



## Microwave sterilization of sliced beef in gravy in 7-oz trays

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### ABSTRACT

This research was to investigate the feasibility for developing a short-time sterilization protocol for a highly inhomogeneous food prepackaged in polymeric trays using 915 MHz microwave (MW) energy. A 915-MHz, single-mode, 10-kW pilot-scale MW system developed at Washington State University was used for this study. The inhomogeneous food consisted of sliced beef and gravy packaged in 7-oz polymeric trays. Specially formulated whey protein gel, matching the beef product in their dielectric properties, was chosen as a model food to emulate the real food for determination of heating patterns and cold spots inside food trays. The heating patterns and cold spots were detected using a chemical-marker-assisted computer vision method. Processing schedules to achieve desired levels of  $F_0$  for 7-oz trays of beef in gravy were established based on temperature histories measured at the identified cold spot location. The developed processing schedules were validated by inoculated pack studies using *Clostridium sporogenes* PA 3679 spores. The results of this study indicate that the 915-MHz single-mode MW sterilization technology is effective for processing of the inhomogeneous food. The procedure established could be used for developing MW sterilization processes for other packaged inhomogeneous foods, such as chicken meat in gravy in trays and salmon in sauce in pouches. The processing data collected could be helpful for industrial scale-up of the MW system.

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### 1. Introduction

Microwave (MW) heating is a result of interaction between alternating electromagnetic field and dielectric material (Orfeuill, 1987). Compared with conventional heating using water or steam as the heating media for package foods, MW energy has the potential to provide more uniform and rapid volumetric heating.

As a unique thermal processing technology, MW heating has been successfully used in food industry, including tempering or thawing of bulk frozen foods (meat, fish, and others), cooking of bacon and sausage, and drying of pasta and vegetables (Bengtsson and Ohlsson, 1974; Hulls and Shute, 1981; Hulls, 1982; Jones, 1992; Schiffmann, 1992). Research on MW treatment of foods has also been reported for disinfecting of insects in agricultural commodities (Wang et al., 2003a), blanching of vegetables, inactivating of enzyme, pasteurization of breads, cured hams and sausage emulsions, sterilization of food products (Decareau, 1985; Venkatesh and Raghavan, 2004).

Two frequencies,  $2450 \pm 50$  and  $915 \pm 13$  MHz, are allocated by the US Federal Communications Commission for MW heating applications (Decareau, 1985; Metaxas and Meredith, 1983). Two thousand four hundred fifty megahertz is widely used in domestic MW ovens and some industrial applications. Two thousand four

hundred fifty megahertz systems have the limitations of small penetration depth ( $\sim 1$  cm) and multi-mode cavities, causing non-uniform and unpredictable heating patterns in food packages. In general, 915-MHz microwaves can penetrate much deeper ( $\sim 3$  cm) in foods, and therefore may provide more uniform heating (Mudgett, 1989). Nine hundred fifteen megahertz systems could be established with a single-mode cavity, which could provide predictable electromagnetic field, resulting in predictable and reproducible heating patterns in foods.

As public concerns over food safety continue to grow and the demand for high quality packaged convenience foods increases, MW heating is drawing much attention of researchers in developing novel pasteurization and sterilization processes for packaged foods. A 915-MHz, single-mode, MW sterilization system for processing packaged foods was developed at Washington State University (WSU) (Tang et al., 2006). It was intended for proving the concept and demonstrating the potential of MW technology in pasteurization and sterilization applications, for developing theories and methodologies in support of technology development, for studying various operation parameters, and for collecting engineering data for industrial scale-up. The system has been used for studying the influence of MW sterilization on quality of various foods, including asparagus (Sun et al., 2007), macaroni and cheese (Guan et al., 2002, 2003), salmon, chicken, rice, scrambled eggs, and mashed potatoes. Most of the MW processed products had superior quality, attractive appearance, and high consumer

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acceptance. This is attributed to the fact that microwave sterilization processes sharply reduced processing time compared with conventional retorting (Guan et al., 2002). During the studies, some necessary processing parameter, such as MW power, flow rate and temperature of circulating water, were established for optimum system operation. Meanwhile, a chemical-maker-assisted computer vision technique for determining heating patterns and cold spots inside MW treated food packages was developed by the WSU MW research group (Pandit et al., 2007a,b).

For scaling-up the MW system for industrial applications, it is necessary to study the MW sterilization and collect processing data for various foods. The objectives of this research were to investigate the technical feasibility of MW sterilization of a highly inhomogeneous food, sliced beef in gravy prepackaged in polymeric trays, and to establish appropriate procedures for developing and validating a MW sterilization process for the inhomogeneous food with the 915 MHz MW technology. Four major steps were taken to achieve the objectives: (1) choosing a model food to emulate the real food for determination of heating patterns, (2) determining cold spots in the food trays, (3) developing schedules for MW sterilization process of the sliced beef in gravy in 7-oz trays, and (4) verifying microbial safety of the MW processed foods by inoculated pack studies.

## 2. Materials and methods

### 2.1. MW sterilization system setup and operation

The pilot MW sterilization system at WSU (Fig. 1) was used in this project. The system consisted of two 5-kW 915-MHz MW generators, waveguides, two MW heating cavities, loading and unloading cavities, a sample tray conveyor system, a water circulation system, and a control and data acquisition system. MW power from the generators was transmitted to the MW heating cavities through waveguides during operation. The loading cavity was used for loading food sample trays and also served as a pre-heating cavity, and the unloading cavity was for unloading processed food trays and served as a cooling cavity. The water circulation system, consisting of two plate heat exchangers, a storage tank, and fixture to introduce compressed air to pressurize water, was used to provide temperature-conditioned pressurized water for pre-heating, auxiliary heating to MW treatment, holding, and cooling of food packages. The control and data acquisition system consisted of a control board, sensors/meters, data loggers, and a computer with a custom built software. It was used to monitor and record operation parameters such as MW powers, sample and water tempera-

tures, and water pressure and flow rate, as well as to control the system operation. Opsens fiber-optic sensors with a data conditioner (Opsens, Quebec, QC, Canada) were used for monitoring food temperature during the process. The sensors were calibrated before process operation over a temperature range from 20 to 121 °C against a heating block, which was calibrated by mercury-in-glass thermometers.

