Optimization of Canning Process Conditions for Fermented African Oil Bean (*Pentaclethra macrophylla* Benth) Seeds in Three Different Media

Victor N. Enujiugha^a* and Charles T. Akanbi^b

^aDepartment of Food Science and Technology, Federal University of Technology, P.M.B. 704, Akure, Nigeria, ^bDepartment of Food Science and Technology, Obafemi Awolowo University, Ile-Ife, Nigeria, *venujiugha@vahoo.com

ABSTRACT

The thermal processing variables for starter culture fermented African oil bean (*Pentaclethra macrophylla* Benth) seed product *ugba* in three media (brine, refined groundnut oil and tomato sauce) and at three temperatures (110° C, 116° C and 121° C) were investigated. Thermal inactivation studies using *Bacillus stearothermophilus* 1518 spores showed that the z-values for the thermal processing were 10.5° C, 12.5° C and 11.0° C for brine-, oil- and sauce-canned samples, respectively. At 121° C processing temperature, the D values of 2.2 min (brine), 3.5 min (oil) and 3.2 min (sauce) would give 5D process times of 11.0, 17.5 and 16.0 min, respectively, as the targeted F values, which are lower than the experimental F values obtained for the three media. Heat penetration studies showed that 116° C, and 51.4-57.2 min at 110° C. Brine-canned samples had the shortest process times at 121° C. The process times by the general method were higher than those by the formula method. The heating rate index f_h was lower in the sauce-canned samples than in the other samples, especially at 121° C; also higher sterilization temperatures resulted in lower f_h values for all the media employed in the canning process. This was attributed to an increase in sterilization value due to faster heat penetration.

Key words: *Pentaclethra macrophylla* Benth, fermented product, canning process, *Bacillus stearothermophilus* spores, processing variables.

1. INTRODUCTION

The traditionally fermented product of African oil bean (*Pentaclethra macrophylla* Benth.) seed, commonly called *ugba*, is a popular condiment and snack along the west coast of Africa, where it is widely consumed across all segments of the population. In the local areas where animal protein and conventional vegetable protein sources are scarce, *ugba* has been effectively utilized in the local diet as a meat analogue and protein source. The proximate chemical composition (g/kg DM) of mature seeds of the African oil bean is as follows (mean \pm s.d., n = 3): crude protein 334.1 \pm 0.9, crude fat 485.0 \pm 1.1, crude fibre 62.8 \pm 2.6, crude ash 23.7 \pm 4.1, carbohydrates as nitrogenfree extracts (NFEs) 89.3 \pm 5.7 (Enujiugha and Agbede, 2000).

The widespread demand for ugba in areas where it is consumed and its restricted availability because of high rate of deterioration pose some problems with regard to its packaging. Packaging in leaves, as is presently carried out in household preparations of ugba, limits both the utilization and the shelf life. The average shelf life of locally prepared ugba is 14±2 days (Enujiugha, 2000). There is no known work in literature on ugba packaging to improve its utilization and shelf life via thermal processing. Canning as a preservation technique has been suggested for ugba processing (Enujiugha and Akanbi, 2002). Canning as a processing and preservation technique offers a unique advantage of preservation of the sensory attributes of the fermented product, with the added elongation of the shelf life.

Ugba is a low-acid food; a product of alkaline fermentation (Enujiugha and Akanbi, 2005). Low acid foods that are heat processed in cans are potentially severe hazard. Spores of proteolytic strains of *Clostridium botulinum*, if they survive the heat process, can germinate and grow in the anaerobic conditions that exist in the can and produce lethal toxin. Some potential spoilage organisms of low acid canned foods are more heat resistant than *C. botulinum*, e.g. *Bacillus stearothermophilus* is a food spoilage organism that grows well at elevated temperatures (40° C to 70° C) (Ng and Schaffner, 1997) and has very heat-resistant spores. Current practice for establishing microbiologically safe thermal processes relies upon heat penetration tests being done under 'worst case' (most adverse) conditions (Codex, 1993). A 'worst case' approach is the only option prior to production, where no data is available for adopting

statistical approaches. Adapting and applying the canning technique to a new product requires an understanding of the lethality parameters and the response of the most thermo-stable spores to the heat treatment. This study was aimed at developing optimal conditions for the thermal processing of fermented African oil bean product in lacquered cans using three different canning media.

