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JOURNAL OF FOOD PROCESSING AND PRESERVATION

D.B. LUNI
EDITOR

FOOD & NUTRITION PRESS, INC.

OLUME 8, NUMBER 1 SEPTEMBER 198

JOURNAL OF FOOD PROCESSING AND PRESERVATION

All articles for publication and inquiries regarding publication should be sent to Dr. D. B. Lund, University of Wisconsin, Department of Food Science, 1605 Linden Drive, Madison, Wisconsin 53706 USA.

All subscriptions and inquiries regarding subscriptions should be sent to Food & Nutrition Press, Inc. P.O. Box 71, Westport, Connecticut 06881 USA.

One volume of four issues will be published annually. The price for Volume 8 is $70.00 which includes postage to U.S., Canada, and Mexico. Subscriptions to other countries are $82.00 per year via surface mail, and $90.00 per year via airmail.

Subscriptions for individuals for their own personal use are $50.00 for Volume 8 which includes postage to U.S., Canada, and Mexico. Personal subscriptions to other countries are $62.00 per year via surface mail, and $70.00 per year via airmail. Subscriptions for individuals should be sent direct to the publisher and marked for personal use.

The *Journal of Food Processing and Preservation* is listed in *Current Contents/Agriculture, Biology & Environmental Sciences (CC/AB).*

The *Journal of Food Processing and Preservation* (ISSN: 0145-8892) is published quarterly by Food & Nutrition Press, Inc.—Office of Publication is 155 Post Road East, Westport, Connecticut 06881 USA. Current issue is September 1984.

Second class postage paid at Westport, CT 06881.

POSTMASTER: Send address changes to Food & Nutrition Press, Inc., P.O. Box 71, Westport, CT 06881.

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Journal of
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VOLUME 8
NUMBER 1

Editor: D. B. LUND

FOOD & NUTRITION PRESS, INC.
WESTPORT, CONNECTICUT 06880 USA

ISSN 0145-8892

Printed in the United States of America

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LETTER FROM THE PUBLISHER

With this first issue of Volume 8 of the *Journal of Food Processing and Preservation,* we welcome Professor Daryl B. Lund as our new Editor. At the same time we wish to sincerely thank Professor Ted P. Labuza who served as Editor during publication of Volumes 1 through 7.

Prof. Labuza did an excellent job throughout his editorship and particularly so in guiding the Journal the first few critical years of its existence. We are forever in his debt and are pleased to report that Prof. Labuza will be involved by continuing to serve on the Editorial Advisory Boards of the *Journal of Food Processing & Preservation* and Food & Nutrition Press.

Prof. Lund is presently Professor and Chairman, Department of Food Science, University of Wisconsin, Madison, Wisconsin. He has served on the Editorial Advisory Board of the *Journal of Food Processing & Preservation* since inception and is thus very familiar with the Journal and its goals. He is the author of 100 technical papers and coeditor of four books in food processing.

Prof. Lund therefore will be able to draw on his excellent background and experience in food processing and preservation in his editorial duties, and we wish him all success in continuing the *Journal of Food Processing and Preservation* as the leading Journal in its field.

JOHN J. O'NEIL
Publisher

LETTER FROM THE EDITOR

Effective with Volume 8, Number 1 of the *Journal of Food Processing and Preservation*, I have assumed the job of Editor. Professor T. P. Labuza served admirably as its editor and developed the Journal into a respected quarterly publication. The papers are peer reviewed and the Journal title page is listed in Current Contents.

This letter is to invite you to submit original research papers or appropiate review articles for consideration. We are anticipating a total time for submission to publication of 6–9 months. Currently, there are no page charges for publication and reprints of your paper can be purchased.

I hope that as you prepare your research results for publication, you will consider the *Journal of Food Processing and Preservation*. Should you have any questions, please contact me. The address for submission of papers is:

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Daryl B. Lund

DEVELOPMENT OF ACIDIFICATION PROCESSING TECHNOLOGY TO IMPROVE COLOR AND REDUCE THERMOPHILIC SPOILAGE OF CANNED MUSHROOMS

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Accepted for Publication: April 10, 1984

ABSTRACT

An acid vacuum hydration process followed by canning in EDTA/brine was evaluated for improving color and reducing thermophilic spoilage of canned mushrooms. Blanching in citric, fumaric or phosphoric acids and vacuum hydration in 0.05 M *citric acid (pH 3.5) improved the color of the canned product significantly over untreated controls. EDTA (500 ppm) in the canning brine further improved color. Application of the citric acid vacuum hydration process to 227 kg quantities of fresh mushroom tissue in a commercial cannery resulted in no loss in canned product yield, significantly improved product color, greatly minimized thermophilic spoilage and did not significantly influence flavor and texture.*

INTRODUCTION

Methods to reduce shrinkage or increase canned product yield of mushrooms while improving color and reducing thermophilic spoilage are of economic importance to mushroom processors. Work conducted at The Pennsylvania State University led to the development of post-harvest treatment to optimize canned product yield of mushrooms (Beelman *et al.* 1973; McArdle and Curwen 1962; McArdle *et al.* 1974; Parrish *et al.* 1974; Beelman and McArdle 1975). These studies led to the recommendation of The Pennsylvania State University-soak, store, vacuum (PSU-SSV) process in which "raw" mushroom tissue was soaked in water for 20 min, stored at 2C for 18 h and then vacuum hydrated prior to blanching, slicing, filling, brining and thermal processing in order to optimize canned pound yields or reduce

[1]Published as Paper No. 6800 Journal Series of The Pennsylvania Agricultural Experiment Station, University Park, PA 16802

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shrinkage (McArdle *et al.* 1974). The color of mushrooms canned in this manner was better than that of mushrooms processed without the PSU-SSV process (Beelman and McArdle 1975). Another factor that required careful investigation was thermophilic spoilage of canned mushrooms. McArdle *et al.* (1978) attributed thermophilic spoilage to the germination and outgrowth of heat-resistant bacterial endospores which survive thermal processing. Thermophilic spoilage occurs when canned mushrooms are improperly cooled after thermal processing or when commercially sterile canned foods are subjected to high temperatures. Deis (1979) demonstrated that addition of citric acid and EDTA to the canning brine could effectively reduce thermophilic spoilage in canned mushrooms. Recent economic pressures on the mushroom processor have necessitated the development of methods that not only improved color and reduced the incidence of thermophilic spoilage but did not adversely affect the canned product yields. Further, such methods have had to be in the realm of acceptibility by regulatory agencies. Therefore, the objectives of this project were to investigate acidification as an aid in processing mushrooms and to evaluate the effects of EDTA in the canning brine on color, canned product yield and thermophilic spoilage of canned mushrooms.

EXPERIMENTAL

Raw Product

Cultivated mushrooms, *Agaricus bisporus* (Lange) Sing, were used in all experiments in this report. Mushrooms were selected from uniform lots of known strain from a commercial production facility and transported to the laboratory. Mushrooms with tightly closed veils and cap diameters ranging from 2.8-4.1 cm were selected and the stems were hand trimmed to within 1 cm of the cap. The trimmed button mushrooms were pooled, accurately weighed into 1 kg experimental samples (initial lot) and placed in numbered kraft bags. Four bagged samples were selected at random for each treatment.

Processing Conditions

All factors influencing canned product yield and quality, other than those under study, were controlled by standardizing processing conditions and comparing treatment results with those of a normal (control) process. Four repetitions of each treatment were performed. All samples were spray washed to remove surface dirt, soaked for 20 min and the soaked tissue was stored at 4C for 18 h prior to blanching to a center temperature of 80C by

immersion in a boiling water bath. The blanched buttons were cooled for 2 min in cold water, drained and filled into plain tin cans (211 × 212) using a fill weight of 120-130 g. A 20 grain NaCl tablet was added to each can. The cans were filled with boiling water and closed using a can closer equipped with an automatic headspacer (1 cm) and steam-flow closure. Thermal processing was conducted in a still retort at 121C for 22 min. The products were held for 2 weeks at ambient temperature prior to evaluation.

The treatments involved acid blanching in 0.05 *M* citric acid (pH 3.5) and soaking in 0.05 *M* citric acid. These treatments were carried out both either in the presence or absence of EDTA. The EDTA concentration was always kept at 500 ppm in the canning brine resulting in an equilibrium EDTA concentration of 200 ppm in the can.

Phosphoric acid, 50% sodium hydroxide and tetrasodium ethylenediamine tetraacetate (EDTA) were all purchased from Fisher Chemicals (Pittsburgh, PA) while citric acid was obtained from Miles Laboratories and fumaric acid from Alberta Gas Chemical Co. Acid solutions were buffered to the desired pH values by the addition of 50% NaOH.

Canned Product Evaluation

Canned product yield was determined by a procedure reported by Beelman and McArdle (1975). Color of the canned products was determined by using an Agtron reflectance meter (Beelman *et al.* 1973). Thermophilic spoilage was determined by incubating cans at 55C for 10 days as suggested by McArdle *et al.* (1978). Criteria for spoilage were swelled containers and containers having a vacuum depression of 3.0 in. or more below the average vacuum of controls and/or containers whose product pH was depressed 0.3 or more below the average pH of controls.

In-Cannery Trial

The optimum process developed in the pilot plant with 1 kg samples was tested in a commercial cannery using 227 kg (500lb) lots of mushrooms. The mushrooms were washed and soaked for 20 min prior to storage at 4C and 95% relative humidity for 24, 48 and 72 h. The washed, stored tissue was subjected to vacuum hydration in a Kroll-Reynolds vacuum hydrator with an average of 4 min residence time under water after breaking the vacuum. The hydrated tissue was blanched, sliced, filled, brined and processed in an FMC sterilmatic continuous rotary cooker to obtain an $F_0 = 12$. Processed cans were cooled and held for 2 weeks prior to analysis for color, yield and thermophilic spoilage. The products were also subjected to triangle sensory tests and hedonic evaluation by trained panelists. All results were analyzed by one way analysis of variance and the means were analyzed by Duncan's Multiple Range Test (SAS 1983).

