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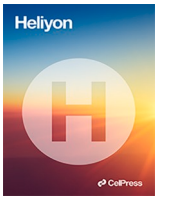

Inactivation kinetics of a surrogate yield conservative predictions of foodborne pathogen reductions from low water activity foods of varying size and composition during low-temperature steam processing

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ABSTRACT

There is a growing interest in using models to predict foodborne pathogen inactivation as a way to validate or verify preventive controls. Unlike liquid foods, solid, low water activity foods (LWAF) are heterogenous in composition and structure and do not transfer heat uniformly. Using models constructed from one food to predict pathogen inactivation on another LWAF is complex and may not always be possible, even if the foods have similar composition. Using models constructed from inactivation kinetics of three foodborne pathogens and a surrogate from vacuum-steam-pasteurized (72 and 82 °C) whole macadamia nuts and dried apricot halves, 3-log reductions were predicted for the same pathogens and foods of reduced size. Model fits (First-order, Weibull, and Gompertz) were significantly impacted by the food type regardless of particle size. Despite the foods being identical in composition with particle size as the only altered characteristic, best-fit models accurately predicted the 3-log reductions only 50% of the time, but the surrogate inactivation models provided conservative predictions for pathogen reductions, highlighting that a surrogate's model may be a suitable tool for predicting pathogen reduction on LWAFs.

1. Introduction

Low-water activity foods (LWAF) are responsible for hundreds of recalls in the United States and Europe each year, as well as dozens of outbreaks of associated foodborne illnesses ([1,2–9]; [10]; [11,12,13,14]). While *Salmonella enterica* subsp. *enterica* is responsible for many of these recalls and outbreaks, *Shiga* toxin-producing *Escherichia coli* (STEC) and *Listeria monocytogenes* are also associated with numerous recalls. While LWAF are not typically considered high-risk foods due to the lack of available water for microbial growth, the ability of pathogens to persist for months to years on LWAF requires additional investigation of associated risks [15–18].

The Risk-Based Preventive Controls for Human Food rule of the Food Safety Modernization Act (FSMA) requires implementation of preventive controls to address pathogen contamination [19]. In-plant validation that utilizes *in situ* processing conditions is the best

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method for evaluation of preventive controls. However, these studies are costly and require specific expertise. It has been proposed that predictive microbiology modeling may inform or, in specific cases, serve as a substitution for in-plant validations. However, limitations of models must be recognized and understood for proper use of these empirical kinetic models [20,21]. Thermal interventions used on LWAF may be limited, since they are heterogenous in composition and do not transfer heat as efficiently as liquid foods.

Primary models should only be applied under the same circumstances under which it was originally developed to describe bacterial inactivation. For example, applying model predictions to a different food, pathogen, or treatment could render predictions irrelevant and unreliable. The objective of this study was to explore the application of primary kinetic models for thermal inactivation of *Salmonella*, Shiga toxin-producing *Escherichia coli* (STEC), and *Listeria monocytogenes*, as well as a surrogate microorganism (*Pediococcus acidilactici*) on vacuum-steam-pasteurized whole macadamia nuts and dried apricot halves at different treatment temperatures (72 or 82 °C). Predictions of 3-log reductions were made from these inactivation models and compared against observed 3-log reductions of the same microorganisms on smaller particles (pieces) of the same vacuum-steamed foods, limiting the experimental variabilities' impacts on the model applications. Additional consideration was given to determine if the model predictions of the surrogate's inactivation (*Pediococcus acidilactici*) could yield similar or more conservative predictions for times necessary to reduce pathogens from low-water activity foods treated with vacuum-steam processing.

2. Materials & methods

In two sets of experiments (replicated at least three times), large pieces (whole macadamia nuts [n = 15], dried apricot halves [n = 17]) and smaller pieces (macadamia nut pieces [n = 17], dried apricot pieces [n = 17]) were inoculated with microorganisms and treated with low-temperature vacuum-assisted steam, and inactivation rates were determined.

Macadamia nut pieces (5–10 mm) and dried apricot halves (Nuts.com, Cranford, NJ) were inoculated with a cocktail of the following: *E. coli* O121:H19 (FNW19M81, wheat flour isolate from 2016 outbreak [FDA]) and O157:H7 F4546 (alfalfa sprout isolate from outbreak); *L. monocytogenes* 1/2a FSL R2-499 (sliced turkey isolate), 1/2b FSL R2-502 (chocolate milk isolate), and 4b (ScottA, milk isolate from 1985 outbreak); *Salmonella enterica* serotype Montevideo (1449, black pepper isolate from 2010 outbreak), Newport (2010 allspice isolate), and Tennessee (K4643, peanut butter clinical isolate from 2007 outbreak); and the nonpathogenic bacterium, *Pediococcus acidilactici* (ATCC 8042). Overnight cultures were prepared in Tryptic Soy Broth (TSB; BD, Sparks, MD) or Lactobacillus MRS broth (*Pediococcus*; BD), streaked onto Tryptic Soy Agar as lawns (TSA; BD), harvested with 0.1% peptone (BD Difco), mixed, and misted onto the whole macadamia nuts and dried apricot halves so that final concentration on the food products was ca. 7–8 log CFU/g after drying. For apricot pieces, the halves were inoculated, dried, and then cut into pieces (8–10 mm) to avoid inoculum internalization through freshly cut surfaces. Additional details for this method were described by Acuff et al. [22].