All low acid foods support the growth of *Clostridium botulinum* and are normally processed on the assumption that they contain spores of this organism. Hence, sterilization value, F_o of 2.78 min. is required to guarantee the safety of such food products. Process times in minutes corresponding to $F_o = 2.78$ min. are usually calculated from the thermal penetration data by two methods: the general (or graphical) method and the nomogram (or mathematical) method (Amr and Yaseen, 1994). The mathematical method is sometimes referred to as the formula method (Pham, 1990). For nonhomogeneous foods, where nonuniform heating is found to occur, the slowest heating point is taken as the location with the smallest F_o value for the heating portion of the canning process (Lu *et al.*, 1991). Both numerical and analytical solutions have been widely used to predict time-temperature behavior of conduction-heating canned foods. All models are based on the solution of the heat diffusion equation with appropriate initial and boundary conditions.

2. MATERIALS AND METHODS

2.1 Materials

Freshly harvested African oil bean seeds (*Pentaclethra macrophylla* Benth) were obtained from local farmers at Nri in Anambra State of Nigeria. Upon receipt, the oil bean seeds were visually inspected and defective seeds were discarded. The seeds were then transported to the laboratory and kept in airtight polyethylene containers in a dry and cool environment until used for analysis. All the chemicals and reagents used in the study were of analytical grade.

2.2 Pure Culture Fermentation of the Seeds and Thermal Processing

The seeds were prepared and fermented as described previously (Enujiugha et al., 2008) using a mixture of *Bacillus subtilis* and *Bacillus licheniformis* at ratio 1:1. The prepared seed slices were inoculated with 10^{-1} dilution of the pure starter culture using sterile syringe at 3% v/w. Fermentation was carried out at 30° C for 4 days. The fermented oil bean seed slices were canned in 300 x 208 kk lac / lac (79 x 63 mm) cans with 300 kk lined ends (Carnaud Metal Box Nig. Plc., Ikeja–Lagos, Nigeria) with three different media (reflecting the use to which the fermented product is put); namely, tomato sauce, refined groundnut oil and brine solution. The tomato sauce used in the present study was prepared from the following ingredients: tomato puree (30% solids), 75%; flour, 5%; sugar, 14.5%; salt, 3%; vinegar, 1%; and spices, 1%; to give a total solids content of 14.0 ± 0.5% (Taiwo *et al.*, 1997). The brine solution used in the study contained 1.5% sodium chloride and 3% sucrose (w/v) as described by Ekanayake and Nelson (1990). The refined groundnut oil used was a commercial brand (Grand Cereals and Oil Mills, Jos, Nigeria) purchased from a local market at Ile-Ife, Nigeria.

2.3 Heat Penetration Studies

The thermal processing of the cans was done using an oil bath at 110° C, 116° C, and 121° C. The temperature at the center of the can was monitored during heating and cooling at 2-min intervals using iron-constantan probes (1.5 mm o.d., type J), connected by thermocouple wires (3.0 mm o.d. sheath and 0.5 mm o.d. wire) of the same type to a multi-channel temperature recorder. The temperature corresponding to various times were plotted on rectangular coordinate graph paper for calculation of process time by the graphical method. The temperature of the product at the centre of the can was plotted against time to obtain temperature-time curves from which the lethality values were computed as described by Jackson and Lamb (1981). Lethality values were plotted against time and the area under the curve was evaluated by counting the squares and using the trapezoidal rule to obtain the sterilizing value F_{0} for the process as well as the process time B. Temperature deficit was plotted against time on semi-log graph, from which necessary data were obtained. The general method was based on the equation $U = F x 10^{(121.1-T)/z}$, where U is the lethal time in minutes at temperature T, F is the lethal time at 121.1° C and z is the slope of the thermal death time (TDT) curve. The lethal rate, which is reciprocal of U, was calculated and used to construct the lethal rate curves from which process parameters were determined.

To calculate by the formula method, heat penetration data were plotted against time on a semi-log graph, the temperature deficit (temperature of heating medium minus cold point temperature) on the log scale. The lag factor for heating (J_h) , slope of the heating curve (f_h) , time in minutes for sterilization at retort temperature (U) and lag factor for cooling (J_c) were determined. The mathematical method is based on the equation $B = f_h (\log J_h I_h - \log g)$,

where B is the process time, I_h is the difference (°C) between retort temperature and food temperature at the start of heating, and g is the difference between retort temperature and the maximum temperature reached by the food at the point of concern. The parameters f_h/U , final temperature deficit g, process time B and final process time (TB) were calculated by the method of Stumbo (1973). Actual process time (TB) was determined by adding process time (B) to the effective heating period during come-up time, i.e. 42% of the come-up time. All the experiments were repeated at least three times and were subjected to statistical analysis, and the standard deviations determined.

