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CONTENTS

RESEARCH NOTE:

HEMAGGLUTINATING ACTIVITY OF LECTINS IN SELECTED VARIETIES OF RAW AND PROCESSED DRY BEANS[1]

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ABSTRACT

*The effects of several processing methods on the hemagglutinating activity of ten varieties of dry beans (*Phaseolus vulgaris L.*) were studied using a serial dilution technique. Lectin activity ranged from 128–512 × 10^4 units/g whole bean flour, with a mean value of 307 × 10^4 units/g bean. Small Red and Kidney bean types had the highest agglutinating activities among the varieties studied. No noticeable changes were observed upon dehulling beans, while germinated beans and protein concentrates had lower hemagglutinating activity. Cooking of beans completely destroyed the lectin activity.*

INTRODUCTION

Lectins (phytohemagglutinins) are glycoproteins with the unique property of binding saccharides and saccharide-containing proteins in a highly specific fashion. Hemagglutinating activity of lectins in *Amaranthus leucocarpus* seeds has been found to affect protein quality (Calderon de la Barca *et al.* 1985). In addition to erythrocyte agglutination, lectins can interact with other types of cells. Lectins make up 2–10% of the protein of many leguminous seeds, and are particularly concentrated in the kidney beans.

Lectins in dry beans have been shown to be tetrameric glycoproteins that exist as a mixture of five hybrids of similar chemical properties but slightly different

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biological activity (Leavitt *et al.* 1977; Jaffe 1983). Unlike soybean lectins, which have little, if any, direct effect on the nutritive value of soy proteins, dry bean lectins inhibit rat growth at levels as low as 0.5% of the diet, with higher levels hastening the onset of death (Turner and Liener 1976; Pusztai and Palmer 1977). In addition, cases of toxicity were reported in humans who had eaten raw or partially cooked Red Kidney Beans (Noah *et al.* 1980).

The effects of several processing methods on the removal of phytic acid, tannins and enzyme inhibitory activities of dry beans have been reported elsewhere (Deshpande and Cheryan 1983, 1984; Deshpande *et al.* 1982). The effects on residual lectin activity in dry beans of these processing methods are described here.

MATERIALS AND METHODS

Dry beans of *Phaseolus vulgaris L.* biotypes were obtained from Roger Brothers Seed Company, Twin Falls, Idaho. The seed colors and types are described in Table 1.

Whole and Dehulled Bean Flours

Whole and dehulled bean flours (60-mesh particle size) were prepared as described by Deshpande *et al.* (1982).

TABLE 1
DRY BEAN VARIETIES AND SEED TYPES SELECTED FOR THE STUDY

Variety	Color	Seed Type
Navy Bean	White	Sanilac
Great Northern	White	D 76067
Small White	White	D 76063
Pink Bean	Pink	Viva
Pinto Beans	Mottled Brown	UI 111
Cranberry	Mottled Pink-Brown	Michigan-Improved
Light Red Kidney	Light Red	Manitou
Dark Red Kidney	Dark Red	Charlevoix
Small Red	Dark Red	UI 36
Black Bean	Dark Black	Black Beauty

Protein Concentrates

The preparation of protein concentrates from the whole beans of Small White, Great Northern, Viva Pink, Cranberry and Light Red Kidney varieties was reported earlier (Deshpande and Cheryan 1984).

Cooking

Intact whole beans (approximately 100 g) were first rinsed with distilled water and then cooked in glass beakers covered with aluminum foil. Bean-to-water ratios were 1:4 (w/v). The cooking temperature was 99°C. Beans were cooked for 15 and 30 min and till they were fully cooked. At the end of the desired period, they were quickly frozen along with the cooking broth, freeze dried, and then ground to obtain 60-mesh flours.

Germination

Whole sound beans (about 50 g) were washed in a 2% solution of sodium hypochlorite and then left soaking in distilled water for 12 h. The soaked beans were washed, spread evenly on cheese-cloth lined glass plates, and were germinated in dark at 37°C. During the germination period, the beans were rinsed twice each day and were sprayed periodically with distilled water. Contaminated seedlings were discarded. Samples were collected at the end of 2, 4 and 6 days and freeze dried. A portion of the 6-day germinated samples was also cooked till soft and freeze dried along with the cooking broth. In both cases, unsoaked and ungerminated beans served as controls.

Hemagglutinating Activity

Samples (100 mg, corrected for moisture content) were extracted with 0.9% sodium chloride (saline) solution for 12 h at 4°C. Trypsinized red blood cells from male sheep were prepared as described by Liener (1955). A 4% suspension of erythrocytes was used for the agglutination test. Starting with 0.1 mL of sample extract, serial two-fold dilutions were made with saline on Falcon 3070 microtiter plates. To each well was then added 0.1 mL of erythrocyte suspension. The plates were gently shaken to mix the contents. After 4 h at room temperature (25°C), agglutination patterns were observed with the aid of a microscope. A series of wells containing only saline and erythrocyte suspension served as negative controls. One hemagglutination unit (HU) was defined as the lowest dilution of hemagglutination occurring under the experimental conditions defined here. The total hemagglutinating activity was calculated as described by Liener and Hill (1953). The results of all hemagglutination analyses are means of four individual determinations per sample. Whenever the replicates varied within one dilution range, the higher dilution was considered for calculating the activity.

RESULTS AND DISCUSSION

The effects of various processing methods on the hemagglutinating activities of lectins of dry bean varieties used in this study are summarized in Table 2. The values obtained for the hemagglutination activity of the whole dry beans were within the ranges reported for several legume species (Nachbar and Openheim 1980; Stein 1976; Thompson *et al.* 1983). In the present study, these values for the whole and dehulled bean lectins ranged from 128–512 $\times$ 10^4 and 128–1024 $\times$ 10^4 units/g of bean flour, with mean values of 307 $\times$ 10^4 and 576 $\times$ 10^4 units/g, respectively. Meaningful comparisons, however, are difficult to make because of the type of erythrocytes used in different studies. Often times, lectins from different sources show agglutinating activity only against specific types of erythrocytes. Small Red and Kidney beans had the highest agglutinating activities among the varieties investigated. Although lectins are found in many commonly eaten foods (Nachbar and Openheim 1980), they are probably most concentrated in legumes.

Unlike phytic acid and enzyme inhibitors (Deshpande *et al.* 1982), no significant increase in lectin activity was observed on dehulling of beans. The mean increase was equivalent to only one serial dilution. A major source of error in the serial dilution technique used for lectin analysis is the identification of the visual agglutination end point. Because of the serial two-fold dilution involved, an error equivalent to one dilution in the estimation of lectin activity is usual. Therefore, a difference in activity equivalent to one dilution is seldom considered significant.

Lectins are known to be quite heat labile. Simple heat processing is usually adequate to destroy the hemagglutinating activity of dry bean lectins. In fact, in optimally cooked beans and those cooked for 30 min, no hemagglutinating activity could be detected. Even in the samples cooked for 15 min, the hemagglutinating activity decreased below detectable levels in most varieties (data not shown here). Only the beans of Red Kidney and Small Red varieties had residual activity (0.5–1.0 $\times$ 10^4 units/g cooked beans). It appears that cooking beans to the point where they may be considered edible is more than sufficient to destroy virtually all of the hemagglutinating activity of lectins. Thompson *et al.* (1983) also observed complete destruction of the hemagglutinating activity of two commercial Kidney bean samples during cooking even before the beans were considered edible. If the uncooked beans are used, for example as in salads, then they could be responsible for the gastrointestinal symptoms occasionally reported with the consumption of Kidney beans (Noah *et al.* 1980).

Germination was quite effective in lowering the lectin activity of dry beans (Table 2). Hemagglutinating activity in the germinated beans decreased by 2–3 fold, while further cooking of the germinated beans lowered the activity below

TABLE 2 .
HEMAGGLUTINATING ACTIVITIES OF RAW AND PROCESSED DRY BEANS

Variety	Whole Beans[a]	Dehulled Beans[a]	Germination (days)[a] 2	4	6	Protein Concentrates[b]
Navy	128	256				
Great Northern	256	512	256	128	64	684 (1183)[c]
Small White	128	128				367 (649)
Cranberry	256	512				672 (1093)
Viva Pink	256	512	256	64	32	689 (1202)
Pinto	256	512				
Light Red Kidney	512	1024				1340 (2451)
Dark Red Kidney	512	1024				
Small Red	512	1024				
Black Bean	256	256	256	128	64	

[a]Values represent HU x 10^4/g bean on a dry weight basis.

[b]Values represent HU x 10^4/g protein on a dry weight basis.

[c]Values in parentheses refer to HU x 10^4 for the respective bean flour controls on per g protein basis.

detectable levels. Although the lectin activity, expressed on per gram protein basis for the protein concentrates, was also slightly lowered, on a unit weight basis, the concentrates, because of their high protein content, would have higher total lectin activity as compared to whole beans. However, further processing of concentrates when used as adjuncts in various food systems should completely eliminate the hemagglutinating activity.

The results of this study suggest that unlike heat stable antinutrients (such as phytate and tannins) and, to some extent, enzyme inhibitors, lectins do not seem to pose any special problems after proper cooking of dry beans for human consumption. However, improperly cooked beans or dehulled beans and their protein concentrates as used in certain food systems may cause minor gastrointestinal disturbances. Any such effects would be relatively small, since beans are but only one component of the human diet. Thus, the antinutritional effects of lectins in inadequately processed bean diets will be diluted or minimized, and, most certainly, very difficult to measure.

REFERENCES

CALDERON DE LA BARCA, A. M., OCHOA, J. L. and VALENCIA, M. E. 1985. Effect of the extraction of hemagglutinin on the nutritive value of *Amaranthus leucocarpus* seeds. J. Food Sci. *50,* 1700.

DESHPANDE, S. S. and CHERYAN, M. 1983. Changes in phytic acid, tannins and trypsin inhibitory activity on soaking of dry beans (*Phaseolus vulgaris*). Nutr. Rep. Int. *27,* 931.

DESHPANDE, S. S. and CHERYAN, M. 1984. Preparation and antinutritional characteristics of dry bean protein concentrates. Qual. Plant. Plant Foods Hum. Nutr. *34,* 185.

DESHPANDE, S. S., SATHE, S. K., SALUNKHE, D. K. and CORNFORTH, D. P. 1982. Effects of dehulling on phytic acid, polyphenols and enzyme inhibitors of dry beans (*Phaseolus vulgaris*). J. Food Sci. *47,* 1846.

JAFFE, W. G. 1983. Nutritional significance of lectins. In *CRC Handbook of Naturally Occurring Food Toxicants,* (M. Rechcigl, Jr., ed.) p. 31, CRC Press, Boca Raton, FL.

LEAVITT, R. D., FELSTED, R. L. and BACHUR, N. R. 1977. Biological and biochemical properties of *Phaseolus vulgaris* isolectins. J. Biol. Chem. *252,* 2961.

LIENER, I. E. 1955. The photometric determination of the hemagglutinating activity of soyin and crude soybean extracts. Arch. Biochem. Biophys. *54,* 223.

LIENER, I. E. and HILL, E. G. 1953. The effect of heat treatment on the nutritive value and hemagglutinating activity of soybean oil meal. J. Nutr. *49,* 609.

NACHBAR, M. S. and OPENHEIM, J. D. 1980. Lectins in the United States diet: A survey of lectins in commonly consumed foods and a review of the literature. Am. J. Clin. Nutr. *33,* 2338.

NOAH, N. D., BENDER, A. E., REAIDI, G. B. and GILBERT, R. J. 1980. Food poisoning from raw red kidney beans. Brit. Med. J. *281,* 236.

PUSZTAI, A. and PALMER, R. 1977. Nutritional evaluation of kidney beans (*Phaseolus vulgaris*): The toxic principle. J. Sci. Food Agr. *28,* 620.

STEIN, M. 1976. Natural toxicants in selected leguminous seeds with special reference to their metabolism and behavior on cooking and processing. Qual. Plant. Plant Foods Hum. Nutr. *26,* 277.

THOMPSON, L. U., REA, R. L. and JENKINS, D.J.A. 1983. Effects of heat processing on hemagglutinin activity in red kidney beans. J. Food Sci. *48,* 235.

TURNER, R. H. and LIENER, I. E. 1976. The effects of the selective removal of hemagglutinins on the nutritive value of soybeans. J. Agr. Food Chem. *23,* 484.

INFLUENCE OF ULTRASOUND ON THE PROPERTIES OF CHYMOSIN AND THE ULTRASTRUCTURE OF ABOMASUM DURING CHYMOSIN EXTRACTION[1]

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ABSTRACT

The effects of ultrasound treatment on the properties of chymosin and the ultrastructure of abomasum during chymosin extraction were investigated. Ultrasound treatment at 36 W/cm² for 80 min did not decrease chymosin activity and its proteolytic activity. However, high intensity ultrasound (157 W/cm²) and the prolonged exposure to high intensity significantly decreased chymosin activity and its proteolytic activity. Ultrasound treatment changed amino acid composition of chymosin. There were significant changes in the mole percent of histidine, proline, tyrosine, methionine, valine, and lysine at the lower specific intensity. Ultrastructure of abomasal tissue was not affected by ultrasound treatment at specific intensity 36 W/cm² for 80 min. However, some mitochondria cristae were damaged and secretory granules were more compact even though their numbers remained the same.

INTRODUCTION

The release of enzymes and proteins from cells and subcellular particles is an outstanding application of ultrasound. Biologically active substances extracted by ultrasound are sometimes more active than thought theoretically possible (Hess and Slade 1955). Ultrasound causes the destruction of the cellular structure by a cavitation effect (Hughes and Nyborg 1962) and increases the activity of substances which are bound in cell structures (Fry 1978). Destruction of animal cells was explained by the development of cavitation inside the cells. In animal

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tissues and extractive media, favorable conditions existed for the development of exterior cavitation (Hughes and Nyborg 1962).

Ultrasound increased the diffusion and osmotic processes, which caused softening of the cell membranes (Tomas and Ronto 1959), and changed the properties and the structure of protein as well as the enzyme activity, particularly in pure protein solution (Zayas and Strokova 1972).

An ultrasound method of chymosin extraction was developed for increased efficiency (Kim and Zayas 1989). The use of ultrasound resulted in a significant reduction of extraction time with increases in activity and yield of chymosin. The optimal parameters for ultrasound extraction have been established: specific intensity, time of ultrasound exposure, temperature of extractive medium, ratio of abomasa to extraction solution and activation time of pro-chymosin.

The application of ultrasound in the extraction process of the insulin production resulted in an increase in the activity and yield of this biologically active substance, as well as a significant decrease in the extraction time (Zayas 1985).

Ultrasound had a selective action on animal tissues; it destroyed some tissues without changing others, and it disrupted the myofibrils with the formation of segments (Zayas and Smolski 1970), causing the destruction of pancreatic tissues (Zayas 1985). During tissue disruption, there was a migration of proteins, minerals, and other materials into extraction medium.

Development of an effective process of chymosin production requires improvement of the extraction process. The full extraction of chymosin from abomasa, increase in yield of chymosin, preservation of its activity, and better quality are the most important problems. The objective of this study was to investigate the influence of ultrasound on the properties of chymosin and the ultrastructure of abomasum during chymosin extraction.

MATERIALS AND METHODS

Sample Preparation

The purified chymosin (Sigma product) containing 98% protein was purchased and kept in the freezer at $-70°C$ until used. Abomasums of one month old calfs were used for experiments. Fresh abomasum was obtained from the slaughter house at Kansas State University and immediately preserved in solution of 2.5% glutaraldehyde in 0.1 M cacodylate buffer, pH 7.4.

Ultrasound Treatment

Chymosin solution, prepared by dissolving 10 g of the purified chymosin (Sigma Product) in 10% NaCl solution previously adjusted to pH 5.9 with HCl, was treated with a Tekmar sonic disrupter, TK 1000 (Tekmar, Cincinnati, OH) at different specific intensities and for different periods of time.

The major components of a typical ultrasonic processor used for chymosin extraction were shown in Fig. 1. The generator (1), or power supply, converts conventional 50 or 60 Hz (cycles/second) alternating current at 120 volts to 20 kHz electrical energy at approximately 1000 V. This high frequency electrical energy is fed to a converter (2), where it is transformed to mechanical vibration. The converter vibrates in a longitudinal direction, and this motion is transmitted to a titanium alloy disruptor horn (3). The horn transmits and amplifies the energy, focusing it into a tip immersed in the extraction solution. Motion of the tip generates the sound waves that propagate through the liquid, generating microbubbles and causing cavitation. Extraction temperature in the container (4) was controlled through a cooling coil (5) connected to a Lauda circulator, K-2/R (Brinkmann, Westbury, NY) used as a cooling device (6).

Chymosin Activity

Chymosin activity was determined according to the methods of McMahon and Brown (1985) and Ernstrom (1958). Milk substrate was prepared by dispersing 12 g low heat, nonfat dry milk (NFDM) in 100 mL 0.01 M $CaCl_2$ solution. The substrate solution was then refrigerated for 20 h to allow time for complete hydration of the NFDM. The substrate solution was warmed to 35°C prior to analysis (30 min), and maintained at that temperature throughout the analysis. One mL chymosin extract for both control and experimental samples was added to 100 mL of the substrate. Milk-clotting time was measured visually, and chymosin activity was calculated as:

$$\text{Chymosin activity (Cu/mL)} = 100\ Ts/Ta \times Cs/Ca$$

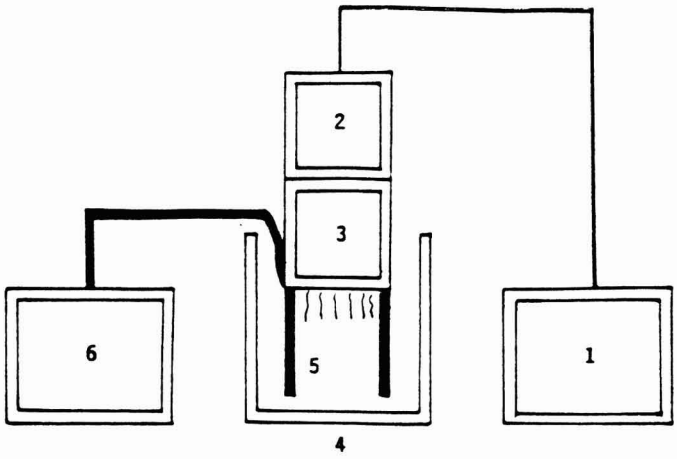

FIG. 1. ULTRASOUND UNIT FOR CHYMOSIN EXTRACTION
1. Ultrasound generator. 2. Converter. 3. Horn.
4. Sample container. 5. Cooling coil. 6. Cooling device.

where Cu = chymosin units; Ts = coagulation time of control chymosin solution; Ta = coagulation time of experimental chymosin solution; Cs = amount of control chymosin solution; Ca = amount of experimental chymosin solution.

Proteolytic Activity

Proteolytic activity of chymosin was determined according to azocasein method (Christen and Marshall 1984). Azocasein solution, prepared by dissolving 1% (wt/vol) azocasein (Sigma product) in sterile 0.02 M phosphate buffer at pH 6.4, was dispensed aseptically into sterile screw-up tubes (2 mL/tube). One mL chymosin solution was added into each tube. After incubation at 32°C for 1 h, the reaction was stopped by adding 3.0 mL aqueous trichloroacetic acid (TCA), 5% wt/vol. A blank was prepared by adding TCA to azocasein without addition of enzyme. After being held for 30 min at 22°C, samples were filtered through Whatman #41 paper and absorbance was measured at 345 nm. Proteolytic activity of chymosin was expressed as absorbance at 345 nm. Mean values obtained from these three replications are presented in Table 1.

TABLE 1.
INFLUENCE OF ULTRASOUND ON THE ACTIVITY OF PURIFIED CHYMOSIN[1]

Method	Chymosin activity, Cu/mg	Proteolytic activity[2]
Control	100.0[a]	0.205[a]
Ultrasound at 36 W/cm^2 for 80 min	95.0[a]	0.197[a]
Ultrasound at 157 W/cm^2 for 10 min	83.5[b]	0.165[b]
Ultrasound at 157 W/cm^2 for 20 min	56.8[c]	0.119[c]

[a,b,c]Means in the same column with different superscripts are significantly different ($P < 0.05$).

[1]Mean values obtained from three replications
[2]Proteolytic activity was expressed as absorbance at 345 nm after 1 h incubation at 32°C

Amino Acid Analysis

The chymosin solution was treated by ultrasound at different specific intensities and for different periods of time. Samples were dialyzed for 48 h against 3 changes (2 liters each) of distilled water. Samples were dried in 0.6 × 5 cm borosilicate glass tubes, evacuated and hydrolyzed with 6 M HCl at 110°C for 22–24 h in a Waters' amino acid hydrolysis vial. The hydrolysates were dried, redried, and derivatized with phenylisothiocyanate (Bidlingmeyer *et al.* 1984). A standard amino acid mixture was also dried, redried, and derivatized in the same way. The derivatized samples were dissolved in 100–200 μL Waters' Pico Tag samples diluents and analyzed by a Pico Tag (Waters) amino analysis column at 46°C using HPLC. Average of four replications are presented in Table 2.

