



# Use of superheated steam to inactivate *Salmonella enterica* serovars Typhimurium and Enteritidis contamination on black peppercorns, pecans, and almonds

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## ABSTRACT

This study investigated the inactivation efficacies and kinetics of superheated steam (SHS) treatment of black peppercorns, pecans, and almonds inoculated with a cocktail of *Salmonella* Typhimurium and *Salmonella* Enteritidis. According to *D*- and *z*-values determined from the Weibull model, *Salmonella* inactivation efficacy increased as the steam temperature increased, and the sensitivity to changes in the SHS temperature was the highest for black peppercorns, followed by pecans and almonds, which is attributable to the differences in the surface properties of the foods. Furthermore, *Salmonella* inoculated on black peppercorns, pecans, and almonds was completely inactivated within 3, 13, and 8 s, respectively, by the 180 °C SHS treatment, and the moisture content, color, and texture did not deteriorate in any sample. Consequently, these data provide the basis for the application of SHS treatment to inactivate *Salmonella* on low-moisture foods without a reduction in quality.

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## 1. Introduction

Black pepper is one of the most popular spices used as a seasoning due to its flavoring properties (Zweifel and Stephan, 2012). Globally, almonds are second after cashew nuts in tree nut production, with 2,560,000 tons being produced in 2010 (Ng et al., 2014). The consumption of pecans has also increased because of their nutrient content. These three commodities all have low moisture contents and are frequently used in ready-to-eat foods without further cooking, which contributes to the survival of *Salmonella* on contaminated foods before ingestion. As a result, various salmonellosis outbreaks have frequently occurred as the

popularity of these commodities increases. For example, black peppers resulted an outbreak in 2010, and 272 persons in 44 states and Washington, DC, became ill (Jeong and Kang, 2014). *Salmonella*-contaminated pecans were recalled in 2010 (FDA, 2010), 2014, and 2015 (FDA, 2015), although the pecans did not induce an outbreak. Additionally, salmonellosis outbreaks from almonds have caused many problems in different nations (Isaacs et al., 2005). Specifically, 205 cases of salmonellosis were associated with consumption of whole raw almonds in the United States and Canada. Among the different *Salmonella* serovars, *S. Enteritidis* and *S. Typhimurium* are the two primary etiologic agents of salmonellosis worldwide (Carrasco et al., 2012). Therefore, *S. Enteritidis* and *S. Typhimurium* contamination on black peppers, pecans, and almonds must be inactivated before their distribution and sale.

*Salmonella* inactivation by conventional thermal processing is not commonly used on black peppers, pecans, and almonds because the volatile flavor components in spices and the inner

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structures of tree nuts are sensitive to heat (Abou-Elkhair et al., 2014; Pan et al., 2012). Although several alternative non-thermal processing methods, such as cold plasma, high pressure, and electron beam irradiation, have been suggested for the reduction of *Salmonella* populations on almonds, there are several constraints of the suggested methods, such as high installation cost, requirement for additional drying step, and significant loss of sensory quality during further storage (Pan et al., 2012; Sánchez-Bel et al., 2008). In cases of spices including black peppercorns, methods of irradiation and fumigation with ethylene oxide have been developed to inactivate the microbial loads. However, the use of ethylene oxide is restricted due to the carcinogenic properties, and irradiation processing also has a problem with poor consumer acceptance, even though this method of *Salmonella*-decontamination of dried spices is permitted in many countries (Kim et al., 2012). Meanwhile, various techniques approved by the Food and Drug Administration have been introduced to control *Salmonella* on black peppercorns, pecans, and almonds, including treatments with steam, hot water, hot oil, and propylene oxide. Nevertheless, these techniques have disadvantages, such as a long treatment time, quality deterioration, and the requirement of an additional drying step (Pan et al., 2012). Therefore, successful *Salmonella* inactivation techniques are required to overcome these disadvantages.

Superheated steam (SHS) is a steam that has additional sensible heat energy that increases the temperature above the saturation point at a given vapor pressure (Cenkowski et al., 2007). SHS has various merits over other thermal treatments, including a fast heat transfer rate due to condensation and gas radiation, an accelerated drying rate, and an oxygen-free environment (Bari et al., 2010). The procedure for SHS treatment for reducing bacteria on the foods is divided as follows. In the first stage, SHS in contact with the foods is condensed due to the temperature difference between the SHS and the foods, and the condensed water surrounds the surface of the foods (Konishi et al., 2004). During this stage, the temperature of the surface rises to approximately 100 °C, and heat from the SHS is conducted to the surface and inactivates a portion of bacteria on the surface. Next, the water surrounding the food surface absorbs the additional heat from steadily flowing SHS and evaporates from the outer layer of the surface (Iyota et al., 2001). During this stage, most of the heat added from the SHS flow is consumed not to increase the temperature of the foods but to evaporate the water, and the temperature of the surface of the foods is maintained approximately 100 °C due to the evaporation of the water. After all of the condensate has evaporated, the surface of the foods re-contacts the SHS flow and absorbs the sensible heat from the SHS. Thereby, the SHS flow transfers convective heat to the food surface and raises the surface temperature to more than 100 °C and to the temperature of the SHS. At the same time, the foods are steadily dried at a constant rate (Chen et al., 2000), and bacterial contamination on the surface of the foods is directly contacted by the SHS without any inhibition by the condensed water. Consequently, SHS has strong potential to eliminate pathogens contaminating food surfaces, regardless of the irregular surface structure of the foods (Cenkowski et al., 2007).