In order to provide stable and reproducible processes, the system was warmed for about 10 min with 80 °C water flowing through the cavities before the first test run. Following the warming up, the temperature of circulating water at the point of heating exchanger was set to 125 °C to obtain a 122 °C temperature at the inlet of the cavities. In this study, the MW power output for each MW heating cavity was set at 2.7 kW, and the circulated water was controlled at 41 psia and 40 L/min.

In a sterilization process, this system was operated following a pre-determined processing schedule, which defined the processing time for each of the four processing periods: pre-heating, MW heating, holding, and cooling. Firstly, sample trays preloaded on the conveyor mesh belt were pre-heated to 60 °C with hot water (122 °C) in the loading section (left end cavity in Fig. 1). Secondly, the trays were moved through the MW heating cavities and treated by both MW energy and hot water (122 °C) with a selected time-speed schedule. The sample trays were then held in the holding cavity (right end cavity in Fig. 1) to achieve a desired  $F_0$  after MW treatment. Finally, the trays were cooled until the center temperature reached below 75 °C with tap water and unloaded from the system.

### 2.2. Food material

The product of interest for MW sterilization in this study was Sysco Block & Barrel sliced roasted beef (Sysco Corporation, Houston, TX) in Nestle Trio low sodium gravy (Nestle Foodservices, Glendale, CA) packaged in trays. Trio gravy was prepared according to package instructions except the weight proportions were as follows: 200 g gravy mix in 1600 ml water. The gravy mix had more than 20 ingredients such as modified food starches, wheat gluten protein, sugar, soybean oil, and salt.

### 2.3. Test of beef shrinkage during heat treatment

In preliminary tests for MW processing of Sysco sliced beef in Nestle Trio gravy, it was observed that beef samples shrunk significantly (about 35% area reduction and 40% weight reduction) after MW sterilization. The shrinkage caused the beef samples to move,

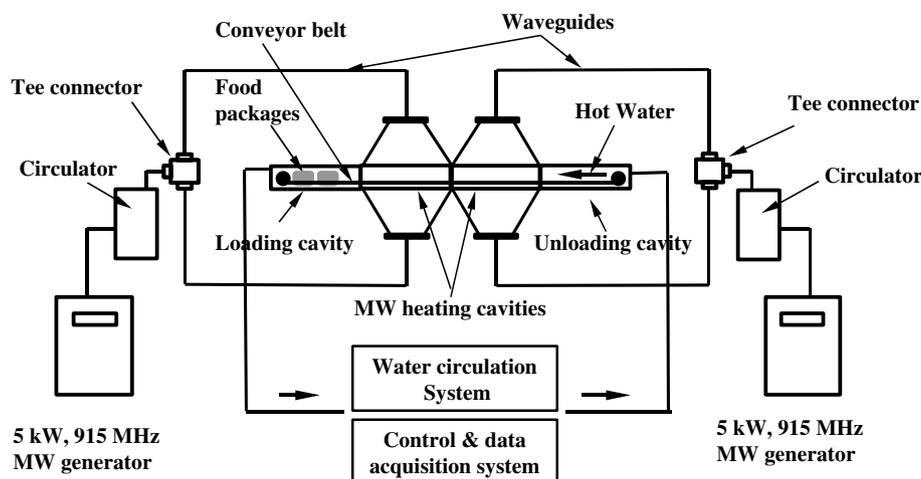


Fig. 1. Schematic diagram of pilot MW sterilization system at Washington State University.

and cold spots did not maintain fixed locations inside trays during MW processing. This made it difficult to record the temperature profiles of cold spots with temperature sensors. Therefore, pre-cooked or pre-shrunk beef was used for the development of MW processing schedules.

Shrinkage of beef was studied by heating sliced beef samples in boiling water with a 0.5% salt addition for different times between 0 and 40 min. The salt addition was to limit the change in dielectric properties of the beef sample. Preliminary dielectric property tests indicated that pre-cooking of beef in boiling water lowered both the dielectric constant and the dielectric loss of the sample, and that an addition of salt in the boiling water could reduce the decrease in dielectric property data of the pre-cooked beef sample. The weight and area of the sample were measured prior to and after the heat treatment. The weight loss and area reduction of the sample were then calculated. It was decided to use beef samples treated for 4 min in the boiling water with 0.5% salt for MW processing schedule development.

#### 2.4. Measurement of dielectric properties of beef and model food samples

The heating pattern and cold spot inside the packaged real food (beef in gravy) is difficult to detect when using the real food itself. Whey protein gel (WPG) was used as a model food to emulate the real food for determining heating patterns and cold spots in trays. WPG can imitate physical characteristics of the real food such as weight, shape, and thickness. It can be easily formulated to match the real food in their dielectric properties, which are critical parameters related to MW heating. WPG can also be added with chemical marker precursor, D-ribose (Sigma–Aldrich Inc., St. Louis, MO), for chemical marker M-2 formation inside the model food during thermal processing. The M-2 formation is dependent on the accumulated thermal processing; the heating pattern inside the sample could be detected by analysis of the M-2 distribution inside the sample.

To determine the formulation of the model food, dielectric properties of pre-treated Sysco sliced beef samples and WPG samples with various compositions were measured using a Helwett-Packard 85070B open-ended coaxial probe connected to an Agilent 4291B Impedance Analyzer (Agilent Technologies Inc., Palo Alto, CA). Detailed description of the measurement system and its operation was provided by Wang et al. (2003b). The composition of WPG varied with the content (20%, 30%, and 40% on wet basis) of Whey Alacen 878 (New Zealand Milk Products Inc., Lemoyne, PA) and the level of salt (0%, 0.3%, and 0.5%). One percent of chemical marker precursor D-ribose was added in the WPG samples for the purpose of heating pattern and cold spot determination. To make WPG samples, ingredients (whey protein concentrate, salt, D-ribose, and water) were mixed into uniform slurry, and a gel was ob-

tained by cooking the slurry in a water bath at 80 °C for 40 min. The set WPG blocks were cut into the same geometry as that of the sliced roast beef. Based on the testing results, the WPG with 35% Whey Alacen 878, 1% D-ribose and 0% salt was determined as the model food to emulate the pre-treated sliced beef in heating pattern and cold spot detection.