2.4 Thermal inactivation studies

Spore suspensions of Bacillus stearothermophilus 1518 were prepared by the method of Lynt et al. (1977), using Cook and Brown medium. This medium has the following composition (in g/l): Bacto-Tryptone (Difco Laboratories, Detroit, Mich., USA), 3.0; Bacto-Peptone (Difco), 6.0; yeast extract (Difco), 3.0; Lab Lemco beef extract (Oxoid Ltd., London, England), 1.5; and agar (Difco), 15.0 for slants. Stock cultures were maintained on agar slants on the medium of Cook and Brown at 5°C. A loopful of slant culture was suspended into 0.05M phosphate buffer (pH 7.0) and heated at 75°C for 10 min, then plated on Nutrient Agar (Difco) and incubated for 18 to 20 hours at 55°C. The resulting lawn of growth was rinsed with sterile distilled water and removed from the agar surface by shaking gently with sterile glass beads (5.00 mm) (Ng et al., 2002). Three ml of culture suspension were dispensed onto 250 ml Cook and Brown medium (as above, except containing 25.0 g/l agar) in a 2.8 l Fernbach flask. After 4 days incubation at 55°C, the lawn of growth on the Cook and Brown medium was removed with 10 ml of sterile distilled water and small glass beads as above, and the combined suspensions from 25 Fernbach flasks containing a mixture of cell forms were centrifuged at 1,000 x g for 30 min at 5°C. The supernatant was decanted, and the residue was washed by suspension in chilled sterile water and recentrifugation. This washing procedure was repeated twice. Subsequently, the washed residue was suspended in 200 ml of 0.05 M potassium phosphate (pH 7.0) containing lysozyme (100 μ g/ml) and incubated with stirring at 37°C for 1 h. Enzyme-treated spores were washed with sterile water four times to remove vegetative debris (Feeherry et al., 1987).

After the final washing, the spores were suspended in Sorensen's M/15 phosphate buffer (to give 1.73×10^8 spores ml⁻¹) and stored at 4°C until used (Wallace *et al.*, 1978; Taiwo *et al.*, 1997). The spore suspension had no detectable vegetative cells, no spores in sporangia, and less than 5% germinated spores (Feeherry *et al.*, 1987). The filled cans containing the microbial spores were heat-processed at 110° C, 116° C and 121° C using the oil bath. Sterilization times were 5, 10, 15, 20, 25, 30, 35, 40, 45, 50 and 55 minutes. Survivor cell enumeration was conducted by surface plating appropriate 10-fold peptone water dilutions of the spores onto nutrient agar (NA). The plates were incubated at 55° C for 48 h. Survivor counts were reported in terms of \log_{10} cfu/ml and were plotted against exposure times to derive the survivor curve per heating temperature. The survivor curve equation was determined through linear regression. The *D* values of the composite inoculum were determined as the number of unit time needed for the survivor curve to traverse 1 \log_{10} cycle and were graphically equal to the negative reciprocal of the slope of the curve (Gabriel, 2007). The z-values were determined by constructing decimal reduction time curves (mean log *D*-value versus temperature), where $z = \text{slope}^{-1}$ (the temperature change necessary to effect a 10-fold change in the *D*-value) (McCormick *et al.*, 2003).

2.5 Statistical Analysis

Data collected from the study were subjected to analysis of variance (ANOVA) and differences among means were separated using Duncan's multiple range test (Duncan, 1955); significances were accepted at 5% level ($p \le 0.05$). The statistical software used was SPSS 10.0 for windows.

3. RESULTS AND DISCUSSION

3.1 D-, z- and F-values from thermal death time curves

The thermal death time (TDT) curves for the *Bacillus stearothermophilus* spores are shown on Figs 1 to 3. The trend of destruction of the spores assumed a linear relationship, with initial slight curvilinear structures for the lower temperatures (110° C and 116° C). At 121° C, the trend of destruction was a straight relationship, whereby successive increase in process time resulted in exponential destruction of the spores. This is in agreement with the observation of Feeherry *et al.* (1987) that there was at least 95% inactivation of the spore population of *Bacillus stearothermophilus* when the spores were heated at 121.1° C for 6min in a model aqueous system. Some researches (Jonsson *et al.*, 1977; Navani *et al.*, 1970) have demonstrated that the survival curves of spores heated at temperatures below 121° C frequently consist of an initial curvilinear portion succeeded by a logarithmic decline.