Despite the merits of SHS on the reduction of contaminated bacteria on food surfaces, the inactivation studies of foodborne pathogenic bacteria using SHS have been rare, and there were only studies of almonds (Ban and Kang, 2016; Bari et al., 2010). Moreover, in the reported studies for the inactivation of *Salmonella* on almonds using SHS, the inactivation kinetics and mechanisms of the SHS treatment remained unclear, and the use of SHS to inactivate *Salmonella* on black peppercorns and pecans has not been reported to date. In this study, SHS was used to inactivate a cocktail of *S. Typhimurium* and *S. Enteritidis* contamination on the surfaces of black peppercorns, pecan halves, and raw almond kernels.

*Salmonella* inactivation kinetics for SHS as a function of temperature and time were compared with the inactivation kinetics for saturated steam (SS). Furthermore, the effect of the steam treatment on quality maintenance was evaluated by measuring the moisture content and color of all samples, the piperine content of black peppercorns, and the texture of almonds.

## 2. Materials and methods

### 2.1. *Salmonella* strains and inoculum preparation

*Salmonella* Typhimurium (ATCC 19585) and *S. Enteritidis* (PT 30 ATCC BAA-1045 and NCCP 12236) were obtained from the bacterial culture collection at Seoul National University (Seoul, Korea). Stock cultures were stored at –80 °C in 0.7 mL of tryptic soy broth (TSB; Difco, Becton Dickinson, Sparks, MD, USA) and 0.3 mL of 50% (v/v) glycerol.

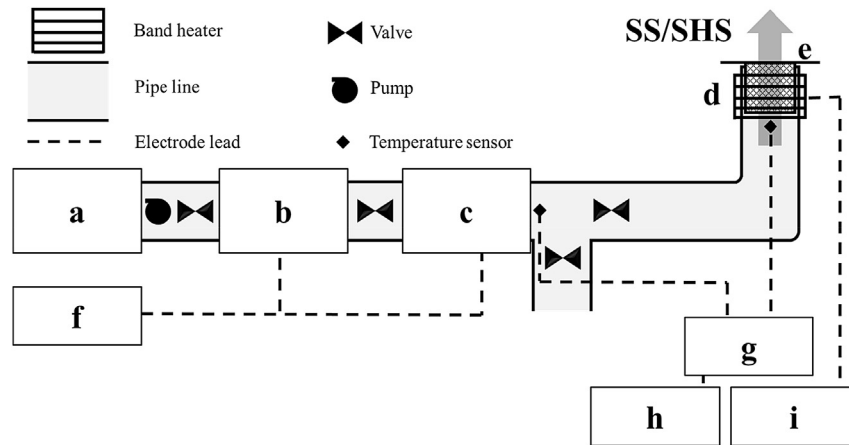
An inoculum of the *Salmonella* cocktail used in this study was prepared using a modification to a previously described method (Pao et al., 2006). Fifty microliters of the thawed stock cultures were transferred to 5 mL of TSB and incubated at 37 °C for 24 h. Next, these cultures were streaked onto tryptic soy agar (TSA; MB Cell, Los Angeles, CA, USA), incubated at 37 °C for 24 h, and stored at 4 °C. One colony proliferated on TSA was transferred to 5 mL of TSB and incubated at 37 °C for 24 h. After incubation, 0.2 mL of each culture was spread onto TSA (0.2 mL per plate) and incubated at 37 °C for an additional 24 h to produce a bacterial lawn. The three types of *Salmonella* lawns were harvested using a sterile plastic spreader with sterilized peptone water (2 mg mL<sup>-1</sup>; 9 mL per plate) and mixed thoroughly to obtain 27 mL of a homogeneous inoculum with a cell density of 9.73 ± 0.12 log colony-forming units (CFU) mL<sup>-1</sup>, which was later confirmed by plate counts on xylose lysine decarboxylase agar (XLD; Difco, Becton Dickinson, Sparks, MD, USA) to prepare the inoculum used to inoculate samples.

### 2.2. Sample preparation and inoculation

Black peppercorns, pecans, and almonds were purchased at a local market (Seoul, Korea), absence of contamination with *Salmonella* was verified, and the materials were subsequently stored at 4 °C before use. Before inoculation, 12 g of black peppercorns, 18 halves of pecans, and 54 kernels of almonds were each placed in a separate sterile polyethylene bag and later warmed to 25 °C for 1 h. The contents of each bag were mixed with the 27 mL of the inoculum of *Salmonella* cocktail, agitated by thorough hand massaging for 2 min to distribute the inoculum uniformly on the samples, and dried overnight at 25 °C until the moisture content (black peppercorns, pecans, and almonds: 12.4 ± 0.3, 4.3 ± 0.3, and 6.0 ± 0.2% dry weight) was not significantly different from the non-inoculated samples. Uniform inoculation levels of 7.34 ± 0.21, 7.24 ± 0.33, and 7.34 ± 0.14 log CFU g<sup>-1</sup> of *Salmonella* on black peppercorns, pecans, and almonds, respectively, were confirmed by plate counts on XLD agar.