#### 2.5. Chemical-marker-based computer vision method for heating pattern analysis

Chemical marker M-2 (4-hydroxy-5-methyl-3(2H)-furanone) is formed in a low-acid media (pH > 5) during a heat process by Maillard (non-enzymatic browning) reactions between D-ribose and amino acids (lysine, arginine, histidine, and methionine) (Kim et al., 1996; Prakash et al., 1997). The M-2 formation in the WPG samples depended on the thermal process level usually described by thermal lethality at 121.1 °C,  $F_0$  (in min). The value of  $F_0$  at a point in a thermal-processed product is calculated based on the temperature history at the point as follows:

$$F_0 = \int_0^t 10^{(T-T_r)/z} dt \quad (1)$$

where  $T$  is the measured temperature (°C);  $T_r$  is the reference temperature (121.1 °C);  $z$  has a value of 10 °C for sterilization; and  $t$  is the heating time (min).

The distribution of M-2 inside the sample correlating with brown color intensity can be detected by a computer vision method. The relationships among the color value equivalent to grayscale value, M-2 yield, and  $F_0$  for WPG samples heated at 121 °C were established by Pandit (2006) as follows:

- (1) Relationship between M-2 yield ( $y_1$ , mg/g sample) and  $F_0$  ( $x_1$ , min):

$$y_1 = -0.0038x_1^2 + 0.1233x_1 + 0.2735, \quad R^2 = 0.98 \quad (2)$$

- (2) Relationship between color value ( $y_2$ ) and M-2 yield ( $y_1$ , mg/g sample):

$$y_2 = 201.7y_1 - 48.052, \quad R^2 = 0.99 \quad (3)$$

- (3) Relationship between color value ( $y_2$ ) and  $F_0$  ( $x_1$ , min):

$$y_2 = -0.6716x_1^2 + 22.343x_1 + 21.79, \quad R^2 = 0.97 \quad (4)$$

Since beef slices in gravy could potentially move inside the tray during MW processing, oval WPG slabs that resembled the geometry of beef slices were placed at three different positions in the 7-oz polymeric trays (Rexam Plastics Containers, Union, MO) to simulate the shape and movement of the beef sample (Fig. 2a,b,c). A tray filled with WPG was used as control. The thickness of the slab was controlled at  $12.5 \pm 0.5$  mm. Sixty grams of gravy was added to

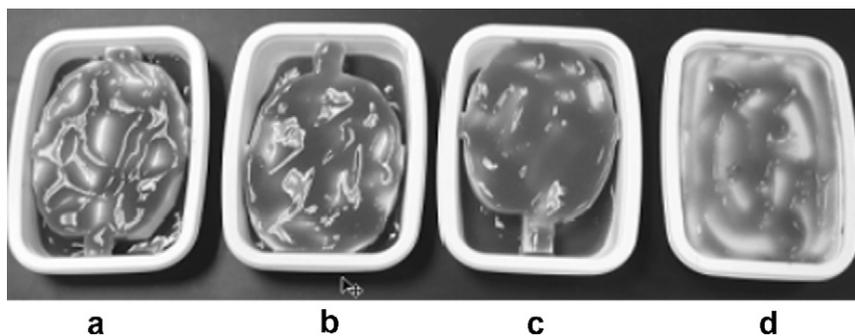


Fig. 2. Positioning of WPG samples.

the oval WPG slabs ( $150 \pm 1.0$  g), and 25 g to the un-cut WPG sample ( $200 \pm 1.0$  g) (Fig. 2d). The trays were sealed with Truitt film (Truitt Bros. Inc., Salem, OR) using a custom tray sealer under an 18-in. Hg vacuum.

Based on preliminary tests with recorded temperature at the cold spot and validated by a computation simulation model (Chen et al., 2007), a processing schedule was chosen for processing of the WPG-in-gray sample trays at 2.7 kW power level in the WSU MW sterilization system. Upon reaching  $121^\circ\text{C}$  at the cold area of the sample, microwave heating was stopped followed by cooling with tap water. For each test run, five trays were treated at the same time. The second tray in the batch was used for the heating pattern detection.

Heating patterns and cold spots in the MW treated WPG samples were determined using the computer vision system (Fig. 3) described in Pandit et al. (2007a). The system consisted of a Paterson light pod (Paterson Photographic Inc., Douglassville, GA), four 26-W helical bulbs (General Electric Co., Schenectady, NY), a Nikon digital camera D70 (Nikon Instrument, Melville, NY), and a Dell computer. The system was equipped with softwares: Nikon Capture 4.0 (Nikon Instrument, Melville, NY), Adobe Photoshop 8.0 (Adobe Systems Incorporated, San Jose, CA), and IMAQ Vision Builder 6.1 (National Instruments Corporation, Austin, TX). The computer vision method involved the following steps:

- (1) Taking digital images using the Nikon Digital Camera and Nikon Capture 4 software.
- (2) Processing the digital images and creating photo packages using the Adobe Photoshop 8.0 software.
- (3) Analyzing heating patterns using the IMAQ Vision Builder 6.1 software.
- (4) Determining color values using the “Quantify” function of IMAQ Vision Builder 6.1 software.
- (5) Locating cold spots using the “Measurement” function of IMAQ Vision Builder 6.1 software.

Details of the computer vision method are described in Pandit et al. (2007a,b). Using this method, cold spots were located in the middle layer of the sample and at the points 18 mm away from the front and back edges of the tray. Since the model food and real food samples have similar dielectric properties, thermal properties, and shapes, the model and real food should have similar heating patterns and same locations of cold spots.