RESULTS AND DISCUSSION

In preliminary experiments, mushrooms from five different growers were evaluated in order to obtain mushrooms from a source that consistently had high incidence of thermophilic spoilage. These evaluations were based on processing mushrooms from the different sources and incubating 20 cans of processed mushrooms from each source at 55C for 10 days and evaluating each can for thermophilic spoilage. The incubated cans were cooled to room temperature and can vacuum, brine pH and brine odor were recorded. Spoilage was expressed as a percentage of 20 cans and is shown in Table 1. Growing source 1 had the highest incidence of thermophilic spoilage at 83% followed by growing source 5. In all further experiments, mushrooms were obtained from growing source 1.

The effects of pH of blanching water on color, canned product yield and thermophilic spoilage were investigated using 0.05 *M* fumaric acid solutions buffered at various pH values; the results are shown in Table 2. As pH of the blanch water became more acidic, the equilibrium pH of the canned product was lowered. This effect was particularly noticeable below pH 4.0. Canned product yields were not significantly different from untreated controls and were not influenced by either acid blanching or EDTA. The color of the canned product was significantly improved in the presence of EDTA in the canning brine. Further improvement in color could be observed due to blanching of mushrooms at pH 4.5 or below. Based on this experiment a pH 3.5 was chosen as the optimum value for color, yield and equilibrium pH. The equilibrium pH of mushrooms blanched at pH 3.5 and canned in the presence of 500 ppm EDTA in the canning brine was only about 0.5 pH units less than the treated controls. Incorporation of EDTA in the canning brine has been reported to improve the color of canned garbanzo beans (Daoud *et al.* 1977). Similar light color due to presence of EDTA in the canning brine has

Table 1. Evaluation of different mushroom sources on potential thermophilic spoilage of canned mushrooms. Values are means of four replicates

Grower #	Thermophilic Spoilage (%)
1	83 A*
2	8 B
3	0 B
4	9 B
5	75 A

*Means followed by the same letter are not significantly different at p = 0.05.

Table 2. The influence of pH of blanch water and the incorporation of EDTA in canning brine on yield and color quality of canned mushrooms. Blanch water contained 0.05 *M* fumaric acid. Values shown are mean of 4 replications

pH of Blanch Water	Equilibrium pH of Canned Product		Canned Product Yield (%)		Agtron Color (% Reflectance)	
	No EDTA	EDTA	No EDTA	EDTA	No EDTA	EDTA
Control (water)	6.20	6.25	72.1A*	72.6A	24.4F	29.9CDEF
5.5	6.31	6.40	71.7A	72.4A	24.1F	30.9CDEF
5.0	6.11	6.29	71.3A	70.3A	25.6EF	32.4BCDE
4.5	5.98	6.03	72.3A	72.0A	27.3DEF	39.5AB
4.0	5.89	5.92	73.2A	70.2A	28.4DEF	40.0AB
3.5	5.67	5.71	72.6A	71.6A	33.6ABCD	38.8AB
3.0	5.45	5.56	71.6A	71.9A	34.5ABCD	39.6AB
2.5	5.12	5.07	71.8A	72.5A	36.1ABC	40.9A

*Means followed by the same letter are not significantly different at p = 0.05.

been reported with canned peas (Flora 1980). The choice of fumaric acid in the initial experiment was arbitrary but studies by Supran *et al.* (1966) and Dray and Powers (1966) had indicated this acid to be most effective among various acids tested with pimentos and tomatoes.

In another experiment, 0.05 *M* concentrations of fumaric, citric and phosphoric acids buffered to pH 3.5, with and without EDTA, were valuated for their effects upon selected quality characteristics of canned mushrooms (Table 3). Overall, the best results were obtained with citric acid and citric acid with EDTA. Color was improved, thermophilic spoilage was eliminated and yield was not affected. However, equilibrium pH values were low compared with other acid treatments. Fumaric acid appeared to be superior to citric acid, since the equilibrium pH of the canned product was closer to that of the control, i.e., the pH differential between the control with no EDTA (pH 6.15) and fumaric blanched with no EDTA (pH 5.68) was 0.47 pH units whereas a differential for comparable citric acid treated sample was 0.77 pH units. Fumaric acid, however, caused the mushroom buttons to assume a brown mottled appearance that was not observed in the citric acid blanched samples.

Acid blanching has some practical problems. It could conceivably lead to significant corrosion problems or may be difficult to accomplish in steam blanchers. This led us to consider an alternate approach. McArdle *et al.* (1974) have demonstrated that vacuum soaking of mushroom tissue improved canned product yields and color. Therefore, treatments used in acid

Table 3. Influence of blanching in 0.05 *M* fumaric, citric and phosphoric acid solutions buffered to pH 3.5 and addition of EDTA in the canning brine on color, yield, pH and thermophilic spoilage of canned mushrooms. Values represent means of four replicates

Acid	EDTA (ppm)	Agtron Color (% reflectance)	Canned product Yield (%)	Equilibrium pH	Thermophilic Spoilage (%)
Tap water	0	27.3 D*	68.9 AB	6.15 A	62.5 C
Tap water	500	36.8 BC	70.5 A	6.21 A	29.2 B
Fumaric	0	34.8 C	66.9 B	5.68 C	0 A
Fumaric	500	43.7 A	68.5 AB	5.73 C	0 A
Citric	0	42.8 A	69.3 AB	5.48 D	0 A
Citric	500	44.6 A	68.6 AB	5.49 D	0 A
Phosphoric	0	29.5 D	69.7 AB	5.94 B	4 A
Phosphoric	500	40.1 AB	69.8 AB	5.97 B	8.3 A

*A higher value denotes a lighter "desirable color
** Means followed by the same letter are not significantly different at p=0.05

blanching were evaluated using vacuum hydration (Table 4). Color was improved (lighter) in all instances where citric acid was used. Further, only treatments that contained citric acid eliminated the potential for thermophilic spoilage. When EDTA was added either in the canning brine or in the vacuum hydration step, spoilage rates of 25% and 33%, respectively, were observed. When vacuum hydration was omitted as a process step and EDTA was added in the blanch water or in the canning brine, thermophilic spoilage rates of 12.5 and 25% were observed, respectively. Vacuum hydration in citric acid followed by water blanching and addition of EDTA to the canning brine was considered to be the best treatment for color improvement and thermophilic spoilage control. This treatment resulted in minimal reduction in canned product yield. Also, corrosion of equipment (e.g., blanchers) would probably be minimized as well.

Vacuum hydration requires special equipment and such capital expenditure could prevent its use by "small" processors. Therefore, experiments were conducted to study the effects of atmospheric soaking for various periods in solutions of EDTA and citric acid, singly and in combinations. Results are shown in Table 5. These results demonstrated that atmospheric soaking of mushrooms for up to 120 min did not improve color nor did it prevent thermophilic spoilage in the canned product. Vacuum hydration to incorporate citric acid followed by the addition of 500 ppm EDTA in the can brine was effective in improving color and preventing thermophilic spoilage, yet it did not reduce canned product yield significantly. Citric acid in all experiments was buffered to pH 3.5 by the addition of 50% NaOH.

Table 4. Effects of vacuum hydration and blanching in water, 0.05 *M* citric acid (pH 3.5) and or 500 ppm EDTA solutions on yield, pH, color and thermophilic spoilage of canned mushrooms. Values are means of four replicates

Medium of Vacuum Hydration	Medium of Blanching	Medium of Canning	Canned Product Yield (%)	Equilibrium pH	Agtron Color (% Reflectance)	Thermo-philic Spoilage
Tap water	Tap water	Brine	73.7 A*	6.37 A	22.9 E	33 A
EDTA	Tap water	Brine	73.3 A	6.37 A	29.0 D	33 A
Citric acid	Tap water	Brine	68.8 B	5.05 D	43.3 B	0 C
Citric acid & EDTA	Tap water	Brine	68.6 B	5.08 CD	46.9 AB	0 C
Citric acid	Tap water	EDTA in Brine	70.3 B	5.14 C	47.0 A	0 C
Tap water	Tap water	EDTA in Brine	75.6 A	6.37 A	29.4 D	8.3 BC
__a	EDTA	Brine	67.9 B	6.30 A	26.3 DE	12.5 BC
__	Citric acid	Brine	67.8 B	5.85 B	36.3 C	0 C
__	Citric acid, EDTA	Brine	68.6 B	5.82 B	41.6 B	4.2 BC
__	Citric acid	EDTA in Brine	68.3 B	5.91 B	42.1 AB	0 C
__	Tap water	EDTA in Brine	68.4 B	6.33 A	32.5 D	25 AB

a__ = no vacuum hydration followed

*Means followed by the same letter are not significantly different at p = 0.05.

Table 5. Effect of atmospheric soaking, vacuum hydration and blanching with EDTA (500 ppm) and 0.05 *M* citric acid pH 3.5 on yield, pH, color and thermophilic spoilage of canned mushrooms. Values are means of four replicates

Treatment	Soak Time (min)	Soak Medium		EDTA in can	Canned Product Yield (%)	Canned Product pH	Agtron Color (% reflect.)	Thermo-philic Spoilage (%)
		EDTA	Citric acid					
I. Soak	20	+[a]	-[b]	-	73.7ABC*	6.29C	25.1CDE	100A
	20	-	+	-	75.2AB	6.32ABC	24.0DE	96A
	20	+	-	-	74.1AB	6.31ABC	24.1DE	100A
	20	-	+	+	72.7BCD	6.35A	27.9CD	75AB
	60	+	-	-	73.8ABC	6.32ABC	27.1CDE	100A
	60	-	+	-	74.4AB	6.31ABC	23.7DE	100A
	60	+	-	-	74.8AB	6.32AB	24.0DE	96A
	60	-	+	+	75.7A	6.34AB	27.9CD	83AB
	120	+	-	-	74.5AB	6.34AB	28.5C	83AB
	120	-	+	-	73.4ABC	6.33AB	23.4E	79AB
	120	-	-	-	73.1ABC	6.34AB	24.4CDE	89.5A
	120	-	+	+	72.9ABCD	6.34AB	27.6CD	100A
II. Vacuum hydration in citric acid				+	71.0CD	4.90D	43.6A	0C
III. Acid blanched in citric acid				+	70.3D	6.05C	36.8B	4C

[a] + = present in treatment

[b] - = absent in treatment

*Means followed by the same letter are not significantly different at p = 0.05.