A laboratory-scale, low-temperature, vacuum-assisted steam delivery system was engineered in a Biosafety Level-2 pilot plant. Newkirk et al. [23] reported the general design and use of the system, which included a canner cooker (25-QT canner cooker; All American®, 25 QT. #925, Manitowoc, WI) with a customized, valve-controlled steam inlet. Amendments were made to connect the system to a medium-pressure steam line and a manually regulated reducing station that lowered steam to <69 kPa [22].

Foods were steam treated (30 g) in modified silicone baking cups (Wilton, Stock 415–9424, Naperville, IL). Pieces of nuts and dried fruits were treated at 72 °C for 0, 0.5, 1, 2, 5, 8, 14, and 20 min and 82 °C for 0, 0.5, 1, 1.5, 2, 2.5, 3.5, and 5 min. Previously, apricot halves were steam treated for the same times and temperatures, but whole macadamia nuts were treated for up to 38 min at 72 °C, and up to 12 min at 82 °C to achieve significant log reductions [22]. Following the steam treatments, the 30-g samples were placed in chilled 0.1% peptone, serially diluted, and plated onto Bile Esculin Agar (BEA; Criterion, Hardy Diagnostics, Santa Maria, CA) for the enumeration of *Pediococcus acidilactici* and on two plates of TSA that were overlayed with a layer of selective agar after 4 h of incubation to recover injured pathogen cells (Xylose Lysine Tergitol-4, Criterion, Hardy Diagnostics, Santa Maria, CA) for *Salmonella* and STEC enumeration, and Modified Oxford agar (Acumedia, Neogen, Lansing, MI) with supplement (Dalynn Biologicals, Calgary, AB) for *L. monocytogenes* enumeration. Overlayed TSA plates and BEA plates were incubated at 35 °C for 24 and 48 h, respectively.

Log reductions were calculated from initial and final bacterial concentrations (log CFU/g) of each treatment and transformed to convey survival, relative to starting populations. Datasets of survivability for each of the pathogens treated with both temperatures on the whole macadamia nuts and apricot halves were used to evaluate the fit of three models that have been used to describe bacterial inactivation on foods: first-order, Weibull, and Gompertz. Replicates of each treatment were pooled and the nlstools and nlsMicrobio packages in R (Version 3.6.1) and RStudio, Inc. (Version 1.1.456, Boston, MA) were used for model construction.

The first-order model parameters were estimated from the compiled data [24]:

$$log\ N_t - log\ N_0 = -kt \tag{1}$$

where N_t is the surviving bacterial population at time, t; N_0 is the initial bacterial population; and k is the inactivation rate (log CFU/g min^{-1}).

The Weibull model was used as an alternative to identify inactivation trends that were not linear [24]:

$$log\ N_t - log\ N_0 = -k \bullet t^{\beta} \tag{2}$$

where $k = k^{'}/\log_e 10$ and is a rate parameter for the reduction, and β indicates whether the shape is a concave upward (<1), downward (>1), or linear (=1) inactivation curve.

The Gompertz model describes inactivation trends that present as a sigmoidal curve with the following modified equation [25]:

Table 1
First-order (k [log CFU/g min^{-1}]), Weibull (k (log CFU/g min^{-1}) and β) and Gompertz (A, μ_M [log CFU/g min^{-1}], λ [min]) model parameter coefficients and statistical results (RMSE [log CFU/g], AIC) for STEC, *Salmonella*, *L. monocytogenes*, and *Pediococcus acidilactici* on steamed dried apricot halves and pieces and whole macadamia nuts and pieces treated (72 and 82 °C).