#### 2.6. Experiments for processing schedule development

Preliminary tests suggested that an acceptable proportion of pre-shrunk beef to gravy in 7-oz trays was about 1.25 parts of beef to 1 part of gravy. Total weight of the product for one tray was

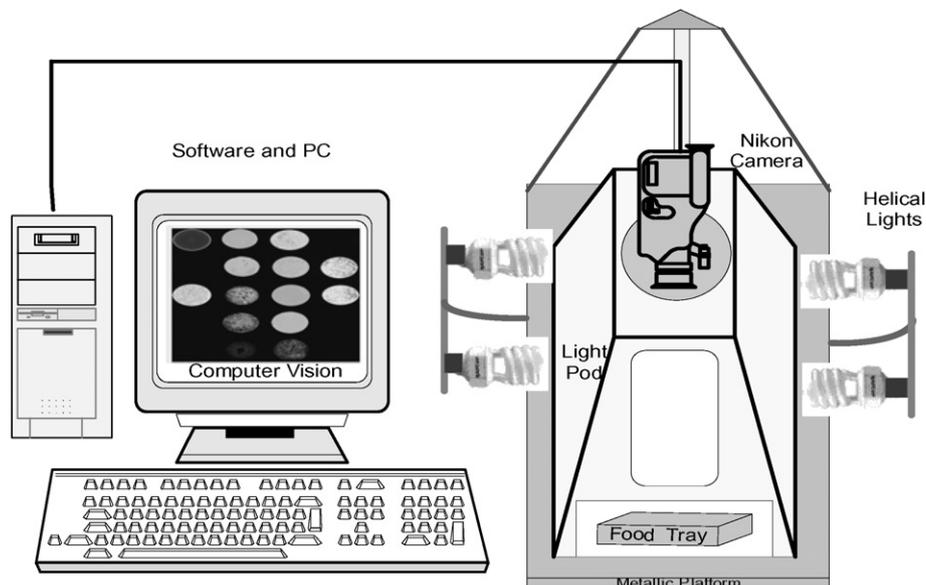


Fig. 3. Computer vision system (Pandit et al., 2007a).

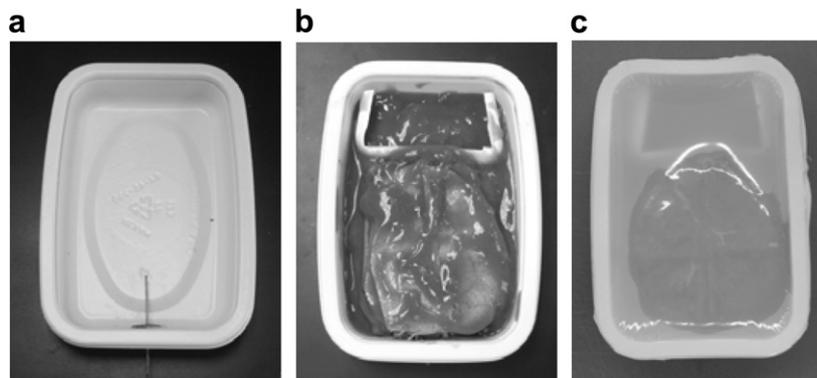


Fig. 4. Preparation of beef in gravy sample trays. (a) 7-oz polymeric tray with a thermo-well, (b) tray filled with beef and gravy, and (c) tray sealed with a film.

180 ± 1.0 g (100 ± 0.5 g beef, 80 ± 0.5 g gravy, 12–13 mm in thickness). In filling the tray, about 1/2 of the gravy was evenly spread on the bottom of the tray, then the pre-treated beef was placed on the gravy, the rest of the gravy was spread on the top and edges (Fig. 4). The beef sample was placed close to the front edge of the tray, and a support frame cut from a polymeric tray was used to keep the beef sample from moving inside the tray. A thermo-well made from polyimide tubing (MicroLumen Inc., Tampa, FL) was pre-fixed through the tray wall and placed between the two central beef slices. The thermo-well was used for inserting a fiber-optic temperature sensor to the cold spot pre-determined with the model food WPG. Sample trays were sealed with a lick film in a custom tray sealer for 3 s at 385 F and under 16-in. Hg vacuum. The trays were conditioned a couple of hours in a low-temperature storage room (3 ± 1 °C) before testing.

To determine processing schedules, experiments were conducted by operating the system manually. Five trays were treated in each run. A fiber-optic probe monitored the temperature at the cold spot in the second tray. The temperature reading and corresponding  $F_0$  were shown on the computer screen during processing. Trials were conducted with different processing conditions to develop a schedule for processing samples to reach an expected final  $F_0$  value, 3 min, at the cold spot. In processing using the selected schedule for  $F_0 = 3$  min, sample trays were firstly pre-heated to 60 °C at the cold spot with 122 °C water, moved at an adequate speed through the MW heating cavities for the cold spot of the food to reach 121 °C by the combination of 2.7 kW MW power and hot water, then held to make  $F_0$  to reach around 1 min, and finally cooled down to 75 °C by tap water to gain additional  $F_0$  of about 2 min. The schedules for larger  $F_0$  values were developed by extending the holding time based on the schedule for  $F_0 = 3$  min.

### 2.7. Inoculated pack studies for validation of developed processing schedules

*Clostridium sporogenes* (PA 3679, batch No. 307) spores were used in inoculated pack studies for microbial validation of the developed processing schedules. The spore suspension was obtained from the Center for Technical Assistance of the National Food Processors Association (NFPA, Dublin, CA). It was divided into cryogenic sterile vials (Fisher Scientific, Pittsburgh, PA) and kept in a freezer (−20 °C) until use. The initial concentration of the stock suspension was approximately  $2 \times 10^7$  CFU/mL, which was determined by an enumeration procedure. The heat resistances ( $D$  values at 121 °C) of the *C. sporogenes* spores in phosphate buffer,

Sysco sliced roast beef, and Nestle Trio low sodium gravy were determined as 0.72, 0.75, and 0.4 min, respectively.