Electron Microscopy

A 1 g sample of fresh abomasal tissue, cut into 0.3 × 0.7 mm pieces and preserved in solution of 2.5% glutaraldehyde in 0.1 M cacodylate buffer, was placed in a 50 mL container with 25 mL 10% NaCl solution previously adjusted to pH 5.9 with HCl. The sample was treated by ultrasound at specific intensity 36 W/cm^2 for 80 min, which were the optimal parameters of chymosin extraction

TABLE 2.
INFLUENCE OF ULTRASOUND ON AMINO ACID COMPOSITION OF THE PURIFIED CHYMOSIN[1,2]

Amino Acid	Control	Ultrasound at 36 W/cm^2 for 80 min	Ultrasound at 157 W/cm^2 for 10 min
Asp	15.22^a	15.92^{a,b}	16.50^b
Glu	14.86^a	14.68^a	14.88^a
Ser	7.41^a	7.48^a	7.12^a
Gly	7.42^a	7.61^a	7.46^a
His	1.91^a	1.75^b	1.69^b
Arg	4.09^a	3.80^a	3.85^a
Thr	7.11^a	7.07^a	7.03^a
Ala	6.83^a	7.04^a	7.14^a
Pro	5.32^a	4.67^b	4.51^b
Tyr	3.22^a	2.73^b	2.81^b
Val	6.30^a	6.64^b	6.68^b
Met	1.34^a	1.10^b	1.16^b
Ile	3.15^a	3.32^{a,b}	3.46^b
Leu	6.46^a	6.59^{a,b}	6.81^b
Phe	4.90^a	4.78^a	4.31^b
Lys	4.46^a	4.56^b	4.84^c

^{a,b,c}Means in the same row with different superscripts are significantly different ($P < 0.05$).
[1]Mole percent
[2]Average of four replications

established in the previous study (Kim and Zayas 1989). Small pieces of abomasal tissue, trimmed to approximately 0.1 mm^3 treated by ultrasound and untreated, were fixed in 1–1.5 mL 3% glutaraldehyde solution in 0.1 M cacodylate buffer (dimethyl arsenate) at pH 7.4 and stored for at least 3 h against one change at 4°C. Samples were rinsed 3 times (10 min each) with cold 0.2 M sucrose solution in 0.1 M cacodylate buffer. After rinsing, samples were retrimmed to about 0.05 mm^3. Samples were then postfixed for 30 min in cold 1% osmium tetraoxide solution in 0.1 M cacodylate buffer at pH 7.4 and repostfixed again for 30 min in the same osmium tetraoxide solution. After fixation, samples were rinsed 3 times (5 min each) with distilled water and warmed to 22°C. Samples then were dehydrated (10 min each) in increasing concentrations of acetone (30, 50, 70, 80, 95 and 100%). The dehydrated samples were then infiltrated through propylene oxide: Luft's epon (2:1, 1:1, 1:2) for a minimum of 2 h each and then through 100% epon for at least 8 h. Samples were embedded in a flat Beem mold and cured for 48 h at 60°C. Gold-silver color sections were cut with glass knives on a Reichert Om-2 ultramicrotome (Am. Optical Corp., Buffalo, NY) and counterstained with uranyl acetate and lead citrate for 10 min each. A Phillips 201 transmission electron microscope (TEM) operated at 60 KV was used to examine the sections.

Statistical Analysis

Analysis of variance and least significant differences were used to ascertain significant effects at the 5% level. Regression analysis was conducted by the method of Steel and Torrie (1980) for simple regression models.

RESULTS AND DISCUSSION

Influence of Ultrasound on the Properties of Chymosin

Influence of ultrasound on a solution of purified chymosin is shown in Table 1. Ultrasound at moderate specific intensity (36 W/cm^2) did not decrease chymosin activity and its proteolytic activity, whereas at high level of specific intensity (157 W/cm^2) of ultrasound, chymosin activity and its proteolytic activity significantly decreased. Prolonged exposure (20 min) at the high specific intensity (157 W/cm^2) significantly decreased the chymosin activity and its proteolytic activity compared to the 10 min treatment.

The specific effect of high intensity ultrasound on chymosin activity could be related to two factors. At first, there is significant temperature increase of the extractive mixture that is very difficult to control even in the laboratory conditions. However, increase of the temperature is not a reliable parameter of ultrasound heat effect on enzymes, because local temperature inside cavitational

bubbles could be significantly higher. Second, phenomenon that should be taken into consideration is an extremely high pressure inside the cavitational bubbles especially at the moment of disruption.

The higher activity of nonpurified (abomasal) chymosin obtained by ultrasound treatment in a previous study (Kim and Zayas 1989) might have been due to the higher level of chymosin extraction from abomasa or the protective effect of foreign substances (mostly proteins) that were extracted together with chymosin. Foreign substances, particularly proteins, may have not only a protective effect during chymosin extraction by ultrasound, but also a positive effect on the quality of chymosin preparation, especially its activity. Protective effect of foreign substances, including proteins during ultrasound extraction was suggested as a result of electrophoretic studies of insulin extracted by ultrasound (Zayas 1986). Additional studies related to the protective role of foreign proteins in extractive mixture are needed.

The negative effect of prolonged exposure of high intensity ultrasound on chymosin activity can be related to inactivation of active sites in enzyme molecules. Another possible effect under these extreme conditions is partial thermal denaturation and partial inactivation of the active sites of enzyme, which results in its partial inactivation.

Ultrasound itself did not increase the chymosin activity and its proteolytic activity. Foreign substances such as proteins (Donovan and Beardslee 1975), sugars (Back *et al.* 1979) and polysaccharides (Imeson *et al.* 1977) stabilize proteins against thermal denaturation. Changes were found in the properties and the structure of proteins as well as the enzyme activity after ultrasound treatment, particularly if the pure protein solution did not contain particles of animal tissue or foreign proteins (Zayas and Strokova 1972). Dubbs (1966) reported that ultrasound had a selective effect in intracellular enzymes, not affecting some, especially serum cholinesterase, while inactivating others such as aminopeptidase. He also found that prolonged ultrasound treatment caused progressive smearing and disappearance of a dilute zone of serum protein in starch gel electrophoresis. According to Weissler (1960), high intensity ultrasound created small quantities of free radicals which could denature proteins and other molecules. According to Santamaria and Levi (1952), without a protective atmosphere or reducing agent present, ultrasound can inactivate enzymes by oxidation of sulfhydryl or other sensitive groups in the enzyme molecule. Oxidase tends to be more sensitive than reductase and amylase. Trypsin and polyphenoloxidase are rapidly inactivated in the presence of air; pepsin is somewhat more resistant. The presence of a reducing agent prevents biocatalytic inactivation of hyaluronidase. Protective effect of a reducing agent was not confirmed by Alliger (1975), who reported that ribonuclease in the presence of a reducing agent was inactivated by OH radicals during ultrasound treatment.

Amino Acid Composition of Chymosin

Amino acid composition of the chymosin treated by ultrasound is shown in Table 2. In this study, the concentration of amino acids was expressed as mole percent. There was a difference in amino acid composition of the purified chymosins treated by ultrasound and untreated ($P < 0.05$). The decrease in the mole percent of some amino acids was observed. Ultrasound treatment at the specific intensity of 36 W/cm^2 for 80 min decreased the mole percent of histidine, proline, tyrosine and methionine. The difference in valine and lysine content was significant although more data on amino acid composition are necessary. Ultrasound treatment at the very high specific intensity (157 W/cm^2) for 10 min influenced the mole percent of phenylalanine, histidine, proline, tyrosine, methionine, aspartic acid, valine, and lysine.

According to Weissler (1960), ultrasound may disrupt or create some amino acids. In the presence of oxygen, solutions of trytophan, tyrosine, and phenylalanine underwent ultrasonic degradation and formed intermediate products with a yellowish-brown color. Spectrophotometry data indicated rupture of the aromatic rings in these compounds. Elpiner and Sokolskaya (1958) reported that these and other free radical changes can be avoided by processing in a gas atmosphere of carbon dioxide or hydrogen. Under prolonged ultrasonic action without protective agents, amino acids might degrade to other complex organic compounds, and in some cases, to other amino acids. A sonicated solution of histidine revealed a chromatographic spot of aspartic acid as well as histidine, probably caused by the breaking of the imidazole ring of histidine (Elpiner and Sokolskaya 1958).

This study established that ultrasonic treatment of the purified chymosin without protective agents might degrade some amino acids such as histidine, tyrosine and phenylalanine. More experimental data are needed to evaluate ultrasound effect on amino acid composition of purified chymosin, treated by high intensity ultrasound.

Influence of Ultrasound on the Ultrastructure of Abomasum During Chymosin Extraction

The ultrastructures of abomasal tissue treated by ultrasound and the control are shown in Fig. 2 and 3, respectively. Ultrasound treatment for up to 80 min at 36 W/cm^2 specific intensity caused no significant changes in the ultrastructure of abomasal tissue. The degree of organelle damage after 80 min ultrasound treatment was surprisingly minimal. Some mitochondrial cristae were disrupted, especially in lumen and secretory granules more compact even though their numbers remained the same. All organelle membranes and plasma membranes were intact.

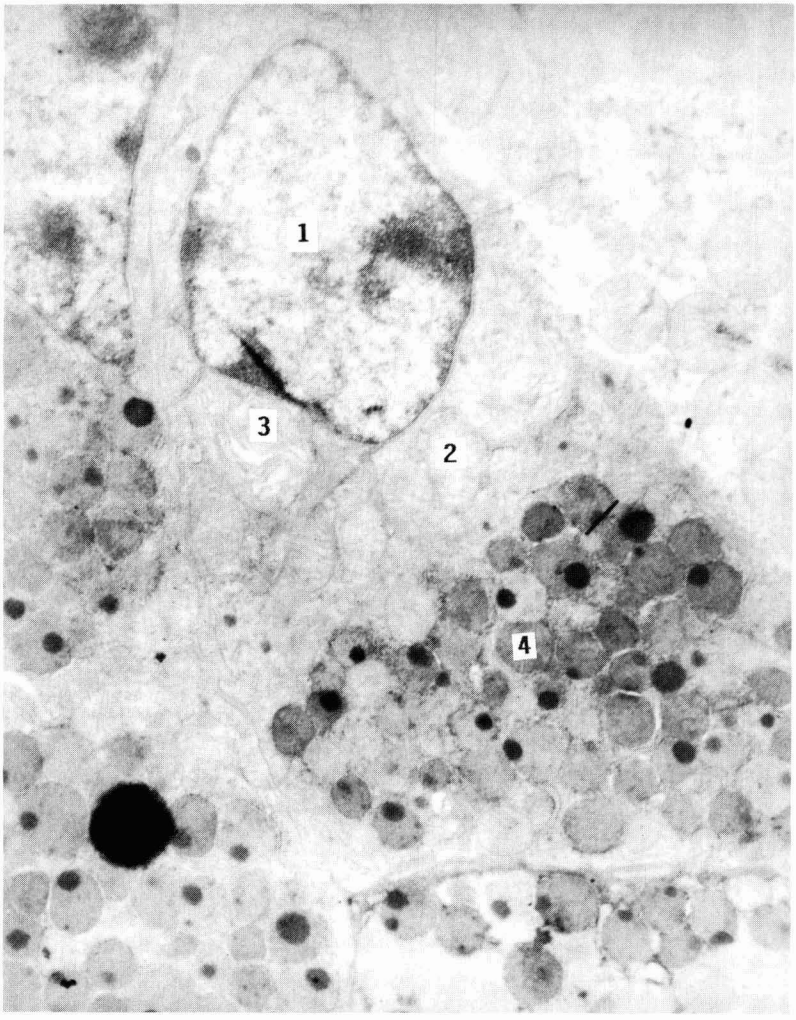

FIG. 2. ULTRASTRUCTURE OF ABOMASUM TREATED BY ULTRASOUND AT 36 W/CM^2 AND 25°C FOR 80 MIN (17475X)
1. Nuclei. 2. Mitochondria. 3. Golgi complex. 4. Secretory granule.

Even though the disruption of abomasal tissue was not severe in this study, there was clear evidence that ultrasound damaged the cellular structure of abomasal tissue, thus enhancing the extraction of soluble proteins, including chymosin. Alliger (1975) found that ultrasound rapidly disrupted mitochondria with fine membrane fragmentation and changed the sedimentation pattern.

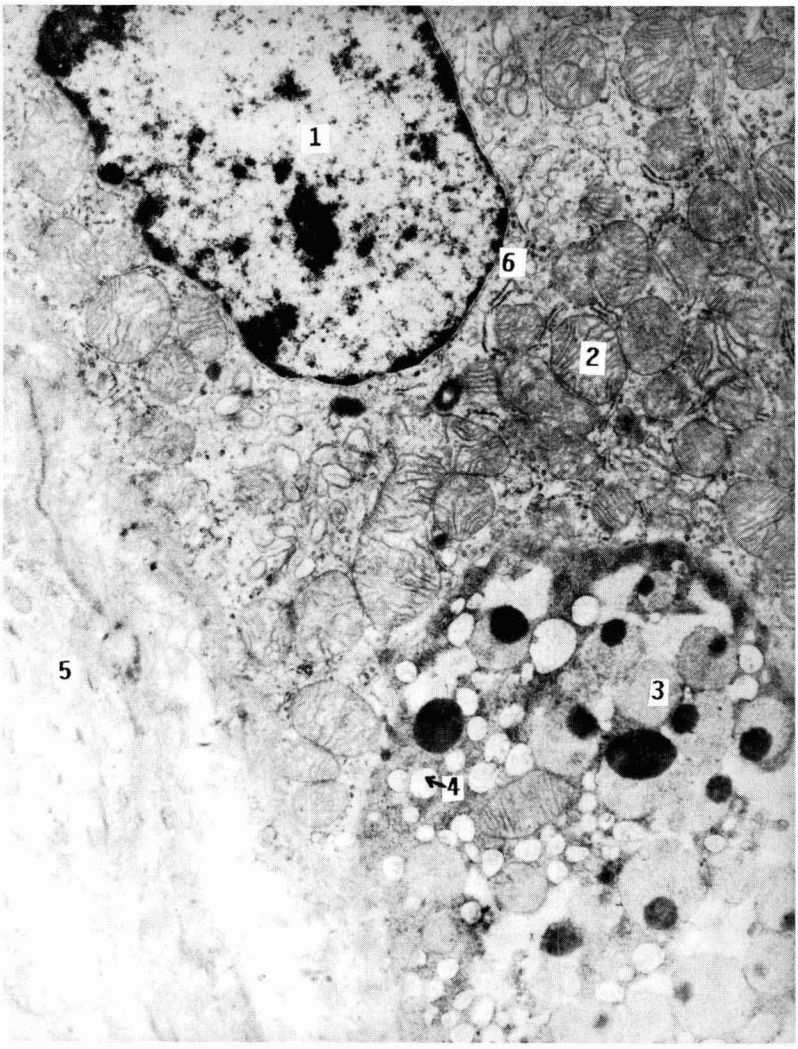

FIG. 3. ULTRASTRUCTURE OF UNTREATED ABOMASUM (17475X)
1. Nuclei. 2. Mitochondria. 3. Secretory granule. 4. Vacuole. 5. Collagen. 6. RER.

Reynolds *et al.* (1978) reported that ultrasound resulted in the disruption of muscle fiber, especially as seen in longitudinal section. The large increase in endomysial space in cross section indicated that the separation of muscle fiber was caused by ultrasound treatment. Theno *et al.* (1977) reported that tumbling or massaging resulted in the increased salt-soluble protein extraction and the cellular disruption. These factors were both increased with ultrasound. The his-

tological structure of abomasum is significantly different from that of muscular tissue and in this study, it was difficult to make comparisons with muscle parts on the microscope photographs.

CONCLUSIONS

Effect of ultrasound on the purified chymosin depends on its specific intensity. Ultrasound treatment of the purified chymosin at specific intensity 36 W/cm^2 for 80 min did not decrease the activity of the chymosin and its proteolytic activity. However, ultrasound treatment at high intensity (157 W/cm^2) and the exposure for 10 and 20 min decreased chymosin activity and its proteolytic activity. There was a significant difference in amino acids composition of the purified chymosins treated by ultrasound and untreated. Ultrasonic treatment of the purified chymosin without protective agents might degrade some amino acids. Difference in amounts of several amino acids were found in chymosins treated by ultrasound at specific intensities of 36 W/cm^2 for 80 min and 157 W/cm^2 for 10 min. There were no changes in the ultrastructure of abomasal tissue after ultrasound treatment at specific intensity 36 W/cm^2 for up to 80 min. Some mitochondrial cristae were damaged and secretory granules were more compact, even though their numbers remained the same. Ultrasound has a significant potential to be recommended for extraction of chymosin from abomasum.

REFERENCES

ALLIGER, H. 1975. Ultrasonic disruption. Am. Lab. *7,* 75.

BACK, J. F., OAKENFULL, D. and SMITH, M. B. 1979. Increased thermal stability of proteins in the presence of sugars and polyols. Biochemistry *18,* 5191.

BIDLINGMEYER, B. A., COHEN, S. A. and TARVIN, T. L. 1984. Rapid analysis of amino acids using precolumn derivatization. J. Chromatog. *336,* 93.

CHRISTEN, G. L. and MARSHALL, R. T. 1984. Selected properties of lypase and protease of *Pseudomonas fluorescens* 27 produced in four media. J. Dairy Sci. *67,* 1680.

DONOVAN, J. W. and BEARDSLEE, R. A. 1975. Heat stabilization produced by protein-protein association. Differential scanning calorimetric study of heat denaturation of trypsin-soybean trypsin inhibitor and trypsin-ovomucoid complexes. J. Biol. Chem. *250,* 1966.

DUBBS, C. A. 1966. Ultrasonic effects on isoenzymes. Clin. Chem. *12,* 181.

ELPINER, I. E. and SOKOLSKAYA, A. V. 1958. Effect of ultrasound on some proteins and amino acids in relation to native of gas present. Doklady Akad. Nauk. USSR *123,* 659.

ERNSTROM, C. A. 1958. Heterogeneity of crystalline rennin. J. Dairy Sci. *41*, 1663.

FRY, F. J. 1978. *Ultrasound, Its Application in Medicine and Biology*. Elsevier Scientific Pub. Co., New York.

HESS, E. and SLADE, H. 1955. An electrophoretic examination of cell free extracts from various serological types of group A hemolytic strep. Biochim. Biophys. Acta *16*, 346.

HUGHES, D. E. and NYBORG, W. L. 1962. Cell disruption by ultrasound. Science *138*, 108.

IMESON, A. P., LEDWARD, D. A. and MITCHELL, J. R. 1977. On the nature of the interaction between some anionic polysaccharides and proteins. J. Sci. Food Agr. *28*, 661.

KIM, S. M. and ZAYAS, J. F. 1989. Processing parameters of chymosin extraction by ultrasound. J. Food Sci. *54*, 700.

McMAHON, D. J. and BROWN, R. J. 1985. Evaluation of formagraph for comparing rennet solutions. J. Dairy Sci. *65*, 1639.

REYNOLDS, J. B., ANDERSON, D. B., SCHMIDT, G. R., THENO, D. M. and SIEGEL, D. G. 1978. Effects of ultrasonic treatment on binding strength in cured ham rolls. J. Food Sci. *43*, 866.

SANTAMARIA, A. and LEVI, F. 1952. Hyaluronidase inactivation by ultrasonic waves. Enzymologie *15*, 285.

STEEL, R.G.D. and TORRIE, J. H. 1980. *Principles and Procedures of Statistics*, 2nd ed. McGraw-Hill Book Co., New York.

THENO, D. M., SIEGAL, D. G. and SCHMIDT, G. R. 1977. Meat massaging techniques. In *Proc. Meat Ind. Res. Conf.* p. 53. Mar. 24–25. Am. Meat Sci. Assoc. and Am. Meat Fnd. Chicago, IL.

TOMAS, G. and RONTO, G. 1959. Effect of ultrasound on the diffusion of the cyanateionsa in frog muscles. Acta Biol. Acad. Sci. Hungary *3*, 45.

WEISSLER, A. 1959. Formation of hydrogen peroxide by ultrasonic waves: free radicals. J. Am. Chem. Soc. *81*, 1077.

WEISSLER, A. 1960. Biochemical effects of ultrasound: Destruction of tryptophan. J. Acoust. Soc. Am. *32*, 1499.

ZAYAS, J. F. 1985. Effect of ultrasound treatment on the extraction of insulin. Biotech. Bioeng. *XXVII*, 1223.

ZAYAS, J. F. 1986. Properties and quality characteristics of crystalline insulin extracted by ultrasound. Biotech. Bioeng. *XXVIII*, 349.