### 2.3. Saturated steam and superheated steam treatments

Custom-made SS/SHS decontamination machinery was designed and built as shown in Fig. 1. The machinery consisted of a water reservoir, a 15-kW steam boiler, a 5-kW superheater, and a steam treatment chamber unit connected with an insulated stainless-steel pipeline, including a condensate water bypass line. A steam treatment cell (diameter, 120 mm; height, 155 mm), which was inserted into the insulated steam treatment chamber unit, was made of stainless-steel mesh (pore size, 254 μm) to ensure that the steam flow was not interrupted, and a band heater with



**Fig. 1.** Schematic diagram of the custom-made saturated steam (SS)/superheated steam (SHS) decontamination machinery. (a) Water reservoir, (b) steam boiler, (c) superheater, (d) outer reacting chamber unit, (e) inner reacting cell, (f) power control unit, (g) temperature monitoring system, (h) temperature data processing system, and (i) temperature controller for the band heater.

temperature controller was wrapped around the chamber unit to prevent dew condensation and heat loss. The steam temperatures at the superheater and the treatment chamber were monitored and recorded by separate temperature sensors and a data processing system. Steam was steadily generated at  $3.0 \pm 0.1 \text{ m s}^{-1}$  for all experimental temperature conditions: 100 °C (SS); and 120, 140, 160, and 180 °C (SHS). The steam equipment was pre-operated for at least 2 h to steadily generate the steam at pre-determined temperatures before samples were subjected to the steam treatment. Sufficient sample material was generated to produce a single layer of 2 g of black peppercorns, 3 halves of pecans, or 9 kernels of almonds on the bottom of the steam treatment cell. Next, maintaining the steady generation of steam, the treatment cell was inserted into the treatment chamber and treated with 100, 120, 140, 160, or 180 °C steams for 1–45 s. Particularly during the treatment time, the temperature of the steam at 180 °C was maintained within  $\pm 0.9$  °C (Fig. S1 in the Supplementary Material). After the steam treatment for the pre-determined time, the samples in the treatment cell were immediately removed and later stored in a desiccator at 25 °C prior to the subsequent measurements.

#### 2.4. Surviving cell enumeration

The steam-treated samples were placed into a stomacher bag (Labplas Inc., Sainte-Julie, Quebec, Canada) containing peptone water ( $2 \text{ mg mL}^{-1}$ ) at a 9-fold volume of the sample weight and homogenized using a stomacher for 2 min at speed level 9 (WS-400; Shanghai Zhisun Equipment Co. Ltd., Shanghai, China). After homogenization, 1 mL sample aliquots were serially diluted 10-fold with 9 mL of peptone water ( $2 \text{ mg mL}^{-1}$ ), and 1 mL of the resulting dilutions was later spread onto XLD agar plates. The plates were incubated for  $\geq 18$  h at 37 °C with the expectation that *Salmonella* cells could not resuscitate on XLD because of the injury caused by the steam treatment (Bari et al., 2010). After the incubation, the colonies (CFU) were enumerated.

The entire samples treated with steam that inactivated *Salmonella* below the detection limit ( $1 \text{ log CFU g}^{-1}$ ) were placed into a stomacher bag containing TSB with a 9-fold volume of the sample weight, homogenized using a stomacher for 2 min, and incubated for 24 h at 37 °C to show the complete inactivation of *Salmonella* species inoculated on the samples. After incubation, 1 mL of the sample aliquots was spread onto XLD agar plates and further incubated for 24 h at 37 °C. After this further incubation, we

determined whether the resuscitated *Salmonella* could form black colonies. Consequently, complete inactivation was defined as a lack of black colonies after this enrichment procedure.

#### 2.5. Moisture content, color, and texture measurements

AACC method 44-15a (AACC-International, 1999) was used to measure the moisture content of black peppercorns, pecans, and almonds with oven drying at 105 °C for 24 h. After cooling in a desiccator at 25 °C for 60 min, the samples were weighed, and the moisture content was calculated on a dry weight basis and expressed as % dry weight.

The color of black peppercorns was measured with a portable chromameter (Model CR-400; Konica Minolta Sensing Inc., Osaka, Japan) using the granular materials attachment (CR-A50; Konica Minolta Sensing Inc., Osaka, Japan), whereas the surface color of pecans and almonds was measured at random locations. Chromaticity results were recorded as  $L^*$  (lightness),  $a^*$  (redness), and  $b^*$  (yellowness) (Hunter, 1975).

A compression test was conducted to determine the initiation force required to fracture almonds. The samples were positioned on the center of the base in a texture analyzer (TA-XT2i; Texture Technologies, Scarsdale, NY, USA). The measurement was conducted using an aluminum cylindrical probe (diameter, 50 mm) that was pressed on the sample until it reached 50% deformation at a speed of  $0.5 \text{ mm s}^{-1}$ . In the force-deformation curve, the force value observed at the first sudden drop in force was obtained and defined as the fracturability.