Pre-treated sliced beef (heated in boiling water with 0.5% salt) in gravy in 7-oz trays was used for the inoculated pack studies. Sample trays were prepared following the sample preparation procedure described above, except for injecting spore suspension into the sample and without placing a thermo-well for the temperature probe. Testing spore suspension containing approximately  $1.3 \times 10^7$  CFU/mL was prepared by diluting stock spore suspension in sterile cold water. Then, 0.1 mL of testing spore suspension was injected into the beef sample at the potential cold spot, which was 18 mm away from the front edge of the beef sample in the middle layer. The cold spot was suggested by the computer vision analysis of MW processed WPG samples that simulated the pre-treated sliced beef. The preparation provided an inoculum level of approximately  $1.3 \times 10^6$  spores/tray.

Four trays with a dummy tray placed at the left end were processed in each run using the MW sterilization system with the developed processing schedules (Table 1). The process was repeated in triplicate using each of the three different processing schedules. In addition, eight trays were not MW treated and used as control samples. Target processing with the  $F_0 = 6$  min schedule was equivalent to an 8-log reduction process for PA 3679 spores based on the  $D$  value (0.75 min) of the spores in Sysco sliced roast beef. The processing was to inactivate the inoculated PA 3679 spores ( $1.3 \times 10^6$  spores/tray) to allow about 1% possibility of a spore surviving in a tray. Under-target processing with the  $F_0 = 3$  min schedule (4-log reduction process) was selected to allow certain survival of inoculated PA 3679 spores after MW processing. Over-target processing with the  $F_0 = 12$  min schedule (16-log reduction process) was used to destroy the PA 3679 spores completely in the inoculated trays.

The end-point method was used to observe the lethality of the processed products (Guan et al., 2003). With this method, the processed trays were incubated at 32 °C and observed every 5 days for 1 month. The trays were further observed for additional 2 months. When bulging was detected, the tray was considered to be positive, which means that at least one *C. sporogenes* spore survived treatment. Trays showing no signs of bulging after 3 months were scored as having zero viable spores.

## 3. Results and discussion

### 3.1. Shrinkage of sliced beef during heat treatment

Fig. 5 shows the characteristic of beef shrinkage in boiling water with 0.5% salt. Most of the shrinkage (about 70%) took place in the

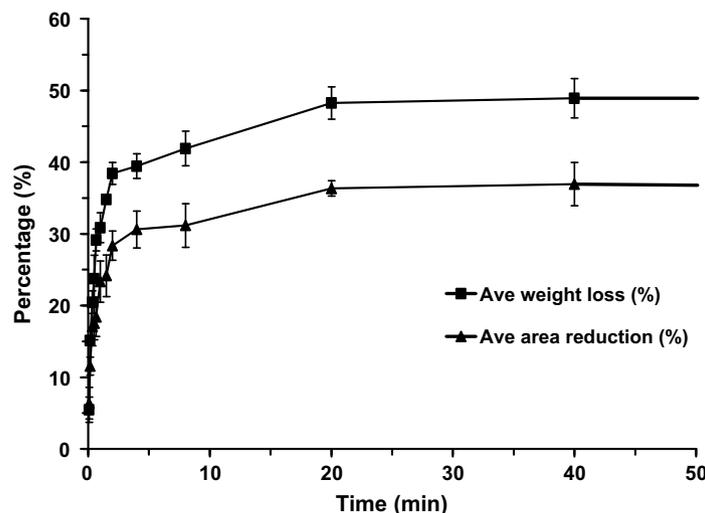


Fig. 5. Weight loss and area reduction of beef samples during heating in boiling water with 0.5% salt.

first four minutes; the sample weight and area reduced slowly between 4 and 20 min, and did not change after 20 min. The 4-min heating in the boiling water was considered adequate to set the geometry of beef samples for MW processing schedule development. Similar method was used in the food industry to reduce shrinkage and control solid content in thermally processed beef and other meat products.

3.2. Dielectric properties of products and selection of model food

Dielectric properties (dielectric constant  $\epsilon'$  and dielectric loss factor  $\epsilon''$ ) of a material determines the behavior of the material in radio frequency or MW fields in terms of dielectric heating; they indicate the ability of the material to reflect, transmit, and absorb

energy from radio frequency waves or microwaves (Nelson and Kraszewski, 1990).  $\epsilon'$  is related to the ability of the material to store electromagnetic energy and  $\epsilon''$  is a measure of the ability of the material to dissipate electrical energy into heat. They are critical parameters that affect coupling and distribution of electromagnetic energy during the heating process (Mudgett, 1986). Dielectric properties of a material vary with the composition such as salt content and moisture content.

For detecting heating patterns and cold spots with a model food to simulate a real product, the dielectric properties of the model food and real food should match. A comparison of dielectric properties between WPG and pre-treated beef samples are presented in Figs. 6 and 7. The dielectric properties of WPG changed when varying the sample composition. Both the dielectric loss factors and the