If the citric acid solutions were unbuffered, the equilibrium pH values were higher but the thermophilic spoilage rate was high and color improvement was minimal (Table 6). Therefore, it became apparent that vacuum hydration in citric acid followed by blanching and addition of 500 ppm EDTA in the can brine were optimal.

The last aspect of this study dealt with verifying the process on a production scale in a mushroom cannery. The data shown in Table 7 are based on 227 kg (500 lbs) quantities of fresh mushroom tissue. Data from the in-cannery trial substantially verified the benefits of the citric acid vacuum-hydration treatment combined with EDTA in the canning brine. Color of treated, processed mushrooms was lighter than that of the untreated controls, canned product yields were not adversely affected, equilibrium brine pH was in the range of 5.87-5.94 and lastly thermophilic spoilage was drastically reduced. The in-cannery trial was conducted when the cannery was undergoing extensive reconstruction and several pieces of equipment such as the briners and headspace regulators were not operating in an optimal manner. Hence, initial temperature after sealing was 27C (80F) which in turn affected can vacuums. Additionally, leaky briner valves led to lower salt and EDTA concentrations than those used in pilot plant studies which enhanced thermophilic spoilage and influenced sensory data.

Results of the taste panel evaluation using triangle testing are shown in Table 8. Panelists were able to discriminate between control (process with no vacuum hydration) samples and samples hydrated in water or citric acid solutions. A statistically significant number of panelists preferred control and acid hydrated samples over water hydrated samples. Control samples were obtained from a local supermarket and were known to be processed in a cannery not using vacuum hydration. Both water and citric acid hydrated samples were produced in the in-cannery trial and contained lower amounts of salt. It is felt that the lack of salt in treated samples may have resulted in decreased preference for these samples.

Flavor and texture were evaluated by panelists using magnitude estimation techniques. Results are shown in Table 9. Flavor of water-hydrated mushrooms was ranked significantly below that of control, and control and acid-hydrated mushrooms had equally pleasing flavor. The texture scores of mushrooms processed by any of the three processes were not significantly different from one another.

The acceptability of color and the overall acceptability of mushrooms processed by three different processes and products imported from Taiwan and the People's Republic of China were evaluated by the panelists and the results are shown in Table 10. These results indicate that color improvement caused by vacuum hydration with citric acid and EDTA in the canning brine was significantly higher than in water hydrated mushrooms. Further, the color of the products processed with vacuum hydration in citric acid was as

Table 6. Effect of soaking in unbuffered acidified 0.05 *M* citrate solution, vacuum hydration, acid blanching and EDTA on yield, pH, color and thermophilic spoilage of canned mushrooms. Values are means of four replicates

TREATMENTS					Canned Product Yield (%)	Equilibrium pH	Agtron Color (% reflectance)	Thermo-philic Spoilage (%)
Tap Water Soak for 20 min	0.05M Citric Acid Soak for 20 min	Vacuum hydration	Acid Blanch	EDTA in Brine				
+[a]	-[b]	-	-	-	69.7CD*	6.25 A	24.5 D	37.4 A
+	-	-	-	+	70.1BCD	6.30 A	30.5 C	37.4 A
-	+	-	-	-	67.4D	6.24 A	24.5 D	33.2 AB
-	+	-	-	+	67.7D	6.31 A	29.5 C	29.1 AB
-	+	+	-	-	72.5ABC	6.21 A	25.5 D	33.0 AB
-	+	+	-	+	71.7ABC	6.23 A	34.1 B	8.3 BC
+	-	+	+	-	72.9AB	5.37 B	38.8 A	4.2 C
+	-	+	+	+	73.9A	5.36 B	39.5 A	0 C

[a] + = Treatment used

[b] - = Treatment not used

*Means followed by the same letter are not significantly different at p = 0.05.

Table 7. Data from in-cannery trial. Effect of scale up of process on color, canned product yield, equilibrium brine pH and thermophilic spoilage. Values are means of four replicates

Treatment[a]	Agtron Color (% reflectance)	Can Product Yield (%)	Equilibrium Brine pH	Thermophilic Spoilage (%)
No Storage, No Vacuum	34.8 B*	68.1 C	6.33 B	90 A
24h Storage, Water Vacuum	36.3 B	64.2 C	6.33 B	21 C
24h Storage, Citric Acid Vacuum	48.8 A	75.5 A	5.94 C	4.2 D
72h Storage, Water Vacuum	39.4 B	73.6 AB	6.38 A	56.9 B
72h Storage, Citric Acid Vacuum	42.3 A	73.4 AB	5.87 D	5.6 D

[a]Treatments: Storage was at 4°C, 95% RH post 20 min washing and soaking; vacuum refers to vacuum hydration; brine contained 500 ppm EDTA when citric acid was used in vacuum hydration.

*Means followed by the same letter are not significantly different at p = 0.05.

Table 8. Results of taste panel evaluation of mushrooms by triangle testing

Replicate	Pair tested	No. Panelists able to discriminate	Preference for		
			Control	Acid	Water
	Control X Acid				
1		7/11*	5 NS	2 NS	
2		5/11 NS	4 NS	1 NS	
3		7/11*	4 NS	2 NS	
Total		19/33*	13 NS	6 NS	
	Control X Water				
1		8/11*	8*		0
2		9/11*	9*		0
3		10/11*	10*		0
Total		27/33*	27*		0
	Water X Acid				
1		8/11*		8*	0
2		9/11*		9*	0
3		8/11*		8*	0
Total		25/11*		25*	0

* Significant at p = 0.05

NS Not significant

good as the color of the imported products. The overall acceptability of experimentally processed samples was lower than imported products and this may again be attributed to a lack of sufficient salt in experimentally processed products.

SUMMARY AND CONCLUSIONS

Based upon observations made during pilot plant and in cannery studies the following can be summarized.

(1) Color improvement of processed mushrooms can be achieved by either blanching the tissue in 0.05 *M* organic acid solutions or by vacuum hydration in such solutions prior to blanching.

(2) Best results were obtained using vacuum hydration in 0.05 *M* citric acid buffered to pH 3.5.

(3) The inclusion of 500 ppm EDTA in the canning brine in conjunction with vacuum hydration in 0.05 *M* citric acid led to improved color of the product and minimized thermophilic spoilage but did not affect canned product yield.

(4) A pilot plant process developed using 1 kg samples was acceptable under commercial cannery conditions and with various production volumes.

Table 9. Results of taste panel evaluation of canned mushrooms using magnitude estimate testing

Treatment	Mean estimates[1]
	Flavor
Control	0.92 A
Acid hydration	0.94 A
Water hydration	0.74 B
	Texture
Control	0.91 A
Acid hydration	0.90 A
Water hydration	0.79 A

[1]A higher value indicates better flavor/texture. Means having same letters are not significantly different. (p = 0.05).

Table 10. Sensory color and overall acceptibility of experimental and imported mushrooms (where 1 = dislike extremely and 10 = like extremely)

Treatment	Mean hedonic score[1]	
	Color	Overall
Control	6.30 B	6.21 BC
Water hydrated	5.91 B	5.45 C
Acid hydrated	7.12 A	5.94 C
Taiwan product	7.21 A	6.76 B
Peoples Republic of China product	7.10 A	7.76 A

[1]Means followed by the same letter are not significantly different (p = 0.05).

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ADHESION CHARACTERISTICS OF ISOLATED WAX SUBSTANCES FROM FRUIT CUTICLES RELATED TO OSMOTIC DEHYDRATION OF WHOLE FRUITS I. EFFECT OF TEMPERATURE AND PRELIMINARY TREATMENT

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Accepted for Publication: May 9, 1984

ABSTRACT

Adhesion behavior of cuticular wax isolated from cherries, plums and grapes, in a 70% sucrose solution, is the subject of discussion in the present work. Criterion is suggested for estimation of the effectiveness of preliminary treatment and temperature conditions in osmotic treatment of whole fruits. An apparatus is described for the determination of the contact angle θ_{slv} according to the method of the lying drop. Calculations are made of the energy characteristics, the energy of wetting W_{slv} and the adhesion work A_{slv}, of the three phase contact, isolated cuticular wax/70% sucrose solution/air, at temperatures from 20°C to 80°C.

INTRODUCTION

Osmotic dehydration of whole fruit is a process closely related to mass transfer through the fruit cuticle. Mass transfer through a solid/liquid phase depends on the nature of the phase contact or the so-called adhesion interaction of the two condensed phases. In osmotic dehydration of whole fruit, three phase contacts arise, namely, the superficial cuticular layer/hypertonic solution/air (S/L/V). As Holloway (1970) reports, wetting of the superficial cuticular layer of plant tissues is subjected to the same basic physiochemical dependences valid for each three phase contact. The adhesion work A_{slv} and the wetting energy W_{slv} are the energy characteristics of the S/L/V system. The contact angle θ_{slv} and the superficial tension of the liquid phase σ_{lv} are the experimentally measurable parameters. Over 75% of the hydrophobic plant tissues have contact angles with water within the range of 100° to 170° (Challen 1960; Hall 1966; Trongton and Hall 1967). The external cuticular layer of green plant parts, including those of fruits, is a

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complex mixture of alcohols, ketones, styrenes, triterpenoids, etc. (Holloway 1969a, 1969b; Martin 1970). In fruits, their percent ratio depends on the plant kind and the stage of ripeness.