#### 2.6. Measurements of the piperine content

Piperine was extracted from 0.5 g of ground black peppercorns in ethanol by reflux in a hot water bath for 2 h in the dark. After extraction, the cooled extracts were passed through 0.45- $\mu\text{m}$  membrane filters and diluted appropriately for further HPLC analysis. The samples were analyzed using an HPLC system including a separation module (Waters 2695; Milford, MA, USA), an analytical column (Venusil XBP C18, 5  $\mu\text{m}$ , 100 Å,  $4.6 \times 250 \text{ mm}$ ; Bonna-Agela Technologies, Newark, DE, USA), and a Waters 996 Photodiode Array Detector (Milford, MA, USA). The sample (10  $\mu\text{L}$ ) that had been injected into the column was isocratically eluted using an acetonitrile-ethanol (50:50) mixture at a flow rate of  $1.0 \text{ mL min}^{-1}$  and quantified using a calibration curve generated with a piperine

standard ( $\lambda$ , 345 nm).

### 2.7. Statistical analyses

The kinetic parameters and the  $z$ -values were estimated using a nonlinear regression iteration procedure and a linear regression procedure in the SigmaPlot 10.0 Windows version (IBM Co., Armonk, NY, USA), respectively. The analysis of variance (ANOVA) used for the nonlinear regression is detailed in the [Supplementary Material](#). All results for moisture content,  $L^*$ ,  $a^*$ ,  $b^*$ , and fracturability of samples were analyzed using Tukey's significant difference tests at a probability level of  $p < 0.05$  in the IBM SPSS statistical software package version 23.0 (IBM Co.). All data are represented as averages of at least three independent experiments or measurements.

## 3. Results and discussion

### 3.1. Inactivation of *Salmonella* on the food samples

The survival of the inoculum of *S. Typhimurium* and *S. Enteritidis* on black peppercorns, pecans, and almonds after SS/SHS treatments is shown in [Fig. 2](#). After complete inactivation by the steam treatments, the levels of *Salmonella* inoculated on black peppercorns, pecans, and almonds were reduced to 6.34, 6.24, and 6.34 log CFU  $g^{-1}$  (initial inoculum levels: 7.34, 7.24, and 7.34 log CFU  $g^{-1}$ , respectively; detection limit: 1 log CFU  $g^{-1}$ ). In all of the samples, non-survival of *Salmonella* after the SHS treatment for the complete inactivation was demonstrated during the further enrichment procedure. Treatment times required for complete inactivation by SS were 25, 35, and 45 s for black peppercorns, pecans, and almonds, respectively, indicating that the SS treatment had difficulty inactivating *Salmonella*. The samples most difficult to treat were almonds followed by pecans and black peppercorns. At 120, 140, 160, and 180 °C, the SHS treatment times required for complete inactivation were 17, 13, 6, and 3 s for black peppercorns; 23, 19, 15, and 13 s for pecans; and 40, 30, 13, and 8 s for almonds, respectively, indicating that the inactivation efficacy of *Salmonella* improved with increasing SHS temperature.

As observed in [Fig. 2](#), *Salmonella* reduction curves were nonlinear and contained a shoulder at many of the SHS temperatures, regardless of sample type, whereas the curves for SS-treated samples were comparatively linear. The shoulders were observed in the ranges of 4–8 and 2–4 s at 120 and 140 °C SHS for black peppercorns ([Fig. 2a](#)), 4–8, 4–8, 4–8, and 2–4 s at 120, 140, 160, and 180 °C for pecans ([Fig. 2b](#)), and 8–25, 8–20, and 4–8 s at 120, 140, and 160 °C for almonds ([Fig. 2c](#)). These nonlinear survival curves for *Salmonella* that contain a shoulder are observed frequently and likely depend on the *Salmonella* strain, inoculation level, and enumeration and heating methods ([Bermúdez-Aguirre and Corradini, 2012](#); [Smelt and Brul, 2014](#); [Valero et al., 2014](#)). However, because the *Salmonella* strain, inoculation level, and enumeration and heating methods were fixed in this study, the nonlinearity required another explanation. At the early stage of the SHS treatment, condensed water was observed on the surface of the samples, despite the heating by the band heater ([Fig. 1](#)), and we assumed that this water condensation might be correlated with the shoulders in the survival curves. As shown in the changes in the moisture content of the 120 °C SHS-treated samples ([Fig. 3](#)), the shoulder periods of black peppercorns, pecans, and almonds ([Fig. 2](#)) were matched to the periods of each moisture content peak at 4–8, 4–8, and 8–25 s, respectively. Therefore, the thermal energy from SHS was spent primarily on re-evaporating the condensed water, rather than inactivating *Salmonella* on the sample surfaces, during the shoulder period. As the SHS temperature increased, the

increase in the heat transfer rate shortened this period ([Chen et al., 2000](#)). After the surface temperatures of black peppercorns, pecans, and almonds rapidly reached 100 °C, they were maintained for a certain period and later increased to 128.7, 150.3, and 160.7 °C within 3, 13, and 8 s of the 180 °C SHS treatment, respectively ([Fig. S2](#) in the [Supplementary Material](#)). However, for black peppercorns and almonds, the period for which the 100 °C temperature was maintained was too short to observe the shoulders shown in [Fig. 2a](#) and [c](#).