Food	Temp	Bacteria	First-Order				Weibull				Gompertz				
			k	*D*-val	RMSE	AIC	*k*	*β*	RMSE	AIC	*A*	μ_M	*λ*	RMSE	AIC
Apricot Halves	72	*E. coli*	−0.41	2.43	1.00	70.96	−0.96	0.69	0.79	60.85	−7.22	−0.62	0.54	**0.78**	**60.78**
		L. mono	−0.40	2.53	1.32	84.46	−1.24	0.58	0.98	70.89	−6.41	−0.70	0.23	**0.92**	**68.66**
		Ped.	−0.21	4.70	0.80	60.21	−0.71	0.55	**0.64**	**50.62**	−3.67	−0.27	−1.00	0.71	56.64
		Sal.	−0.44	2.29	1.45	89.01	−1.39	0.57	1.04	73.99	−7.13	−0.79	0.36	**0.88**	**66.91**
	82	*E. coli*	−1.57	0.64	1.35	85.66	−2.15	0.75	1.28	83.75	−6.45	−4.33	0.99	**0.96**	**71.04**
		L. mono	−1.51	0.66	1.22	80.51	−2.19	0.70	1.09	75.95	−6.07	−3.18	0.63	**0.89**	**67.23**
		Ped.	−1.16	0.86	0.82	61.68	−1.60	0.74	0.75	58.18	−5.38	−1.50	0.15	**0.73**	**57.65**
		Sal.	−1.57	0.64	1.43	88.24	−2.37	0.67	1.27	83.58	−5.94	−11.07	1.34	**0.80**	**62.03**
Whole Macada-mia Nuts	72	*E. coli*	−0.18	5.44	0.85	60.96	−0.69	0.60	**0.64**	**48.43**	−6.68	−0.21	−1.19	0.78	58.66
		L. mono	−0.14	7.29	0.67	49.60	−0.69	0.51	**0.40**	**26.99**	−4.81	−0.15	−2.26	0.55	42.61
		Ped.	−0.10	10.32	0.69	51.30	−0.46	0.53	**0.61**	**46.36**	−3.66	−0.10	−2.69	0.67	51.70
		Sal.	−0.13	7.54	0.68	50.51	−0.59	0.55	**0.48**	**35.89**	−4.59	−0.16	−1.53	0.60	46.31
	82	*E. coli*	−0.54	1.85	1.14	83.72	−1.23	0.63	0.98	**76.49**	−5.20	−1.00	0.49	**0.96**	76.62
		L. mono	−0.52	1.94	0.99	73.57	−1.21	0.61	0.78	62.54	−5.01	−0.94	0.43	**0.76**	**62.04**
		Ped.	−0.37	2.73	0.76	62.49	−0.73	0.68	**0.69**	**58.63**	−3.71	−0.57	0.34	0.72	61.81
		Sal.	−0.44	2.27	0.95	73.95	−0.86	0.70	**0.88**	**71.03**	−4.48	−0.68	0.35	0.92	74.13
Apricot Pieces	72	*E. coli*	−0.39	2.54	0.92	88.73	−0.77	0.75	**0.83**	**82.70**	−7.88	−0.49	0.33	0.87	86.83
		L. mono	−0.38	2.65	1.03	95.42	−1.00	0.64	0.77	77.86	−6.33	−0.60	0.30	**0.75**	**77.45**
		Ped.	−0.21	4.81	0.59	59.74	−0.31	0.86	**0.58**	**60.41**	−5.26	−0.23	0.70	0.61	64.24
		Sal.	−0.39	2.59	0.94	86.98	−0.95	0.67	**0.71**	**71.09**	−6.92	−0.54	0.17	0.74	73.74
	82	*E. coli*	−1.60	0.62	1.26	105.49	−1.89	0.87	1.26	106.14	−6.78	−4.04	0.99	**0.92**	**87.50**
		L. mono	−1.56	0.64	1.45	113.86	−2.14	0.75	1.37	111.38	−6.00	−10.62	1.37	**0.62**	**63.47**
		Ped.	−1.11	0.90	0.84	77.75	−1.16	0.97	0.85	79.66	−4.75	−2.39	0.91	**0.71**	**69.78**
		Sal.	−1.60	0.62	1.38	110.75	−2.13	0.77	1.31	108.63	−6.28	−7.74	1.29	**0.70**	**70.31**
Macada-mia Nut Pieces	72	*E. coli*	−0.33	3.01	0.85	73.51	−0.89	0.64	**0.68**	**61.85**	−11.12	−0.31	−0.82	0.79	71.13
		L. mono	−0.24	4.11	0.93	78.49	−1.15	0.43	**0.50**	**44.29**	−3.87	−0.39	−1.28	0.64	58.92
		Ped.	−0.24	4.23	1.04	84.86	−1.11	0.43	**0.72**	**64.63**	−3.80	−0.37	−1.28	0.82	72.93
		Sal.	−0.27	3.69	0.89	75.78	−1.08	0.49	**0.57**	**51.55**	−6.00	−0.25	−2.87	0.70	64.66
	82	*E. coli*	−1.30	0.77	0.99	79.30	−2.15	0.61	**0.77**	**66.39**	−5.44	−1.85	0.01	0.81	70.07
		L. mono	−1.41	0.71	1.20	92.86	−2.42	0.57	**0.94**	**79.65**	−5.75	−2.03	−0.01	0.96	82.12
		Ped.	−0.80	1.25	0.84	72.69	−1.50	0.50	0.64	58.32	−3.03	−1.55	0.18	**0.59**	**54.35**
		Sal.	−1.03	0.97	1.07	86.32	−1.91	0.51	**0.86**	**74.71**	−4.02	−1.55	−0.07	0.89	77.78

Best fit values are bolded.

$$log\ N_t - log\ N_0 = A \bullet exp\left\{ - exp\left[\frac{\mu_M \bullet e}{A}(\lambda - t) + 1\right]\right\} \quad (3)$$

where μ_M (log CFU/g min^{-1}) is the maximal value of inactivation, λ is the lag time (min), and *A* is the asymptote.

Akaike's Information Criteria (AIC) and root mean squared error (RMSE) were used to evaluate model fit of all datasets, with lower values indicating better fit.

The objective of this study was to determine if inactivation models from whole food particles (macadamia nuts and apricot halves) could predict inactivation times for smaller pieces of the same foods, so times necessary for 3-log reductions were predicted from inactivation data of larger pieces [22]. These "predicted" times were compared against the actual "observed" times for 3-log reductions of bacteria on pieces (predicted vs. observed). Raw error (min) was calculated as the difference between the predicted and observed times, while relative error was the ratio of the raw error to the predicted times of 3-log reductions on smaller pieces of the same food [26]:

RE = (observed – predicted)/predicted

The relative error determined accuracy of model predictions: acceptable (−0.30-0.15), fail-safe (−1.0 to −0.31), fail-dangerous (>0.15). These ranges were based on previously used intervals for model evaluation [26]. While fail-safe predictions between −1.0 and −0.30 were not considered accurate, they were considered excessively safe for predicted times necessary for 3-log reductions.