ZAYAS, J. F. and SMOLSKI, N. T. 1970. Changes in ultrastructure of muscular tissue resulting from ultrasonic tenderization. Proc. XVI. Eur. Congr. Meat Res. Workers. Sofia, Bulgaria. *II*, 1141.

ZAYAS, J. F. and STROKOVA, N. D. 1972. Influence of ultrasound on properties of meat proteins. Proc. XVIII Eur. Congr. Meat Res. Workers. Canada. *II*, 206.

SALTING BEHAVIOR OF YELLOWTAIL (*TRACHURUS MCCULLOCHI* NICHOLS)

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ABSTRACT

*Wet salting of low-fat yellowtail (*Trachurus mccullochi *Nichols) in 5%, 10%, 15%, 21% and saturated brine solutions at 25°C and 35°C, and desalting of salt-saturated fish, was carried out and assessment made of the salt content, moisture content, water activity and rate of water transfer. Brine concentration, fish shape and brine temperature had a significant effect ($P < 0.01$) on both equilibrium salt content (X_s^+) and rate of salt uptake (k_s). In whole and split fish, the rate of salt uptake increased with an increase in brine concentration, while moisture loss occurred only in fish brined in from 15% up to saturated brine. In whole fish, the rate of salt loss during the desalting process was nearly the same as the rate of salt uptake during the salting process, while in split fish the rate of salt loss was faster than the rate of salt uptake.*

INTRODUCTION

Salting is a traditional method of preserving fish which is still used extensively in many tropical countries because of the simplicity of the process, the low production cost and its ability to be combined with drying and smoking or other methods in order to satisfy consumers' habits and requirements. In combination with the drying process, salting also assists in reducing the water activity (a_w) of the fish to assist preservation before drying. If the weather conditions are not suitable for sun drying, or if the drying capacity of a mechanical dryer is insufficient, e.g., when a large quantity of fish is caught at the same time, then fish is frequently stored in saturated brine for lengthy periods. However, in-

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creasing the salt content of the fish produces a slower rate of reduction of moisture content and a higher final moisture content during drying (Berhimpon *et al.* 1990). Light salting, on the other hand, is limited by the possibility of spoilage during an extended processing step. The choice of salt level, therefore, depends on the length of processing delays and the nature of the process to be applied to the product. In recent years there has been a preference for lower salt fish (Buckle *et al.* 1988).

The aims of this study were to determine the minimum process requirements for a salted fish suitable for drying. Three aspects were considered: equilibrium salt content, the rate at which equilibrium is reached and the changes in a_w of the fish during salting and desalting. This study was part of a detailed investigation aimed at improving the quality of traditionally processed dried-salted fish in South-east Asia.

MATERIALS AND METHODS

Raw Materials

Fresh yellowtail (*Trachurus mccullochi* Nichols) in ice were purchased from the Kingsford Fish Market, Kingsford, NSW, within 24 h of capture. The weight range for individual fish was 59–76 g, the length range 17–20 cm, and the proximate composition ranges were moisture 72.0–72.3%, fat 0.67–0.70% and protein 21.9–23.4% (wet weight basis, WWB). The fish were iced during transportation to the laboratory where they were immediately washed with tap water and then wet salted. Coarse refined and kiln dried salt (Pacific Salt Pty Ltd, NSW, purity 99.7% NaCl) was used to prepare 5% (16.3% saturation), 10% (33.8% saturation), 15% (52.5% saturation), 21% (76.6% saturation) and 26.5% w/w(100% saturation) brines.

Salting

Fish were divided into three batches: the first batch was left as whole fish, the second batch was split longitudinally and spread along the backbone, and the third batch was filleted (skin on). Brining of whole and split fish was conducted in plastic containers at 25° and 35°C. Brining of fillets was conducted in glass containers at 25°C and 35°C. Brine concentrations were adjusted by salt addition and monitored with a salt analyzer (PTI-54, CHK Engineering, Sydney).

To brine the whole and split fish, 20 containers each containing 28 fish were held for 50 h at the given temperature with occasional stirring, during which samples were taken for analysis of salt and moisture contents and a_w. To brine the fillets, 10 containers each containing 20 fillets were held for 3.5 h at the given temperature with occasional stirring, during which samples were taken for measurement of water transfer.

Desalting

Whole and split fish were brined in saturated brine at 25°C for 96 h to ensure equilibrium was reached (Berhimpon *et al.* 1990). Saturated fish were then desalted in running tap water at ambient temperature (approx. 22°C) and samples taken up to 40 h and for split fish up to 30 h for analysis of salt and water contents and a_w.

Chemical Analyses

Moisture content was determined by a vacuum oven method (102° ± 2°C for 24 h); protein content by automated Kjeldahl procedure (Kjel-Foss Model 16210-A/SN, A/SN Foss Electric, Denmark); fat content by the Soxhlet extraction method (AOAC 1984); salt content by titration with silver nitrate (FAO 1981); a_w by a relative humidity meter (Hygroskop DT, Rotronic Ag, Zurich) calibrated against saturated salt solutions providing the following relative humidities at 25°C: $MgCl_2 \cdot 6H_2O$, 33.2%; K_2CO_3, 43.2%; $Mg(NO_3)_2 \cdot 6H_2O$, 53.4%; $NaNO_2$, 64.4%; NaCl, 75.8%; KCl, 84.2%; K_2SO_4, 96.9% (Wexler and Hasegawa 1954; Young 1967; Labuza 1984); and water transfer was determined according to the method of Fougére (Fougére 1952; Deng 1977).

Mathematical Modelling and Statistical Analyses

The following equations (1 and 2), developed by Zugarramurdi and Lupin (1980), were used to determine the rate of salt uptake (k_s) and water diffusion (k_w):

$$X_s = X_s^o \cdot e^{-k_s t} + X_s^+ \cdot (1 - e^{-k_s t}) \quad (1)$$

$$X_w = X_w^o \cdot e^{-k_w t} + X_w^+ \cdot (1 - e^{-k_w t}) \quad (2)$$

where: X_s/X_w = salt/water content (g/100g nonsalt solids basis, NSSB) at time, t
X_s^o/X_w^o = salt/water content (g/100g NSSB) at time zero
X_s^+/X_w^+ = salt/water content (g/100g NSSB) at equilibrium
k_s/k_w = specific rate constants for salt uptake/water loss, and
t = time (h).

To obtain the fit data for water transfer, the model (Eq. 3) proposed by Deng (1977) was used:

$$X = C(1 - e^{-kt}) \quad (3)$$

where: X = weight change (%) at time t,
C = weight change (%) at equilibrium,
k = rate constant.

The equilibrium salt and water contents (X_s^+ and X_w^+) and the specific rate constants (k_s and k_w) were determined by a nonlinear least square procedure using the SAS package (SAS Institute Inc. 1985). The X_s^+, X_w^+, k_s and k_w values of the regression equation were then analyzed as dependent variables, by a two-way analysis of variance procedure using the SPSS package (Kim and Kohout 1975).

RESULTS AND DISCUSSION

Salting

Figure 1 shows that in whole and split fish, the rate of salt uptake increased with an increase in brine concentration to reach constant values dependent upon brine concentration. The higher the brine concentration the higher the equilibrium salt content and the faster the fish reached equilibrium, in agreement with previous work on yellowtail (Berhimpon *et al.* 1990).

Brine concentration, fish shape and brine temperature individually had a significant effect ($P < 0.01$) on both X_s^+ and k_s values (Table 1). Split fish attained equilibrium salt contents higher and faster than those of whole fish, and fish, brined at 35°C, whether whole or split, attained equilibrium salt content higher and faster than those of fish brined at 25°C. Brine concentration and fish shape in combination were found to have a significant effect ($P < 0.01$) on X_s^+ and k_s

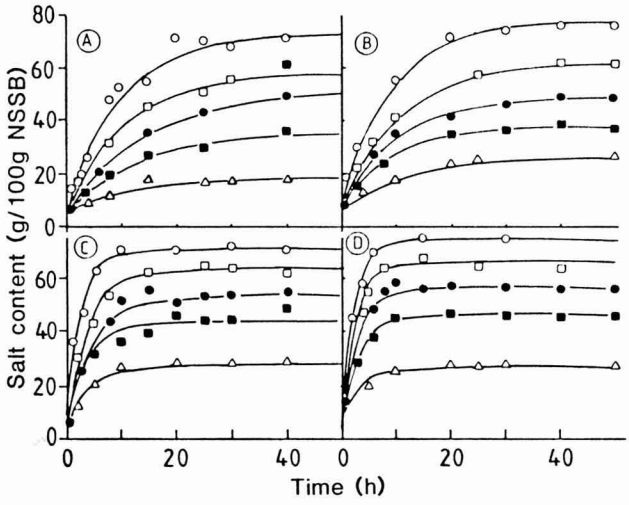

FIG. 1. SALT CONTENTS OF YELLOWTAIL DURING BRINING
(A) Whole fish at 25°C. (B) Whole fish at 35°C. (C) Split fish at 25°C. (D) Split fish at 35°C: 5% Brine (△). 10% Brine (■). 15% Brine (●). 21% Brine (□). 26.5% Brine (○).

TABLE 1.
EQUILIBRIUM SALT CONTENT (X_s^+) AND SPECIFIC RATE CONSTANT (k_s) FOR SALT UPTAKE OF YELLOWTAIL

			Parameter					
			1		2		3	
BC	FS	BT	X_s^+	k_s	X_s^+	k_s	X_s^+	k_s
5	W	25	18.5	0.09	18.6	0.07	18.5	0.08
		35	25.4	0.10	27.0	0.08	26.1	0.09
	S	25	27.9	0.20	28.4	0.21	28.1	0.21
		35	27.3	0.26	27.7	0.30	27.5	0.28
10	W	25	35.5	0.08	36.0	0.07	35.7	0.07
		35	36.0	0.12	37.5	0.13	36.7	0.13
	S	25	44.4	0.25	44.0	0.24	44.2	0.25
		35	47.1	0.27	45.5	0.31	46.3	0.29
15	W	25	51.3	0.07	52.6	0.06	51.9	0.07
		35	47.8	0.12	49.2	0.10	48.4	0.11
	S	25	53.9	0.20	55.0	0.20	54.5	0.20
		35	56.3	0.34	57.6	0.30	57.0	0.32
21	W	25	56.2	0.11	61.8	0.08	59.0	0.09
		35	59.8	0.10	62.2	0.10	61.0	0.10
	S	25	63.2	0.26	65.5	0.23	64.3	0.24
		35	65.5	0.38	67.6	0.30	66.6	0.38
26.5	W	25	72.6	0.10	73.0	0.10	72.8	0.10
		35	77.6	0.11	78.3	0.11	77.9	0.11
	S	25	71.2	0.39	71.2	0.37	71.2	0.38
		35	77.1	0.36	74.0	0.40	75.4	0.40

BC,FS & BT= Brine concentration(%w/w), fish shape (whole or split) and brine temperature (°C), respectively
W and S = Whole and split fish,
1 and 2 = Replication 1 and 2; and 3= X_s^+ and k_s values calculated from average of experimental data of 2 replications; coefficient of correlation ranged from 0.98 to 0.99.

values, while the combined effect of brine temperature and concentration was significant ($P < 0.01$) only for the k_s value; other interactions had no effect on X_x^+ and k_s values.

Comparing the results with those of experimental values in Table 2, Table 1 indicates that k_s values for fish brined in 15% and 21% brine were slightly lower

TABLE 2.
EXPERIMENTAL VALUES OF k_s AND X_s^+ FOR SALTED FISH

Species	Fish size (g)	Brine temp. (°C)	Brine conc. (%w/w)	k_s	X_s^+ (g/100g NSSB)	Reference
Yellowtail (*Trachurus mccullochi* Nichols)	47-65	22-23	15	0.09	54.3	Berhimpon *et al.* (1990)
			21	0.06	69.0	
			26.5	0.13	72.0	
Sardine (*Sardinops neopilchardus*)	49-78	25	26.5	0.05	57.5	Poernomo (1986)
	61	19-22	26.5	0.07	0.73*	Wuttijumnong(1987)
	48-64	25	26.5	0.04	74.4	Utomo (1985)
		30	26.5	0.07	79.9	
		35	26.5	0.09	71.0	
Indian oil sardine (*Sardinella longiceps*)	51	19-22	26.5	0.04	0.83*	Wuttijumnong(1987)

* g/g salt free fat free dry basis (sfffdb).

than those reported previously, but were the same for fish brined in saturated brine. Yellowtail absorbed salt at a faster rate than did high fat fish (2.5–3.1% fat WWB) (Poernomo 1986).

Figure 2 shows that in whole and split fish, water loss occurred in fish brined in 15% or higher brines, and increased with an increase in brine concentration. In 5% and 10% brine, split fish absorbed water, although the salt content of the fish increased during brining to reach the concentration of the brine. Conversely, whole fish lost water in the first few hours followed by a gradual increase in water content, and the salt content increased during brining to reach concentrations of 4.2% and 8.2% (WWB), respectively. Equilibrium moisture contents and specific rate constants for moisture loss/uptake can be seen in Table 3.

According to Zugarramurdi and Lupin (1980), skin acts as a membrane in whole fish, and salt diffuses into the fish by a mechanism of dialysis. Water diffuses to the outside due to the difference in osmotic pressure between brine

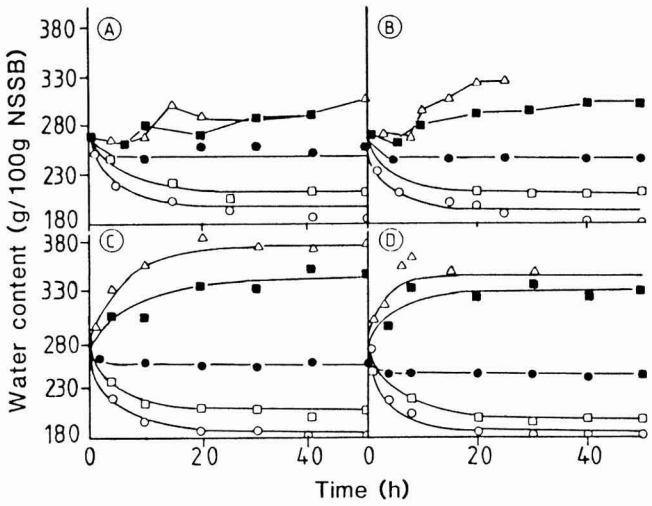

FIG. 2. MOISTURE CONTENT OF YELLOWTAIL DURING BRINING
(A) Whole fish at 25°C. (B) Whole fish at 35°C. (C) Split fish at 25°C. (D) Split fish at 35°C: 5% Brine (△). 10% Brine (■). 15% Brine (●). 21% Brine (□). 26.5% Brine (○).

and fish muscle solution. Osmotic equilibrium is reached when solute pressures on both sides of the skin become equal and water diffusion ceases, and simultaneously the salt transference to the fish ceases. In this case, the salt concentration inside the fish is lower than that in the brine.

The swelling of the fish at the conclusion of salting in unsaturated brine also occurred after 35 h in 15% brine (Berhimpon *et al.* 1990). According to Zaitzev *et al.* (1969), the swelling is due to the difference in mobility of the molecules and ions of water and to those of salt. Water diffusion continues until it is compensated for by the elasticity of the fish tissue and the hydrophilic properties of proteins. When that happens, the salt concentration is not yet equal to that of the brine and salt diffusion continues. As a result, the osmotic equilibrium is upset, and a small amount of water is returned to the fish. Crean (1961), working with cod, found that a critical internal salt content exists in the region of 8% (WWB), above which the protein is denaturated rapidly and salt gain is accompanied by water loss. Zaitzev *et al.* (1969) found that when Black Sea sprat was soaked in brine solutions of various concentrations, levels of up to 12% caused flesh swelling and weight increase, whereas at salt concentrations in excess of 12% the fish protein became denaturated and lost its water holding capacity.

Figure 3 shows clearly that at 25°C, fillets brined in 5% and 10% brine increased in weight, while fillets brined in solutions from 15% up to saturated brine decreased in weight. At 35°C, fillets brined in 10% brine also decreased in weight, and the weight loss of the fillets in various brine concentrations

TABLE 3.
EQUILIBRIUM MOISTURE CONTENT (X_w^+) AND SPECIFIC RATE CONSTANT (k_w) FOR MOISTURE LOSS/UPTAKE OF YELLOWTAIL*

BC**	FS	BT	Parameter	
			X_w^+	k_w
5	W	25	-+	-
		35	-	-
	S	25	375.2	0.15
		35	344.3	0.30
10	W	25	-	-
		35	-	-
	S	25	340.7	0.11
		35	328.8	0.19
15	W	25	246.9	0.79
		35	245.5	1.48
	S	25	252.4	0.82
		35	243.5	1.22
21	W	25	210.4	0.17
		35	211.8	0.27
	S	25	207.2	0.25
		35	199.7	0.19
26.5	W	25	196.1	0.26
		35	192.6	0.31
	S	25	185.8	0.25
		35	186.4	0.30

* Calculated from average of experimental data of 2 replications.

**See Table 1 for explanation.

+Equation 2 was unable to fit the data.

increased with increased temperature. This indicates that increased brine temperature affected the diffusion of water and the denaturation of protein, which caused the flesh to release water.

Figure 4 shows that a_w decreased faster for fish salted in higher brine concentrations. Split fish brined in saturated brine at both 25° and 35°C reached a minimum a_w of 0.75. Split fish had an ultimate a_w lower than that of whole fish only in fish brined in 21% and saturated brines. Bacteria generally do not grow in products with an a_w of less than about 0.88, but certain haloduric and halophilic bacteria will grow in cured fish with an a_w to 0.75 (FAO 1981).

Table 4 shows the development of off-odour and spoilage during salting of fish. Fish brined in 5% and 10% brines are still liable to spoil during brining

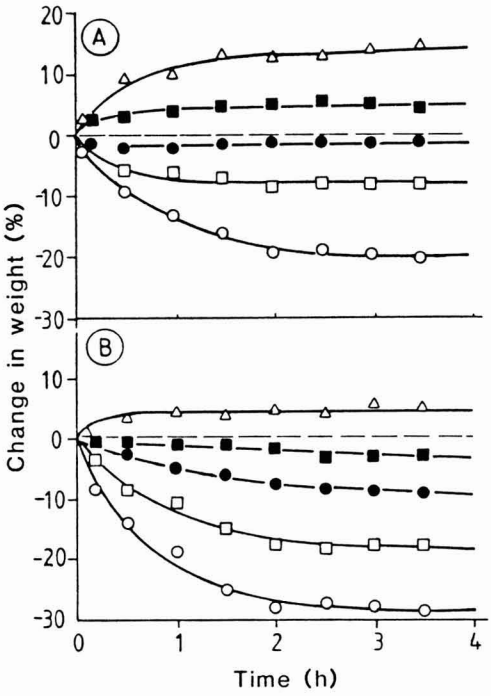

FIG. 3. WATER TRANSFER IN YELLOWTAIL FILLET DURING BRINING
(A) 25°C. (B) 35°C: 5% Brine (△). 10% Brine (■).
15% Brine (●). 21% Brine (□). 26.5% Brine (○).

for 50 h, while fish brined in 15% or higher salt solution did not spoil during 50 h brining. The growth of potentially pathogenic organisms and/or toxin production during the brining of fish in low salt brines requires further study.

The lowest brine concentration that can be used to reduce moisture content, and hence a_w, to a satisfactory level is about 15%. Brine concentrations lower than 15% are useful only for producing a mildly salty taste in the finished product and to inactivate most of the putrefactive bacteria. These aims, however, also can be satisfied by brining the fish in saturated brine for a short time. Brining at 35°C gives an advantage only at high brine concentrations for short brining times, since at lower brine concentrations putrefaction proceeds faster than does the preservation effect of salt.

Desalting

Figure 5 shows that salt loss, water uptake and increase in a_w occurred faster in split fish than in whole fish in the early stages of desalting. Since salt and

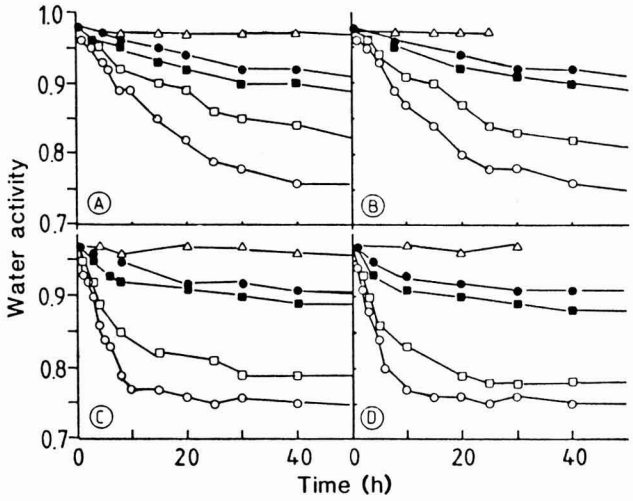

FIG. 4. WATER ACTIVITIES OF YELLOWTAIL DURING BRINING
(A) Whole fish at 25°C. (B) Whole fish at 35°C. (C) Split fish at 25°C. (D) Split fish at 35°C: 5% Brine (△). 10% Brine (●). 15% Brine (■). 21% Brine (□). 26.5% Brine (○).

water diffusion result from differences in osmotic pressure of the fish and the medium, the model (Eq. 1 and 2) suggested by Zugarramurdi and Lupin (1980) was used to describe the desalting data. Table 5 presents the brining parameters obtained using the model. In whole fish, the rate of salt loss approximated the rate of salt uptake of the fish brined in saturated brine, while the rate of water uptake was faster than the rate of water loss. In split fish, the rates of salt loss and water uptake were faster than the rates of salt uptake and water loss of split fish brined in saturated brine.

