shown in the article but obtained from the study by Gómez-López et al. (2014) under identical experimental conditions),
- a reduction of >3 log *E. coli* O157 is aimed for, i.e. a residual of 1 mg/L of free chlorine or 50 mg/L of PAA + LA (1:40) is used.

The required PAA dosage to satisfy the COD demand is negligible compared to the dosage required to cope with the water refreshing, whereas the vice versa is observed for free chlorine (Fig. 8). Although the COD per water volume decreases, the total COD load remains the same and is merely "spread out" over larger water volumes.

4. Discussion

4.1. Peracetic acid disinfection in oxidant demand free conditions

PAA was applied in two conditions of low oxidant demand water, i.e. in tap water and oxidant demand free buffer. In both cases, an initial shoulder effect was observed. Concerning PAA, not much is known about the inactivation rate in oxidant demand free conditions. PAA has mostly been researched in a wastewater context due to its high stability in the presence of organic matter. However, these studies can provide some information regarding the inherent disinfection efficiency of PAA. For example, Dell'Erba et al. (2004) disinfected *E. coli* in wastewater by adding 4 mg/L which decayed to about 3.2 mg/L in 30 min contact time. As such, *E. coli* and total coliforms were exposed to more than 3.2 mg/L of PAA during the experiment. In order to reach 1 log reduction

Table 3
Prediction quality of the Geeraerd models for *E. coli* O157 inactivation with PAA + LA in tap water.

PAA (mg/L)	kmax	SL	r^2	RPD
14	21.4 ± 2.0[a]	0.23 ± 0.07	0.91	2.78
19	22.6 ± 5.7	0.16 ± 0.08	0.89	2.48
24	21.7 ± 4.1	0.10 ± 0.06	0.95	3.43

[a] Standard error of mean.

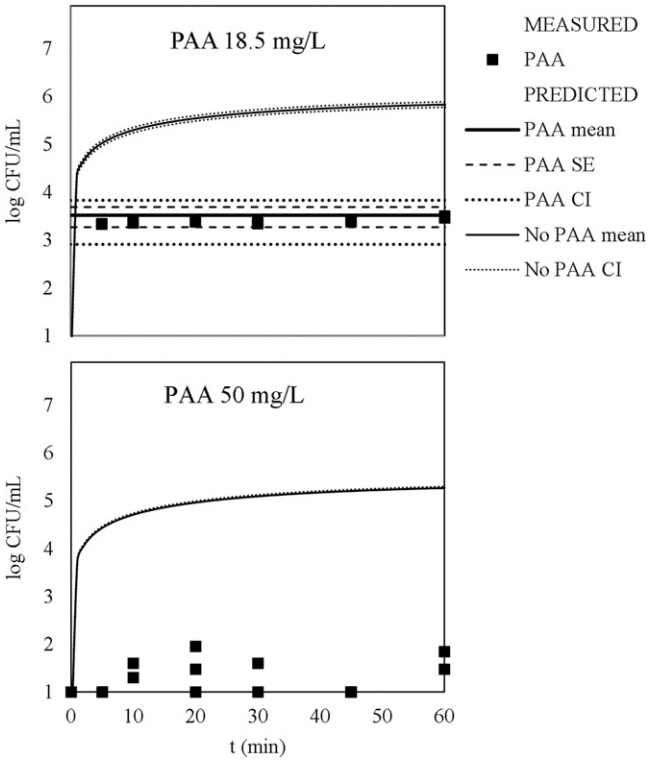


Fig. 6. Model for *E. coli* O157 contamination during process wash water disinfection in the washing tank with PAA + LA, SE = standard error of mean, CI = 95% confidence interval.

of *E. coli* and total coliforms, about 15 min of contact time were necessary. Although such a reasoning simplifies the process by not considering particle shielding, higher resistance of wild-type strains etc., it is an indication of the relatively slow disinfection rate of PAA compared to ozone (von Gunten, 2003), free chlorine (Van Haute et al., 2013), and chlorine dioxide (LeChevallier et al., 1988). The small k value (Model 0, Table 2) compared to that in the Hom modeling of *E. coli* O157 with free chlorine (Van Haute et al., 2013) expresses the considerably slower inactivation kinetics of PAA (+LA).