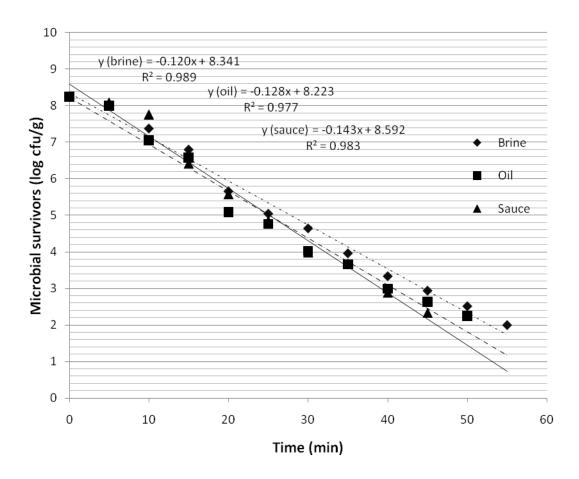


Fig. 1: The survivor curves for Bacillus stearothermophilus spores at 110° C

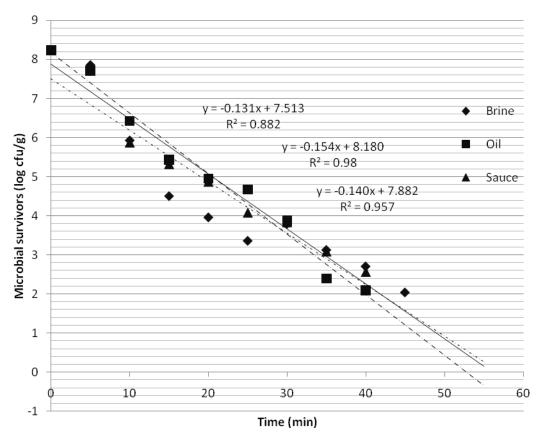


Fig. 2: Survivor curves for *Bacillus stearothermophilus* spores at 116°C

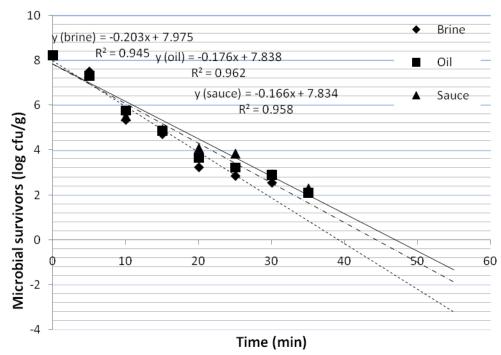


Fig 3: Survivor curves for Bacillus stearothermophilus spores at 121°C

The curvilinear portions may result from the effect of spore clumping, an activation process countering the inactivation process, or the existence of an intermediate state of thermal susceptibility between the normal viable state and death. The three canning media used (brine, sauce and oil) exhibited different patterns of response to the three treatment temperatures (110° C, 116° C and 121° C). The spore populations survived longer in the brine medium than in the other media at lower temperatures (110° C and 116° C), but survived for longer period in sauce and oil than in brine at 121° C. This could be attributed to the high conductive capacity of the medium containing stressful substances such as sugar and salt. Cook and Gilbert (1969) and Labbe (1979) have demonstrated that heated spores of *Bacillus stearothermophilus* are sensitive to NaCl in the medium. In this regard, it has been suggested that the presence of 1 to 3% NaCl may play an important role in the control of spoilage of low-acid (pH \geq 4.6) canned foods by thermophilic bacteria (Cook and Gilbert, 1969).

The experimentally-determined D- and z-values for the microbial survivors at the three temperatures (110°C, 116°C, and 121°C) and in the three canning media used are shown in Table 1. The D-values ranged from 11.2 to 12.0 min for the samples processed at 110° C; from 5.5 to 6.7 min for the samples processed at 116° C; and from 2.2 to 3.5 min for the samples processed at 121°C. These values fall within the range obtained by other workers (Taiwo et al., 1997; Wallace et al., 1978; Mallidis and Scholefield, 1985). The thermal resistance of the Bacillus stearothermophilus spores in the present study as characterized by D and z values indicate that these spores are extremely heat resistant. This agrees with the observation of Feeherry et al. (1987) who used aqueous suspensions of Bacillus stearothermophilus and concluded that a 12D process for Clostridium botulinum would not provide a 5D process as used for Bacillus stearothermophilus in NCA (1968). The D values reported in this study were within the range of D values reported for the five strains of Bacillus stearothermophilus at different temperatures by Jackson and Lamb (1981). The samples processed in brine had higher D values at 110°C and 116°C than those processed in oil and sauce at the same temperatures, revealing fewer survivors in the oil- and sauce-canned samples at lower temperatures. However at 121°C, the *Bacillus stearothermophilus* spores showed the least thermal resistance in the brine- canned samples, as evidenced from the lower D value obtained than in any of the other media. This same trend is also observed in the calculated D values indicating a highly lethal effect of brine on the spores at sterilization temperature of 121° C. The lower D values at 121° C (both experimental and calculated) indicate a higher lethal rate at this temperature than at 116°C. The results also indicate that canning in brine at 121°C for 35

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min would appear to be the optimized condition for fermented African oil bean seed slices. Canning the fermented product in oil or sauce would require a time of 40 min at 121°C.