The results suggest the possible advantage of desalting heavily salted fish before drying in order to increase the rate of fish drying. Since the salt-induced denaturation of protein is an irreversible process, it can be predicted that water absorbed by denaturated flesh during desalting would remain largely as free water that should evaporate readily during drying.

Based on the results, processors of salted-dried fish have a number of options available depending on the market they are supplying and the level of technology at their disposal. A low salt product with a limited shelf-life could be produced in a short period of time and would satisfy consumer preference, but mechanical drying followed by sophisticated packaging and storage systems would be required. Alternatively, a high salt fish with a long shelf-life would require simple processing and packaging technology but would have limited consumer accept-

TABLE 4.
DEVELOPMENT OF OFF-ODOUR AND SPOILAGE OF YELLOWTAIL DURING SALTING IN VARIOUS BRINE SOLUTIONS

Sample	Observation*
Whole fish	
5% brine at 25°C	Off-odour after 30h, spoilage after 40h
5% brine at 35°C	Off-odour after 15h, spoilage after 25h
10% brine at 25° & 35°C	Off-odour started to develop after 50h
Split fish	
5% brine at 25°C	Off-odour after 20h, spoilage after 40h
5% brine at 35°C	Off-odour after 15h, spoilage after 30h
10% brine at 25° & 35°C	Off-odour started to develop after 50h
>15% brine at 25° & 35°C	No off-odour during 50h

* Off-odour refers to development of earthy, ammoniacal odour; spoilage refers to development of bacterial slime and tissue softening.

ability. The second option is the current practice in Indonesia mainly because of its low capital intensity.

Further studies are in progress on the effect of salt content (salting and desalting) and drying parameters on the physical, chemical and sensory properties of dried-salted yellowtail.

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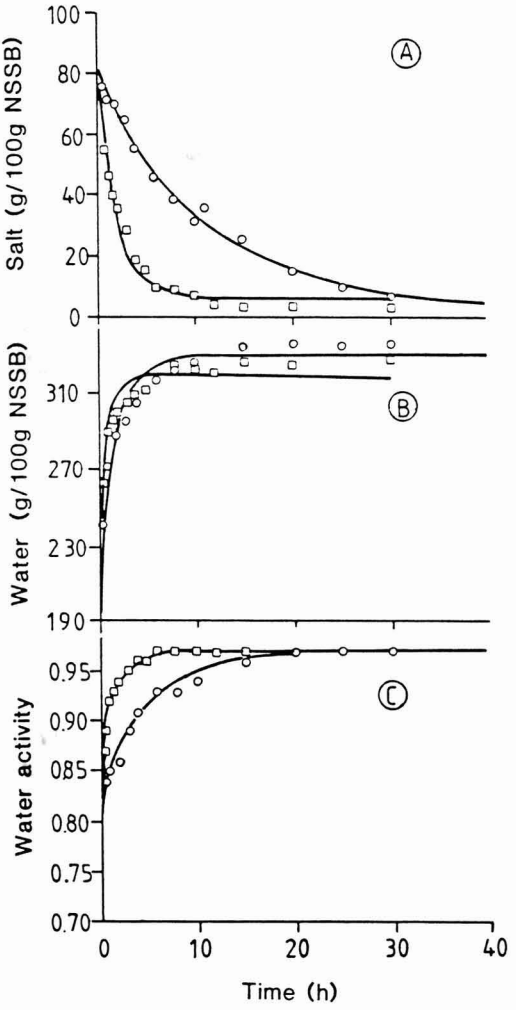

FIG. 5. SALT AND WATER CONTENTS, AND WATER ACTIVITIES OF YELLOWTAIL DURING DESALTING
(A) Salt content. (B) Water content. (C) Water activity: whole fish. (○) Split fish (□).

REFERENCES

AOAC. 1984. *Official Methods of Analysis,* 14th ed., p. 291. Association of Official Analytical Chemists, Washington, D.C.

BERHIMPON, S., SOUNESS, R. A., BUCKLE, K. A. and EDWARDS, R. A. 1990. Salting and drying of yellowtail (*Trachurus mccullochi* Nichols). Int. J. Food Sci. Technol. *25,* 409–419.

TABLE 5.
PARAMETERS OF THE DESALTING OF YELLOWTAIL

Fish shape	Parameter					
	X_s^+	k_s	R^2	X_w^+	k_w	R^2
Whole	4.1	0.10	0.99	330.4	0.55	0.99
Split	6.3	0.53	0.98	318.8	1.33	0.99

X_s^+/X_w^+ = equilibrium salt/water content (% NSSB).

k_s/k_w = specific rate constant for salt loss/water uptake.

R^2 = coefficient of correlation.

BUCKLE, K. A., SOUNESS, R. A., PUTRO, S. and WUTTIJUMNONG, P. 1988. Studies on the stability of dried salted fish. In *Food Preservation by Moisture Control,* (C. C. Seow, T. T. Teng and C. H. Quah, eds.) pp. 103–115, Elsevier Applied Science, London.

CREAN, P. B. 1961. The light pickle salting of cod. J. Fish. Res. Board Can. *18*(5), 833–844.

DENG, J. C. 1977. Effect of freezing and frozen storage on salt penetration into fish muscle immersed in brine. J. Food Sci. *42*(2), 348–351.

FAO. 1981. *The Prevention of Losses in Cured Fish.* FAO Fish. Rep. 279. Food and Agriculture Organization, Rome.

FOUGÉRE, H. 1952. Water transfer in codfish muscle immersed in sodium chloride solutions. J. Fish. Res. Board Can. *9*(8), 388–392.

KIM, J. and KOHOUT, F. J. 1975. Analysis of variance and covariance: subprograms anova and oneway. In *Statistical Package for the Social Science,* 2nd ed., (N. H. Nie, C. H. Hull, J. G. Jenkins, K. Steinbrenner and D. H. Bent, eds.) pp. 398–433, McGraw-Hill Book Co., New York.

LABUZA, T. P. 1984. *Moisture Sorption: Practical Aspects of Isotherm Measurement and Use,* pp. 65–66. American Association of Cereal Chemists, St. Paul, MN.

POERNOMO, A. 1986. Salting behaviour of sardines. J. Post Harvest Fish. Res. *51,* 23–33.

SAS INSTITUTE INC. 1985. *SAS User's Guide: Statistics,* 5th ed. SAS Institute, Cary, NC.

UTOMO, B.S.B. 1985. Effect of temperature and pH on the salting kinetics of sardines. M. App. Sc. Thesis, School of Food Science and Technology, The University of New South Wales, Kensington, NSW.

WEXLER, A. and HASEGAWA, S. 1954. Relative humidity-temperature relationship of some saturated salt solutions in temperature range 0° to 50°C. J. Res. Nat. Bur. Stand. *53*(1), 19–26.

WUTTIJUMNONG, P. 1987. Studies on moisture sorption isotherms, salting kinetics and drying behaviour of fish. Ph.D. Thesis, School of Biological Technologies, The University of New South Wales, Kensington, NSW.

YOUNG, J. F. 1967. Humidity control in the laboratory using salt solutions. A review. J. Appl. Chem. *17,* 241–245.

ZAITZEV, V., KIZEVETTER, I., LAGUNOV, L., MAKAROVA, L., MINDER, L. and PODSEVALOV, V. 1969. *Fish Curing and Processing*. Translated from the Russian by A. De Merendol, p. 208. MIR Pub., Moscow.

ZUGARRAMURDI, A. and LUPIN, H. 1980. A model to explain observed behaviour on fish salting. J. Food Sci. *45,* 1305–1311.

PHYSICO-CHEMICAL PROPERTIES AND ACCEPTABILITY OF FLOUR PREPARED FROM MICROWAVE BLANCHED SWEET POTATOES

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ABSTRACT

Flour was prepared from microwave blanched "Jewel" sweet potatoes. For comparison, flours were also prepared from steam blanched and unblanched sweet potatoes. Microwave blanched flour had higher yield (18%) than steam blanched (13%). There were differences ($p < 0.05$) in moisture, ash, starch, sugar and thiamin content among the three flours. No differences in oil and water holding capacity or viscosity were observed between the microwave and steam blanched flours. There were no differences in L-values among flours, but a-values were smaller for microwave blanched and b-values were greater for unblanched flours ($p < 0.05$). Visual observation indicated that blanched flours were deeper in yellow-orange color than unblanched flour. Three bakery products, bread, cake and cookies, were prepared. The sensory scores of products containing microwave blanched flour were higher than that of products containing unblanched or steam blanched flour. The products were highly acceptable.

INTRODUCTION

Sweet potatoes (*Ipomoea batatas*) are an important crop grown extensively in the Southern United States. Sweet potatoes are consumed not only in baked form but can also be used as ingredient in ice cream, pie fillings, pancakes, breads and icing (Winarno 1980). Flours were previously prepared from sweet potatoes after drying without blanching. Such flour deteriorates quickly in flavor and quality (Dawson 1951). Fruits and vegetables are steam or water blanched prior to freezing and dehydration in order to improve color and texture (Noble and Gordon 1964; Odland and Eheart 1975). Steam blanching of sweet potatoes was reported by Spadaro and Patton (1961) and by Wadsworth *et al.* (1966).

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The use of microwave for food processing has been increasing rapidly in recent years (Smith 1984). Recently, peanut flour was prepared by combination of microwave-vacuum oven drying techniques (Pominski and Vinnett 1989). Martin prepared flour from microwave baked sweet potatoes (1984). Purcell and Walter (1988) investigated carbohydrate components in sweet potatoes baked by microwave and convection heating. Little information is available on microwave blanching nor the properties of flour prepared from microwave blanched sweet potatoes. The objectives of this study were to determine the nutritive composition, physical properties and sensory quality of microwave blanched sweet potato flour and to compare with steam blanched and unblanched sweet potato flours.

MATERIALS AND METHODS

Sweet Potato Flour

"Jewel" sweet potatoes were obtained from the George Washington Carver Agricultural Experiment Station, Tuskegee University, Tuskegee, AL. After harvest, storage roots were cured at 30°C and relative humidity (RH) of 80–85% for 1 week, then stored for 1 month at 15°C and 80% RH.

Storage roots weighing 250–450 g were washed, peeled and sliced 2–3 mm thick. They were divided into 3 batches. Each batch consisted of 4–5 kg. One batch was steamed for 10 min. The condition of microwave heating was tested prior to the experiment. To determine the time required to reach 90°C or higher, sweet potato roots of known weight were heated at various times. A thermometer was inserted immediately after heating to measure the inner temperature. After establishing time-weight relationship, this factor (10 min for 800 g) was used when larger amounts of sweet potato roots were heated for blanching. Another batch was not blanched. The three batches of sweet potatoes blanched or unblanched were dried in a "Harvest Maid" dehydrator (Alternative Pioneering System, Inc., Minneapolis, MN) at 50–60°C for 24 h or until crispy (7–8% moisture). The dried slices were first ground with a Wiley's mill, followed by a ball mill (Norton Chemical Process Products Division, Akron, OH) to a fine powder. Sixty-100-mesh-size portions were collected by using sieves. The flour was packaged in 0.05 mm thick polyethylene bags, sealed and stored at 4.4°C until use. The percentage yield was calculated as weight of dry flour/weight of fresh potatoes × 100.

Chemical Analysis

Moisture, protein, fat and ash were determined according to AOAC (1980). Riboflavin and thiamin were determined fluorometrically (AOAC 1980), and beta-carotene by the spectrometric method (Reddy and Sistrunk 1980). Starch was determined by an acid hydrolysis method (Lu *et al.* 1986). Sugars were

analyzed by a HPLC system (LDC/Milton Roy, Rivera Beach, FL) equipped with a refractomonitor. An Amino 5-s carbohydrate column (Bio Rad) was employed. The solvent system consisted of 75% acetonitrile and 25% water at a flow rate of 1.5 mL/min at 20°C (Lu *et al.* 1986). A Perkin Elmer atomic absorption spectrophotometer 3080 (Perkin Elmer's Corp., Norfork, CT) was employed to determine Na, K and Fe (Anon. 1971).

Physical Properties

Flours were analyzed for water and oil binding capacity (Beuchat 1977). An Oswald viscometer was employed to measure relative viscosity of 0.1% solution at 20°C. A Minolta Chroma Meter II (Minolta Corp., Ramsey, NJ) was used to measure the color difference values, "L", "a" and "b."

Preparation of Baked Products

Three products, bread, cake and cookies were prepared. Table 1 shows the formulation for all three products. To prepare bread, the ingredients were mixed into dough. The dough was placed into a well-greased bowl, covered with a moist cheesecloth and allowed to rise at 26–29°C for 1½ h. The dough was punched, folded over and allowed to rise for an additional 50 min. Loaves were baked at 204°C for 40 min. The cake was prepared by first mixing butter and sugar until light and fluffy with an Oster 5 speed mixer (Oster, Milwaukee, WI) at speed 4 for 5 min. Four eggs were added to creamed mixture. The dry ingredients, sifted all purpose flour, sweet potato flour, baking powder and nutmeg, were mixed thoroughly using a Waring hand mixer at speed 3 for 3 min (Crown Distributors, Edison, NJ). The batter was poured into greased floured pans (18.75′ × 4.5′ × 2.75′) and heated at 176°C for 40–50 min. The method used in the cake was adapted for the cookies. The dough was dropped by teaspoonfuls on cookie sheets and baked for 10 min at 176°C. The size of cookies was approximately 5 cm diameter and 1 cm thick.

Sensory Evaluation

Evaluation of bread, cake and cookies was conducted at separate times. A small piece of sample (about 30 g) was served with a cup of water. A 9-point scale was employed to determine preference, with 9 points being the highest score (liked extremely) and 1 point being the lowest (disliked extremely) (ASTM 1968). Thirty-three panelists, consisting of students, staff and faculty at Tuskegee University (Tuskegee, AL), evaluated the products.

Statistics

Data were analyzed by analysis of variance. Means were separated by Duncan's Multiple Range Test at the 5% level (Steel and Torrie 1980).

TABLE 1.
SWEET POTATO PRODUCTS FORMULATION

Ingredient	BREAD	Amount
Wheat Flour		298.0g
Sweet Potato Flour		127.7g
Yeast		7.0g
Sugar		11.9g
Salt		8.3g
Water		500.0ml

Ingredient	CAKE	Amount
Butter		227.0g
Sugar		336.0g
Wheat Flour		198.6g
Sweet Potato Flour		85.1g
Baking Powder		12.9g
Salt		8.3g
Nutmeg		2.3g
Eggs		224.0g

Ingredients	COOKIES	AMOUNT
Butter		227.0g
Sugar		252.0g
Wheat Flour		178.8g
Sweet Potato Flour		76.6g
Baking Soda		5.2g
Salt		8.3g
Raisins		180.5g
Eggs		112.0g
Vanilla		5.0ml

RESULTS AND DISCUSSION

The yield of flour was 13.13% for steam blanched, 18.65% for microwave blanched and 18.96% for unblanched sweet potatoes. Some soluble components, i.e., sugars and minerals, might have dripped down during steaming process. Therefore, yield was smaller for steam blanched than for unblanched and microwave blanched sweet potatoes. Unblanched dried slices were easiest to grind to flour, followed by microwave blanched slices, then steam blanched slices.

The steam blanched slices appeared slightly chewy and rubbery while unblanched and microwave blanched slices were brittle and crumbly.

Starch is the major component of sweet potatoes. The starch content ranged from 58.4 to 66.86%. Starch content was greater for unblanched than for blanched flour ($p < 0.05$) (Table 2). A portion of starch is converted to sugar during the blanching process because of activation of amylases; therefore, the starch content of blanched fours was slightly decreased. Martin (1984) reported that flour prepared from microwave baked sweet potatoes had 40–60% starch.

The major sugars in the blanched flours were sucrose and maltose, while sucrose was the major sugar in the unblanched flour. Appreciable amounts of glucose and fructose were also present. No maltose was detected in unblanched flour (Table 3). Total sugars were highest for steam blanched flour, followed by microwave blanched, then unblanched flour. Blanching decreased starch and increased sugar content in sweet potatoes.

There were no differences in protein, fat, beta-carotene or sodium among the three flours assayed ($p < 0.05$). Ash content was slightly greater in unblanched than in blanched flours ($p < 0.05$). Potassium was greater in microwave blanched and unblanched than in steam blanched flour. Smaller potassium value of steam blanched flour might be caused by dripping loss due to steaming. Slightly greater iron content in steam blanched flour than unblanched and microwave blanched flour might come from the contamination from the steaming equipment. Less thiamin and riboflavin were observed in steam blanched than in microwave blanched and unblanched flours ($p < 0.05$). This could be a time-temperature effect. Thiamin and riboflavin are destroyed when exposed to high temperature for a long period of time (Chichester 1973). Except for starch and sugar content, the composition of microwave blanched flour was similar to that of unblanched flour, and slightly greater in thiamin and riboflavin and potassium than steam blanched flour.

Color is an important attribute in food. The yellow-orange color of sweet potatoes is due mainly to the presence of beta-carotene (Martin 1984). There were no difference in "L" value, but "a" value was lowest and "b" value was highest for unblanched flour (Table 4). This agreed with our visual observation in that unblanched flour had light pale-yellow color while steam and microwave blanched flours had deep yellow-orange color indicating that blanching inactivated oxidizing enzymes; therefore, color was retained.

There was no difference in pH value and viscosity of flours, but water absorption ability was greater, while oil absorption was smaller for both blanched than for unblanched flour. This could be due to differences in starch and sugar content among these flours. Both steam and microwave blanched flour had greater sugar content and smaller starch content than unblanched flour. The results of the sensory evaluation are presented in Table 5. The scores of breads were 6.33 for steam blanched, 6.48 for microwave blanched, and 6.36 for unblanched.

TABLE 2.
NUTRIENT COMPOSITION OF SWEET POTATO FLOUR[1]

	Steamed blanched	Microwave blanched	Unblanched
Moisture (%)	5.85 ± 0.17^{c}	8.20 ± 0.17^{a}	6.50 ± 0.17^{b}
Protein (%)	6.20 ± 0.11^{a}	6.30 ± 0.32^{a}	7.40 ± 0.48^{a}
Fat (%)	1.33 ± 0.27^{a}	1.50 ± 0.01^{a}	1.35 ± 0.30^{a}
Ash (%)	3.03 ± 0.03^{a}	3.66 ± 0.03^{a}	3.91 ± 0.17^{a}
Starch (%)	58.40 ± 0.04^{b}	60.00 ± 1.26^{b}	66.86 ± 1.81^{a}
B-Carotene	17.81 ± 2.23^{a}	18.56 ± 2.09^{a}	20.80 ± 1.92^{a}
Thiamin (mg)	0.008 ± 0.00^{a}	0.012± 0.00^{b}	0.14 ± 0.00^{b}
Riboflavin (mg)	0.55 ± 0.09^{a}	0.95 ± 0.04^{b}	0.93 ± 0.16^{b}
Iron (mg)	0.19 ± 0.02^{a}	0.11 ± 0.01^{b}	0.10 ± 0.00^{b}
Sodium (mg)	119.0 ± 0.80^{a}	114.8 ± 0.10^{a}	117.9 ± 0.52^{a}
Potassium (mg)	399.4 ± 0.28^{a}	436.6 ± 0.42^{b}	483.8 ± 0.36^{b}

Means with the same superscript in the same row are not significantly different at the 5% level

[1]Means ± S.E. on three determinations. Dry weight basis.

TABLE 3.
SUGAR CONTENT OF SWEET POTATO FLOUR[1]

Sample	Sucrose (%)	Maltose (%)	Glucose (%)	Fructose (%)	Total Sugars
Steam blanched	10.74 ± 0.81[a]	10.36 ± 0.92[a]	2.13 ± 0.27[a]	1.96 ± 0.18[a]	25.19
Microwave blanched	9.04 ± 2.54[a]	8.66 ± 2.5[a]	1.30 ± 0.08[b]	1.40 ± 0.11[b]	20.34
Unblanched	11.45 ± 1.10[a]	----	1.38 ± 0.16[b]	1.15 ± 0.06[b]	13.98

Means with the same superscript in the same column are not significantly different at 5% level.