Heat maps of the relative error between each combination of model predictions of 3-log reduction times were constructed to visualize trends between predictive and observed times (R [Version 3.6.1, library[pheatmap]). The maps indicated with color gradients the accuracy model predictions by comparing predicted (vertical axis) and observed (horizontal axis) values in the context of the fail-safe (green) and fail-dangerous (red) intervals.

3. Results

Vacuum-steam processing treatments (72 and 82 °C) of macadamia nuts (whole and pieces) and dried apricots (halves and pieces) successfully inactivated 3–5 log CFU/g of STEC, *L. monocytogenes*, and *Salmonella* spp. Log-linear, first-order models were used to calculate *D*-values of each microorganism on each food for the two treatment temperatures (Table 1). Pathogen $D_{72°C}$-values were

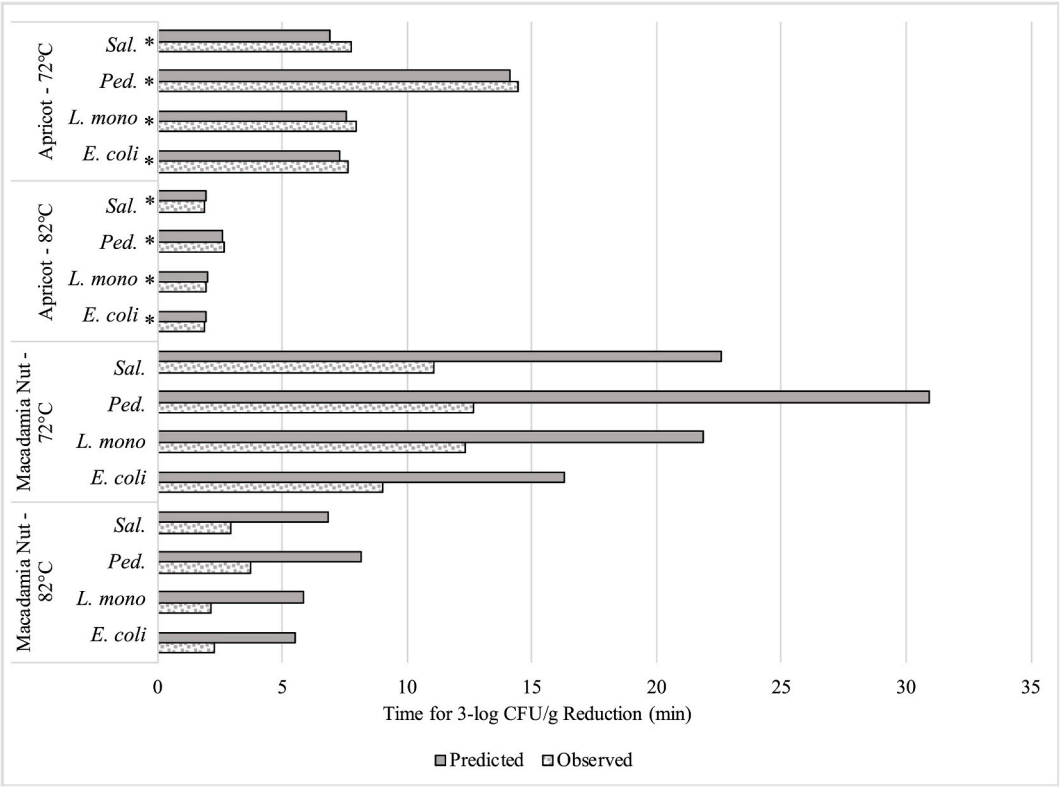

Fig. 1. Predicted and observed times from first-order models of microbial inactivation for macadamia nuts and pieces and apricot halves and pieces. Comparisons of predicted vs. observed times for 3-log reductions were made by first-order models of *Salmonella*, *Pediococcus*, *L. monocytogenes*, and *E. coli* on whole macadamia nuts (predicted) vs. macadamia nut pieces (observed) and apricot halves (predicted) vs. apricot pieces (observed) vacuum-steamed at 72 and 82 °C. * indicates relative error values within the acceptable range (−0.30-0.15).

between 2.3 and 2.6 min (apricots halves and pieces) and 3.0–7.5 min (macadamia nut pieces and whole nuts), and $D_{82^{\circ}C}$-values for apricot and macadamia nut samples were 0.6 min (apricot) and 0.7–2.7 min, respectively. The *D*-values of *Pediococcus acidilactici* were larger than those of the pathogens for all treatments on each food type and size, highlighting its suitability as a surrogate for these processing conditions. Nonlinear primary inactivation models of vacuum-steam-treated macadamia nuts and dried apricots (whole and pieces) were constructed (Table 1). For both dried apricot halves and pieces, the Gompertz model, in general, best described the bacterial inactivation for each bacterium based on RMSE and AIC. Alternatively, bacterial inactivation models for whole macadamia nuts and pieces were best fit by a Weibull model with upward concavity. The fits of first-order models for both food types were not vastly different from those of the Weibull and Gompertz, but they were not ranked as best fit in any case.