Another possible speculation is that the inoculated *Salmonella* might be washed out with the condensed water or blown away by the steam flow of 3.0 m  $s^{-1}$  during the steam treatment. Thus, this speculation was examined using the data shown in [Table S1](#) ([Supplementary Material](#)). This result verified that the *Salmonella* inactivation observed in this study resulted solely from the thermal effect of the steam treatment. Meanwhile, the purpose of this study was to assess *Salmonella* inactivation and was not related to the uniformity of the steam, but the uniformity was demonstrated unintentionally ([Fig. S3](#) in the [Supplementary Material](#)). Therefore, based on these results, the steam transferred thermal energy evenly onto the sample surface without washing or blowing effects.

### 3.2. Inactivation kinetics of *Salmonella* on the food samples

Many researchers experience problems in fitting a linear model to the survival curves of microorganisms ([den Besten et al., 2006](#); [Peleg, 2006](#)). The Weibull model has been adapted to fit the survival curves more accurately and overcome these problems ([van Boekel, 2002](#)):

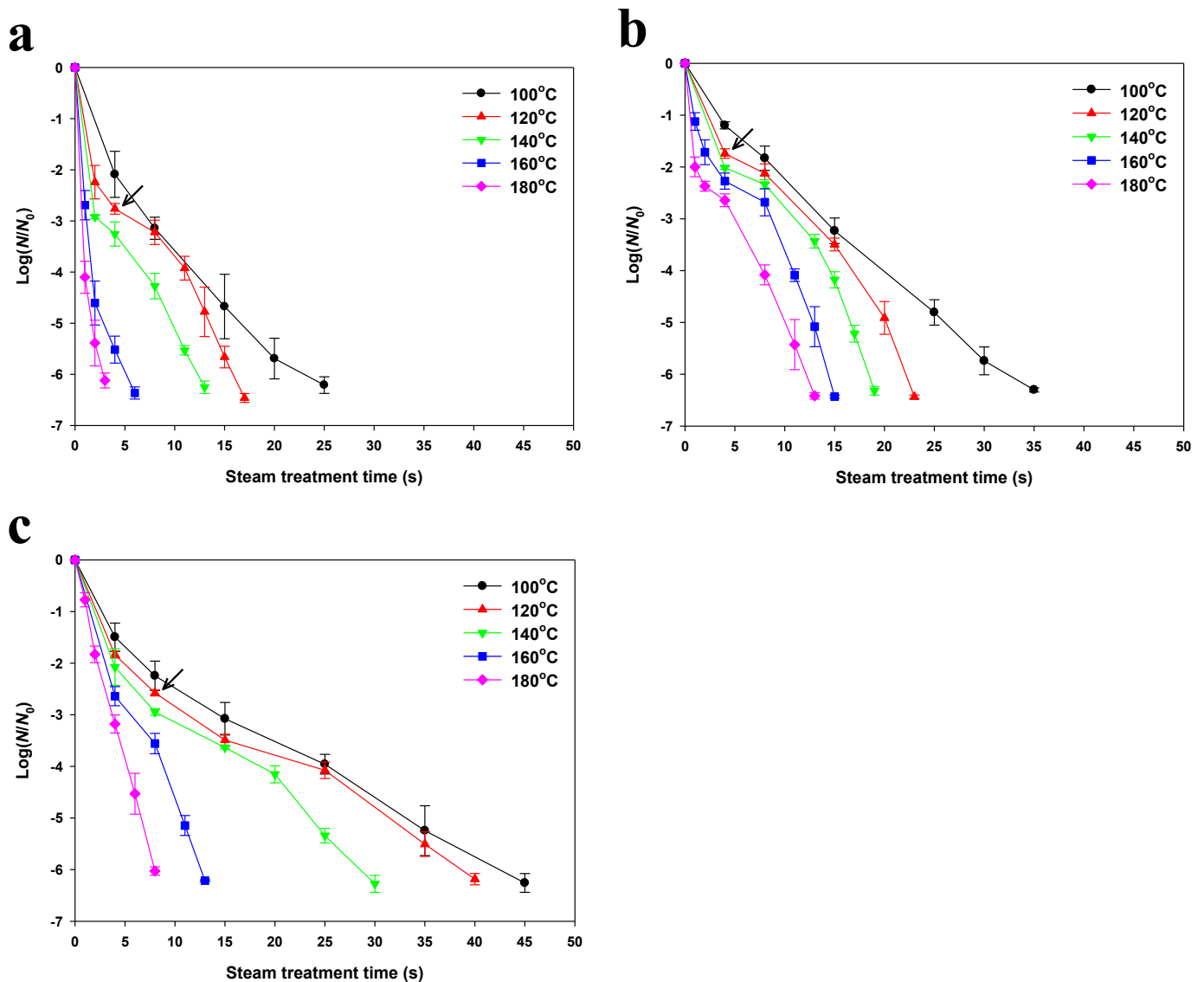
$$\text{Log}(N/N_0) = -(t/\alpha)^\beta$$

where  $\alpha$  is the scale parameter,  $\beta$  is the shape parameter, and  $t$  is the treatment time.