[1]Means ± S.E. on three determinations.

TABLE 4.
PHYSICAL PROPERTIES OF "JEWEL" SWEET POTATO FLOUR

Functional Properties	Steam blanched	Microwave blanched	Unblanched
Water absorption	4.03 ± 0.27[a]	3.90 ± 0.15[a]	2.67 ± 0.06[a]
Oil absorption	1.80 ± 0.11[a]	1.86 ± 0.06[a]	2.46 ± 0.00[b]
pH	5.66 ± 0.04[a]	5.72 ± 0.01[a]	5.79 ± 0.00[b]
Relative Viscosity Conc. (0.1%)	1.10 ± 0.01[a]	1.20 ± 0.08[a]	1.28 ± 0.07[a]
Color L	88.05 ± 0.75[a]	88.90 ± 0.50[a]	87.65 ± 0.95[a]
a	3.15 ± 0.05[a]	2.65 ± 0.25[a]	1.80 ± 0.00[b]
b	27.40 ± 0.50[b]	27.55 ± 0.15[b]	29.55 ± 0.55[a]
Visual perception	orange	orange	light orange

Means with the same superscript in the same row are not significantly at the 5% level.

TABLE 5.
SENSORY EVALUATION SCORES

Product	Steam blanched	Microwave blanched	Unblanched
Bread	6.33 ± 0.33	6.48 ± 0.28	6.36 ± 0.33
Cake	8.15 ± 0.12	8.21 ± 0.17	7.03 ± 0.24
Cookie	7.93 ± 0.18	7.96 ± 0.18	7.06 ± 0.29

Mean ± S.E.

These scores were close, and the effect of incorporating blanched flour was not clearly demonstrated. The breads all left a characteristic lingering sweet potato flavor in the mouth when tasted. For cake and cookies, the scores were higher for both steam and microwave blanched than for unblanched. For cake, the scores were 8.15 for steam blanched and 8.21 for microwave blanched while that of unblanched was 7.03. The result indicates that the cakes incorporated with blanched flours were ranked between "like moderately" and "like very much." The result of cookies was similar to that of cakes. The scores were 7.93 for steam blanched, 7.96 for microwave blanched while that of unblanched was 7.06, indicating that panelists liked the blanched sweet potato flour products.

All the bakery products tested had 30% of wheat flour substituted with sweet potato flour. Sweet potato flour can be used to replace up to 30% wheat flour without changing acceptability of product. In many developing countries where sweet potato is a major staple and where flour is imported, partial substitution of wheat flour with blanched sweet potato flour would be economically desirable and organoleptically acceptable.

Processing of sweet potatoes into flour increases not only the storage ability but also further diversifies sweet potato usage. Considering factors such as yield, ease of process and energy efficiency, microwave blanching is preferred to the traditional steam blanching sweet potatoes. Further study is needed to determine the effect of storage conditions on the quality of microwave blanched sweet potato flour.

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REFERENCES

ANON. 1971. Analytical Method of Atomic Absorption Spectrophotometry. Perkin Elmer's Corp., Norwalk, CT.

AOAC. 1980. *Official Methods of Analysis,* 31st ed. Association of Official Analytical Chemist, Washington, D.C.

ASTM. 1968. Manual of Sensory Testing Methods, STP 434. Am. Soc. Testing Materials, pp. 22–24.

BEUCHAT, L. R. 1977. Functional and electrophoretic characters of succinylated peanut flour proteins. J. Agr. Food Chem. *25,* 258.

CHICHESTER, C. O. 1973. Nutrition in food processing. World Rev. *16,* 318.

DAWSON, P. R. 1951. Sweet potato for food and feed. USDA. Yearbook of Agriculture, p. 204.

LU, J. Y., WHITE, S., YAKUBU, P. and LORETAN, P. A. 1986. Effects of gamma radiation on nutritive and sensory qualities of sweet potato roots. J. Food Qual. *9,* 425.

MARTIN, F. W. 1984. Techniques and problems in small scale production of flour from sweet potato. J. Agr. Univ. Puerto Rico *68*(4), 423.

NOBLE, I. and GORDON, J. 1964. Effects of blanching method on ascorbic acid and color of frozen vegetables. J. Am. Diet. Assoc. *44,* 120.

ODLAND, D. and EHEART, M. S. 1975. Ascorbic acid, mineral and quality retention in frozen broccoli blanched in water steam and ammonia steam. J. Food Sci. *40,* 1004.

POMINSKI, J. and VINNETT, C. H. 1989. Production of peanut flour from microwave vacuum dried peanuts. J. Food Sci. *54,* 187.

PURCELL, A. E. and WALTER, W. M. 1988. Comparison of carbohydrate components in sweet potatoes baked by convection heating and microwave heating. J. Agr. Food Chem. *36,* 360.

REDDY, N. N. and SISTRUNK, W. A. 1980. Effect of cultivar, size, storage and cooking method on carbohydrate and some nutrients of sweet potato. J. Food Sci. *45,* 221.

SMITH, R. D. 1984. Microwave Power in Industry. Thermo Energy Corp., Palo Alto, CA. for Electric Power Research Institute, Palo Alto, CA.

SPADARO, J. J. and PATTON, E. L. 1961. Precooked dehydrated sweet potato flakes. Food Eng. *33*(7), 46.

STEEL, R. G. and TORRIE, J. H. 1980. *Principles and Procedures of Statistics.* McGraw-Hill Book Co., Inc., New York.

WADSWORTH, J. I., KOLTER, S. P., GALLO, A. S., ZIEGLER, G. M. and SPADARO, J. J. 1966. Instant sweet potato flakes; factors affecting drying rate on double drum drier. Food Technol. *20,* 815.

WINARNO, F. G. 1980. Sweet potato processing and byproducts utilization in the tropics. In the *Proceeding of the 1st International Symposium on Sweet Potato.* pp. 373. Tainan, Taiwan.

EFFECTS OF HEATING AND POSTMORTEM AGING ON PHYSICAL PROPERTIES OF SQUID MANTLE

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ABSTRACT

Physical and tensile properties of Loligo and Illex squid mantles were evaluated during refrigerated storage and after various heat treatments. A major decrease in ultimate tensile strength of the mantle occurred between 50° and 60°C in a direction parallel to the longitudinal axis of the mantle. After storage for more than 4 days at 4°C, the ultimate tensile strength of the mantle decreased significantly in the longitudinal but not the transverse direction. In addition, heating of mantle (100°C for 2 min) stored for more than 4 days led to greater strength loss than observed in heated, nonstored mantle, indicating that refrigerated storage produces changes in collagen that make it more susceptible to the effects of high temperature. Strength loss in the transverse direction after heating was observed in mantle of Loligo but not Illex.

INTRODUCTION

The texture of squid mantle is quite unique and has prompted several studies of various species (Ward and Wainwright 1972; Gosline and Shadwick 1983; Shadwick and Gosline 1984). The state of freshness prior to freezing has been reported to have an influence on the texture of cooked squid after prolonged frozen storage. Samples of Illex showing distinct discoloration of both skin and muscle layers under the skin (an indication that the squid was not fresh) (Takahashi 1965) had sensory "hardness" scores and Warner-Bratzler force readings

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about 30% less than those of fresh squid (Kolodziejska *et al.* 1987). Holding the squid prior to cooking for 18 h at room temperature or 2 h at 35°C did not affect the texture of the cooked squid, although it did cause significant proteolysis of myosin as judged by SDS-PAGE (Kolodziejska 1985). It has been shown that the ultimate tensile strength of squid mantle is greater in the direction parallel to the long axis of the squid body than in the perpendicular direction (transverse) (Otwell and Hamann 1979b; Kuo *et al.* 1990). It was also reported that heating the mantle at 60°C caused a significant decrease in the ultimate tensile strength in the longitudinal direction of Illex and Loligo (Kuo *et al.* 1990), which could have been due to melting of the collagen fibers predominantly oriented in that direction (Otwell and Hamann 1979a). The objective of this work was to compare the response of two North Atlantic squid, *Illex illecebrosus* and *Loligo pealei,* to refrigerated storage, heating, and the combination of storage and heating with respect to their tensile and other properties.

MATERIALS AND METHODS

Squid was obtained from day boats in Gloucester, MA, and transported on ice to the laboratory in Amherst, MA. Mantles with lengths of 14.4 ± 3.3 cm for *Loligo pealei* and 14.2 ± 3.8 cm for *Illex illecebrosus* were chosen for this study. They were cleaned (skin, tentacles, and viscera removed) and cut open along the dorsal surfaces which yielded a flat, approximately triangular shaped "fillet" of squid meat, whereupon it was packaged in sealed plastic bags, and stored at −40°C.

Thawed mantles were either tested at room temperature (about 25°C) or cooked prior to testing. Samples were heated in distilled water (squid to water ratio 1/60 weight ratio) at the specified temperatures for 2 min and then immediately immersed in distilled water at room temperature for 10 min.

The procedures of specimen preparation and mechanical testing using an Instron are described in detail elsewhere (Kuo *et al.* 1990).

Moisture content was determined by the AOAC method (AOAC 1984). Mantle pH measurements were taken from a blend of macerated squid in distilled water (1/10 weight ratio) homogenized for 3 min. Weight loss and changes in the physical dimensions were monitored before and after cooking. Density was determined as the weight of sample divided by the volume of water displaced at room temperature. Extractable hydroxyproline (HOP) was obtained by treatment of macerated tissue with 0.1 M phosphate buffer, pH 7.0 at 26° or 70°C followed by centrifugation (Hatae *et al.* 1986). The HOP content was determined by a colorimetric method (Woessner 1961) after hydrolyzing the tissue with 6 M HCl for 3 h at 130°C.

All analyses were performed in triplicate on each squid and at least three squid mantles were used in each treatment. Data are expressed as mean ± standard

deviation. The significance of the differences was evaluated using the Duncan Multiple Range Test and Student's t-test at a 95% level of confidence.

RESULTS

Effects of Heat Treatment

The ultimate tensile strength of *Loligo pealei* and *Illex illecebrosus* cooked mantles as a function of treatment temperature in both the longitudinal and transverse directions is shown in Fig. 1 and 2, respectively. In both species decreases in ultimate tensile strength in the longitudinal direction occurred between 50° and 60°C. The slight trend towards strength recovery above 60°C is not statistically significant. There was no significant effect of temperature on the ultimate tensile strength in the transverse direction in the Illex mantles. In those of Loligo the strength in this direction increased when heated to 80° and 100°C.

The effects of heating Loligo and Illex mantle at 60°C and 100°C for 2 min are shown in Table 1. When squid mantle is heated in water, it shrinks and curls. The longitudinal shrinkage is believed to be due to contraction of collagen fibers in the tunic and the transverse shrinkage to contraction of muscle fibers (Takahashi 1965). The curling is always in the direction of the longitudinal axis of the mantle. It is obviously caused by greater contraction of the outer tunic relative to that of the inner tunic. Since the muscle cells near both surfaces are

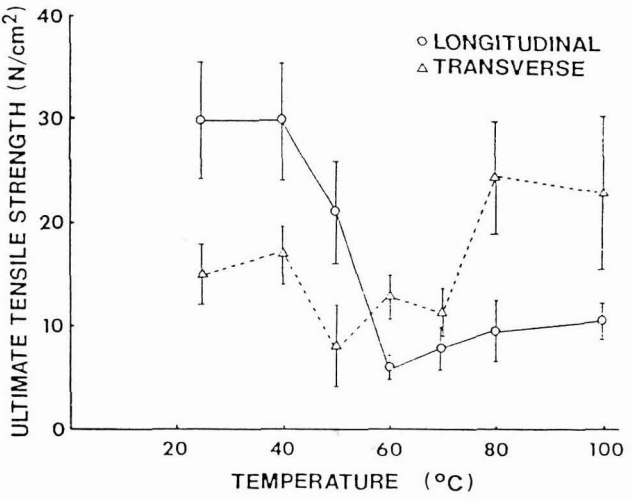

FIG. 1. RELATIONSHIP BETWEEN ULTIMATE TENSILE STRENGTH AND TEMPERATURE OF HEATING OF *LOLIGO PEALEI*
Samples were placed for 2 min in water at the indicated temperature.

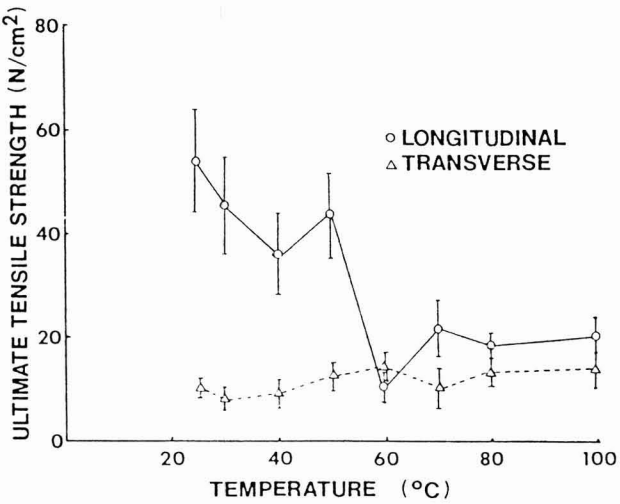

FIG. 2. RELATIONSHIP BETWEEN ULTIMATE TENSILE STRENGTH AND TEMPERATURE OF HEATING OF *ILLEX ILLECEBROSUS*
Samples were placed for 2 min in water at the indicated temperature.

TABLE 1.
EFFECT OF COOKING TEMPERATURE ON SQUID MANTLE[1]

	Loligo		Illex	
	60°C (%)	100°C (%)	60°C (%)	100°C (%)
Weight loss	5±1	30±3	9±1	36±2
Transverse shrinkage	8±2	22±5	5±2	27±5
Longitudinal shrinkage				
outer tunic side	23±6	58±4	45±3	58±2
inner tunic side	9±3	12±5	12±4	23±4
Thickness increase	15±4	22±12	9±2	9±7
Density increase	29±1	38±11	28±8	42±3
Moisture content	84.9±1.9	76.5±2.5	79.1±0.5	68.7±4.5

[1]Cooked for 2 min at the specified temperature.
The density of raw mantle of both species was 1.1±0.1 g/cm^3.
The moisture content of raw Loligo was 82.8±0.5% and of the raw Illex 80.2±1.7%.

the same morphologically, this difference is probably due to structure, composition, and/or orientation of the collagen fibrils. This reinforces our previous suggestion that the profound changes in tensile strength in the longitudinal direction of heated squid mantle relates to changes in collagen (Kuo *et al.* 1990). Increase of mantle thickness with concomitant weight loss is another manifestation of heat-induced contraction. The type of geometric changes in the mantle were of the kind expected of a material undergoing asymmetric contraction. The same pattern was observed in the two squid species.

Effects of Refrigerated Storage

The changes in ultimate tensile strength of mantle stored at 4°C are shown in Table 2. In both species a significant loss of strength in the longitudinal direction occurred at approximately the same time, i.e., between 4 and 7 days for Loligo and between 4 and 5 days for Illex. No significant changes in the transverse direction were observed in either species.

Stored mantles were heated in water at 100°C for 2 min. Their moisture content, weight loss, and strength in the longitudinal and transverse directions are given in Table 3. As expected (Otwell and Hamann 1979b; Kuo *et al.* 1990), the ultimate tensile strength in the longitudinal direction decreased upon heating. The effect was larger after 4 days of storage than that observed with mantle stored for 4 days or less. In heated Loligo mantle there was also a loss of strength in the transverse direction in unison with that of the longitudinal direction. In Illex, in contrast, no significant loss of strength in the transverse direction was observed in heated mantle during the 7 day storage period.

TABLE 2.
ULTIMATE TENSILE STRENGTH (N/cm^2) OF RAW SQUID MANTLE STORED AT 4°C

	Loligo		Illex	
Days	longitudinal	transverse	longitudinal	transverse
0	36 ± 8^a	13 ± 3^c	61 ± 10^a	20 ± 4^c
2	33 ± 5^a	14 ± 3^c	57 ± 11^a	17 ± 3^c
4	31 ± 6^a	13 ± 4^c	55 ± 10^a	18 ± 4^c
5	--	--	40 ± 5^b	15 ± 3^c
7	21 ± 4^b	14 ± 4^c	42 ± 6^b	14 ± 3^c

Values with different superscripts within each species are significantly different ($p\leq0.05$).

TABLE 3.
EFFECT OF HEATING AT 100°C FOR 2 MIN ON RAW SQUID MANTLE PREVIOUSLY STORED AT 4°C

	Days at 4°C					
	0	2	4	5	7	12
Loligo						
Moisture (%)	72.9±0.2	---	74.7±0.3	---	77.0±0.8	81.0±0.2
Weight loss (%)	48±3	---	47±2	---	38±4	24±3
Ultimate tensile strength (N/cm^2) L	15 ± 2^a	---	13 ± 2^a	---	9 ± 2^b	5 ± 1^c
T	23 ± 6^c		21 ± 4^c		16 ± 2^a	15 ± 3^a
pH of raw squid	6.6		7.0		7.5	8.2
Illex						
Moisture (%)	74±0.3	76±0.5	76.8±0.5	78.8±0.5	81.4±0.8	---
Weight loss (%)	46±3	44±3	42±2	41±4	37±3	---
Ultimate tensile strength (N/cm^2) L	33 ± 6^a	34 ± 7^a	32 ± 5^a	24 ± 3^b	20 ± 3^b	---
T	14 ± 3^c	15 ± 3^c	14 ± 4^c	13 ± 5^c	13 ± 4^c	---
pH of raw squid	6.8	7.1	7.3	7.3	7.5	---

Moisture content for Loligo before heating was 87.9±1.5% and for Illex 83.8±0.3%
Weight loss of heated mantle is expressed as a percentage of raw mantle.
Values with different superscripts within each species are significantly different ($p \leq 0.05$).
L- value in longitudinal direction; T- value in transverse direction.

The moisture content of cooked stored mantle tissue increased with time of storage. This was also reflected in a smaller weight loss (Table 3). During storage the pH of the raw mantle progressively increased. Comparison of shrinkage data of fresh and stored mantles indicated that storage of raw mantle at 4°C for up to at least 7 days had no effect on cooked mantle dimensions.

Since tensile properties of raw squid mantle are regulated in part by collagen fibers (Kuo *et al.* 1991), the amount of extractable hydroxyproline with storage time was also determined (Fig. 3). The results were inconclusive, as no major changes in HOP contents were observed after extraction at either 26° or 70°C.

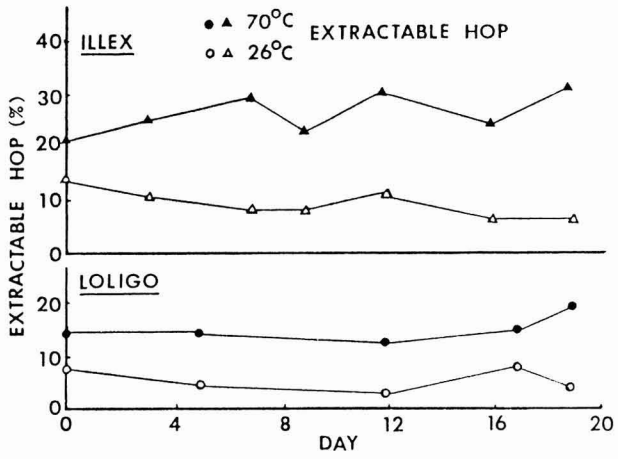

FIG. 3. CHANGES IN 26°C- AND 70°C-EXTRACTABLE HYDROXYPROLINE (HOP) OF SQUID MANTLE STORED AT 4°C

DISCUSSION

Although the role of collagen in regulating tenderness and other mechanical properties of postmortem muscle tissue is well accepted, its specific effects in textural changes that occur during postmortem storage are not fully understood. It has been difficult to show that important changes in the collagen molecules occur during storage or to relate any molecular changes to textural modifications. Likewise, no major changes in the proportion of extractable hydroxyproline were found in this work with squid. Nevertheless, the observed strength loss in the longitudinal direction of the mantle of both squid species indicates that there were changes during storage and that collagen molecules were probably involved. Similar observations were reported by Otwell and Hamann (1979b). Other work (Kuo *et al.* 1991) has shown that treatments that modify collagen fibers reduce the strength of raw squid mantle in the longitudinal direction. That the strength loss was primarily due to modification of collagen fibers is further supported in this work by the lack of effect of postmortem storage on squid mantle strength in the transverse direction. Other work (Kuo *et al.* 1991) has indicated that when the contractile proteins were modified, the strength of mantle oriented transverse to the longitudinal axis decreased.