	D-	values (min)		
Sample	110° C	116° C	121°C	z-values (° C)
Brine-canned	12.0±0.2 (8.81)	6.7±0.5 (7.26)	2.2±0.3 (5.27)	10.5±0.7
Groundnut oil-canned	11.5±0.5 (8.33)	6.0±0.4 (6.49)	3.5±0.1 (5.68)	12.5±1.1
Tomato sauce-canned	11.2±0.1 (7.61)	5.5±0.7 (7.04)	3.2±0.3 (5.89)	11.0±0.9

Table 1: Experimental D- and z-values for the microbial survivors

Values are expressed as the means of three replicate determinations (mean \pm s.d.). Figures in parenthesis are the calculated D-values.

The z value of 10.5° C obtained for the brine-canned samples in this study (Table 1) was not significantly different (p > 0.05) from the calculated z value of 10.33° C. Different z values in the range 6.6 - 12.3° C have been reported for spore suspensions of *Bacillus stearothermophilus* (Mallidis and Scholefield, 1985; Feeherry *et al.*, 1987; Rodrigo and Martinez, 1988). The range of z values obtained in this study for the three canning media ($10.5 - 12.5^{\circ}$ C) is within the generally reported range. The z value is usually expressed as the number of degrees Celsius required to bring about a tenfold change in the death time or death rate (NCA, 1968).

Table 2 gives the values for the experimental and calculated F. F is the number of minutes required to destroy a given number of organisms at a given temperature. The F value is used to compare the sterilizing values of different processes. Pflug and Odlaug (1978) in their review of z and F values to ensure the safety of low-acid canned foods concluded that using a single z value of 10° C is still the best recommended for calculating low-acid canned food sterilization processes. In the present study, the experimental F values were higher than the calculated or targeted F values. In establishing a new processing schedule, a 5D for the test organism is equivalent to or greater than a 12D process for *Clostridium botulinum* (Wallace *et al.*, 1978). A 12D process for *Clostridium botulinum* results in 2.6 - 3.0 min process at 121.1°C (Pflug and Odlaug, 1978). In the present study, at 121°C processing temperature, the D values of 2.2 min (brine), 3.5 min (oil) and 3.2 min (sauce) would give 5D process times of 11.0, 17.5 and 16.0 min, respectively, as the targeted F values, which are lower than the experimental F values obtained for the three processing media at the same temperature. This suggests that the process times employed in this study would yield a commercially safe product.

	Table 2:	Experimental F-values ((min)) derived from	the decimal	reduction times*
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	F-י		
Sample	110° C	116° C	121° C
Brine-canned	74.88±1.06	41.54±0.18	12.52±1.20
	(60.0)	(33.5)	(11.0)
Groundnut oil-canned	69.00±0.91	36.96±2.03	21.56±0.15
	(57.5)	(30.0)	(17.5)
Tomato sauce-canned	66.19±1.10	31.24±0.33	19.01±0.17
	(56.0)	(27.5)	(16.0)

*Derived from survivor curves as F = D (log a – log b) (NCA, 1968) Values are expressed as the means of three replicate determinations (mean ± s.d.). Figures in parenthesis are calculated F-values (F = 5D).

The results of this study indicate that lower temperatures would result in longer process times and this may not be good for some of the seed nutrients as well as the maintenance of commercial sterility. For better nutrient retention, the process time should be short, even if it means raising the temperature of processing. The best processing conditions derived from the present study are those at 121°C.

3.2 Process times by general and formula methods

The thermal process times for fermented oil bean seed slices in the three media at three temperatures according to general (graphical) method are presented in Table 3. At sterilization temperature of 121° C, F_o value was equal to 5.69D for brine-canned samples, 6.16D for oil-canned samples, and 5.94D for sauce-canned samples. Since the experimental F values were higher than the calculated (or targeted) F values, as earlier mentioned in this report, to ensure complete microbiological safety under commercial processing conditions, thermal process equivalent to D values higher than 5.69, 6.16 and 5.94 min in the respective media is advisable. The process times decreased with increasing processing temperatures. The rate of decrease was highest in brine-canned samples, followed by the oil-canned samples, respectively, at the sterilization temperature of 121° C. The same samples had process times of 49.4 min, 45.5 min and 46.2 min, respectively, at the temperature of 116° C. These values are within the range reported for some other food products by other workers (Wallace *et al.*, 1978; Amr and Yaseen, 1994; Taiwo *et al.*, 1997). The graphical method of process calculation is an accurate method according to Nath and Ranganna (1983), as no assumptions about the nature of heating or cooling curves are involved. However, this method can be used only when the processing conditions such as can size, and initial and processing temperatures are identical to the conditions used in the heat penetration studies (NCA, 1968). The formula method overcomes these disadvantages.