The survival curves shown in [Fig. 2](#) were fitted using the Weibull model, and the fitted curves and parameter values are shown in [Fig. S4](#) ([Supplementary Material](#)) and [Table 1](#), respectively. The appropriateness of the Weibull model was evaluated using ANOVA ([Table S2](#) in the [Supplementary Material](#)). The model was appropriate for fitting the curves and reflected the shoulders of the curves properly, which validated the use of this model for determining  $D$ - and  $z$ -values. In this instance,  $D$ -values were calculated as  $\alpha(2.303)^{1/\beta}$  using the Weibull model ([van Boekel, 2002](#)), and  $z$ -values were determined from the fitted lines shown in [Fig. 4](#). As shown in [Table 1](#),  $D$ -values for all samples decreased as the steam temperature increased, and  $z$ -values were higher in the order of almonds (146.26 °C), pecans (104.71 °C), and black peppercorns (47.06 °C). Based solely on the results presented in [Table 1](#),  $D$ -values determined from the model were reasonable because the steam at higher temperatures inactivated more *Salmonella* on the samples.

Because a larger  $D$ -value indicated a reduced efficacy of the steam treatment in inactivating *Salmonella*, the efficacies of the SS/SHS treatments were lower for treating pecans and almonds than for treating black peppercorns ([Table 1](#)). Moreover, the differences in  $z$ -values indicated differences in the sensitivity of *Salmonella* as a function of the changes in the steam temperature. The sensitivity among SHS-treated samples was high, with black peppercorns showing the greatest sensitivity (47.06 °C) followed by pecans (104.71 °C) and almonds (146.26 °C). These results revealed a difference among the samples rather than the steam treatments. Pecans have narrow and deep valleys on the surface that hinder the heat transfer of SS/SHS, unlike black peppercorns and almonds. In contrast, almonds have many micron-size cavities and crevices on the surface ([Fig. S5](#) in the [Supplementary Material](#)), which could shelter *Salmonella* from SS/SHS or impede the re-evaporation of the





**Fig. 2.** Survival curves of the *Salmonella* inoculated on (a) black peppercorns, (b) pecans, and (c) almonds after saturated steam and superheated steam treatments (detection limit, 1 log CFU g<sup>-1</sup>; arrows indicate the initiation points of drying effect by the superheated steam at 120 °C).

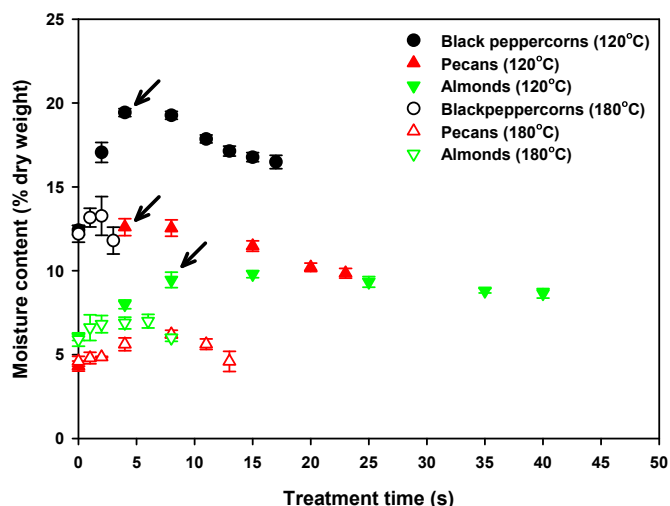
condensed water in the initial stage of the SHS treatment. This impediment for almonds was indirectly verified by the tail on the *Salmonella* survival curve (Fig. 2c) and by the change in moisture content in response to the 120 °C SHS treatment (Fig. 3), which was absent in the samples treated with 180 °C SHS at greater energy. Indeed, almonds are notorious for the difficulties associated with using conventional thermal techniques to inactivate *Salmonella* while maintaining quality (Lee et al., 2006). Thus, various thermal treatments have been investigated to inactivate *Salmonella* on almonds. In previous studies of thermal treatment for almonds, *D*- and *z*-values for a 121 °C hot-air treatment for *S. Enteritidis* PT 30 were 1500 s and 26.1 °C, respectively (Pan et al., 2012). However, the *D*- and *z*-values for hot-water (60, 70, 80, and 88 °C) treatments for *S. Enteritidis* PT 30 were 156, 72, 45, and 23 s and 35 °C, respectively (Harris et al., 2012). These *D*- and *z*-values are greater than the values for other foods (Silva and Gibbs, 2012) and indicated the difficulties in inactivating *Salmonella* on almonds. Fortunately, based on the *D*-value, *Salmonella* inactivation was immensely improved by the SS/SHS treatments compared with

treatments reported in previous studies, despite the difference in serotypes and the large *z*-values (146.26 °C).