An effect of storage on ultimate tensile strength in the longitudinal direction was observed in heated squid samples (Table 3). The loss in the heated samples occurred after the same storage time that caused loss of strength in raw samples. Thus, the change(s) which occurred in the collagen during storage affected the tensile properties of both raw and heated squid. The data suggest that the strength

loss in heated mantles was not caused by complete melting and disorganization of the collagen. On the contrary, a significant amount of structure must have remained after heating. Otherwise, heating should have reduced the tensile strength to a single low value regardless of whether the sample was aged.

During storage the pH of the squid tissue increased. The moisture content of the corresponding cooked samples also increased, and their weight losses decreased. It is likely that these changes are interrelated. Muscle proteins bind more water as the pH increases, thus the greater retention of water in squid cooked after aging.

An examination of the effect of temperature on the ultimate tensile strength showed that the strength loss in the longitudinal direction in both squid species was complete at about 60°C (Fig. 1 and 2), confirming the results of Otwell and Hamann (1979b). This may be attributed to melting of the collagen fibers whose orientation is generally in the longitudinal direction (23–27° off the major axis). No such strength loss was observed in the transverse direction, confirming a previous suggestion that muscle cells contribute more to the tensile properties in this direction than do collagen fibers (Kuo *et al.* 1991) . Data scatter made it difficult to draw other conclusions. Scatter was primarily due to sample-to-sample variation, as is usually the case with animal materials, although experimental factors such as difficulty in accurately controlling specimen dimensions was also a factor.

Storage for 7 days at 4°C had no effect on the strength of heated Illex mantle in the transverse direction, but it decreased that of Loligo. Whether this is an indication that there are inherent structural differences between the mantles of the two species is a question that cannot be answered without further study.

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REFERENCES

AOAC. 1984. Official Methods of Analysis of the Association of Official Analytical Chemists, 13th ed. p. 431. Association of Official Analytical Chemists, Washington, D.C.

GOSLINE, J. M. and SHADWICK, R. E. 1983. Molluscan collagen and its mechanical organization in squid mantle. In *The Mollusca, Vol. 1. Metabolic Biochemistry and Molecular Biomechanics,* (P. W. Hochachka, ed.) pp. 371–398, Academic Press, New York.

HATAE, K., TOBIMATSU, A., TAKEYAMA, M. and MATSUMOTO, J. J. 1986. Contribution of the connective tissue on the texture difference of various fish species. Bull. Japan. Soc. Sci. Fish. *52*, 2001–2007.

KOLODZIEJSKA, I. 1985. The proteins of squid meat. Prox. XXXI Europ. Meeting of Meat Res. Workers, Albena, pp. 175–178.

KOLODZIEJSKA, I., SIKORSKI, Z. E. and SADOWSKA, M. 1987. Texture of cooked mantle of squid *Illex argentinus* as influenced by specimen characteristics and treatments. J. Food Sci. *52*, 932–935.

KUO, J.-D., PELEG, M. and HULTIN, H. O. 1990. Tensile characteristics of squid mantle. J. Food Sci. *55*, 369–371, 433.

KUO, J.-D., HULTIN, H. O., ATALLAH, M. T. and SUN PAN, B. 1991. Role of collagen and contractile elements in tensile strength of squid mantle. J. Agric. Food Chem. (In press).

OTWELL, W. S. and HAMANN, D. D. 1979a. Textural characterization of squid (*Loligo pealei* Leseur): scanning microscopy of cooked mantle. J. Food Sci. *44*, 1629–1635, 1643.

OTWELL, W. S. and HAMANN, D. D. 1979b. Textural characterization of squid (*Loligo pealei* L.): instrumental and panel evaluations. J. Food Sci. *44*, 1636–1643.

SHADWICK, R. E. and GOSLINE, J. M. 1984. The role of collagen in the mechanical design of squid mantle. In *Biology of Invertebrate and Lower Vertebrate Collagens*, (A. Bairati and R. Garrone, eds.) pp. 299–304, Plenum Press, New York.

TAKAHASHI, T. 1965. Squid meat and its processing. In *Fish as Food, Vol. IV*, (G. Borgstrom, ed.) pp. 339–354. Academic Press, New York.

WARD, D. V. and WAINWRIGHT, S. A. 1972. Locomotory aspects of squid mantle structure. J. Zool. London *167*, 437–449.

WOESSNER, J. F. 1961. The determination of hydroxyproline in tissue and protein samples containing small proportions of this amino acid. Arch. Biochem. Biophys. *93*, 440–447.

A CONTROL SYSTEM FOR ACHIEVING CORRECT HEAT STERILIZATION PROCESSES: A ONE-DIMENSIONAL APPROACH TO PACKAGED CONDUCTION HEATING OF FOODS

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ABSTRACT

A procedure was developed for an on-line control system to achieve the correct heat sterilization for conduction heating of food. The procedure collects a series of time/temperature data via a sensing probe during the early stages of the heating phase and analyzes the collected data for parameters of conduction heating. From this data, it predicts the time/temperature relationship for the remainder for the heating phase and during cooling, integrates the lethal rate, and determines an appropriate time to start cooling to achieve a desired process lethality. This procedure has been further validated via laboratory experiments. This procedure estimated the true location of the sensing probe inserted in the food. Locating the sensing probe on the container outside surface saved considerable computer time for calculation and facilitated setting up the probe.

INTRODUCTION

Thermally sterilized food is one of the most economical and safest food products currently available on the market. The sterilizing value at the thermal center of food is used as a criterion to evaluate thermal processes (Hayakawa 1983a). The sterilizing value is very dependent on the time at which cooling is started because it governs the duration of the high temperature period in the phase of heating as well as cooling (Naveh *et al.* 1983). In the case of sterilizing temperature sensitive foods, the quality of which is easily reduced by overcooking, some method to accurately predict the time/temperature relationship at the thermal center is required in order to decide the appropriate time to start cooling, which accurately gives the desired process lethality.

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Transient state temperature distribution in a canned or other packaged food undergoing heat sterilization processes may be accurately represented by the analytical or numerical solution of the Fourier's equation for heat conduction, provided that the thermophysical properties and boundary conditions such as heat conductivity and heat capacity as well as the surface heat transfer coefficient are known. Unfortunately, however, these physical constants are not widely available (Hayakawa 1983b). To determine the precise values for thermal conductivity of a particular food product, as well as the surface heat transfer coefficient, often requires a separate, lengthy and detailed analysis.

In this paper, a procedure is developed which collects a series of time/temperature data via a sensing probe from the early stages of the heating phase and analyzes the collected data for the parameters responsible to predict the time/temperature relationship for the remainder of heating and cooling phase. Using this data it integrates the lethal rate, and determines an appropriate time to start cooling to achieve a desired process lethality.

The approach of this paper may seem similar to those of inverse heat conduction analyses. In some inverse heat conduction analyses, thermophysical properties have been estimated from a known temperature history (Angstroem 1864; Cezairliyan 1971; Iida 1982). In these analyses, measurements were so designed as not to be affected by boundary conditions such as surface heat transfer.

Another group of inverse conduction analyses, estimates boundary conditions from a known temperature history at a site inside the body, using known thermophysical properties (Stolz 1960; Burggraf 1964; Beck 1970, 1985).

The approach of this paper does not take the form of either of the inverse analyses described above. We estimate, from a known temperature history in the early stage of heating, the location of the sensing probe and some parameters which are combinations of thermophysical properties and/or coefficients related to a boundary condition. We attempt, using these parameters, to predict the remaining time/temperature history from which correct heat sterilization can be performed.

THEORETICAL CONSIDERATIONS

Heat Transfer Equation

We assume that a canned or packaged conduction heating food is sterilized in a retort with hot water. After an appropriate duration of heating, t_q, the hot water is discharged and replaced with cooling water. A step-functional temperature change of the surrounding medium is assumed. The shape of food is assumed to be a rectangular thin slab (2D in thickness) which satisfies one-dimensional heat conduction.

Transient state temperature distributions in these solid bodies may be represented by the solution of following Fourier's equation for heat conduction as follows.

$$\rho\, C_p \frac{\partial T}{\partial t} = \lambda \frac{\partial^2 T}{\partial x^2} \tag{1}$$

The initial and boundary conditions are:

$$T = T_i \qquad t=0 \tag{2}$$

$$\pm \lambda \frac{\partial T}{\partial x} = -\, h(T - T_h) \qquad t > 0 \qquad x = \pm D \tag{3}$$

In terms of dimensionless variables

$$\eta = \frac{(T - T_h)}{(T_i - T_h)} \,, \quad \theta = \frac{t}{\tau} \,, \quad \xi = \frac{x}{D}$$

where $\tau = \rho C_p D^2/\lambda$, Fourier's equation becomes

$$\frac{\partial \eta}{\partial \theta} = \frac{\partial^2 \eta}{\partial \xi^2} \tag{4}$$

subject to the conditions

$$\eta = 1 \qquad \theta = 0 \tag{5}$$

$$\pm \frac{\partial \eta}{\partial \xi} = -\, B_i\, \eta \qquad \theta > 0 \qquad \xi = \pm 1 \tag{6}$$

where $B_i = h\, D/\lambda$.

The analytical solution of Eq. (4) is given by

$$\eta = \sum_{n=1}^{\infty} [A_n \exp(-P_n^2\, \theta) \cos (P_n\, \xi)] \tag{7}$$

where P_n is an n-th root of following equation,

$$P_n \tan(P_n) = B_i \tag{8}$$

and A_n is given by

$$A_n = \frac{\int_0^1 \cos(P_n\, \xi)\, d\xi}{\int_0^1 \cos^2(P_n\, \xi)\, d\xi} = \frac{2 \sin(P_n)}{P_n + \sin(P_n) \cos(P_n)} \tag{9}$$

From Eq. (7), (8) and (9), η is a function of two independent variables, θ, ξ, and a parameter B_i.

$$\eta = \frac{T - T_h}{T_i - T_h} = \eta(\theta, \xi, Bi) = \eta\left(\frac{t}{\tau}, \frac{x}{D}, Bi\right) \tag{10}$$

When T_h, T_i and D are known, Eq. (10) gives the time/temperature relationship at a location x as a function of two system parameters τ and B_i. In other words, what is required to specify the time/temperature relationship is not the values of the individual physical properties which appear in Eq. (1), but the system parameters τ and B_i. In this procedure, we have dealt with T_h, T_i and D as the known values due to their ease of measurement. Thus leaving the problem of how to estimate the system parameters τ and B_i as well as the location of the probe x, which is detailed below.

Estimating System Parameters and the Location of the Probe

In this system, a temperature sensing probe is placed in a sample food. The collection of a series of time/temperature data sets, at the location of the sensing probe, begins when we start a thermal sterilizing process.

$$(t_1,T_1),(t_2, T_2),(t_3,T_3), \cdots\cdots\cdots ,(t_k,T_k)$$

These time and temperature data sets may be reduced to dimensionless forms:

$$\theta_j = \frac{t_j}{\tau}, \qquad \eta_j = \frac{(T_j - T_h)}{(T_i - T_h)}, \qquad j=1,2,3,\cdots\cdots,k.$$

We attempt to estimate two system parameters τ, B_i and moreover the location of sensing probe ξ, by analyzing these data sets. To do this initially we use some estimated values for the parameters τ_o, B_{io} and ξ_o and substitution of these values into Eq. (10) gives an approximate value for the temperature at the true location of the probe.

$$\eta_{jo} = \eta(\theta_{jo}, \xi_o, B_{io}) = \eta(t_j/\tau_o, \xi_o, B_{io}) \tag{11}$$

This approximation may be improved to give η' by taking the first and second terms of the Taylor series concerning each parameter increment $\delta\tau$, δB_i and $\delta\xi$ as follows:

$$\eta' = \eta(\theta_{jo},\xi_o,B_{io}) + \frac{\partial\eta}{\partial\tau}\delta\tau + \frac{\partial\eta}{\partial Bi}\delta Bi + \frac{\partial\eta}{\partial\xi}\delta\xi \tag{12}$$

The difference between this improved approximation and a value observed with the probe is defined as E_j which is described as:

$$E_j = \eta' - \eta_j = R_j + \delta\eta \tag{13}$$

where

$$R_j = \eta(\theta_{oj}, \xi_o, Bi_o) - \eta_j \tag{14}$$

$$\delta\eta = \frac{\partial\eta}{\partial\tau}\delta\tau + \frac{\partial\eta}{\partial Bi}\delta Bi + \frac{\partial\eta}{\partial\xi}\delta\xi \tag{15}$$

The increments, $\delta\tau$, δB_i and $\delta\xi$, may be determined so that the sum of E_j^2 to be minimized; this occurs when following conditions are satisfied:

$$\begin{aligned}
\frac{\partial\sum_{j=1}^{k}\{E_j^2\}}{\partial\tau} &= 2\sum_{j=1}^{k}\{R_j + \delta\eta\}\frac{\partial(\delta\eta)}{\partial\tau} = 0 \\
\frac{\partial\sum_{j=1}^{k}\{E_j^2\}}{\partial Bi} &= 2\sum_{j=1}^{k}\{R_j + \delta\eta\}\frac{\partial(\delta\eta)}{\partial Bi} = 0 \\
\frac{\partial\sum_{j=1}^{k}\{E_j^2\}}{\partial\xi} &= 2\sum_{j=1}^{k}\{R_j + \delta\eta\}\frac{\partial(\delta\eta)}{\partial\xi} = 0
\end{aligned} \tag{16}$$

Discarding second and higher derivatives and rearranging we have:

$$\begin{aligned}
&\sum_{j=1}^{k}\left(R_j\frac{\partial\eta}{\partial\tau}\right) + \sum_{j=1}^{k}\left(\frac{\partial\eta}{\partial\tau}\frac{\partial\eta}{\partial\tau}\right)\delta\tau + \sum_{j=1}^{k}\left(\frac{\partial\eta}{\partial\tau}\frac{\partial\eta}{\partial Bi}\right)dBi + \sum_{j=1}^{k}\left(\frac{\partial\eta}{\partial\tau}\frac{\partial\eta}{\partial\xi}\right)\delta\xi = 0 \\
&\sum_{j=1}^{k}\left(R_j\frac{\partial\eta}{\partial Bi}\right) + \sum_{j=1}^{k}\left(\frac{\partial\eta}{\partial Bi}\frac{\partial\eta}{\partial\tau}\right)\delta\tau + \sum_{j=1}^{k}\left(\frac{\partial\eta}{\partial Bi}\frac{\partial\eta}{\partial Bi}\right)dBi + \sum_{j=1}^{k}\left(\frac{\partial\eta}{\partial Bi}\frac{\partial\eta}{\partial\xi}\right)\delta\xi = 0 \\
&\sum_{j=1}^{k}\left(R_j\frac{\partial\eta}{\partial\xi}\right) + \sum_{j=1}^{k}\left(\frac{\partial\eta}{\partial\xi}\frac{\partial\eta}{\partial\tau}\right)\delta\tau + \sum_{j=1}^{k}\left(\frac{\partial\eta}{\partial\xi}\frac{\partial\eta}{\partial Bi}\right)dBi + \sum_{j=1}^{k}\left(\frac{\partial\eta}{\partial\xi}\frac{\partial\eta}{\partial\xi}\right)\delta\xi = 0
\end{aligned} \tag{17}$$

When R_j and the derivatives of η with respect to each parameter in Eq. (17) are available, the increments, $\delta\tau$, δB_i and $\delta\xi$, used in improving the approximation can be obtained by solving Eq. (17) numerically. R_j may be obtained directly from Eq. (14) using a measured η_j value. Derivatives of η with τ, B_i and ξ may be obtained analytically (see Appendix I). An approximation for this derivative is available by substituting θ_{jo}, ξ_o, τ_o and B_{io} into the derivatives obtained as follows:

$$\frac{\partial \eta}{\partial \tau} \sim \sum_{n=1}^{\infty} \left[\frac{P_{no}^2 \, \theta_o}{\tau_o} A_{no} \exp(-P_{no}^2 \, \theta_o) \cos (P_{no} \, \xi_o) \right] \qquad (18)$$

where P_{no} is an n-th root of Eq. (8) with an approximate value B_{io}. The derivatives with respect to other parameters $\partial\eta/\partial B_i$, $\partial\eta/\partial\xi$, may be approximated in the same manner. The improved approximation for parameters τ, B_i and ξ are:

$$\tau' = \tau_o + \delta\tau, \qquad B_i' = B_{io} + \delta B_i, \qquad \xi' = \xi_o + \delta\xi .$$

Repeating this procedure several times may help us access to true values τ, B_i and ξ.

The Time to Start Cooling and Lethality Evaluation

After a duration of time of heating, t_q, the hot water is discharged and cooling with cold water starts. A step functional temperature change of the surrounding water from T_h to T_c is assumed. We also assume that the surface heat transfer coefficient is constant and equal during the heating and cooling phase. The time/temperature relationship within the food during cooling may be calculated by Fourier's equation using the parameters τ, B_i, and ξ, estimated in the previous section. Substituting $t = t_q$ in Eq. (7), the initial temperature profile in the cooling phase is given as:

$$T_q(x) = T_h + (T_i - T_n) \sum_{n=1}^{\infty} [A_n \exp(-P_n^2 \, t_q/\tau) \cos(P_n \, x/D)].$$

The solution is as follows (see Appendix II):

$$T = T_C + (T_C - T_h) \sum_{n=1}^{\infty} [C_n \exp\{-P_n^2 \, (t - t_q) / \tau\}] \qquad (19)$$

where,

$$C_n = A_n \, [\{(T_i - T_h)/(T_C - T_h)\} \exp(-P_n^2 \, t_q / \tau) - 1] \qquad (20)$$

We employ a sterilizing value, F_p, to evaluate a heat sterilizing process. The sterilizing value is estimated by integrating the lethal rates, L, with respect to the processing time over the period of heating as well as cooling.

$$F_p = \int_0^{t_q} L\,dt + \int_{t_q}^{t_e} L\,dt \tag{21}$$

The lethal rate used is a function of temperature, T, z value, and a reference temperature, T_r, as follows:

$$L = 10^{(T-T_r)/z} = \exp\left(\frac{T - T_r}{z} \ln 10\right) \tag{22}$$

Substituting Eq. (7), (19) and (22) to Eq. (21):

$$F_p = \int_0^{t_q} \exp\left[\{T_h + (T_i - T_h) \sum_{n=1}^{\infty} (A_n \exp(-P_n^2\, t/\tau)) - T_r\}(\ln 10)/z\right] dt$$

$$+ \int_{t_q}^{t_e} \exp\left[\{T_c + (T_c - T_h) \sum_{n=1}^{\infty} (C_n \exp(-P_n^2\, (t-t_q)/\tau)) - T_r\}(\ln 10)/z\right] dt \tag{23}$$

The time to start cooling, t_q, may be determined through a trial and error procedure, so that the obtained sterilizing value favorably agrees with the sterilizing value aimed at.

EXPERIMENTALS

A slab of acrylic resin and mannan jelly food were used as sample materials. A slab of acrylic resin was used as a food substitute because (1) it was easy to place a sensing probe accurately at the location aimed at, (2) no convection occurred, (3) repeated use was allowed, (4) thermal properties of the resin are similar to those of food. A slab (ca. 10.0 mm thick) was cut into a square (114 mm $\times$ 114 mm) with an aspect ratio of 11.4 which assured one-dimensional heat transfer. Two copper-constantan thermocouples made from thin sheet (0.055 mm thick) were inserted into each slab; one was placed on the midplane in the slab which measured the temperature at the geometric center. Another thermo-

couple was placed on a selected level which worked as a sensing probe to estimating the system parameters (Fig. 1).

A slab of mannan jelly, konnyaku, was used as an immobilized water model food. Mannan jelly was sandwiched with a couple of tinplates and fixed with bolts and nuts. A copper-constantan thermocouple wire (0.08 mm diameter) was inserted into the mannan jelly with its junction located at its geometrical center. Another thermocouple was soldered onto the outside surface of a tinplate (Fig. 2).

A cylindrical vessel (300 mm diameter, 300 mm height) filled with boiling water was heated to keep the temperature of hot water at a saturate temperature. Another cylindrical vessel (300 mm diameter, 300 mm height) filled with a continuous flow of tap water, which was allowed to overflow, was used to cool the food sample.

A food sample with the attached thermocouples was suspended in the hot water vessel at the start of the experiment. After a duration of heating t_q, the food sample was pulled out and put into the cooling vessel. The body was fixed in the water so that its surface was kept vertical.

The output voltage from the thermocouple was amplified and converted to digital signals through a data logger of a model THERMODAC 32 (Eto Co.). The digital signals were fed into a personal computer of a model NEC PC98XL2 (32 bits 16 MHz, CPU 80386 with MPU 80387) through a port RS-232C (Fig. 3). A data processing program written in BASIC language was used.