Models from the larger food particles (whole macadamia nuts, dried apricot halves) were used to predict times for 3-log bacterial reductions, which were compared against the observed times required for 3-log bacterial reductions on the corresponding smaller food particles (macadamia nut pieces, dried apricot pieces) (Figs. 1–3). Raw and relative errors were determined (Figs. 1–3 and Supplemental Tables 2–4). Inactivation models for apricot halves had greater prediction accuracy than those of macadamia nuts overall. First-order models of dried apricot halves were accurate predictors of 3-log reductions of respective bacteria on apricot pieces 100% of the time (Fig. 1, Supplemental Table 2), Weibull models accurately predicted reductions on apricot pieces 63% of the time (Fig. 2, Supplemental Table 3), and Gompertz models accurately predicted times for 3-log reductions only 50% of the time (Fig. 3, Supplemental Table 4). All 3-log reduction predictions from macadamia nut pieces were in the fail-safe interval for reductions on macadamia nut pieces (<-0.30; Figs. 1–3, Supplemental Tables 2–4).

Heat maps displayed the relative error calculated from each model combination to visually highlight comparisons in the predicted and observed values (Figs. 4–6), showing fail-safe (green) to fail-dangerous (red) relative error values regardless of model fit. At both 72 and 82 °C, first-order model predictions between apricot halves, apricot pieces, and macadamia nut pieces were relatively similar and showed that times required to reduce bacteria by 3-log were close to one another, yielding small relative error (evident by green-white colors, Fig. 4). Red-orange colors were primarily associated with whole macadamia nut models and *Pediococcus* models, both of which required longer steam treatments for 3-log reductions and subsequently had high relative error. Heat maps of predicted values from Weibull and Gompertz models (Figs. 5 and 6) had similar trends and groupings, though with slightly less accuracy than what was observed from the first-order models.

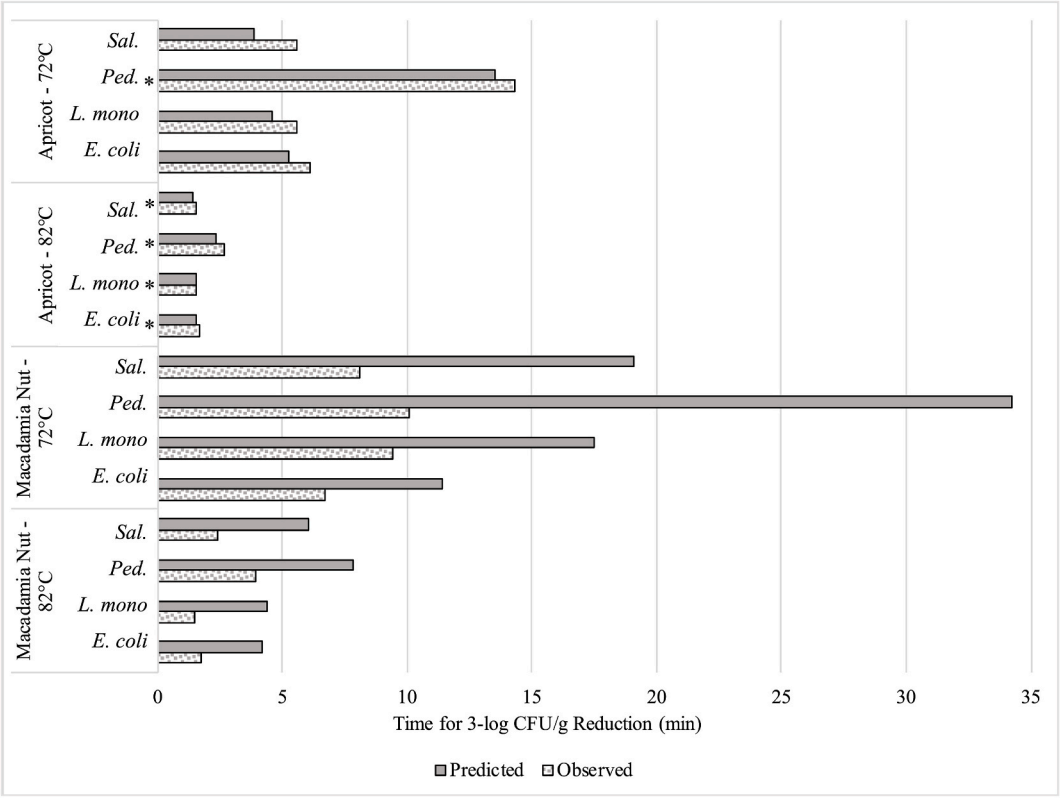

Fig. 2. Predicted and observed times from Weibull models of microbial inactivation for macadamia nuts and pieces and apricot halves and pieces. Comparisons of predicted vs. observed times for 3-log reductions were made by Weibull models of *Salmonella*, *Pediococcus*, *L. monocytogenes*, and *E. coli* on whole macadamia nuts (predicted) vs. macadamia nut pieces (observed) and apricot halves (predicted) vs. apricot pieces (observed) vacuum-steamed at 72 and 82 °C. * indicates relative error values within the acceptable range (−0.30-0.15).