Various methods and conditions for the application of osmotic dehydration to fruit canning are described in the technical literature (Ponting *et al.* 1966; Farkas and Lazar 1969; Jackson and Mohamed 1971). The hydrophobic character of the fruit cuticle, defined as $\theta_{slv} > 90°$, is the basic reason for the low rate of osmotic drying in whole fruits. Consequently, it is advisable to apply preliminary treatment to fruits which will reduce the value of θ_{slv}. We carried out investigations on the adhesion characteristics of fruit cuticular wax isolated from cherries (*Cerasus vulgaris,* CV), plums (*Prunum domestica,* PD) and grapes (*Vitis vinifera,* VV) while studying the processes connected with osmotic dehydration of whole fruits. The present report includes the results obtained from the investigations of preliminary treatments and temperature on the adhesion characteristics of isolated fruit cuticular wax.

MATERIALS AND METHODS

The experiments were conducted with ripe cherries, plums and grapes. The fruits were subjected to a hypertonic 70% sucrose solution. The effect of preliminary treatment on osmotically treated fruits was further investigated by determining adhesion parameters in the temperature range 20° to 80°C. The following preliminary treatments were applied:

(1) Blanching in boiling water (90°-96°C) for 30 s

(2) Blanching in a 2% aqueous solution of ethyl oleate for 30 s

(3) Blanching in a 2% aqueous solution of ethyl oleate and 3% K_2CO_3 for 3 s

(4) Cold treatment in ethyl oleate for 5 min.

The preliminary treatments were achieved in a water bath equipped with a thermostat. By this method, the whole fruit surface was uniformly treated. The ratio of fruit to solution was 1:7. The treated fruits were immediately washed in hot water and cooled to room temperature. After superficial drying, they were subjected to osmotic dehydration.

The process of osmotic dehydration was conducted at 50°C. The treated fruits were fixed on steel needles spaced about 1.5 cm apart prior to osmotic dehydration. As a result, the possibility of increase in concentration of the hypertonic solution was eliminated and a uniform course of osmosis was assured. The process of osmotic dehydration for cherries and grapes was 3 h; while for plums, it was 4 h. After the appropriate time, the samples were washed in hot water (70-80°C), tempered, air-dried at 20°C and weighed.

The percent decrease in fruit mass (M%), as a result of osmotic dehydration, was calculated from (Eq. 1):

$$M = \frac{M_o - M_x}{M_o} \times 100 \quad (1)$$

Where:

M_o = initial fruit mass in g, defined with a precision of ± 0.05 g

M_x = fruit mass after osmosis in g, defined with the precision of ± 0.05 g

The effect of the preliminary treatments of the fruits on the adhesion relations of the cuticular waxes in contact with the hypertonic solution (70% sucrose) was studied through measurement of the contact angle θ_{slv}. The three phase contact, where θ_{slv} is formed, was measured on an arbitrarily formed film of isolated cuticular waxes. These films were extracted from the fruits after the preliminary treatments. The arbitrarily formed film of wax substances was prepared in the following way. Five ml of a chloroform solution of cuticular wax was applied to a well-cleaned and horizontal optically integral glass plate. The concentration of the solution was m%, and the area of the glass plate was S cm^2. After evaporation of the solvent on the glass surface, a film of arbitrarily crystallized cuticular waxes was formed. The process was conducted at room temperature. The quantity of wax per unit area must be equal to that of the fruit cuticle. Therefore, the following condition was met (Eq. 2):

$$q = \frac{V \times m}{S \times 100} \quad (2)$$

Where:

q = quantity of cuticular wax per cm^2 of the surface of the fruit studied

The magnitude of q for each kind of fruit was determined by a weight measurement in the following way. A certain number of fruits were subjected to four consecutive extractions with chloroform at room temperature. The duration of each extraction was 10 s. According to Dudmon and Grucarevik (1962) and Martin (1970), under these conditions the superficial cuticular wax layer is extracted. This is the layer which participates in the formation of the three phase contact angle θ_{slv}. The chloroform solution thus obtained was distilled and dried to constant weight at a temperature not exceeding 40°C.

The contact angle θ_{slv} was determined using an equipment whose principle block circuit is indicated in Fig. 1. In all known methods reported so far,

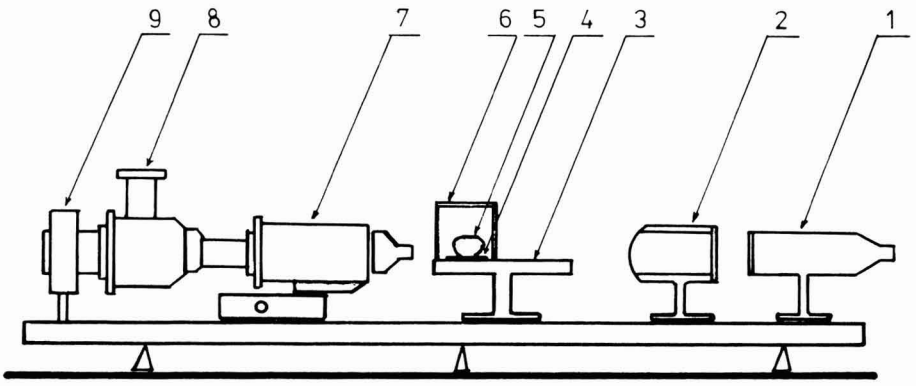

FIG. 1 BLOCK-CIRCUIT OF THE EQUIPMENT USED FOR THE DETERMINATION OF θ_{slv} BY THE METHOD OF THE LYING DROP; 1. LIGHT SOURCE, 2. CONDENSOR, 3. POLISHED METAL PLATE WITH BUILT-IN ELECTRIC HEATER, 4. GLASS PLATE COVERED WITH A FILM OF THE CUTICULAR WAX STUDIED, 5. DROP OF THE LIQUID PHASE INVESTIGATED, 6. GLASS CUVETTE, 7. HORIZONTAL MICROSCOPE, 8. EYEPIECE, 9. PHOTOCAMERA

points on the drop outline are used to estimate θ_{slv}. These points are removed from the phase contact. This restriction is due to experimental difficulties related to obtaining a clear image of the drop profile in the region of the phase contact. The precision of calculating θ_{slv}, in such an approach, is within the range of 1 to 10%. These calculations are also complicated due to the influence of gravity on the formation of the lying drop (Fig. 2). Its contribution to the general behavior of the drop grows as the distance from the phase contact increases. In the region of the phase contact, (when $h \rightarrow 0$, See Fig. 2), the outline of the lying drop coincides with the one it would have in the absence of a gravitational field.

Our observations indicated that this coincidence was precise enough when $h \leqslant 10^{-4}$ m for water phases and drops of $d \geqslant 5 \times 10^{-3}$ m. This means that if we viewed a clear image of the drop with $h = 0.1$ mm from the level of the three phase contact, direct estimation of θ_{slv} could be made. For this purpose, it was necessary to observe the region round the phase contact magnified at least 100 times.

We used a horizontal microscope in combination with a photocamera. The contact angle was determined from photos of the phase contact. Total magnification was 130X. So that we could simply determine the point of the three phase contact K_{slv} and the plane of the solid phase, we provided conditions under which we could observe a region of the drop in the phase contact together with the reflected image. Figure 3 indicates the image of the phase contact observed for different values of θ_{slv}. The tangents NK_{slv} and

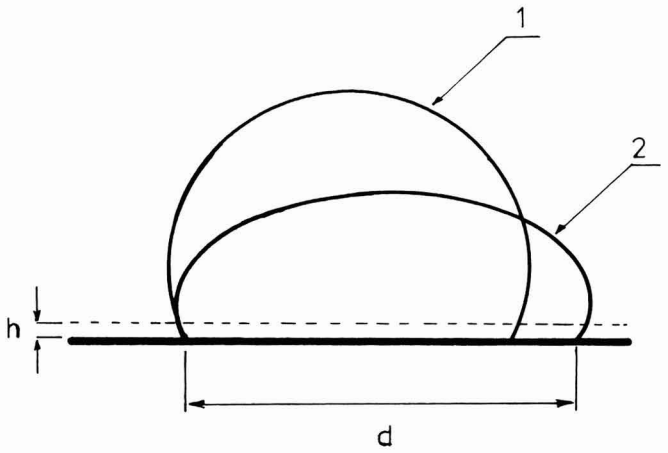

FIG. 2. DROP LYING ON HORIZONTAL SURFACE; 1. WITHOUT GRAVITATION, 2. WITH GRAVITATION

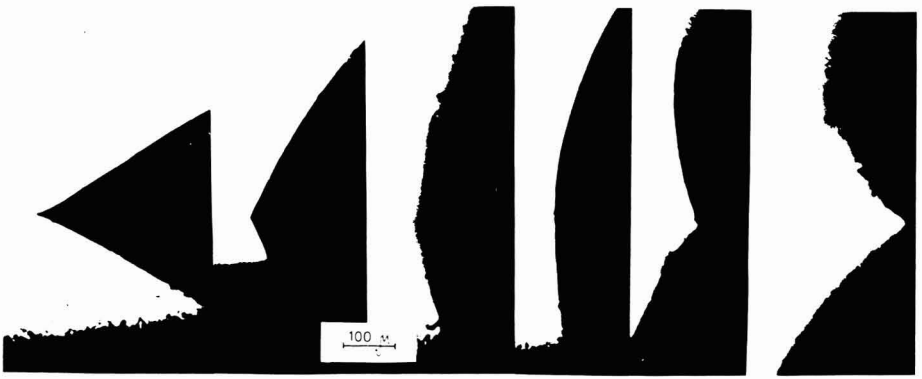

FIG. 3. PHOTOS OF THE THREE PHASE CONTACT S/L/V TAKEN BY THE METHOD OF THE LYING DROP FOR DIFFERENT VALUES OF $\theta_{s/v}$

MK_{slv} (Fig. 4) define angle α. As a result, for a contact angle larger than 90°, $\theta_{slv} = (180 - 1/2\,\alpha)$, while for a contact angle smaller than 90°, $\theta_{slv} = 1/2\,\alpha$.