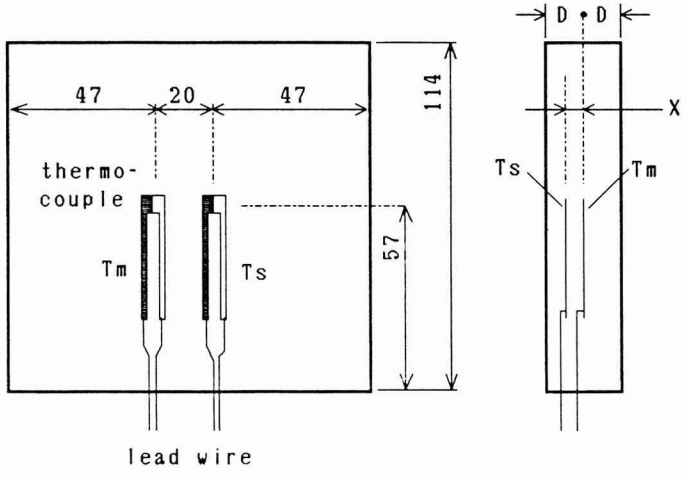

FIG. 1. A PLAN AND ELEVATION VIEW OF AN ACRYLIC RESIN SLAB WITH THERMOCOUPLES
Ts: sensing probe for estimating system parameters,
T_m: measuring temperature at geometric center.

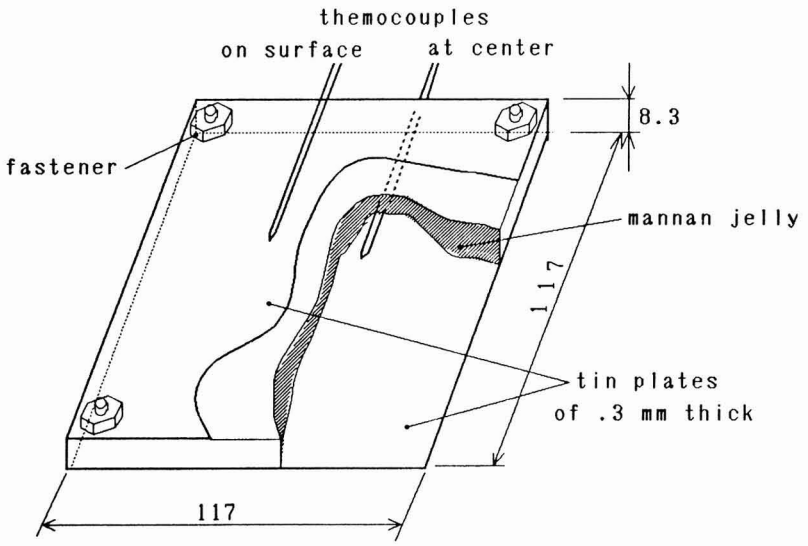

FIG. 2. A SLAB OF MANNAN JELLY SANDWICHED WITH A COUPLE OF TINPLATES

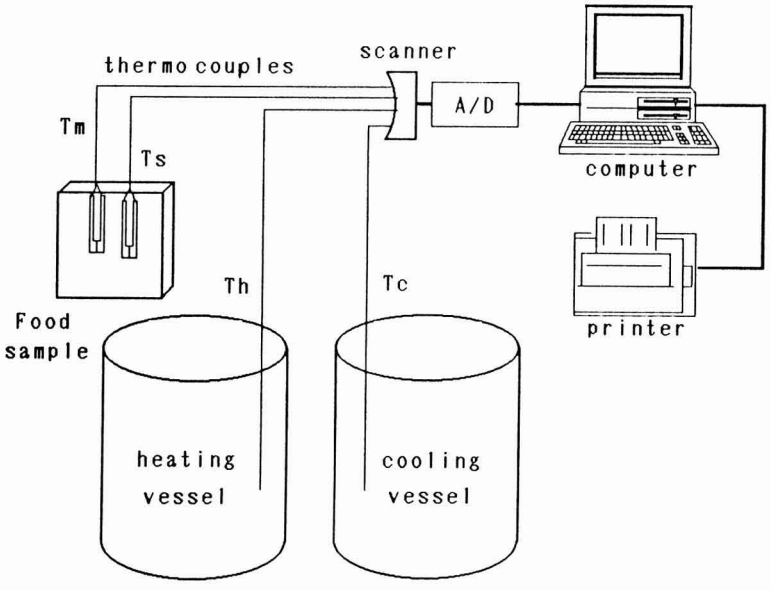

FIG. 3. A SCHEMATIC DIAGRAM OF EXPERIMENTAL APPARATUS

In commercial thermal processes with hot water, 121.1°C is a popular reference temperature for calculating sterilizing value and 125°C is a popular hot water temperature, which is 3.9°C higher than the reference temperature. In this experiment, however, hot water sterilization was conducted by use of boiling water in open air (ca. 99.6°C). In order to fulfil the simulation for sterilizing with hot water at 125°C, we used 95.7°C as the reference temperature, being 3.9°C lower than the hot water temperature. In addition to this, we used a modified Thermal Death Time curve for *C. botulinus* spores as shown in Fig. 4 for the sterilizing value calculation in this experiment.

RESULTS AND DISCUSSION

Predicting Time Temperature Relationship and Sterilizing Value

A typical result of experiments using an acrylic resin slab as food substitute is shown in Fig. 5A. The x marks denote the temperatures collected with the sensing probe: 21 points with 10 second interval in this case. When all the preset number of temperature data were collected, the computer started iterating calculations to estimate the parameters τ, B_i and ξ. The iteration process continued until it gave increments, $\delta\tau$, δB_i and $\delta\xi$ of less than 0.001%. The progress of improvement in approximating parameters τ and B_i was monitored through the

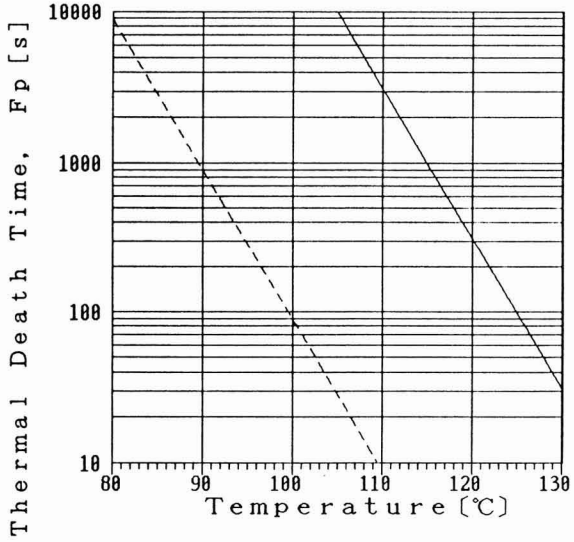

FIG. 4. A MODIFIED THERMAL DEATH TIME CURVE (BROKEN LINES) USED IN THIS EXPERIMENTAL WORK
Solid line indicates the Thermal Death Time for *C. botulinum* spores.

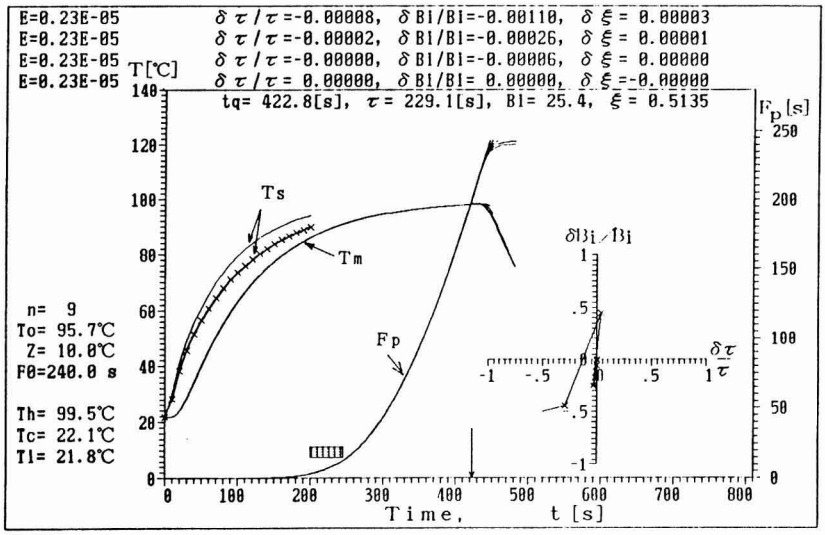

FIG. 5A. A COMPUTER DISPLAY, WHICH SHOWS THE PROGRESS OF COMPUTER CALCULATION, IN AN EXPERIMENT USING AN ACRYLIC SLAB WITH A SENSING PROBE LOCATED AT $\xi = 0.523$

The temperatures collected by the computer are denoted with x marks. The extent of the progress in improving parameter approximation is shown on the top. The variance in nonlinear multiple regression, denoted with E, is shown at the upper left. The increments for the parameters used in the iteration process are plotted on the right. After it completed estimating parameters, τ, B_i, and ξ, the computer predicted the temperature history at the geometric center of the sample, T_m, and corresponding lethal rate integral, F_p, in a trial and error search for the appropriate t_q value. The appropriate t_q finally obtained is indicated with an arrow at the bottom. The striped bar at the bottom denotes the time period used by the computer for calculation.

display as is shown on the right in Fig. 5A. Using these estimated parameters, the computer predicted the progress of the temperature rise and integrated the lethal rate L with respect to time from zero to an upper limit time. We defined t_H and t_L as the upper limit time with which the integral came up to 95% and 70% of the sterilizing value aimed at, respectively. We used $(t_H + t_L)/2$ as an initial value for t_q, the time to start cooling, whilst searching for an appropriate t_q using the Regula Falsi method.

The trial-and-error process to search for an appropriate t_q value was monitored through the computer display (Fig. 5A) on which the predicted temperature of heating and cooling phases, as well as the corresponding lethal rate integral, were plotted against the processing time.

When it successfully found an appropriate time t_q, the computer display, after it was reset, showed the final result of the prediction (Fig. 5B): the solid lines show the temperature history measured at the geometric center of the sample and the sterilizing values calculated from it. Both of the solid lines favorably

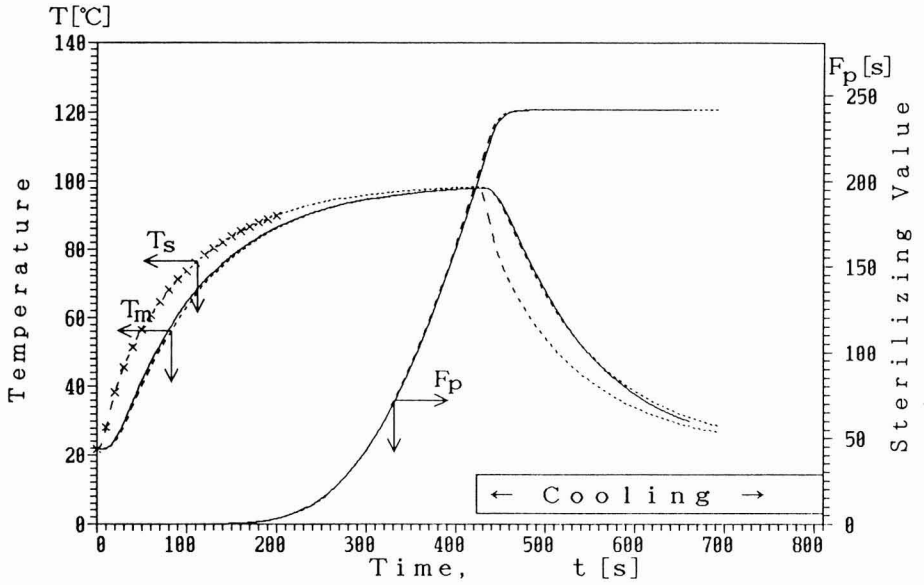

FIG. 5B. TIME/TEMPERATURE RELATIONSHIP AND LETHAL RATE INTEGRAL BOTH MEASURED AND PREDICTED USING AN ACRYLIC SLAB (HALF THICKNESS: 4.93 mm) WITH A SENSING PROBE LOCATED AT $\xi = 0.523$

Key x denotes the measured temperature with the probe which was collected by the computer and was used for estimation. The dotted lines denote predicted values, while the solid lines are measured values (T_s: temperature at the location of sensing probe, T_m: temperature at geometric center, F_p: lethal rate integral with respect to time from 0 to t).

agree with the dotted lines which denote the temperature and the sterilizing values predicted by this system. The period for cooling determined by the computer is shown at the bottom of Fig. 5B.

The time period used by the computer for calculation is denoted by a striped bar at the bottom in Fig. 5A. This computer time, used in predicting the parameters τ, B_i and ξ and searching for the appropriate time t_q, was around 50–100 seconds in total even though we used BASIC language, a computer language which is the slowest in processing. This computer time was short enough to perform an on-line control of the sterilization for thin foods of as little as 10 mm thickness.

Selected parameters used for prediction as well as those predicted are listed in Table 1. The thermal processing was successfully performed giving a sterilizing value which very closely agreed to predicted value. The predicted value for τ agreed to within 1% of the value calculated using the thermophysical properties for an acrylic resin. The probe location was also favorably predicted by this method.

TABLE 1.
PARAMETERS USED FOR PREDICTION AND THOSE PREDICTED

Sample				
Material		acrylic resin	acrylic resin	mannan jelly
Probe Location	ξ [-]	0.518 -0.529	0.156 -0.169	1.00
System Parameter Calc.	τ [s]	230 [1]	216 [1]	126 [2]
Parameters measured and used for prediction				
Half Thickness	D [mm]	4.93	4.78	4.15
Hot Water Temperature	T_h [°C]	99.5	99.1	99.2
Cooling Water Temperature	T_c [°C]	22.1	22.5	22.8
Parameters used for prediction				
Sterilizing Value Aimed at	F_p [s]	240	240	240
Reference Temperature	T_r [°C]	95.7	95.7	95.7
z Value	[°C]	10	10	10
Number of Data Collected	k [-]	21	21	21
Interval of Data Collection	Δt [s]	10	10	10
Number of Terms for Appro.	n [-]	9	9	9
Parameters predicted				
Time to Start Cooling	t_q [s]	423	426	296
System Parameter	τ [s]	229	223	116
Probe Location	ξ [-]	0.514	0.276	-
System Parameter	B_i [-]	25.4	23.5	10.5
Sterilizing Value Performed	F_p [s]	241	236	241

[1]: Calculated using physical properties for acrylic resin: C_p=1740 J/(kg K), ρ= 1270 kg/m^3, λ = 0.233 W/(m K).

[2]: Calculated using physical properties for water: C_p=4184 J/(kg K), ρ=999 kg/m^3, λ = 0.57 W/(m K).

A typical result of experiments using an acrylic slab which had a sensing probe closely located to the middle of slab (ξ = 0.16) is shown in Fig. 6. Selected parameters used for prediction as well as those predicted are listed in Table 1. A good agreement in F_p and τ is shown. However, the predicting of the location of the probe failed. The error in ξ was 11% of the half thickness.

This result suggests that it is hard to predict the location of the sensing probe precisely when the probe is located closely to the geometric center. This is because the temperature gradient shows little increase, and hence errors in location have little effect on predicting the temperature at the geometric center as well as the sterilizing value. This result also suggests that the location of the sensing probe does not need to be precisely predicted for the use of sterilizing value estimation when the probe is located near to the center of a sample.

Locating the Probe on the Container Outside Surface

A result of the experiment using mannan jelly is shown in Fig. 7. In this experiment the sensing probe was placed on the outside surface of a tinplate with which the mannan jelly was sandwiched.

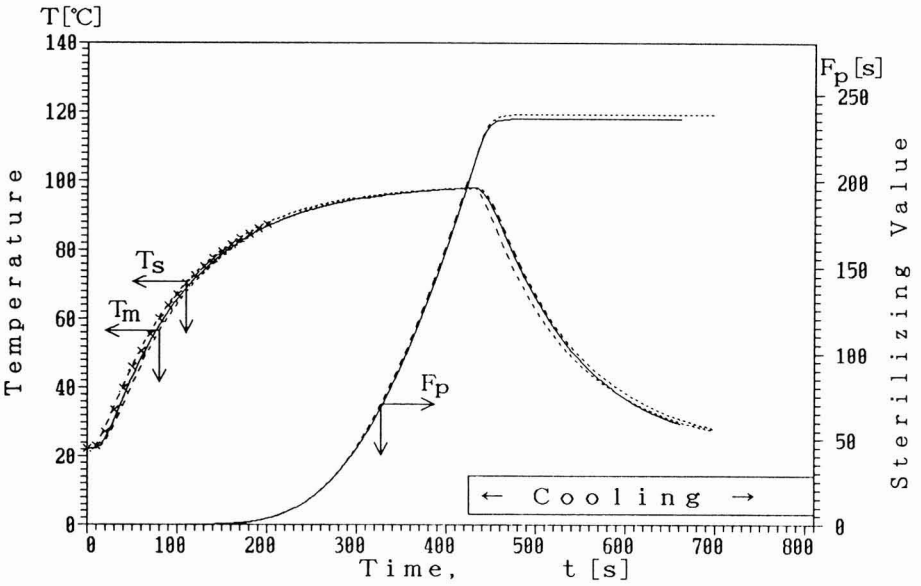

FIG. 6. TIME/TEMPERATURE RELATIONSHIP AND LETHAL RATE INTEGRAL BOTH MEASURED AND PREDICTED USING AN ACRYLIC SLAB (HALF THICKNESS: 4.78 mm) WITH A SENSING PROBE LOCATED AT $\xi = 0.162$

Key x denotes the measured temperature with the probe which was collected by the computer and was used for estimation. The dotted lines denote predicted values, while the solid lines are measured values (T_s: temperature at the location of sensing probe, T_m: temperature at geometric center, F_p: lethal rate integral with respect to time from 0 to t).

When the probe is located on the top surface of the sample slab, i.e., $\xi = 1$, Eq. (7) and (9) are simplified as follows:

$$\eta = \sum_{n=1}^{\infty} [D_n \exp(-P_n^2 \theta)] \qquad (7)'$$

where

$$D_n = A_n \cos(P_n) = \frac{2 B_i}{B_i^2 + B_i + P_n^2} \qquad (9)'$$

These equations were used in the experiment described in Fig. 7. Selected parameters used for and determined by the predictions are listed in Table 1. A good agreement in the sterilizing value as well as the parameter τ was found.

This successful result is particularly noteworthy because setting the probe at the surface of container ($\xi = 1$) has the following advantages:

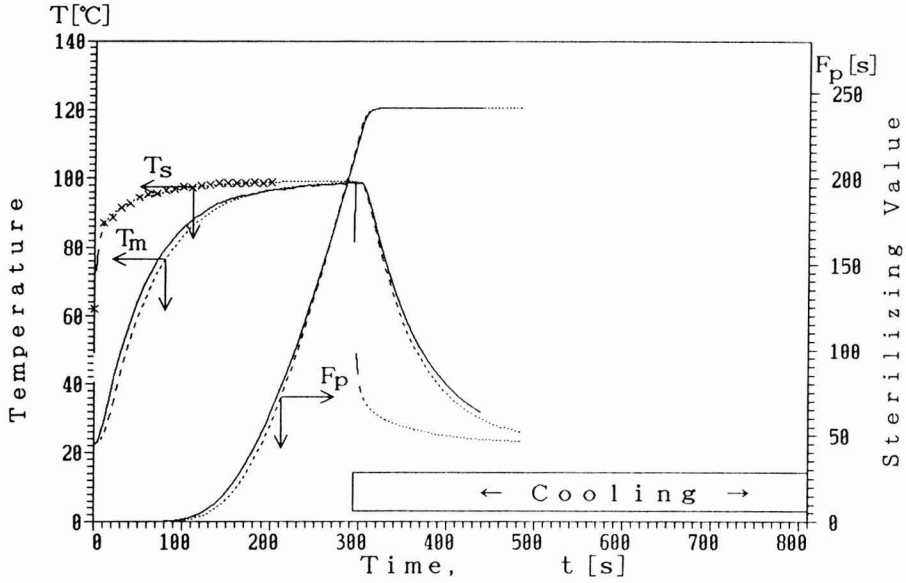

FIG. 7. TIME/TEMPERATURE RELATIONSHP AND LETHAL RATE INTEGRAL BOTH MEASURED AND PREDICTED USING A MANNAN JELLY SLAB (HALF THICKNESS: 4.15 mm) WITH A SENSING PROBE LOCATED AT OUTSIDE SURFACE

Key x denotes the measured temperature with the probe which was collected by the computer and was used for estimation. The dotted lines denote predicted values, while the solid lines are measured values (T_s: temperature at the location of sensing probe, T_m: temperature at geometric center, F_p: lethal rate integral with respect to time from 0 to t).

(1) Eq. (7)′ considerably saves computer time during iteration.

(2) Facilitate the setting up the sensing probe, i.e., no need to insert it into a container. All that is required is to solder or glue it on the outside surface.

It should be emphasized, however, that locating the probe on the container outside surface is not applicable to those cases in which B_i is very large or the materials from which the container was made have a fairly smaller thermal diffusivity than that of food.