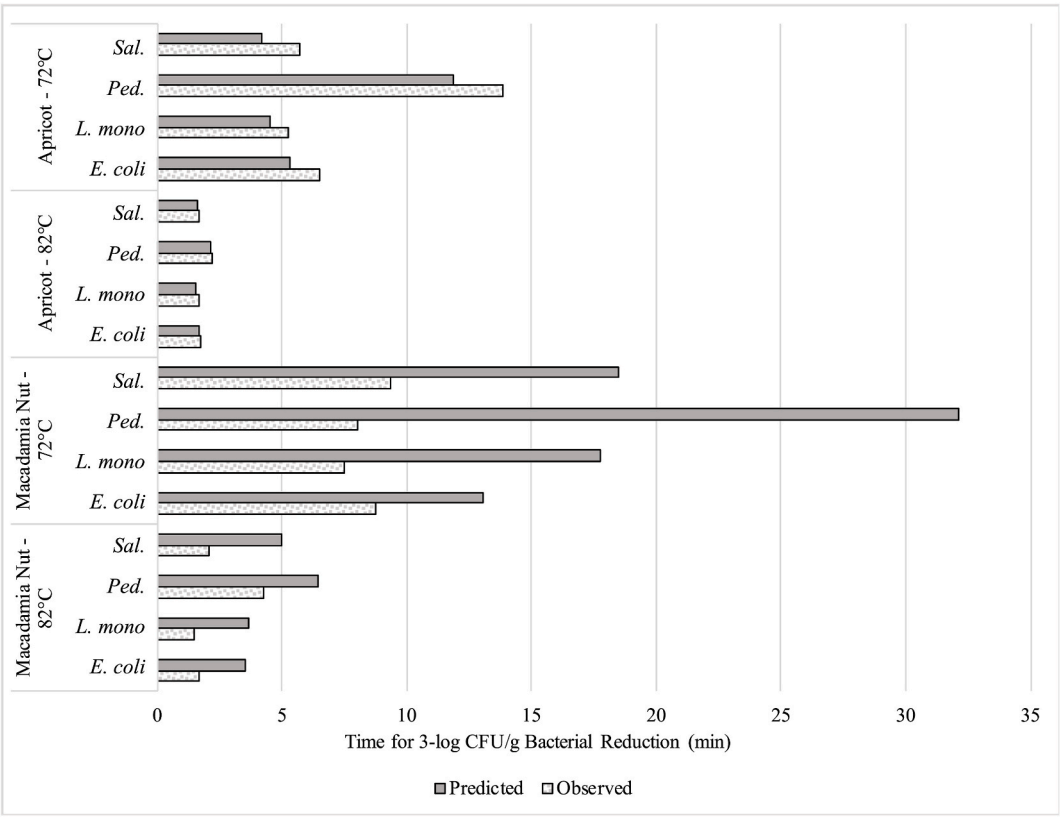

Fig. 3. Predicted and observed times from Gompertz models of microbial inactivation for macadamia nuts and pieces and apricot halves and pieces. Comparisons of predicted vs. observed times for 3-log reductions were made by Gompertz models of *Salmonella*, *Pediococcus*, *L. monocytogenes*, and *E. coli* on whole macadamia nuts (predicted) vs. macadamia nut pieces (observed) and apricot halves (predicted) vs. apricot pieces (observed) vacuum-steamed at 72 and 82 °C. * indicates relative error values within the acceptable range (−0.30-0.15).

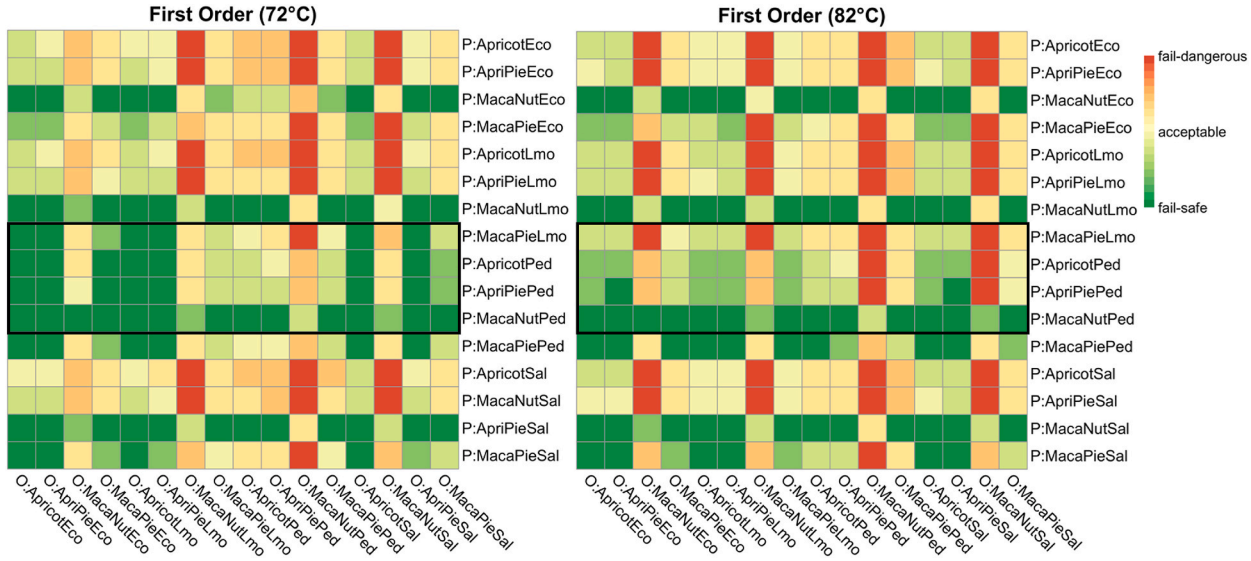

Fig. 4. Heat maps of relative error between predicted and observed times from first-order models of microbial inactivation. Predicted (vertical) and observed (horizontal) times were those required for 3-log reductions of microorganisms based on first-order model parameters 72 and 82 °C treatments. The acceptable relative error ranged from −0.30 (fail-safe; green) to 0.15 (fail-dangerous; red). Nomenclature of each line dictates predicted or observed, type of food, and microorganism (e.g., P:MacaPieSal is predicted time for 3-log reduction of *Salmonella* on macadamia nut pieces.). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