The parameter values that result from fitting the models to the *E. coli* O157 inactivation data in oxidant demand free conditions provide information concerning the disinfection rate of the studied microorganism or otherwise stated in the context of fresh(−cut) produce wash water, concerning the necessary concentration of the disinfectant residual. A first approach could be to use a rule of thumb: e.g. a reduction of

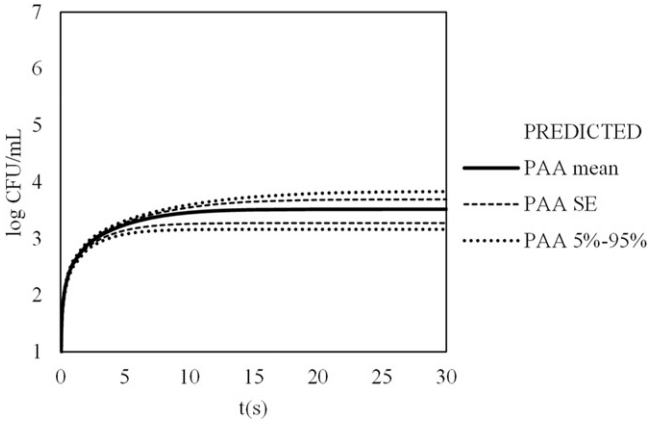


Fig. 7. Model output for first 30 s during process wash water disinfection in the washing tank with PAA + LA (18.5 ± 1.1 mg/L PAA).

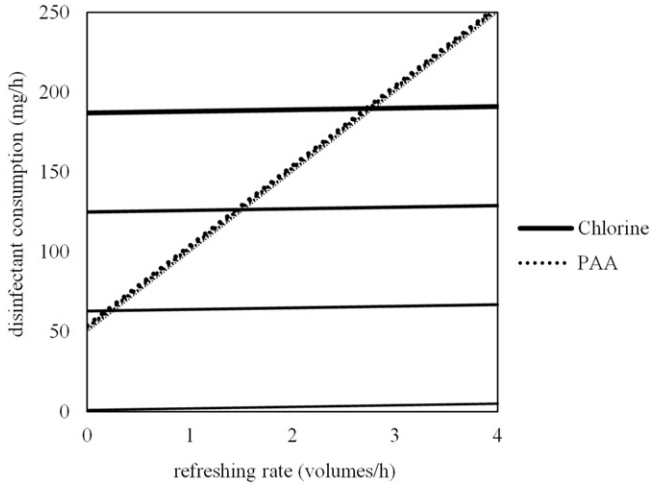


Fig. 8. Consumption of disinfectant in function of influx of COD and refreshing rate; influx of COD in the range 0–1500 mg O_2/h; increasing line thickness denotes increasing influx of COD.

3 log CFU/mL within 30 to 60 s is desired to avoid microbial cross-contamination, which can be derived from fitting the Hom model or Geeraerd model to the inactivation data of the target microorganism in oxidant demand free conditions. A second approach is to implement these parameter values (Table 3) in the dynamic wash water disinfection model (Eq. (14)) in order to consider both the inflow, outflow and death of the target microorganism in the washing tank as was done in this study.