	Process temperature	Brine	Oil	Sauce
Decimal reductions'	* 110° C	6.24	6.00	5.91
	116° C	6.20	6.16	5.68
	121° C	5.69	6.16	5.94
Process time (min)	110° C	57.2	54.9	51.4
	116° C	49.4	45.5	46.2
	121° C	39.3	41.6	42.7

Table 3: Process times (min) according to the graphical method for different decimal reductions

*Values are expressed as the F/D ratio (Nath and Ranganna, 1983)

The results of process time calculations based on the mathematical (formula) method are presented in Table 4. The Ball's process times (B) were in the range 49.82 - 54.12 min for the samples processed at 110° C; 42.41 - 47.11 min for the samples processed at 110° C; 42.41 - 47.11 min for the samples processed at 110° C; and 38.11 - 40.56 min for the samples processed at 121° C. The results indicate that cans processed at higher temperatures required lower process times than those processed at the lower temperatures. This is in agreement with the results from studies elsewhere (Wang *et al.*, 1988; Amr and Yaseen, 1994). The process times by the general method were slightly higher than those of the formula method; this was also the observation of Amr and Yaseen (1994). The heating rate index f_h was lower in the sauce-canned samples than in the other samples, especially at 121° C. This could be attributed to an increase in sterilization value due to faster heat penetration. Higher sterilization temperatures resulted in lower f_h values, which agrees with the works of Amr and Yaseen (1994) and Taiwo *et al.* (1997). In the calculation of heat penetration by the formula method, the heating rate index f_h is assumed to be equal to the cooling rate index f_c . The results of the lag factors and temperature deficit indicate a very short come-up time due to the fact that the temperature of the heating medium (oil) only slightly reduced upon the introduction of the sealed cans. This was made possible with the use of oil bath, and not vertical retort, in the heat penetration studies. It is usually recommended that the come-up time should generally be kept as short as possible consistent with good commercial practice (NCA, 1968).

Table 4: Thermal process parameters at the three temperatures according to formula method

Parameter	Brine	Oil	Sauce
Heating rate index f_h (min)	27.83±0.03 ^{a*}	25.10±0.17 ^b	23.51±1.03 ^b
Heating lag factor $J_{\rm h}$	1.13 ± 0.12^{a}	0.88 ± 0.11^{b}	1.07 ± 0.16^{a}
Cooling lag factor $J_{\rm c}$	$1.97{\pm}0.06^{a}$	1.50 ± 0.02^{a}	1.33 ± 0.09^{a}
Temperature deficit g (°C)	$0.43{\pm}0.05^{a}$	$0.39{\pm}0.02^{b}$	$0.44{\pm}0.04^{a}$
Process time B (min)	54.12 ± 1.20^{a}	50.09±1.15 ^a	49.82±1.50 ^a

(a) Values for process parameters at 110° C

(b) Values for process parameters at 116° C

Parameter	Brine	Oil	Sauce
Heating rate index $f_{\rm h}$ (min)	27.15±1.20 ^{a*}	21.04±1.15 ^b	22.50±0.90 ^b
Heating lag factor $J_{\rm h}$	$0.98{\pm}0.03^{b}$	1.23 ± 0.02^{a}	1.20 ± 0.05^{a}
Cooling lag factor $J_{\rm c}$	2.22 ± 0.12^{a}	1.38 ± 0.03^{b}	1.27 ± 0.15^{b}
Temperature deficit g (°C)	$0.95{\pm}0.03^{a}$	0.91 ± 0.10^{a}	1.12±0.03 ^a
Process time B (min)	47.11 ± 1.32^{a}	42.41 ± 1.52^{a}	45.07±1.33 ^a