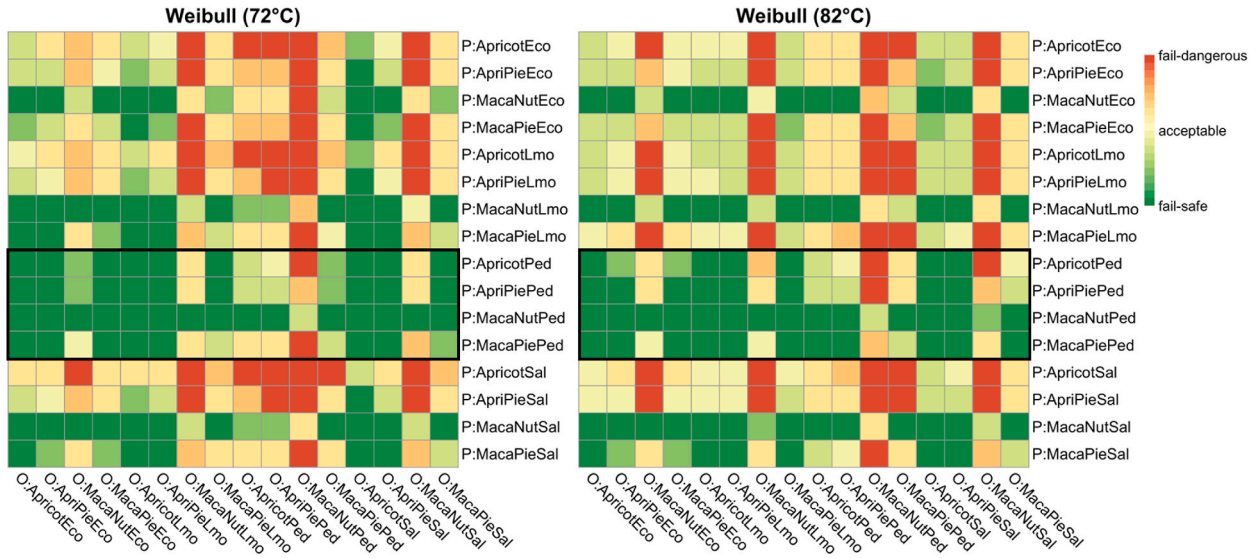

Fig. 5. Heat maps of relative error between predicted and observed times from Weibull models of microbial inactivation. Predicted (vertical) and observed (horizontal) times were those required for 3-log reductions of microorganisms based on Weibull model parameters 72 and 82 °C treatments. The acceptable relative error ranged from −0.30 (fail-safe; green) to 0.15 (fail-dangerous; red). Nomenclature of each line dictates predicted or observed, type of food, and microorganism (e.g., O:MacaNutLmo is observed time for 3-log reduction of *L. monocytogenes* on whole macadamia nuts.). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

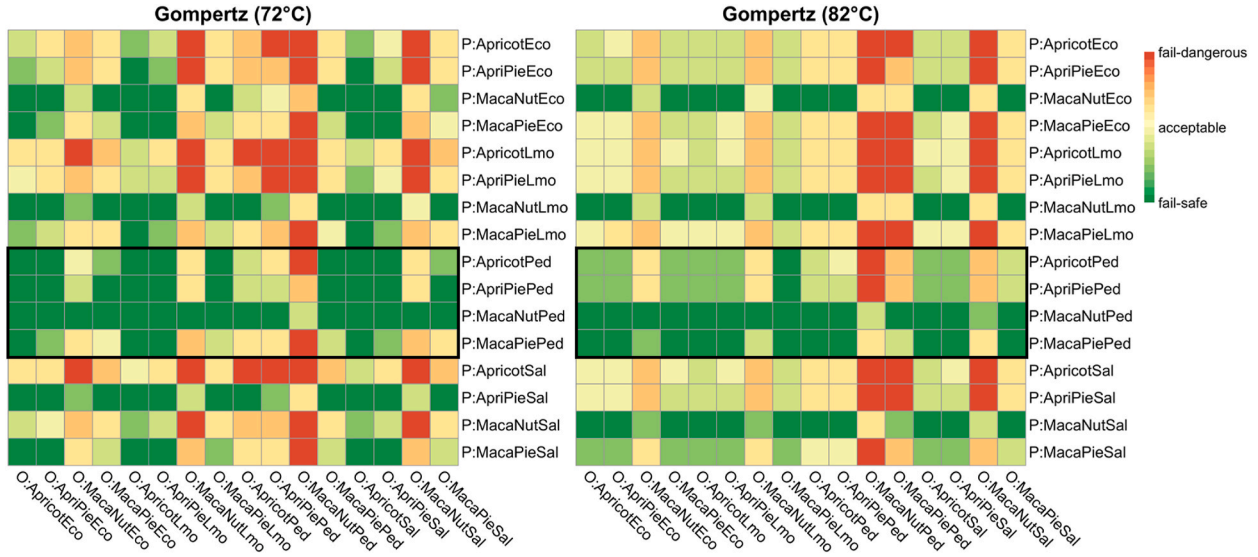

Fig. 6. Heat maps of relative error between predicted and observed times from Gompertz models of microbial inactivation. Predicted (vertical) and observed (horizontal) times were those required for 3-log reductions of microorganisms based on Gompertz model parameters 72 and 82 °C treatments. The acceptable relative error ranged from −0.30 (fail-safe; green) to 0.15 (fail-dangerous; red). Nomenclature of each line dictates predicted or observed, type of food, and microorganism (e.g., O:ApriPieEco is observed time for 3-log reduction of *E. coli* on apricot pieces.). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