4.2. Peracetic acid process water recycling of fresh-cut lettuce wash water

In this study, the UV_{254}(F) absorption is the variable with most influence on initial and time dependent decay of PAA. A validation with IPW was needed in order to find the correlation between UV_{254}(F) and PAA decay. In the SPW, COD and turbidity correlate with UV_{254}(F) because the different SPWs are derived from the same matrix (iceberg lettuce) and differ only in dilution (Fig. 2). A sharp initial decay of disinfectant concentration often occurs for chemical oxidants in wastewater disinfection, including PAA. This has been attributed to particles, reduced inorganic species such as iron and manganese, microorganisms, volatilization and reaction of the disinfectant with water (Falsanisi et al., 2006; Hoff, 1986; john et al., 2005). Free chlorine decay (and *E. coli* O157 inactivation) correlated with COD but not the UV_{254}(F) of leafy vegetables wash water (Van Haute et al., 2013). There is considerable variability in the COD:UV_{254}(F) ratio dependent on the type of produce that is washed, even among different leafy vegetables. For example, the SPW derived from spinach has a much higher COD:UV_{254}(F) ratio than the SPW derived from iceberg lettuce. When equal amounts of organic matter are transferred to the water when washing iceberg lettuce and spinach, the PAA decay would be lower in wash water from iceberg lettuce than from spinach. On the other hand, for free chlorine decay this would not be the case, because the free chlorine decay correlates with a more general indicator for organic matter, i.e. COD. When assessing the necessary disinfectant dose to perform a disinfection treatment, the water quality variable of interest will depend on the disinfectant.

The disinfection behavior of PAA was similar in SPW/IPW as in tap water, i.e. a shoulder effect followed by a log-linear inactivation. This behavior is not universal however, as values of $m < 1$ in Hom models, i.e. no shoulder but tailing, have been observed e.g. in wastewater for *E. coli* and total coliforms (Dell'Erba et al., 2004; Falsanisi et al., 2006; Liberti et al., 1999). This occurred with high initial PAA demand (Falsanisi et al., 2006), as well as with low initial demand followed by low disinfectant decay (Dell'Erba et al., 2004), and as such rapid disinfectant decay cannot fully account for the observed decreased disinfection rate in function of time in those studies. A possible explanation would be the presence of subpopulations with different resistance against PAA within a species (*E. coli*) or varying resistance between species within a microbial group (total coliforms), clumping of microbial cells, or particle association of microbial cells (Gyürék and Finch, 1998). As in the current study all *E. coli* O157 cells were derived from two strains that were grown in ideal conditions, such variety among the population might have been absent.

E. coli O157 inactivation during process water recycling with PAA + LA in SPW could be estimated with the PAA concentration (initial decay and gradient of PAA decay) and the contact time (modified Hom model) or through PAA (+LA) concentration and UV_{254}(F) to predict the kmax and SL values for the Geeraerd model. The *E. coli* O157 inactivation in IPW was lower than expected from the UV_{254}(F) and PAA concentration values and the pH was shown to be influential on the inactivation, presumably because the acid form of the PAA has somewhat greater microbial inactivation (Kitis, 2004). This can also serve as an explanation for the improvement in disinfection efficacy at these low concentrations of LA (i.e. 28 to 82 mg/L) which in itself have no significant disinfectant potential against *E. coli* or *Listeria* spp. (Ho et al., 2011; Virto et al., 2006), but which did lower the pH, i.e. increase the relative abundance of the acid form of PAA.

Both the *E. coli* O157 inactivation rate and disinfectant decay rate in leafy vegetables wash water are considerably higher in the case of free chlorine (Van Haute et al., 2013) compared to PAA + LA. Due to the stability of PAA in the wash water, a disinfectant residual can be maintained by adding relatively low doses which interact with the microorganisms during relatively long contact times to achieve disinfection as is being used in wastewater disinfection (Collivignarelli et al., 2000; Dell'Erba et al., 2004). In dynamic wash water disinfection a rapid microbial inactivation is of paramount importance to counter microbial cross-contamination, which is not necessarily the case for process water recycling. Therefore, PAA is an excellent disinfectant for process water recycling, requiring low doses. However, the longer contact times might require a larger washing tank.

4.3. Peracetic acid dynamic wash water disinfection

For the process wash water disinfection in the washing tank, the model for PAA + LA accurately predicted the microbial wash water contamination. However, the used models do not incorporate certain phenomena.