(c) Values for process parameters at 121°C

Parameter	Brine	Oil	Sauce
Heating rate index f_h (min) Heating lag factor J_h Cooling lag factor J_c Temperature deficit g (°C) Process time B (min)	$\begin{array}{c} 23.25{\pm}0.17^{a^{*}}\\ 0.76{\pm}0.04^{b}\\ 4.84{\pm}0.06^{a}\\ 1.73{\pm}0.02^{a}\\ 38.11{\pm}1.22^{b} \end{array}$	$\begin{array}{c} 20.10{\pm}0.12^{b}\\ 0.88{\pm}0.02^{b}\\ 1.75{\pm}0.05^{b}\\ 1.53{\pm}0.06^{a}\\ 38.60{\pm}1.53^{b} \end{array}$	$\begin{array}{r} 19.55{\pm}1.02^{b}\\ 1.63{\pm}0.01^{a}\\ 1.59{\pm}0.10^{b}\\ 1.32{\pm}0.01^{a}\\ 40.56{\pm}1.38^{a} \end{array}$

Each value is expressed as the mean±s.d. of three determinations

*Values with the same letters in superscript along a row are not significantly different (p > 0.05)

4. CONCLUSION

The thermal processing requirements for fermented African oil bean seed product in lacquered can have been evaluated in this study. The z-values were 10.5° , 12.5° and 11.0° C for brine-, oil- and sauce-canned samples, respectively. The corresponding D values at 121° C processing temperature were 2.2 min (brine), 3.5 min (oil) and 3.2 min (sauce). The targeted F values were higher than the experimental F values, indicating a commercially safe product. Process times according to graphical method were in the range 39.3 - 42.7 min at 121° C, 45.5 - 49.4 min at 116° C, and 51.4 - 57.2 min at 110° C. These process times were higher than those by the mathematical method, which is desirable in terms of energy use and plant throughput.

5. **REFERENCES**

Amr, A. S. and E. I. Yaseen. 1994. Thermal processing requirements of canned chickpea dip. International Journal of Food Science and Technology 29: 441-448

Codex. 1993. Recommended International Code of Hygiene Practice for Low Acid and Acidified Low Acid Canned Foods. CRC/RCP 23-1979, Rev. 2. Rome: Food and Agriculture Organization of the United Nations

Cook, A. M. and R. J. Gilbert. 1969. The effects of sodium chloride on heat resistance and recovery of heated spores of *Bacillus stearothermophilus*. *Journal of Applied Bacteriology* **32**: 96-102.

Duncan, D. B. 1955. Multiple range and multiple F tests. *Biometrics* 11: 1-42.