### 3.3. Quality of the food samples after steam treatment

Quality values for black peppercorns, pecans, and almonds after complete *Salmonella* inactivation by SS/SHS treatments are shown in Table 2. High-temperature steam treatments for 60 s have not been shown to affect the phenolic compound content in many food commodities (Bolling et al., 2010). In this study, SS/SHS treatments were administered for ≤45 s. Additionally, the initial piperine content (57.6 ± 3.5 mg g<sup>-1</sup>) in black peppercorns was similar to the content (55.0 ± 3.8 mg g<sup>-1</sup>) measured after a 180 °C SHS treatment for up to 25 s, unlike a 100 °C SS treatment for 960 s reported in a previous study (Waje et al., 2008). Based on this result, short-term SS/SHS treatments might not affect the piperine content in black peppercorns.

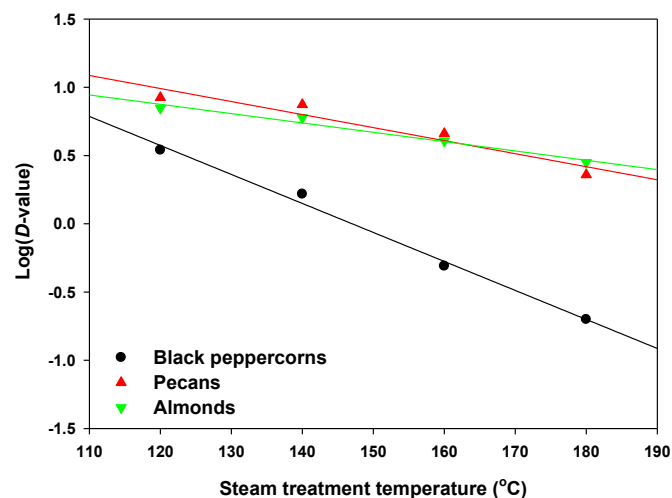
The moisture content of foods after decontamination processes is a critical quality value because a high moisture content can



**Fig. 3.** Changes of moisture content (% dry weight) of black peppercorns, pecans, and almonds under the superheated steam treatment at 120 and 180 °C (arrows indicate the initiation points of drying effect by superheated steam).

induce food deterioration during storage (Perera, 2005). Therefore, if the moisture content increases during steam treatment, then the additional moisture should be eliminated using a subsequent drying process. As shown in Table 2, the moisture content of all steam-untreated samples was like the moisture content of the samples following SHS treatments at 160 and 180 °C but differed from the moisture contents of the samples following SS/SHS treatments at 100, 120, and 140 °C. These observations reflected the faster drying effects of the 160 and 180 °C SHS treatments on the samples due to higher heat energies. Based on both the moisture content and speed of complete decontamination, the 180 °C SHS treatment was the best treatment among the steam treatments examined in this study. The trends in the fracturability values in almonds were comparable to the moisture content, which was attributed to the sensitivity of its inner meshed structure to the thermal treatment (Young et al., 2004). Little change in color ( $L^*$ , lightness;  $a^*$ , redness; and  $b^*$ , yellowness) was observed for all samples. Moreover, the sensory qualities (appearance, flavor, and taste) of black peppercorns, pecans, and almonds were not significantly affected by the complete inactivation of *Salmonella* using the 180 °C SHS treatment ( $p > 0.05$ ).

In summary, *Salmonella* spp. on black peppercorns, pecans, and almonds was completely inactivated by the 180 °C SHS treatment



**Fig. 4.** The z-value determination based on D-values obtained from Weibull models ( $\text{Log}(N/N_0) = -(t/\alpha)^\beta$ ) fitting the survival curves for a cocktail of *Salmonella* inoculated on black peppercorns, pecans, and almonds after superheated steam treatment.

within 3, 13, and 8 s, respectively, without a deterioration in quality. According to a previous study (Shah et al., 2017), *S. Enteritidis* PT 30  $> 6.10 \log \text{CFU g}^{-1}$  was completely inactivated on black peppercorns using a vacuum steam treatment at 75 °C for 1 min. In a study with pecans (Beuchat and Mann, 2011),  $5 \log \text{CFU g}^{-1}$  of a *Salmonella* cocktail with five serotypes was inactivated by oil roasting at 138 °C for 2 min. The 180 °C SHS treatment used in this study reduced the times required for complete *Salmonella* inactivation on black peppercorns and pecans to 3 and 13 s, respectively. In a study with almonds (Ban and Kang, 2016), the use of SHS at 200 °C led to  $> 6.0 \log$  reductions of *S. Typhimurium* and *S. Enteritidis* PT 30 after 15 s, with no change in quality. Although the inactivation time was 8 s at 180 °C in this study, this discrepancy in the inactivation time was primarily attributable to the different SHS systems used. Hence, the SHS treatment effectively inactivated *Salmonella* contamination on black peppercorns, pecans, and almonds.

#### 4. Conclusions

In this study, *Salmonella* on black peppercorns, pecans, and almonds was completely inactivated within 40 s, regardless of the

**Table 1**

Parameter values and D-/z-values determined from Weibull models ( $\text{Log}(N/N_0) = -(t/\alpha)^\beta$ ) fitting the survival curves for a cocktail of *Salmonella* inoculated on black peppercorns, pecans, and almonds after saturated and superheated steam treatments.