For the purposes of the present study, it was also necessary to know the surface tension of the hypertonic solution, σ_{LV}. The surface tension is used in an expression for estimation of the wetting energy, W_{slv}. Data as reported by Dobzyckiego (1973) were used for determination of σ_{LV} of the sugar solutions; for the determination of the surface tension of water, data reported by Hodgman (1959) were utilized. In order to establish the values of $\sigma_{LV}^{70\%}$ temperature intervals of 20° to 80°C, the following conditions were imposed.

1. For a sugar solution at a concentration of 60 to 70% (Eq. 3):

$$\left(\frac{d\,\sigma_{LV}}{dc}\right)_t = \text{const.} \tag{3}$$

2. Within the temperature range of 20 to 80°C for pure water and the 70% sugar solution, the following equation is valid (Eq. 4):

$$\left(\frac{d\,\sigma_{LV}}{dt}\right)0\% = \left(\frac{d\,\sigma_{LV}}{dt}\right)70\% \tag{4}$$

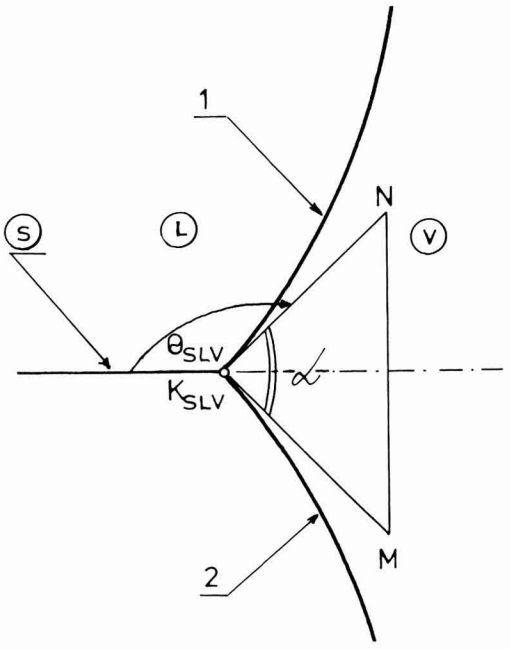

FIG. 4. DRAWING OF THE IMAGE OF THE THREE PHASE CONTACT OBSERVED WITH THE EQUIPMENT GIVEN IN FIG. 1.; 1. OUTLINE OF THE DROP, 2. REFLECTED IMAGE

A polished metal plate with an electric heating component provided heat to the sample under investigation (Fig. 1, position 3). A drop of temperature-resistant silicone oil was set between the metal plate and the glass plate on which the arbitrarily formed film of cuticular wax substances was deposited. Thus, a high exchange of heat flow between the heating plate 3 and the object of investigation 6 was secured. The film of silicon oil between the polished metal plate and the glass plate ensured a good reflected image of the drop (Fig. 3).

RESULTS AND DISCUSSION

Table 1 gives the θ_{slv} values for solutions of various sucrose concentrations and cuticular wax substances of fruits at 20°C which were not subjected to preliminary treatments.

All contact angles were larger than 90° indicating that surfaces of the investigated fruits were hydrophobic. Therefore, in order to raise the osmotic dehydration efficiency, it was necessary to perform a preliminary hydrophilization of the fruits. The highest hydrophobic character was manifested by the wax substances derived from grapes cuticles (130-150°) while those of cherries and plums gave θ_{slv} values of about 100°. These results satisfactorily correlated with the technical data reported on the hydrophobic character of plant cuticle (Holloway 1969b).

The discontinuous nature of the external cuticular wax layer affected the adhesive behavior of the plant tissue. The magnitude of the contact angle

Table 1. Contact angle of the isolated cuticular wax/sucrose solution/air phase contact at 20°C.

FRUIT	CONTACT ANGLE (θ_{slv}) FOR SUCROSE CONCENTRATION OF (%)			
	0	40	60	70
Cherries	100.5	94*	100.0	101.0
Grapes	131.6	128.1	122.3	150.3
Plums	95.0	106.5	101.0	131.0

*The value was reported at 24°C

was substantially dependent on the structure and nature of the wax coating. Holloway (1969b, 1970), when studying the contact angle of water obtained from the green parts of over 40 plant varieties, established that only 60% of the wetting energy was determined by the chemical composition of the wax substances. Holloway carried out the experiments with smooth films formed by constraint of extracted cuticular wax. Thus, the effect of the structural factor of the wax surface was eliminated in the adhesion relations.

In our opinion, the method of arbitrarily formed film, adopted in the present work, provided a possibility for partial recovery of the structural characteristics of the cuticular surface. This fact was confirmed by the higher values of θ_{slv}, which we obtained, compared to those of Holloway (Table 1).

The equations of Young-Dupre (Good and Stromberg 1979a; Duncan and Staw 1980) relate the energy characteristics of the three phase contact A_{slv} and W_{slv} to the measurable parameters of a real system, the contact angle θ_{slv} and the surface tension of the liquid phase σ_{lv} (Eq. 5).

$$\sigma^{\circ}_{sv} = \sigma_{sl} + \sigma_{lv} \cos\theta + \pi^{\circ} \quad (5)$$

where:

σ_{lv}, σ_{sl}, σ°_{sv} - equilibrium surface tensions for the corresponding two phase systems: liquid/gas (l/v), solid/liquid (s/l) and solid/gas (s/v)

π° - surface pressure of the vapors of the liquid phase under pressure P°, adsorbed onto the solid phase film

In low-energy condensed phases, π° is negligible (Zimon 1974; Pugachevich *et al.* 1982). Therefore, Eq. 5 could be written as follows (Eq. 6):

$$\sigma_{sv} - \sigma_{sl} = \sigma_{lv} \cos\theta \quad (6)$$

The difference $(\sigma_{SV} - \sigma_{SL})$ is the wetting energy or the energy released in the presence of the liquid/solid phase contact.

Figure 5(a, b, c) shows the relationship $W_{slv} = f(t^{\circ})$ for the fruit cuticular waxes in contact with a 70% sucrose solution studied after various preliminary treatments, and the curves 1, 2, 3, 4 and 5 demonstrate the effect of each preliminary treatment on fruits. The continuous line gives the terminal value of the wetting energy when $\theta = 0$, i.e., $W^{max}_{slv} = f(t^{\circ})$. At 20°C, the three fruit cuticular waxes without any preliminary treatments showed negative wetting energies and formed the following relationship (Eq. 7):

$$W_{VV} : W_{PD} : W_{CV} = -75 : -53 : -8 \quad (7)$$

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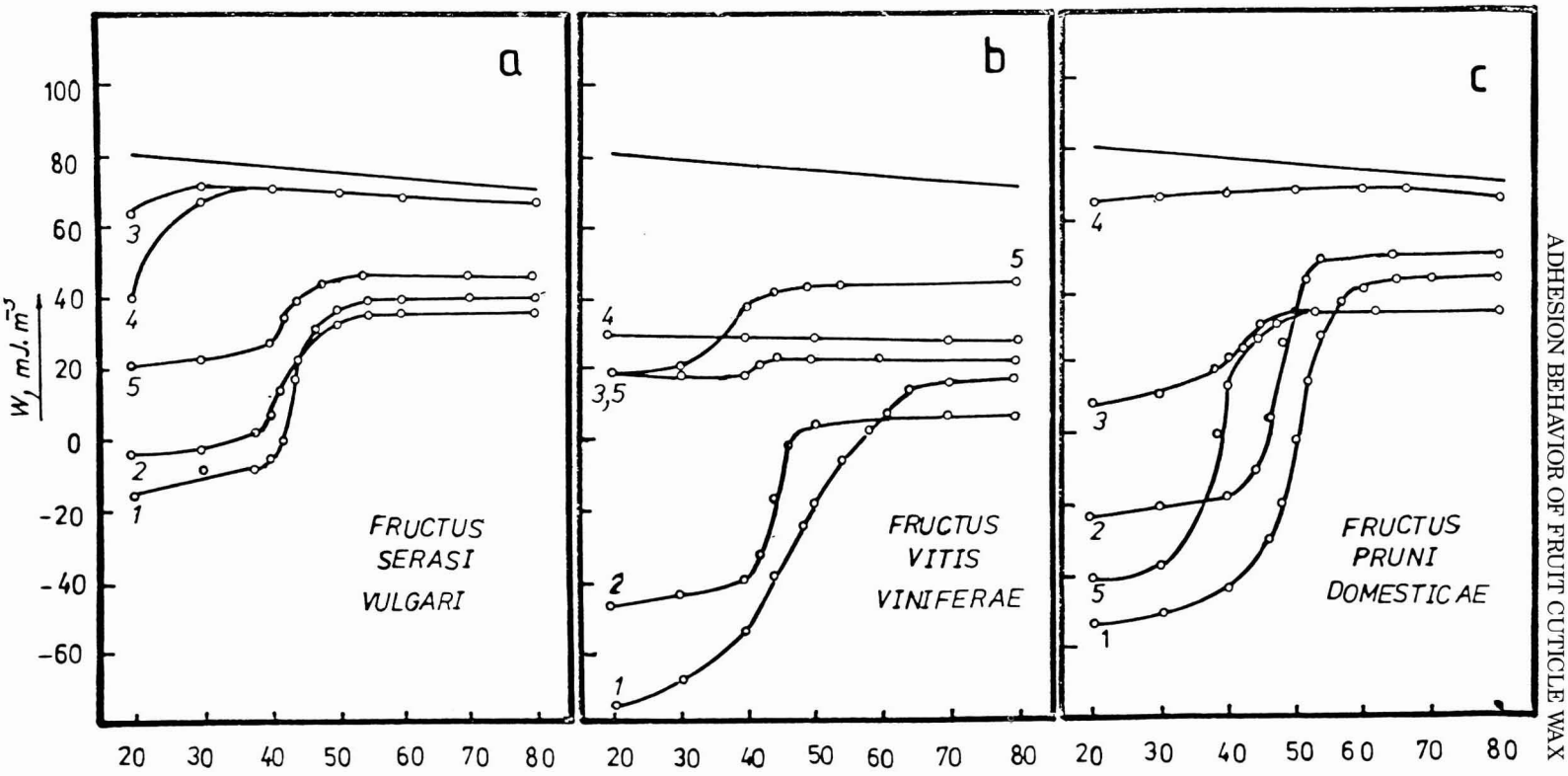