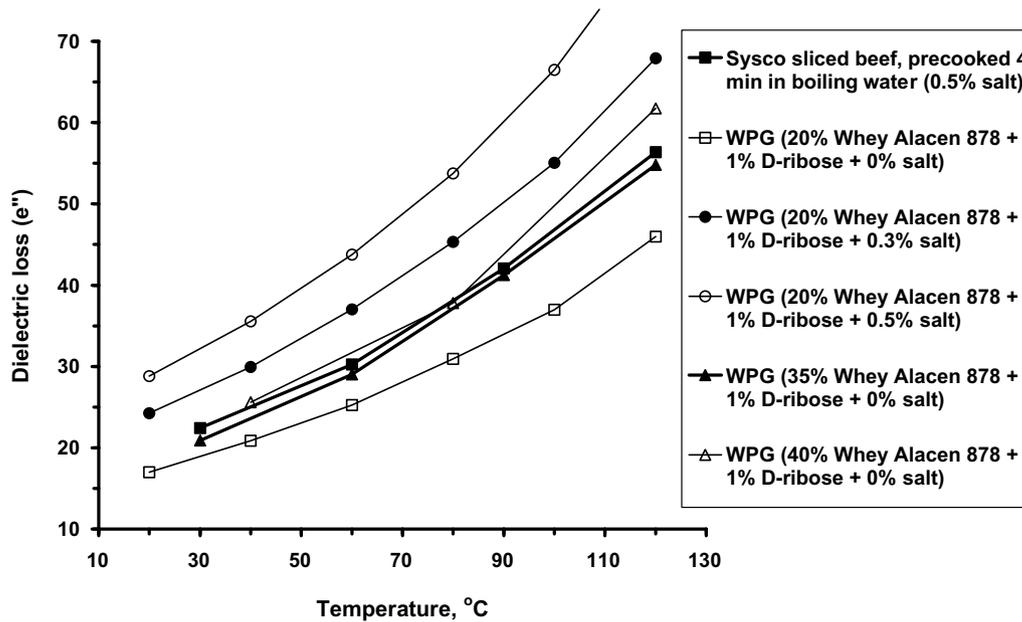


Fig. 6. Dielectric loss factors at 915 MHz for pre-cooked sliced beef and WPG with different compositions.

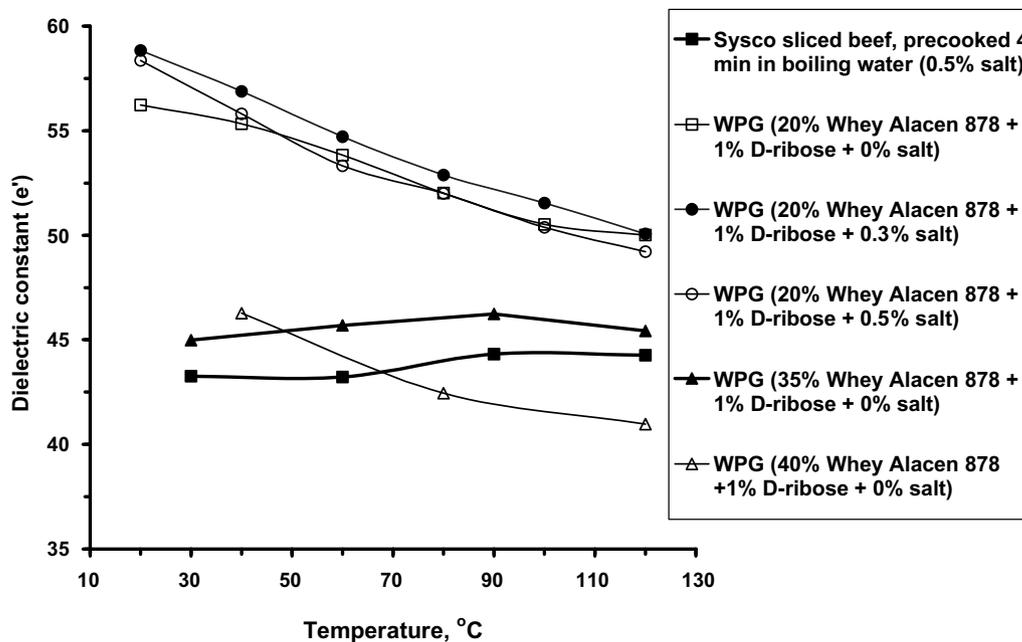


Fig. 7. Dielectric constants at 915 MHz for pre-cooked sliced beef and WPG with different compositions.

dielectric constants of the WPG with 35% Whey Alacen 878, 1% D-ribose, and 0% salt were close to those of the pre-cooked beef. Therefore, the WPG mentioned above was decided as a model food for heating pattern and cold spot determination.

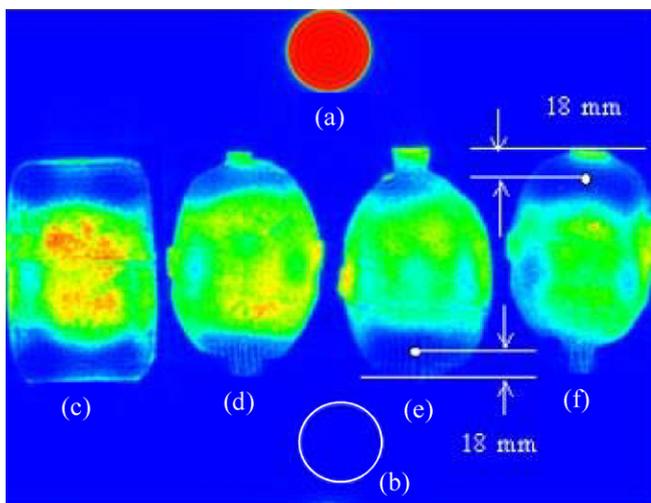
### 3.3. Heating patterns and cold spots

Previous studies (Pandit et al., 2007a) revealed that cold spots (obtaining the least thermal processing) located in the middle layer of the sample. This was caused by the limitations of the microwave power penetration and heat conduction. In this study, only the heating patterns of the middle layer in the MW treated WPG samples were analyzed (Fig. 8). Red and blue colors in two cylindrical samples served as the minimum and maximum values for the color scale used to analyze digital color images of the microwave treated WPG samples. Red represents the highest color value (255) for WPG samples saturated with chemical maker M-2; and blue defines the lowest color value (0) to fresh WPG samples. Although having different shapes, being placed at different positions and sealed in trays with different amounts of gravy, the four WPG samples had similar heating patterns during the MW process. More intensive heating occurred at the central area than at the areas close to the front and rear edges of the sample. Heating patterns were repeatable among the experimental replicates. The lowest and highest color value regions were defined as the cold spot and hot spot, respectively.