4. Discussion

The heterogeneity of the low-water activity food category thwarts goals of a "one-size-fits-all" model for foods with even minimal differences. For example, wheat flour has been extensively used as the food matrix in bacterial inactivation studies due to recent outbreaks, but model fits for inactivation data vary greatly across studies [18,27,28]. These variances may be due to slight differences in the food matrices used in the study, strain type, water activity level, heating method or other experimental methods. The present study controlled for many of these factors by using the same bacterial strains, foods, and treatment conditions, with only the size of the

particle varying between predicted and observed values. The relative error provided insight into the application of models from large food particles to evaluate if they could be used beyond their original design, which showed that the predictive power of the models was not consistent.

Results indicated that models were conservative and predicted many fail-safe times, but were limited in accuracy in these instances. Table 1 highlights the best fit model with bolded values, which were different for the fruits and nuts. Distinct trends of model fit were observed on the basis of macadamia nut vs. apricot, but trends were not observed based on particle size, microorganism type, or treatment temperature. The heatmaps provided visualization of certain trends. Predicted and observed values were grouped on heatmaps based on microorganism ("Eco, Lmo, Ped, Sal"), which highlighted the success of *Pediococcus* models in predicting conservative times for 3-log reductions for all pathogens (numerous green-white squares, emphasized with black outlines). For example, the Gompertz *Pediococcus* inactivation models for apricot halves predicted fail-safe times of 72 and 82 °C steam treatments for 3-log reductions of every pathogen on apricot halves, apricot pieces, and macadamia nut pieces (Figs. 3 and 6).

Dried apricot halves models, particularly first-order, had significant success in providing accurate or fail-safe estimates for 3-log bacterial reductions on apricot pieces. Whole macadamia nut models were less accurate, but were far more conservative, providing overestimated times (fail-safe) necessary for 3-log reductions on macadamia nut pieces, potentially resulting in a safer but overprocessed product. The reason for excessively longer treatments is not fully understood since the whole macadamia nuts and pieces had comparable composition with size being the only difference (confirmed by analyses). However, the inoculation of the macadamia nut pieces was slightly altered. Whereas the apricot halves were inoculated and then cut into smaller pieces, the macadamia nut pieces were purchased as smaller particles and inoculated without further fabrication for logistical purposes.

Models typically describe single pathogen population inactivation, but LWAF have been recalled for contamination of a variety of pathogens, given their agricultural origin and processing environments. Ergo, it would be advantageous for models to predict inactivation of a surrogate for multiple pathogens. The presented work demonstrated that while the reductions of pathogens were not usually significantly different from one another, the surrogate models could conservatively predict fail-safe times necessary for reductions of all tested pathogens. Models describing pathogen inactivation on low-water activity foods (LWAF) have the additional difficulty characterizing the degree of increased thermal resistance induced by desiccation [29–33]. Additionally, LWAF are a broad category with a variety of compositions, which can reduce thermal treatment efficacy [34–37], Therefore, thermal inactivation processes should target multiple pathogens or a surrogate for broader application.

Criticisms of using predictions from primary models highlight limited application, as inactivation estimates could be inaccurate if variabilities (such as changes to pH, temperature, water activity, heterogenous contamination) are not considered [38,39]. Primary models constructed of whole macadamia nuts and dried apricot halves were generally accurate or excessively conservative in predicting times necessary for 3-log bacterial reductions on their smaller particle counterparts. Moreover, food size and composition impacted inactivation trends and predictions. The information presented in the current study can guide researchers in building secondary models, which are more robust with extensive data collection to describe the effects of processing conditions and intrinsic factors (pH, water activity, particle size, composition) on the model parameters [20,40]. In conclusion, using models that predict a validated surrogate's inactivation would be a prudent approach for LWAF processors designing thermal inactivation treatments.

5. Conclusions

Low-temperature vacuum-assisted steam processing is effective in reducing pathogens from macadamia nuts, dried apricot halves, and their respective pieces. Model fits were largely impacted by the food composition, and possibly particle size in the case of macadamia nuts. Furthermore, comparing predictions and observations of 3-log bacterial reductions from each model emphasized the utility of a surrogate. If the surrogate is validated properly, its model may offer increased applicability for designing processes that inactivate multiple target pathogens.

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Author contribution statement

Jennifer C Acuff: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper. Kim Waterman, Jian Wu, Claire Marik Murphy: Performed the experiments; Wrote the paper. Daniel Gallagher: Analyzed and interpreted the data; Wrote the paper. Monica A Ponder: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Data availability statement

Data included in article/supp. material/referenced in article.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2023.e17893.

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