The used *E. coli* O157 cells were in the stationary phase, yet not exposed to heavy external stress. In real life situations however, cells may have been present on the produce for extended periods of time. When the specific growth rate is lower, when nutrient limitation occurs or even starvation, the resistance of bacteria against chemical disinfectants tends to increase (Berg et al., 1982; Cromeans et al., 2010; Doull et al., 1980; Hoff and Akin, 1986; Koivunen, 2007; Lee and Frank, 1990; Lisle et al., 1999; Luh and Marinas, 2007; Taylor et al., 2000).

Also, the main assumption of the model ("the microbial inactivation depends on the disinfectant residual and contact time irrespective of the physicochemical load of the water matrix") is not necessarily valid in water with a high physicochemical load. Microbial inactivation can occur without the presence of observable disinfectant residual. While the disinfectant demand is met, disinfectants react both with microorganisms and other water matrix constituents. This has been observed for ozone (Hunt and Mariñas, 1999; Janex et al., 2000; Xu et al., 2002) and suspected for free chlorine (Van Haute et al., 2013; Winward et al., 2008). The PAA + LA process wash water model however, could predict the *E. coli* O157 inactivation based on the disinfectant residual and contact time. This can be explained by the relatively slow decay of PAA in the presence of organic matter, resulting in lower disinfectant demands of the wash water, and as such lower concentration gradients of the disinfectant (i.e. the impact of the disinfectant dose on the microorganisms is not much different from that of the disinfectant residual),

and overall slower influence of concentration gradients on microbial inactivation.

The dynamic wash water disinfection model (Eqs. (14) and (16)) shows that controlling a wash water disinfection process with PAA is possible based on knowledge of disinfectant residual (and pH). Organic matter in the wash water only influences the disinfectant demand and as such the necessary dose to maintain the disinfectant residual. The model shows that through automatic monitoring of PAA residual and pH, and automatic dosing of PAA (and possibly acids) the microbial concentration in the wash water could be controlled. According to this model combined with the PAA demand equations (Eqs. (17)–(19)). PAA seems only interesting for disinfection of *E. coli* O157 when high COD loads are present and low refreshing rates applied. As these are also the conditions in which the highest concentrations of DBPs would be generated with free chlorine, the two disinfectants seem complementary in this regard. However, in this research, PAA + LA was used. The high required PAA residuals (and as such forty times higher LA dosage) and the expected application of water refreshing to some degree make it at least questionable whether there is any cost-effective advantage of using the combined disinfectant instead of solely PAA (and replace the lactic acid with e.g. food grade HCl for acidification). The comparison of PAA + LA and free chlorine can be extended to other relevant fresh(−cut) produce wash water disinfectants such as chlorine dioxide and ozone by studying these disinfection techniques according to the same experimental setup. As such, this methodology can be used as a comparative tool based on two important process variables: necessary disinfectant residual and disinfectant demand.

5. Conclusion

Models were made to understand both the process water recycling and wash water disinfection processes with PAA + LA. Application of the same methodology on free chlorine (Van Haute et al., 2013) allowed comparison of both disinfectants. The behavior of PAA (+LA) during wash water disinfection is relatively slow compared to free chlorine disinfection, i.e. slower in reaction with water matrix constituents and a slower inactivation of *E. coli* O157. PAA (+LA) seems a better choice than free chlorine for disinfecting wash water with a high physicochemical load. Addition of low PAA dosages (compared to high free chlorine dosages) during process water recycling can achieve sufficient *E. coli* O157 inactivation when long contact times are applied. However, for wash water disinfection, a higher PAA residual is necessary to achieve rapid microbial inactivation and therefore the impact of water refreshing on the necessary disinfectant dose is higher in the case of PAA (+LA) than of free chlorine. The described methodology can be used to study and compare other water disinfectants in fresh(−cut) produce washing operations and as such be useful for industry and governmental agencies.

Acknowledgments

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