FIG. 5. EFFECT OF VARIOUS KINDS OF PRELIMINARY TREATMENT ON THE WETTING ENERGY OF ISOLATED CUTICULAR WAXES IN THE THREE PHASE CONTACT OF FRUIT CUTICULAR WAX/70% SUCROSE SOLUTION/AIR, AT VARIOUS TEMPERATURES; 1. REFERENCE, 2. BLANCHING IN BOILING WATER FOR 30 s, 3. BLANCHING IN 2% AQUEOUS SOLUTION OF ETHYL OLEATE FOR 30 s, 4. BLANCHING IN 2% AQUEOUS SOLUTION OF ETHYL OLEATE AND 3% K_2CO_3 FOR 30 s, 5. COLD TREATMENT IN ETHYL OLEATE FOR 30 s

An increase in the temperature led to changes in the adhesion properties of the fruit cuticular waxes. Up to 50°C, without any preliminary treatments, the fruits maintained the same order of wetting energy, however, important value changes were reported (Eq. 8):

$$W_{VV} : W_{PD} : W_{CV} = -25 : 2.5 : 38 \quad (8)$$

Above 50°C, changes in the order of wetting energies were also observed (Eq. 9):

$$W_{VV} : W_{CV} : W_{PD} = 7.8 : 40 : 41.5 \quad (9)$$

Some of the graphs of the relationship $[W_{slv} = f(t°)]$ are a matter of interest where we could write $\frac{dW_{slv}}{dt} \rightarrow$ const. An example would be the plots obtained for the cuticular wax derived from grapes after blanching in a boiling emulsion of 2% ethyloleate and 3% K_2CO_3 for 30 s (Fig. 5b, curve 4). If the diffusion constant of water, under these conditions, is also temperature independent, or could be assumed as such, then the osmotic dehydration of the grapes, thus treated, could be carried out at room temperature. Slight temperature dependences are indicated by curve 3 in Fig. 5b, curve 4 in Fig. 5c, curves 3 and 4 in Fig. 5a, at temperatures $t >$ 30°C.

The shape of the curves shown in Fig. 5a illustrates that the temperature changes in the adhesion properties of cuticular waxes are negligible from 20 to 35°C and from 60 to 80°C. However, in the majority of cases, within the limits of 40-55°C important changes occurred in the behavior of the heterogeneous s/l/v system. This fact was used to determine the most appropriate temperature conditions in the interval between 50 and 55°C for conducting the osmotic dehydration of the fruits. However, the effect of the preliminary treatments on the relationship $W_{slv} = f(t°)$ should also be considered. In conclusion, it is evident that both factors, temperature and preliminary treatment could be successfully combined to determine the most appropriate technological and energy conditions for osmotic dehydration adjusted to the specific features of the fruits.

The effect of the various kinds of treatment on the adhesion behavior of the s/l/v system could be expressed by the dimensionless factor $F_{x,y...}$, as determined by Eq. 10. In our experimental results, the factor F reflected the effect of two parameters, temperature and preliminary treatment.

$$F_{x,y} = 1/2 \frac{W_{slv}^{max} + W_{slv}}{W_{slv}^{max}} \quad (10)$$

Where:

W_{slv}^{max} is the maximum value of the wetting energy in a given three phase contact when $\theta = 0$.

Taking into account Eq. 6, the factor $F_{x,y}$ and W_{slv} undergo changes depending on the value of θ_{slv}, as follows (Eq. 11):

$$0° \leqslant 0_{slv} \leqslant 90° \qquad 1 \geqslant F_{x,y} \geqslant 0.5 \qquad W_{slv}^{max} \geqslant W_{slv} \geqslant 0 \qquad (11)$$

$$90° \leqslant \theta_{slv} \leqslant 180° \qquad 0.5 \geqslant F_{x,y} \geqslant 0 \qquad 0 \geqslant W_{slv} \geqslant W_{slv}^{min}$$

Where:

W_{slv}^{min} is the minimum wetting energy obtained in the s/l/v phase contact, when $\theta_{slv} = 180°$.

It is obvious that the optimum conditions for osmotic dehydration would be $F_{x,y} = 1$. When $F_{x,y}$ was less than 0.5, the wetting energy in such a system has a negative value and the wetting process is concomitant with heat adsorption.

The level of the plateau in the curves of the relationship $W_{slv} = f(t°)$ at $t° > 60°C$ indicates the efficiency of the preliminary treatment applied to the fruits on one hand, and the ineffectiveness of raising the temperature above 55°C on the other hand. As for the fruit varieties studied, the best results were obtained under the following conditions.

Cherries. Blanching in a boiling emulsion of 2% ethyl oleate with and without 3% K_2CO_3 for 30 s. At 30°C, the wetting energy was raised by 80 mJm^{-2} compared to that in fruits not subjected to a preliminary treatment. Osmotic dehydration using a 70% sucrose solution could successfully be conducted at $t \geqslant 30°C$.

Grapes. Cold treatment in ethyl oleate for 5 minutes. At 50°C, the wetting energy using a 70% hypertonic sucrose solution was 44 mJm^{-2}, i.e. it was increased by 65 mJm^{-2} compared to that in fruits without any treatment. Under the conditions described, the partial osmotic dehydration should be carried out at $t \geqslant 45°C$.

Plums. Blanching in boiling emulsion of 2% ethyl oleate and 3% K_2CO_3 for 30 s. The wetting energy using a 70% sucrose solution at $t = 50°C$ was 69.3 mJm^{-2} and increased by 70 mJm^{-2} compared to the reference sample. At 20°C, this change was approximately 120 mJm^{-2}. Under these conditions, the partial osmotic dehydration could also be conducted at room temperature.

These results illustrate that the temperature conditions adopted and the method of preliminary treatment of cherries and plums could be considered

close enough to the optimum as the factor $F_{x,y} > 0.90$. Concerning grapes, the temperature conditions and the preliminary treatment applied, when conducting the partial osmotic dehydration, were considerably below the optimum. In this case, $F_{x,y} < 0.80$ and further studies were undertaken to find out the optimum values where $F_{x,y} \geq 0.9$.

Table 2 summarizes the values of $F_{x,y}$ obtained with the fruits at 50°C correlated to the preliminary treatment applied. For comparison, Table 2 also includes the values of the adhesion work $A_{slv}^{50°C}$ and the efficiency of the partial osmotic dehydration applied expressed in terms of the reduction of fruit mass M in %. It is evident that the parameters $F_{x,y}$ and $A_{slv}^{50°C}$ correlated well with M%. Here is an opportunity to make a preliminary determination of the efficiency of the given precursory treatment on the fruits and to establish the proper temperature conditions without the necessity of carrying out the osmotic dehydration itself. Therefore, based on the results obtained, we recommend the application of the change in the factor $F_{x,y}$ as an indicative quantitative expression for the determination of the efficiency of various factors affecting the kinetics and the character of the partial osmotic dehydration of whole fruits.

CONCLUSIONS

Adhesion behavior of fruit cuticular waxes isolated from cherries, plums and grapes in contact with a 70% sucrose solution were related to the kind of preliminary treatments. The results revealed that:

(1) The cuticular wax coatings of the fruits studied were hydrophobic. The hydrophobic character was more pronounced in grapes and the contact angle of wetting in an arbitrarily formed film of isolated cuticular waxes was ~ 150°. In plums and cherries, the angle was ~ 134° and ~ 101°, respectively. Therefore, it was necessary to improve the process of osmotic dehydration of these fruits and adequate conditions should be sought to reduce the hydrophobic character so that the contact angle is reduced to less than 90° and the wetting energy $W_{slv} > 1/2\ W_{slv}^{max}$.

(2) Factor $F_{x,y}$ is a convenient criterion for the estimation of the efficiency of a definite treatment on the fruits for reducing their hydrophobic character.

(3) Within the framework of our experiments, it was found that the most appropriate temperature conditions for osmotic dehydration of whole fruits of cherries, plums and grapes ranged from 45° to 55°C when the following preliminary treatments were performed; cherries and plums were placed in a boiling emulsion of 2% ethyl oleate for 30 s which caused factor $F_{x,y}$ to rise to 0.965 and 0.957, respectively, and grapes were treated in ethyl oleate at

Table 2. Adhesion work A_{slv}, factor $F_{x,y}$ reporting the readiness of the fruit cuticular surface for osmotic dehydration and the reduction of the fruit mass M% following osmotic dehydration of the whole fruit using a 70% sucrose solution at 50°C and various kinds of preliminary treatment of the fruits. The data reported on $F_{x,y}$ and A_{slv} were obtained on isolated cuticular fruit waxes

PRELIMINARY TREATMENT OF FRUITS	F%			M%*			A, mJm^{-2}		
	CV	VV	PD	CV	VV	PD	CV	VV	PD
Reference sample without preliminary treatments	74.3	37.9	48.3	15.0	14.4	9.1	112.6	57.5	73.3
Blanching in boiling water for 30 s	72.7	52.6	75.0	30.1	32.3	19.2	110.0	79.8	113.8
Blanching in boiling emulsion of 2% ethyl oleate for 30 s	96.5	65.5	72.7	49.6	48.3	31.4	146.4	100.0	110.3
Blanching in boiling emulsion of 2% ethyl oleate and 3% K_2CO_3 for 30 s	96.5	68.0	95.7	53.4	51.5	36.5	146.4	102.0	145.0
Treatment in natural ethyl oleate at 20°C for 5 min	80.8	79.0	74.3	44.2	45.6	32.3	122.6	120.0	112.0

*The time of osmotic treatment of cherries and grapes was 3 h and of plums 4 h.