- Ekanayake, A. and P. E. Nelson. 1990. Effect of thermal processing on lima bean vitamin B-6 availability. *Journal* of Food Science **55**(1): 154-157.
- Enujiugha, V. N. 2000. Development of a new food paste from *Pentaclethra* species. *Applied Tropical Agriculture* **5**(2): 89-94.
- Enujiugha, V. N. and J. O. Agbede. 2000. Nutritional and anti-nutritional characteristics of African oil bean (*Pentaclethra macrophylla* Benth) seeds. *Applied Tropical Agriculture* **5**(1): 11-14.
- Enujiugha, V. N. and C. T. Akanbi. 2002. Adaptable techniques for the post-harvest processing and preservation of African oil bean seeds. In Proc. Regional Workshop on Promotion of Appropriate Agro-processing Technologies in West Africa, 23-26 October, 164-169. Ile-Ife, Nigeria.
- Enujiugha, V. N. and C. T. Akanbi. 2005. Compositional changes in African oil bean (*Pentaclethra macrophylla* Benth) seeds during thermal processing. *Pakistan Journal of Nutrition* **4**(1): 27-31.
- Enujiugha, V. N., C. T. Akanbi and H. A. Adeniran. 2008. Evaluation of starters for the fermentation of African oil bean (*Pentaclethra macrophylla* Benth) seeds. *Nutrition and Food Science* **38**(5): 451-457.
- Feeherry, F. E., D. T. Munsey and D. B. Rowley. 1987. Thermal inactivation and injury of *Bacillus* stearothermophilus spores. Applied and Environmental Microbiology **53**(2): 365-370.
- Gabriel, A. A. 2007. D-values of composite Salmonella enterica serotypes typhimurium and enteritidis in Philippine native orange juice. Journal of Food Processing and Preservation **31**: 649-658.
- Jackson, A. T. and J. Lamb. 1981. Calculations in food and chemical engineering: Theory, worked examples and problems, 98-102. London, UK: The MacMillan Press Ltd.
- Jonsson, U., B. G. Snygg, B. G. Harnulv and T. Zachrisson. 1977. Testing two models for the temperature dependence of the heat inactivation rate of *Bacillus stearothermophilus* spores. *Journal of Food Science* **42**: 1251-1252,1263.
- Labbe, R. G. 1979. Recovery of spores of Bacillus stearothermophilus from thermal injury. Journal of Applied Bacteriology 47: 457-462.
- Lu, Q., S. J. Mulvaney and F. Hsieh. 1991. Thermal processes for metal cans compared to retortable plastic containers. *Journal of Food Science* 56(3): 835-837.
- Lynt, R. K., H. M. Solomon, T. Lilly and D. A. Kautter. 1977. Thermal death time of *Clostridium botulinum* type E in meat of blue crabs. *Journal of Food Science* **43**: 1022-1025.
- Mallidis, C. G. and J. Scholefield. 1985. The release of dipicolinic acid during heating and its relation to the heat destruction of *Bacillus stearothermophilus* spores. *Journal of Bacteriology* **59**: 479-486.
- McCormick, K., I. Y. Han, J. C. Acton, B. W. Sheldon and P. L. Dawson. 2003. D- and z-values of *Listeria monocytogenes* and *Salmonella typhimurium* in packaged low-fat ready-to-eat turkey bologna subjected to a surface pasteurization treatment. *Poultry Science* 82: 1337-1342.
- Nath, N. and S. Ranganna. 1983. Determination of a thermal process schedule for guava (*Psidium guajava* Linn.) Journal of Food Technology 18(3): 301-316.
- Navani, S. K., J. Scholefield and M. R. Kibby. 1970. A digital computer program for the statistical analysis of heat resistance data applied to *Bacillus stearothermophilus* spores. *Journal of Applied Bacteriology* **33**: 609-620.
- NCA. 1968. Laboratory Manual for Food Canners and Processors, vol. 1, 3rd edn., 166-351. National Canners Association. Connecticut, USA: The AVI Publishing Co.
- Ng, T. M. and D. W. Schaffner. 1997. Mathematical models for the effects of pH, temperature, and sodium chloride on the growth of *Bacillus stearothermophilus* in salty carrots. *Applied and Environmental Microbiology* **63**: 1237-1243.
- Ng, T. M., E. Viard, M. I. Caipo, S. Duffy and D. W. Schaffner. 2002. Expansion and validation of a predictive model for the growth of *Bacillus stearothermophilus* in military rations. *Journal of Food Science* **67**(5): 1872-1878.
- Pflug, I. J. and T. E. Odlaug. 1978. A review of z and F values used to ensure the safety of low-acid canned foods. *Food Technology*, **6**, 63-70.
- Pham, Q. T. 1990. Lethalithy calculation for thermal processes with different heating and cooling rates. *International Journal of Food Science and Technology* **25**: 148-156.
- Rodrigo, M. and A. Martinez. 1988. Determination of a process time for a new product: canned low-acid artichoke hearts. *International Journal of Food Science and Technology* **23**: 31-41.
- Stumbo, C. R. 1973. *Thermobacteriology in food processing*, 2nd edn., 143-154, 255-325. New York, USA: Academic Press, Inc.
- Taiwo, K. A., C. T. Akanbi and O. O. Ajibola. 1997. Establishing Processing Conditions for canning cowpea seeds in tomato sauce. *International Journal of Food Science and Technology* **32**: 313-324.

- Wang, C. R., K. C. Chang and I. Grafton. 1988. Canning quality evaluation of pinto and navy beans. *Journal of Food Science* 53: 772-776.
- Wallace, M. J., K. L. Nordsiden, I. D. Wolf, D. R. Thompson and E. A. Zottola. 1978. Thermal inactivation of *Clostridium sporogenes* PA 3679 and *Bacillus stearothermophilus* 1518 in low-acid home-canned foods. *Journal of Food Science* 43: 1738-1740.

6. NOMENCLATURE

- D = time required at any temperature to destroy 90% of the microbial spores. This is mathematically equal to the reciprocal of the slope of the survivor curve
- z = temperature difference required for a ten-fold change in the decimal reduction time (°C)
- $f_{\rm h}, f_{\rm c}$ = inverse slopes of heating and cooling curves (min)
- $J_{\rm h}$, $J_{\rm c}$ = intercept coefficient of the heat penetration plot during heating and cooling
- g = difference between temperature of heating medium and temperature at a point in the can at the end of heating (°C)
- U = time in minutes for sterilization at temperature of heating medium
- B = Ball's process time in minutes
- a = initial spore population
- b = final spore population
- I_h = difference between temperature of heating medium and food temperature at the start of heating (°C)
- T = operating temperature
- F = the number of minutes required to destroy a given number of organisms at a given temperature
- $F_o =$ mass average sterilizing value (min) of the heat process at 121°C when z = 10°C