Samples	Temperature (°C)	$\alpha$	$\beta$	D-value (s)	z-value (°C)	$R^2$
Black peppercorns	100	1.16	0.60	4.65	47.06	0.99
	120	0.84	0.59	3.46		
	140	0.27	0.46	1.65		
	160	0.07	0.41	0.49		
	180	0.02	0.36	0.20		
Pecans	100	3.53	0.81	9.93	104.71	0.92
	120	3.47	0.94	8.41		
	140	3.20	0.99	7.47		
	160	1.55	0.77	4.58		
	180	0.50	0.55	2.29		
Almonds	100	2.37	0.61	9.20	146.26	0.97
	120	1.51	0.54	7.04		
	140	1.43	0.58	5.99		
	160	1.46	0.82	4.04		
	180	1.13	0.91	2.81		

**Table 2**  
Moisture content, color (L\*, lightness; a\*, redness; and b\*, yellowness), and fracturability of black peppercorns, pecans, and almonds after the complete inactivation of *Salmonella* via saturated and superheated steam treatments<sup>a</sup>.

Samples	Treatments	Moisture content (%, dry weight)	Color			Fracturability (N)
			L*	a*	b*	
Black peppercorn	25 s at 100 °C	28.3 ± 0.5 a	31.6 ± 0.5 b	0.9 ± 0.2 b	2.0 ± 0.4 b	–
	17 s at 120 °C	16.1 ± 1.0 b	32.8 ± 0.4 a	1.5 ± 0.4 a	3.0 ± 0.3 a	–
	13 s at 140 °C	14.6 ± 0.5 c	32.7 ± 0.2 a	1.1 ± 0.0 a	2.9 ± 0.1 a	–
	6 s at 160 °C	12.5 ± 0.9 d	33.1 ± 0.5 a	1.2 ± 0.4 a	3.2 ± 0.6 a	–
	3 s at 180 °C	11.8 ± 0.8 d	33.1 ± 0.8 a	1.2 ± 0.2 a	3.1 ± 0.8 a	–
	Control	12.2 ± 0.5 d	33.1 ± 0.9 a	1.3 ± 0.0 a	3.4 ± 1.0 a	–
Pecan	35 s at 100 °C	10.0 ± 0.5 a	36.5 ± 1.8 ab	15.4 ± 1.2 a	23.6 ± 3.0 ab	–
	23 s at 120 °C	9.6 ± 0.4 a	36.2 ± 3.5 ab	15.8 ± 1.6 a	25.1 ± 4.0 a	–
	19 s at 140 °C	6.4 ± 0.2 b	35.3 ± 2.2 bc	14.2 ± 1.1 b	21.1 ± 2.9 bc	–
	15 s at 160 °C	4.5 ± 0.4 c	32.5 ± 1.4 d	15.3 ± 1.5 ab	19.5 ± 2.9 c	–
	13 s at 180 °C	4.6 ± 0.6 c	34.1 ± 1.8 cd	15.7 ± 1.2 a	21.9 ± 1.5 bc	–
	Control	4.6 ± 0.3 c	35.3 ± 2.8 bc	14.9 ± 1.0 ab	23.3 ± 2.7 ab	–
Almond	45 s at 100 °C	9.4 ± 0.1 a	46.7 ± 3.7 c	15.6 ± 1.4 ab	34.0 ± 2.5 a	67.5 ± 20.1 d
	40 s at 120 °C	8.6 ± 0.2 a	46.8 ± 2.8 c	15.1 ± 1.0 ab	33.4 ± 1.6 a	88.0 ± 23.5 c
	30 s at 140 °C	7.4 ± 0.3 b	46.8 ± 2.6 c	17.6 ± 1.1 a	33.5 ± 2.6 a	108.5 ± 16.4 b
	15 s at 160 °C	6.0 ± 0.3 c	51.0 ± 2.2 a	15.1 ± 1.6 b	32.4 ± 2.3 a	118.6 ± 18.7 ab
	8 s at 180 °C	6.0 ± 0.2 c	48.1 ± 2.4 bc	17.5 ± 1.1 a	32.6 ± 1.6 a	128.9 ± 37.9 ab
	Control	5.9 ± 0.4 c	50.0 ± 3.0 ab	15.0 ± 0.8 b	33.3 ± 2.2 a	147.2 ± 22.0 a

<sup>a</sup>Different letters (a–d) in a column for the same sample are significantly different at  $p < 0.05$ .

use of the SS or SHS treatment. However, the SS treatment was not appropriate for *Salmonella* inactivation because the moisture content was elevated in the samples. Based on the  $D$ - and  $z$ -values determined from the fitting of *Salmonella* survival curves using the Weibull model, the SHS treatment completely inactivated *Salmonella* on black peppercorns, pecans, and almonds, despite differences in sensitivity due to surface structural differences. Additionally, the inactivation efficacy increased as the SHS temperature increased. Consequently, the 180 °C SHS treatment inactivated *Salmonella* completely on black peppercorns, pecans, and almonds within short periods (3, 13, and 8 s, respectively) without any deterioration in quality, as measured by the moisture content, color, and texture. In conclusion, the results of this study provide the basis for applying the minimal SHS process in food industrial settings to control *Salmonella* spp. on various foods without a deterioration in quality.

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## Appendix A. Supplementary data

Supplementary data related to this article can be found at <https://doi.org/10.1016/j.jfoodeng.2017.11.036>.

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