20°C for 5 min with $F_{x,y} = 0.79$. The temperature conditions thus determined for plums and cherries could be considered close enough to the optimum ($F_{x,y} > 0.9$) for their continued application, while for grapes a possibility of a more efficient treatment should be investigated. The application of temperature conditions higher than 60°C is inefficient, in terms of energy, as the wetting energy slightly increases.

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THE EFFECT OF PROCESSING ON THE TRYPSIN INHIBITOR, HEMAGGLUTININ, TANNIC ACID AND PHYTIC ACID CONTENTS OF SEEDS OF TEN COWPEA VARIETIES

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Accepted for Publication: May 7, 1984

ABSTRACT

Trypsin inhibitor (TI) and hemagglutinin activities, tannic acid and phytic acid contents were determined in ten varieties of cowpea. The different varieties were subjected to four processes; cooking, soaking, autoclaving and germination. TI and hemagglutinin activities were completely eliminated by cooking and autoclaving, while tannic acid and phytic acid contents were only partly affected. Cooking reduced tannic acid contents by 31.0-47.3%. Soaking for three days decreased TI activity by a mean of 31.2%, hemagglutinin activity by 19.0%, tannic acid by 13.4% and phytic acid by 24.4%. Maximum germination effects on TI and hemagglutinin activities were obtained in 'Westbreed' and 'Kano 1696' where percentage losses amounted to 57.2 and 57.6, respectively. The phytic acid contents of all varieties were greatly reduced by germination.

INTRODUCTION

Legumes are generally known to contain various natural constituents which affect their nutritional quality. Some of these components are proteins which inhibit specific enzyme activities, as for example the inhibitors of proteases and amylases (Whitaker and Feeney 1972). Others are the hemagglutinins, saponins, tannins and antivitamins (Liener 1969). Cuthbertson (1968) has also reported the presence of phytic acid which interfers with mineral element absorption and utilization, and reacts with proteins to form complex products which have inhibitory effects on peptic digestion (Barre 1956).

Although heat treatment will effectively eliminate most of these undesirable substances, the application of other processes such as soaking, steeping, decorticating and germination have also been effective in reducing them. More importantly, there is less functional as well as nutritional damage to the protein which results from excessive heat treatment.

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This paper is an account of our investigation on the effects of soaking, germination, cooking and autoclaving on the trypsin inhibitor, hemagglutinin, tannic acid and phytic acid contents of ten varieties of cowpeas (*Vigna unguiculata* (L.) Walp) grown in Nigeria.

MATERIALS AND METHODS

The ten varieties of cowpea were obtained from the National Cereals Research Institute, Moor Plantation, Ibadan, Nigeria.

The processing methods employed were soaking, germination, cooking and autoclaving of each of the ten varieties.

About 50g of the dry cowpea grains were first washed with 10% mercuric iodide (w/v) to remove surface contamination and, thereafter, soaked in 200ml distilled water at room temperature (27°C) for 3 days. The samples were then rinsed in distilled water and freeze-dried.

Germination of seeds was carried out in sterile petri dishes lined with wet cotton wool for a period of three days. Samples of sprouting beans were withdrawn at the end of the three days, rinsed in distilled water and freeze-dried.

Cooking of the bean samples was done in pressure cookers at 15 psi for 15 min. The cooked samples were then freeze-dried. The samples to be autoclaved were first milled in the raw form and then autoclaved at 105°C at 15 psi for 20 min.

The freeze-dried samples from the first three processes above were milled to pass through an 0.5 mm sieve, and along with the milled autoclaved samples were separately stored in capped bottles at 4°C until required for analysis.

Analytical Methods

Extraction of trypsin inhibitors (TI) from raw and processed cowpeas was carried out according to the method of Kakade *et al.* (1969) using 0.17M saline, TI activity was determined by the method of Nesheim (1965) in which the inhibitor was mixed with the trypsin before addition to the substrate (0.04M Tris buffer, pH 8.2). Proteolytic activity was then determined after incubation at 37°C for 60 min. Tryptic activity was expressed in terms of trypsin units, one trypsin unit being defined as the amount of trypsin which produced an increase in optical density of substrate solution at 410 Mμ in 10 minutes at 30°C equivalent to 0.01 micromoles of P-nitroaniline per 7 ml final volume of reaction mixture (Erlanger *et al.* 1961).

Hemagglutinin activity was determined by the photometric technique of

Liener (1955), which measures the ability of hemagglutinin extracts to agglutinate rabbit erythrocytes. For the determination of phytic acid, a combination of two methods was used. The extraction and precipitation of phytic acid were performed according to the method of Wheeler and Ferrel (1971) and iron in the precipitate was measured by Makower's (1970) method. A 4:6 Fe/P atomic ratio was used to calculate phytic acid content. Tannic acid was estimated as described by Eggum and Christensen (1975). All the results are the average of duplicate or triplicate analysis.

RESULTS

Trypsin Inhibitor Activity

TI activities in the raw, autoclaved, cooked, soaked and germinated cowpea varieties are presented in Table 1. In the raw whole beans, TI activity

Table 1. Trypsin inhibitor activity in raw and processed cowpea varieties (TUI/mg protein)[1]

Variety	Raw whole beans	Auto-claved beans	% Loss	Cooked beans	% Loss	Soaked beans	% Loss	Germinated beans	% Loss
IGBIRA	19.6	Nil	100	Nil	100	11.3	42.4	8.4	59.1
SAMARU LOCAL	25.4	-	100	-	100	19.6	22.8	12.5	50.8
KANO 1696	22.8	-	100	-	100	16.5	27.6	10.8	52.6
BLACKIE	25.0	-	100	-	100	18.0	28.0	13.0	48.0
ADZUKI	24.3	-	100	-	100	15.3	37.0	10.8	55.6
FARV-13	28.2	-	100	-	100	20.0	29.1	13.6	51.8
WEST BREED	25.7	-	100	-	100	19.7	23.4	11.0	57.2
IFE BROWN	20.5	-	100	-	100	14.3	30.3	11.3	44.9
PRIMA	22.3	-	100	-	100	13.4	39.9	10.2	54.3
NIGERIA B7	23.2	-	100	-	100	15.8	31.9	13.6	41.4
MEAN	23.7	-	100	-	100	16.4	31.2	11.5	51.4
STD. DEVIATION	2.7	-	-	-	-	2.94	6.6	1.65	5.3
% COEFFICIENT OF VARIATION	11.3	-	-	-	-	17.9	21.2	14.3	10.2

[1]Expressed as Trypsin units inhibited (TUI) per mg protein as defined by Kakade et al. (1969). Mean of duplicate or triplicates

ranged between 19.6 and 28.2 TUI/mg protein. Autoclaving and cooking resulted in a complete loss of activity while soaking decreased inhibitor activity by 22.8-42.4%. In the germinated samples, percentage losses were highest in 'Igbira' and 'Westbreed' where TI activities were reduced to 8.4 and 11.0 TUI/mg protein corresponding to 59.1 and 57.2% losses, respectively.

Hemagglutinin Activity

Values for hemagglutinin activity in the raw and processed cowpea varieties are shown in Table 2. Raw whole beans gave a range of 33.5-98.9 Hu/mg protein.

Autoclaving and cooking completely eliminated hemagglutinin activity in all varieties, while germination effected an average loss of 46.3%. Soak-

Table 2. Hemagglutinin activity in raw and processed cowpea varieties (Hu/mg protein)[1]

Variety	Raw whole beans	Autoclaved beans	% Loss	Cooked beans	% Loss	Soaked beans	% Loss	Germinated beans	% Loss
IGBIRA	43	-	100	-	100	36	16.3	20	53.5
SAMARU LOCAL	80	-	100	-	100	65	18.8	45	43.8
KANO 1696	33	-	100	-	100	28	15.2	14	57.6
BLACKIE	93	-	100	-	100	77	17.2	50	46.2
ADZUKI	94	-	100	-	100	82	12.8	60	36.2
FARV-13	45	-	100	-	100	35	22.2	24	46,7
WEST BREED	36	-	100	-	100	27	25.0	18	50,00
IFE BROWN	38	-	100	-	100	29	23.7	25	34.2
PRIMA	98	-	100	-	100	80	18.4	59	39.8
NIGERIA B_7	49	-	100	-	100	39	20.4	22	55.1
MEAN	61	-	100	-	100	49.8	19.0	33.7	46.3
STD. DEVIATION	26.9	-	-	-	-	23.2	3.9	17.8	7.9
% COEFFICIENT OF VARIATION	44.1	-	-	-	-	46.7	20.4	52.9	17.2

[1]Expressed as hemagglutinin units (HU) per mg protein as defined by Liener (1955). Mean of duplicate or triplicate.

ing was less effective than germination and decreases in 'Westbreed', 'Ife Brown', 'Farv-13', 'Blackie' and 'Adzuki' amounted to only 25.0% 23.7%, 22.2% and 12.8% losses in hemagglutinin activity, respectively.