Estimating B_i Values

The success of predicting B_i values, Biot number, in this experiment may be evaluated as follows. Substituting the conditions in this experiment into an empirical equation for the vertical plate natural convection heat transfer, we calculated a Nusselt number of 220 from values of the Grashof number and Rayleigh number 1.03×10^{10} and 1.8×10^{10} respectively (Holman 1981). With this Nusselt number, we can estimate the surface heat transfer coefficient as $h = 1.23 \times 10^3$ W/ (m^2K), which gives $B_i = 26.4$ for acrylic resin sample

(λ = 0.233 W/ (mK)) and B_i = 8.1 for mannan jelly sample (λ = 0.637 W/ (mK)). These estimated B_i values favorably agree with predicted B_i shown in Table 1.

There have been few reports on the surface heat transfer coefficient for packaged foods in commercial retorts. Naveh *et al.* (1983) used h = 1.0×10^3 W/ (m^2K) when they calculated the sterilization value in a cooling phase using water. This value closely approximates to the h value estimated above.

Effect of Errors in Sensing Temperature

As this method analyzes a series of temperature data collected in the early stages of temperature rise that plays a critical role in this system, we examined the effect of random errors, which might occur in measuring temperature, on the system performance. In the case of thermal processing which deals with temperature sensitive foods, temperatures are usually measured very precisely to monitor the duration of the high temperature period, because the sterilizing value is very sensitive to this period. Therefore we postulated 1% as the maximum error which a sensing probe may make.

A numerical experiment was conducted assuming that an acrylic resin slab (half thickness: 5.00 mm) with a sensing probe located at ξ = 0.40 was heated with hot water (99.6°C) and cooled with cooling water (20.0°C). The computer generated rises in temperature by an analytical solution to heat conduction equation where system parameters τ = 188.3 s and B_i = 8.8 were assumed. At the same time the computer imposed a random error (0.1–1.0%) onto the temperature. The computer collected 21 data sets using an interval of 10 s, and analyzed them using the 9 terms in series expansion of Eq. (7).

The results of the experiment, shown in Table 2, indicate that the present procedure was able to predict the true temperature history at the geometric center and hence the sterilization was performed with a sterilizing value very close to the value aimed at, even if the measured temperatures (dimensionless) had up to 1% error. Table 2 indicates also that as the error increases, a decrease in the predicted parameters τ and B_i are observed, while predicting the location of

TABLE 2.
EFFECT OF RANDOM ERRORS IN MEASURED TEMPERATURES ON SYSTEM PERFORMANCE : PARAMETERS ESTIMATED

Errors Imposed $100\,\Delta\eta/\eta$ [%]		0.0	0.1	0.2	0.3	0.5	1.0
Time to Start Cooling	t_q [s]	398	398	398	397	397	396
System Parameter	τ [s]	179	178	175	173	167	151
Probe Location	ξ [-]	0.403	0.402	0.402	0.402	0.403	0.402
System Parameter	B_i [-]	7.8	4.56	7.08	6.74	5.93	4.38
Sterilizing Value Performed	F_p [s]	242	241	240	240	238	236
Sterilizing Value Aimed at	F_p [s]	240	240	240	240	240	240

sensing probe ξ is not affected by the error. When the error was increased to over 1%, the computer iteration sometimes failed to converge. The chance of failure seemed to depend on the values of the initial estimation for the parameters: τ_o, B_{io} and ξ_o. This problem is left, as yet, unresolved.

Effect of Container Thickness

The effect of container thickness on the progress of sterilizing value at cooling phase was examined using numerical experiments and the result is shown in Fig. 8. In this examination, the half thickness of a container D varied from 2.0–6.0 mm, and the physical properties of water were used as those of the food material. Parameters as well as physical properties used are in the caption for Fig. 8.

Figure 8 shows that with thicker containers, larger rises of sterilizing value were observed during the cooling phase. This leads to a conclusion that when the container is thick, we have to predict the time to start the cooling process precisely; fortunately, however, when using thick containers we have enough

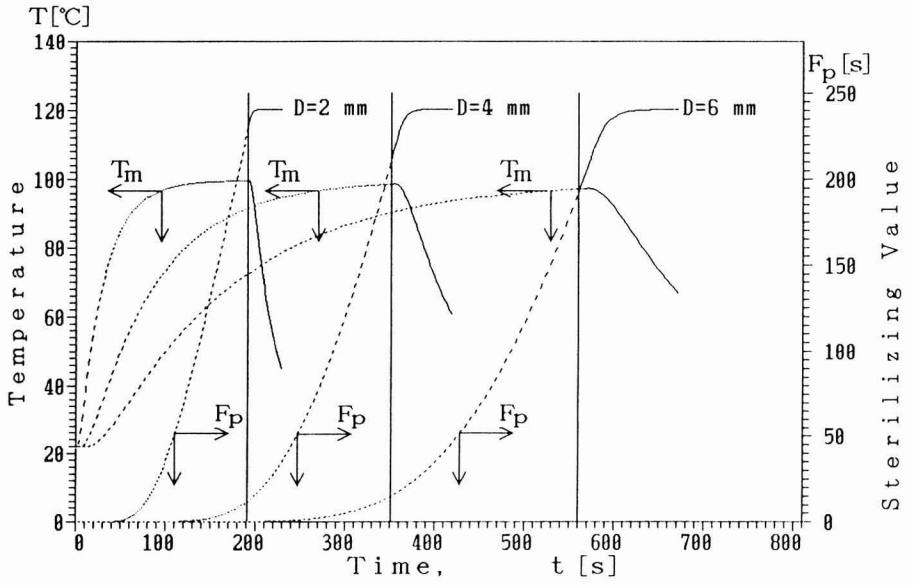

FIG. 8. TIME/TEMPERATURE RELATIONSHIP AND PROGRESS OF LETHAL RATE INTEGRAL AT THE GEOMETRICAL CENTER OF THE SLAB WITH VARIED THICKNESS PREDICTED IN NUMERICAL EXPERIMENTS

Solid lines describe temperature as well as the progress of lethal rate integral in cooling phase, whereas dotted lines show heating phase. The physical properties used were: density = 1000 kg/m^3, surface heat transfer coefficient = 500 W/(m^2K), heat capacity = 4184 J/(kg K), heat conductivity = 0.57 W/(m K). In this experiment 95.7°C was used as the reference temperature. The sterilizing value aimed at was 240.0 s. Hot water temperature = 99.6°C, cooling water temperature = 20.0°C.

time for the calculation. When the container is thin, on the other hand, we have little time for calculation; fortunately, a precise prediction for the time to start cooling is not necessary, as the rise of sterilizing value gained in the cooling phase is small; therefore we do not need to predict the temperature history in advance. All we need in this case is to measure the temperature at the center of container and watch the lethality integral which the computer calculates using the measured temperature. We can start cooling when the lethality integral comes up to a preset sterilizing value.

In this paper a potential application of a new procedure for achieving proper heat sterilization was examined in a one-dimensional approach, assuming a homogeneous initial temperature profile and a step-functional temperature change of the surrounding medium. The principal proposed in this paper may be extended to two- or three-dimensional problems, i.e., finite cylinders or polygon prisms, provided that they have an exact solution for heat conduction equation.

APPENDIX I

Partial derivatives of η with respect to τ, B_i and ξ are given as follows:

$$\eta = \sum_{n=1}^{\infty} [A_n \exp(-P_n^2\,\theta) \cos(P_n\,\xi)]$$

$$= \sum_{n=1}^{\infty} [A_n \exp(-P_n^2\,t/\tau) \cos(P_n\,\xi)]$$

$$\frac{\partial \eta}{\partial \tau} = \sum_{n=1}^{\infty} [(P_n^2\,\theta/\tau)\, A_n \exp(-P_n^2\,\theta) \cos(P_n\,\xi)]$$

$$\frac{\partial \eta}{\partial \xi} = \sum_{n=1}^{\infty} [-P_n\, A_n \exp(-P_n^2\,\theta) \sin(P_n\,\xi)]$$

$$\frac{\partial \eta}{\partial B_i} = \sum_{n=1}^{\infty} \{[\frac{1}{A_n}\,\frac{dA_n}{dB_i} - 2P_n\theta\frac{dP_n}{dB_i} - \xi\,\frac{\sin(P_n\,\xi)}{\cos(P_n\,\xi)}\,\frac{dP_n}{dB_i}]$$

$$\times\ A_n \exp(-P_n^2\theta) \cos(P_n\,\xi)\}$$

where

$$\frac{dP_n}{dB_i} = \frac{P_n}{B_i^2 + B_i + P_n^2}$$

and

$$\frac{1}{A_n}\,\frac{dA_n}{dB_i} = [2B_i + \frac{\{2P_n + (B_i^2 + P_n^2)(P_n / B_i)\}\,P_n}{B_i^2 + B_i + P_n^2}]\,[B_i^2 + P_n^2]^{-1}$$

$$-[2P_n\,B_i + P_n + \frac{(B_i^2 + B_i + 3P_n^2)\,P_n}{B_i^2 + B_i + P_n^2}]\,[B_i^2 + B_i + 3P_n^2]^{-1} P_n^{-1}.$$

APPENDIX II

The heat conduction equation and initial and boundary conditions during cooling may be given as follows using a modified time, $t^* = t - t_q$:

$$\rho C_p \frac{\partial T}{\partial t^*} = \lambda \frac{\partial^2 T}{\partial x^2} \tag{1a}$$

$$T(t^*, x)_{(t^*=0)} = T_q(x) \qquad t^*=0 \tag{2a}$$

$$\pm \lambda \frac{\partial T}{\partial x} = h(T_C - T) \qquad t^* > 0 \qquad x = \pm D \tag{3a}$$

where T_q is a temperature profile at the end of heating which is obtained through substituting $t = t_q$ into Eq. (7).

$$T_q(x) = T_h + (T_i - T_h) \sum_{n=1}^{\infty} [A_n \exp(-P_n^2 t_q/\tau) \cos (P_n x/D)]$$

When we define a dimensionless temperature in cooling phase as $\eta^\circ = (T - T_c)/(T_c - T_h)$, the dimensionless boundary condition during cooling is

$$\pm \frac{\partial \eta^\circ}{\partial \xi} = - B_i \eta^\circ \qquad \theta > 0, \qquad \xi = \pm 1 \tag{6a}$$

The solution is given by

$$\eta^\circ(t^*, x) = \sum_{n=1}^{\infty} [C_n \exp(-P_n^2 t^*/\tau) \cos (P_n \xi)] \tag{7a}$$

The coefficient C_n may be determined as follows so that the temperature profile at the end of heating phase is equal to the initial temperature profile in cooling phase.

The temperature profile at the end of heating phase may be expressed in the dimensionless form for cooling phase as follows:

$$\begin{aligned}
&\eta(t_q, x) \\
&= \{T_h + (T_i - T_h) \sum_{n=1}^{\infty} [A_n \exp(-P_n^2 t_q/\tau) \cos(P_n x/D)] - T_c\} / (T_c - T_h) \\
&= \{(T_i - T_h)/(T_c - T_h)\} \sum_{n=1}^{\infty} [A_n \exp(-P_n^2 t_q/\tau) \cos(P_n \xi)] - 1 \\
&= \sum_{n=1}^{\infty} [Q_n A_n \cos(P_n \xi)] - 1,
\end{aligned}$$

where Q_n is a constant given by

$$Q_n = \{(T_i-T_h)/(T_c-T_h)\} \exp(-P_n^2 t_q/\tau).$$

Since $\eta(t_q,x)$ is equal to $\eta^\circ(t^*,x)_{(t^*=0)}$, we have an equation:

$$\sum_{n=1}^{\infty}[Q_n A_n \cos(P_n \xi)]-1 = \sum_{n=1}^{\infty}[C_n \cos(P_n \xi)].$$

Multiplying both sides of above equations by $\cos(P_n \xi)$ followed by an integration with respect to ξ gives:

$$C_n = \frac{Q_n A_n \int_0^1 \cos^2(P_n \xi)d\xi - \int_0^1 \cos(P_n \xi)d\xi}{\int_0^1 \cos^2(P_n \xi)d\xi}$$

$$= Q_n A_n - \frac{2 \sin(P_n)}{P_n+\sin(P_n)\cos(P_n)} = A_n\{Q_n - 1\}$$

$$= A_n [\{(T_i-T_h)/(T_c-T_h)\} \exp(-P_n^2 t_q / \tau) - 1].$$

NOMENCLATURE

A_n	[−]	n-th Fourier coefficient (in heating phase) defined by Eq. (9)
B_i	[−]	Biot number = hD/λ
C_n	[−]	n-th Fourier coefficient (in cooling phase) defined by Eq. (20)
C_p	[J/(kg K)]	Specific heat capacity of food at constant pressure
D	[m]	half thickness of food
D_n	[−]	n-th Fourier coefficient (in heating phase) defined by Eq. (9)′
E_j	[−]	temperature difference between approximated and measured defined by Eq. (13)
F_p	[s]	sterilizing value
h	[W/(m²K)]	heat transfer coefficient at the surface between container and water
P_n	[−]	n-th eigenvalue in the solution of Fourier's heat conduction equation given by Eq. (8)
R_j	[−]	temperature difference between approximated and measure defined by Eq. (14)
t	[s]	time

t_e	[s]	time at the end of cooling
t_q	[s]	time to start cooling
T_c	[°C]	cooling water temperature
T_h	[°C]	heating water temperature
T_i	[°C]	initial temperature of food
T_m	[°C.]	temperature at geometric center of food
T_r	[°C.]	reference temperature of a thermal death time curve
T_s	[°C.]	temperature at the location of sensing probe
x	[m]	space variable
z	[°C.]	z value of a thermal death time curve
η	[−]	dimensionless temperature = $(T - T_h)/(Ti - T_h)$
θ	[−]	dimensionless time = t/τ
λ	[W/(m K)]	thermal conductivity of food
ξ	[−]	dimensionless space variable = x/D
ρ	[kg/m^3]	density of food
τ	[s]	system parameter = $\rho C_p D^2/\lambda$

REFERENCES

ANGSTROEM, A. J. 1864. Nachtrag zu dem Aufsatze; Neue Methode das Warmeleitungsvermogen der Korper zu bestimmen. Annalen d. Physik u. Chemie (Leipzig) *123,* 628.

BECK, J. V. 1970. Int. J. Heat Mass Transfer *13,* 703.

BECK, J. V. 1985. *Inverse Heat Conduction.* Wiley-Interscience Publication, New York.

BURGGRAF, O. R. Trans. ASME, Ser. C *86,* 373.

CEZAIRLIYAN, A. 1971. Recent development in thermophysical properties research. ASME *12.*

HAYAKAWA, K. 1983a. Basic principles for the mathematical evaluation of thermal process. In *Heat Sterilization of Food* (T. Motohiro and K. Hayakawa, eds.) pp. 1–10, Koseisha-Koseikaku, Tokyo.

HAYAKAWA, K. 1983b. Heat transfer in canned food during heat sterilization processes. In *Heat Sterilization of Food* (T. Motohiro and K. Hayakawa, eds.) pp. 109–120, Koseisha-Koseikaku, Tokyo.

HOLMAN, J. P. 1981. *Heat Transfer,* 5th ed., New York. McGraw-Hill, p. 275.

IIDA, Y. 1982. Proceedings of the International Heat Transfer Conference, vol. 4, pp. 527–432.

NAVEH, D., PFLUG, I. J. and KOPELMAN, I. J. 1983. Transient cooling of conduction heating products during sterilization: Sterilizing values. J. Food Process. Pres. *7,* 275–286.

STOLZ, G. 1960. Trans. ASME, Ser. C *82,* 20.

BOOK REVIEW

Foodborne Diseases. By Dean O. Cliver, Ed. 1991. Academic Press, 1250 Sixth Ave., San Diego, CA 92101. 395 pages.

One might expect that a book entitled *Foodborne Diseases* would discuss food microbiology, and indeed editor Dean O. Cliver's book of that same title does cover this topic. The unusual elements in this book are two additional chapters on diet and disease. At first I was puzzled at their inclusion, but after reading Cliver's introduction I understood his reasoning. He explains it best:

> The average person's life includes more than 75,000 meals—small wonder that most illnesses occur "after eating"! . . . The question of diseases linked to foods is seldom addressed together with foodborne intoxications and infections, yet chronic illnesses such as heart disease and cancer are being associated, perhaps incorrectly, with diet in a way that makes them seem to be the leading foodborne diseases in developed countries.

Cliver also mentions that this text was developed from and for the course taught to advanced undergraduates and graduate students in Bacteriology and Food Science by the faculty of the Food Research Institute. As such, it may be suitable for similar courses taught elsewhere.

The book is logically organized into five parts: Principles, Intoxications, Infections, Illnesses (Diet and Disease) and Prevention. Chapters address all the microbiological usual suspects, as well as a few others including natural toxicants, infrequent pathogens, viruses and parasites. IFT scientific status summaries address some of these other topics, but these chapters compliment rather than replace those summaries. The inclusion of these other topics make this book valuable for its well-rounded coverage of the subject of foodborne disease.

The first three chapters in the principles section set the stage well and place the chapters that follow in the proper context. Transmission of diseases (Chapter 1) and disease processes (Chapter 2) tell the how and the why of foodborne disease. Preservation, sanitation and microbiological specifications are covered in chapter 3.

Most sections on microbial pathogens are well written and several begin with an informative historical perspective that may not be found in other texts. The strong basic research focus of the Institute is evidenced by sections on classification and biochemical characteristics found in many of the chapters. Information on detection and isolation of pathogens may be a useful starting point for students beginning research in this area. Practical advice on prevention and control is contained in each chapter as well.

The sections on illnesses linked to foods (diet and disease) might have stood out from the rest of the book as incongruent, but instead they blend with the other chapters and make a complete picture. These topics may be with us for some time, and the chapters form a useful foundation for future research and debate.

Journal of Food Processing and Preservation **15** (1991) 157-158.

The final part of the book on prevention makes a strong ending to the story. Cliver's chapter on the roles of industry, government and the consumers are superb except for his slight of the USDA Cooperative Extension Service. In many cases the county extension office is the first place a consumer will turn with a question on food safety, and the importance of this support structure should not be ignored. E. M. Foster's final chapter weaves each thread from the text into a final summary.

The shortcomings of the text are few, but may be important to some.

Risk communication skills have been recognized as being an important part of the dialog on food safety. The perception of a risk might be different from the actual hazard it poses, and thus influence a particular decision or behavior. Some mention of risk communication, especially in the concluding segments might have been useful.

This book is not a lengthy, exhaustive reference text like Doyle's *Foodborne Bacterial Pathogens*. The chapters are usually short, and hit the important points. The reference lists that follow each chapter are also quite short, and only list key references on that topic.

Statements are often made in the body of a chapter without citing a specific reference. While references are listed at the end of each chapter, this lack of connection with the text may be an irritation to some. This style makes it difficult to follow up on a particularly interesting idea or statement for further discussion or study.

These points may detract slightly from the overall quality of the text, but this text still stands as a lucid, comprehensive and useful text for teaching an advanced undergraduate or graduate level class on foodborne disease.

DON SCHAFFNER, Ph.D.

$^{N}_{P}$ PUBLICATIONS IN FOOD SCIENCE AND NUTRITION

Journals

JOURNAL OF MUSCLE FOODS, N.G. Marriott and G.J. Flick, Jr.
JOURNAL OF SENSORY STUDIES, M.C. Gacula, Jr.
JOURNAL OF FOOD SERVICE SYSTEMS, O.P. Snyder, Jr.
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Books

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Page two should contain an abstract of not more than 150 words. This abstract should be intelligible by itself.

The main text should begin on page three and will ordinarily have the following arrangement:

Introduction: This should be brief and state the reason for the work in relation to the field. It should indicate what new contribution is made by the work described.

Materials and Methods: Enough information should be provided to allow other investigators to repeat the work. Avoid repeating the details of procedures which have already been published elsewhere.

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In some cases it might be desirable to combine results and discussion sections.

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DEWALD, B., DULANEY, J.T. and TOUSTER, O. 1974. Solubilization and polyacrylamide gel electrophoresis of membrane enzymes with detergents. In *Methods in Enzymology,* Vol. xxxii, (S. Fleischer and L. Packer, eds.)pp. 82–91, Academic Press, New York.

HASSON, E.P. and LATIES, G.G. 1976. Separation and characterization of potato lipid acylhydrolases. Plant Physiol. *57,* 142–147.

ZABORSKY, O. 1973. *Immobilized Enzymes,* pp. 28–46, CRC Press, Cleveland, Ohio.

Journal abbreviations should follow those used in *Chemical Abstracts.* Responsibility for the accuracy of citations rests entirely with the author(s). References to papers in press should indicate the name of the journal and should only be used for papers that have been accepted for publication. Submitted papers should be referred to by such terms as "unpublished observations" or "private communication." However, these last should be used only when absolutely necessary.

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Standard nomenclature as used in the biochemical literature should be followed. Avoid laboratory jargon. If abbreviations or trade names are used, define the material or compound the first time that it is mentioned.

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CONTENTS