The cold spots were located at the points 18 mm away from the front and rear edges of the sample tray. The specified cold spots were considered as the ones of the pre-shrunk sliced beef in gravy, since the WPG and the beef matched each other with dielectric properties and had similar heating behavior in MW processing. The location of cold spot in a real food was validated by Pandit (2006), using a number of fiber-optic temperature sensors, to be the same location of cold spot pre-determined inside the model food.

### 3.4. Schedules for processing of sliced beef in gravy

Processing schedules developed for three thermal lethality levels:  $F_0 = 3, 6,$  and  $12$  min at the cold spot were summarized in Table 1.



**Fig. 8.** Heating patterns and cold spot locations in the middle layer of MW treated WPG samples. (a) Defining color scale – maximum color value; (b) defining color scale – minimum color value; (c) WPG with the shape of the tray; (d) WPG cut to the shape of sliced beef, placed at center of the tray; (e) WPG cut to the shape of sliced beef, placed close to front edge of the tray; and (f) WPG cut to the shape of sliced beef, placed close to back edge of the tray.

**Table 1**

Summary of processing schedules developed for MW sterilization of 7-oz trays of pre-treated sliced beef in gravy

$F_0$ (min)	Pre-heating time (min)	MW heating time (min)	Holding time (min)	Cooling time (min)	$C_0$ (min)
3	4.9	6.93	1/60	5	20
6	4.9	6.93	3	5	33
12	4.9	6.93	6	5	59

$C_0$  (in min) in the table is cook value at the cold spot with a reference temperature of  $100^\circ\text{C}$ . It was calculated using the following equation (Holdsworth, 1997):

$$C_0 = \int_0^t 10^{(T-T_{\text{ref}})/z_c} dt \quad (5)$$

where  $T$  is the measured temperature ( $^\circ\text{C}$ ) at the cold spot;  $T_{\text{ref}}$  is the reference temperature ( $100^\circ\text{C}$ );  $z_c$  has usually a value of  $33.1^\circ\text{C}$ ; and  $t$  is the heating time (min).

The  $D$  value of *Clostridium Botulinum* is generally considered to be 0.25 min at  $121.1^\circ\text{C}$  (Pflug and Odlough, 1978); and the processing achieving  $F_0$  of 3 min may result in 12-log reduction of *C. Botulinum* spores, which is the minimum thermal treatment used in the food industry for low-acid foods (Banwart, 1989). The processing schedule for  $F_0 = 3$  min was determined based on experiments. Fig. 9 shows an example process operated to achieve  $F_0 = 3$  min. The schedules for  $F_0 = 6$  and  $12$  min were established by extending the holding time after MW treatment but keeping the pre-heating, MW heating, and cooling times same as those for the  $F_0 = 3$  min schedule. Fig. 10 shows an example MW process with the processing schedule for  $F_0 = 6$  min.

The developed processing schedules are effective for the MW sterilization of sliced roast beef (without pre-shrinkage) in gravy. Product thickness reduces more for fresh sliced beef than for pre-treated beef during the process. Being processed with a same processing schedule, the beef without pre-shrinkage may receive more thermal treatment than the pre-shrunk beef with the same initial thickness. The more thermal treatment suggests that the MW processed product has a longer shelf life and is safer for consumption than expected. This was confirmed by preliminary processing tests on un-pre-treated sliced beef in gravy with  $F_0 = 6$  schedule; the average  $F_0$  of five test runs was  $7.7 \pm 1.5$  min, which was 1.7 min more than that for pre-shrunk beef in gravy; and the average cook value  $C_0$  was  $34.9 \pm 2.5$  min, 1.9 min more than that for pre-shrunk beef in gravy (Table 1).

For comparison, one processing test was conducted using the MW system as a retort ( $122^\circ\text{C}$  water only, without MW power), in which the trays were treated to the same thermal lethality level as the MW processed samples mentioned above. Test results showed that the time for heating the sample from  $60$  to  $121^\circ\text{C}$  and holding till starting cooling to achieve the set final  $F_0$  (8 min) in the MW process (9.9 min) was half of that in the retort process (19.5 min). The cook value  $C_0$  gained in the MW process was 60% of that gained in the retort process. If the 4 min pre-treatment time was counted as a part of the time for processing the pre-shrunk product, the processing time and the  $C_0$  gain in the MW process were still much lower than those in the retort process. Therefore, the MW sterilization for both pre-shrunk and fresh sliced beef in gravy could shorten processing time and reduce thermal deterioration to the product compared with the traditional retort process.

### 3.5. Inoculated pack studies

Table 2 summarizes the microbial test results for the MW treated and control sample trays that had been inoculated with *C. sporegenes* PA 3679 spores. All the eight control samples swelled

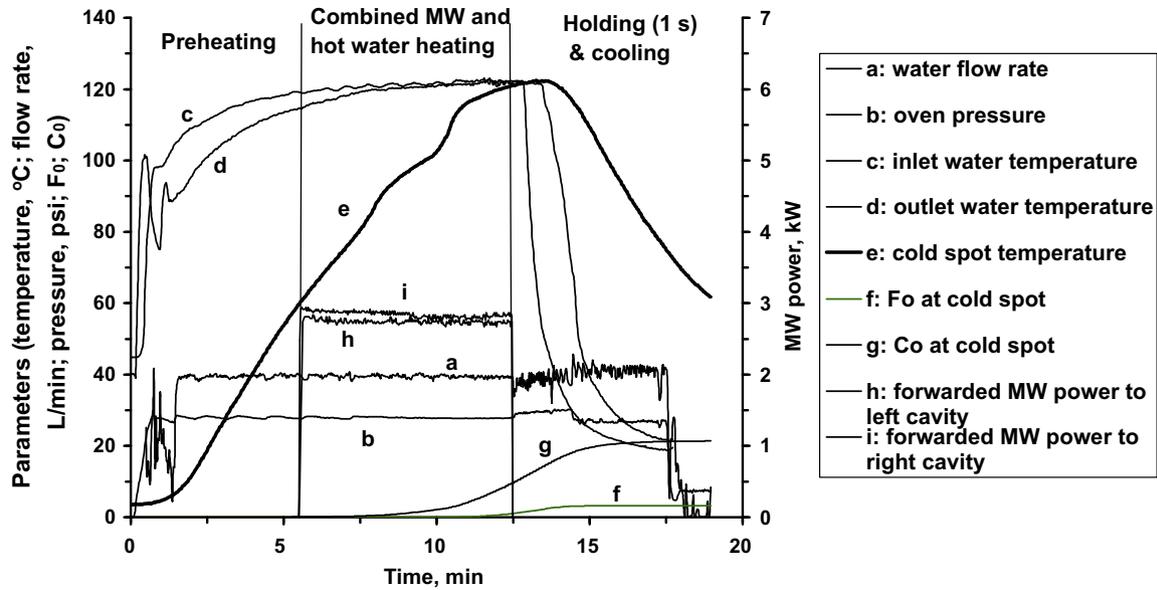


Fig. 9. Example of MW sterilization for 7-oz trays of pre-treated beef in gravy with 2.7 kW MW power and manual operation to achieve  $F_0 = 3$  min.

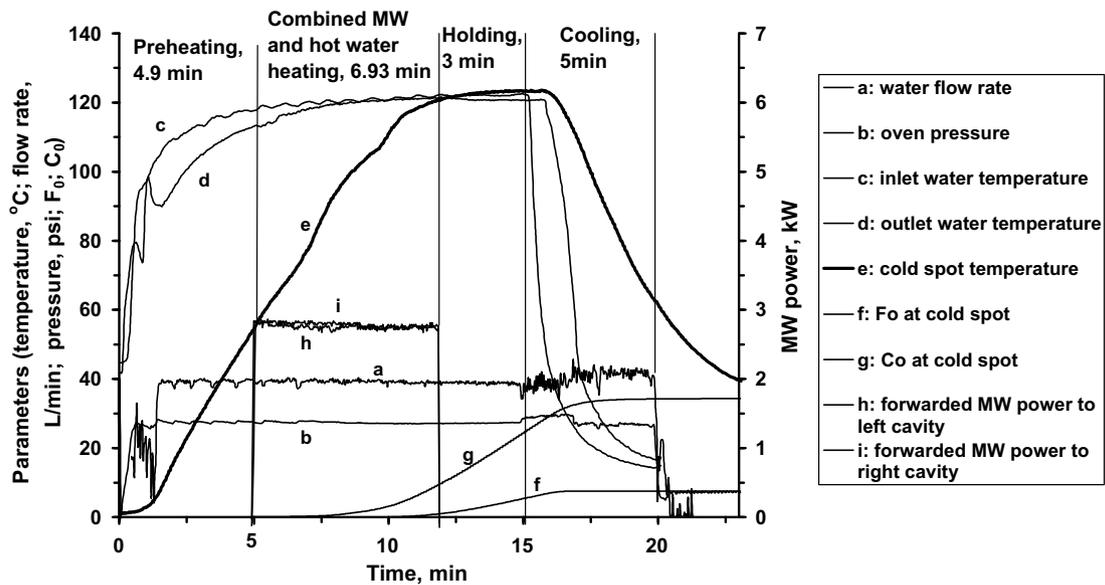


Fig. 10. MW process with the auto-processing schedule for  $F_0 = 6$  min.

Table 2

Results of inoculated pack studies for pre-treated sliced beef in gravy in 7-oz trays inoculated with *C. sporogenes* PA 3679 spores

Inoculation level (spores/tray)	Process level	$F_0$ (min)	Log-reduction value	Number of trays	Number of positive trays <sup>a</sup>
$1.3 \times 10^6$	Control	N/A	N/A	8	8
$1.3 \times 10^6$	Under-target	3	4	12	1
$1.3 \times 10^6$	Target	6	8	12	0
$1.3 \times 10^6$	Over-target	12	16	12	0

<sup>a</sup> Indicated by gas production and characteristic odor (storage period – 3 months).

within 4 days because of gas production from the growth of *C. sporogenes* PA 3679. Bulging was detected in one of the trays processed at  $F_0 = 3$  min. The other trays subjected to the target process ( $F_0 = 6$  min) and over-target process ( $F_0 = 12$  min) showed no evidence of gas production during 3 months of incubation. Therefore,

the microbial validation studies using inoculated packs of sliced beef in gravy suggested that at least one PA 3679 spore survived in one tray after the under-target process, but the target and over-target processes apparently sterilized the PA 3679 spores in the inoculated trays.

The inoculated packed studies results suggested that the method used in this study was feasible for developing MW processing of inhomogeneous foods, and that the developed processing was effective for sterilization of sliced beef in gravy prepackaged in 7-oz polymeric trays.

#### 4. Conclusions

A microwave (MW) sterilization process for sliced beef in gravy prepackaged in 7-oz polymeric trays was developed in this study. For processing schedule development, sliced beef was pre-treated for 4 min in boiling water with 0.5% salt before processing to limit possible movement of samples inside trays and make it possible to monitor temperatures at cold spots. Whey protein gel (35% Whey Alacen 878, 1% D-ribose, 0% salt, and 64% water) was chosen as a model food to emulate the pre-treated sliced beef for heating pattern and cold spot determination. The model and real foods had matched dielectric properties. Cold spots were determined to be 18 mm away from the front and rear edges in the middle layer of the tray using a chemical maker-aided computer vision method. Processing schedules to achieve different lethality levels ( $F_0 = 3, 6, \text{ and } 12 \text{ min}$ ) were established for treatment of the pre-cooked beef in gravy in 7-oz trays with the pilot-scale MW sterilization system developed at Washington State University. The developed processing schedules were verified by inoculated pack studies using *C. sporogenes* PA 3679 spores. The results suggested that the 915-MHz single-mode MW sterilization technology was effective for processing of the inhomogeneous food. The techniques provided in this study may be used for developing MW sterilization processes for other packaged inhomogeneous foods, such as fish in gravy in pouches and chicken meat in gray in trays. The engineering data collected in this study could be used for scaling-up the MW system for industrial application.

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