Tannic Acid

Tannic acid contents, expressed as percentage of bean dry weight, in raw and processed cowpea varieties are presented in Table 3. The ranges are; for raw whole beans, 0.42-0.78%, for autoclaved beans, 0.33-0.67%; for cooked beans, 0.23-0.42%, for soaked beans, 0.37-0.69% and for germinated beans, 0.29-0.56%. Cooking and germination decreased tannic acid contents by 31.0-47.3% and 23.8-37.0% respectively. Autoclaving was not as effective as cooking and germination and losses obtained ranged between 13.8% in 'Kano 1696' and 28.3% in 'Nigeria B_7'.

Table 3. Tannic acid content in raw and processed cowpea varieties (g/100g dry matter)[1]

Variety	Raw whole beans	Auto-claved beans	% Loss	Cooked beans	% Loss	Soaked beans	% Loss	Germinated beans	% Loss
IGBIRA	0.60	0.49	18.3	0.35	41.7	0.54	10.0	0.42	30.0
SAMARU LOCAL	0.46	0.36	21.7	0.26	43.5	0.38	17.4	0.29	37.0
KANO 1696	0.58	0.50	13.8	0.30	31.0	0.50	13.8	0.41	29.3
BLACKIE	0.78	0.67	14.1	0.42	46.2	0.69	11.5	0.56	28.2
ADZUKI	0.48	0.36	25.0	0.27	43.8	0.40	16.7	0.33	31.3
FARV-13	0.63	0.54	14.3	0.35	44.4	0.55	12.7	0.48	23.8
WEST BREED	0.55	0.40	27.3	0.29	47.3	0.46	16.4	0.35	36.4
IFE BROWN	0.42	0.35	16.7	0.23	45.2	0.37	11.9	0.29	31.0
PRIMA	0.66	0.48	27.3	0.36	45.5	0.59	10.6	0.46	30.3
NIGERIA B_7	0.46	0.33	28.3	0.28	39.1	0.40	13.0	0.32	30.4
MEAN	0.56	0.45	20.7	0.31	42.8	0.49	13.4	0.39	30.8
STD. DEVIATION	0.11	0.11	5.9	0.06	4.7	0.11	2.6	0.09	3.8
% COEFFICIENT OF VARIATION	19.6	24.4	28.7	19.4	11.1	22.5	19.5	23.1	12.3

[1]Mean of duplicates or triplicates.

Phytic Acid

The results presented in Table 4 show that germination of cowpea seeds greatly lowered the phytic acid contents in the varieties. The decreases amounted to 51.6% in 'Prima' and 43.4% in 'Adzuki'. Soaking decreased phytic acid content of the seeds by 19.4 to 28.0% and cooking decreased by 7.7-11.7%. Autoclaving effects were very slight and maximum loss did not exceed 7.2% in 'Nigeria B7'.

DISCUSSION

The results obtained in this study suggest a fairly high concentration of antinutritional components in the cowpea varieties assayed. TI activity in the raw whole beans compares favorably with those of the navy bean (Kak-

Table 4. Phytic acid content of raw and processed cowpea varieties (mg/100g dry weight)[1]

Variety	Raw whole beans	Auto-claved beans	% Loss	Cooked beans	% Loss	Soaked beans	% Loss	Germinated beans	% Loss
IGBIRA	314.7	299.3	4.9	287.8	8.5	233.5	25.8	167.8	46.7
SAMARU LOCAL	299.0	280.7	6.1	273.6	8.5	226.8	24.2	150.3	49.8
KANO 1696	290.0	277.8	4.2	260.1	10.3	208.8	28.0	143.3	50.6
BLACKIE	329.6	307.2	6.8	296.9	9.9	250.0	24.1	177.6	46.1
ADZUKI	316.5	300.3	5.1	280.6	11.4	240.9	23.9	179.2	43.4
FARV-13	280.7	267.0	4.9	253.1	9.9	223.6	20.4	156.4	44.3
WEST BREED	300.8	280.5	6.8	273.3	9.2	220.5	26.7	155.6	48.3
IFE BROWN	325.7	304.3	6.6	300.5	7.7	241.8	25.8	169.3	48.0
PRIMA	330.4	314.1	4.9	296.3	10.3	246.8	25.3	160.0	51.6
NIGERIA B7	312.6	290.2	7.2	276.0	11.7	252.0	19.4	168.3	46.2
MEAN	310.0	292.1	5.8	279.8	9.7	234.5	24.4	162.8	47.9
STD. DEVIATION	16.9	15.2	1.0	15.8	1.3	14.3	2.7	11.7	2.7
% COEFFICIENT OF VARIATION	5.5	5.2	18.1	5.1	13.1	6.1	11.0	7.2	5.6

[1]Mean of duplicates or triplicates.

ade and Evans 1965), but are lower than in the soybean (Kakade *et al.* 1972), lima bean (Ologhobo and Fetuga 1982) and *Vicia faba* bean (Rafik El-Mahdy *et al.* 1980). Varietal differences were not evident as indicated by the low percentage coefficient of variation; apart from 'Igbira', 'Ife Brown' and 'Farv-13', all other varieties showed close similarities in TI activities. There was, however, considerable variability in hemagglutinin activity (CV, 44.1%). From the results obtained, it is possible to arrange the different cowpea varieties according to their hemagglutinin activity into low and medium hemagglutinin cultivars. A similar observation with different varieties of soybean has been reported by Kakade*et al.* (1972). The levels of tannic acid obtained are low and are not likely to be of any nutritional significance. Phytic acid, however, appears an abundant constituent and the high contents in 'Prima', 'Blackie' and 'Ife Brown' may affect the nutritional suitability of these varieties.

Among the processing methods studied, cooking and autoclaving were equally effective in eliminating TI and hemagglutinin activities, while soaking and germination were only partially effective. Such heat destruction has been reported for several legumes including the soybean (Liener 1962), black-eye (Richardson 1948), cowpea (Oliveira 1973) and lima bean (Ologhobo and Fetuga 1982). Observations on tannic acid, however, indicated an average of only 42.8% decrease by cooking and 20.7% by autoclaving. For phytic acid, cooking and autoclaving were only slightly effective in decreasing contents. This was expected as the beans were not incubated or steeped before cooking. According to Chang *et al.* (1977), steeping of beans or incubation in water followed by cooking in boiling water increased inorganic P concentration with 50% hydrolysis of bean phytate. This was mainly attributed to the phytase activity during the steeping of seeds or incubation and the effect of heat treatment. In the present study, the beans were not subjected to any other processes prior to cooking or autoclaving, and the reduction in phytic acid content was mainly the effect of heat. The results also indicate that cooking in water was more effective in lowering phytic acid than autoclaving, thus suggesting that the traditional cooking methods are adequate in eliminating some of the undesirable substances.

The phytic acid content of the cowpea varieties were greatly diminished by germination but reduced only slightly upon soaking. This was also true for the decrease in tannic acid contents. Cream and Haisman (1964) had attributed the decrease in phytic acid loss upon soaking legumes to complexing of inositol hexaphosphate with Ca and Mg to form insoluble phytates which could be extracted only with dilute acids. This possibly explains the decrease in phytic acid loss on soaking in water. Tannic acid on the other hand must have been lost through the leaching of a small fraction of hydrolyzable phenolic compounds, located in the seed coats of cowpea varieties

(Elias and Bressani 1979), into the soaking medium. Some amounts of polyphenols have been found in the soaking and cooking waters of *Phaseolus vulgaris*, indicating that large amounts of polyphenols could be eliminated by discarding washing and cooking waters (Fukuda Suzuki 1978). The actual amounts ingested will thus depend on how beans are processed and consumed.

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MOLD INHIBITION IN TORTILLA BY DIMETHYL FUMARATE

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Received for Publication March 2, 1984
Accepted for Publication June 20, 1984

ABSTRACT

Nine batches of tortilla containing 0, 0.05, 0.1., 0.2 and 0.4 g of calcium propionate or dimethyl fumarate per 100 g corn flour were prepared. The tortillas were individually packaged in polyethylene bags, stored at room temperature and observed daily for visible signs of mold growth. Samples from each batch were also subjected to the determination of moisture and water activity. The mold-free shelf-lives of tortillas containing calcium propionate ranged from 2-5 days and those of tortillas containing dimethyl fumarate ranged from 2-11 days.

INTRODUCTION

Tortilla and tortilla products made from corn flour are staple food in Mexico and Central America. They are also becoming popular in the United States. The main drawback in the commercial distribution of tortilla is its very limited shelf-life. Because of its high moisture content, which ranges from 45-48%, tortilla is highly susceptible to microbial spoilage. Average shelf-life of tortilla, under tropical conditions, has been reported to be as low as 12 hr (Rubio 1972 a). Storage temperatures above 25°C, particularly above 30°C, favor the growth of bacteria which produces off odor and ropiness. At temperatures below 25°C, the first sign of spoilage is the appearance of moldy spots (Rubio 1972 a). In the U.S. at present, most of the tortillas are distributed and marketed under refrigerated conditions.

Many attempts have been made to improve the shelf-life of tortilla. Rubio received six patents for various formulations to improve the shelf-stability of tortilla and tortilla products. The additives in the formulations were: aliphatic polycarboxylic acids or their anhydrides; hydrophylic inorganic gels, such as aluminum hydroxide or ferric hydroxide; epichlorohydrin, acetic

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acid, propionic acid and their water soluble salts, diacetates; and methyl, ethyl, propyl and butyl esters of parahydroxybenzoic acid (Rubio 1972 a, b, 1973, 1974 a, b, 1975).

Pelaez and Karel (1980) developed intermediate moisture tortillas by incorporating various humectants such as glycerol, salt, glucose, sucrose, corn syrup and corn solids in addition to mycostatic agent potassium sorbate. They reported that with proper packaging the intermediate moisture tortilla may be protected from microbial growth for 30 days. However, the addition of their humectants and the special packaging considerably increased the cost of tortilla.

Another approach to mold inhibition in tortilla has been the combination of low levels of sorbates or propionates in the dough with an external sorbate spray after cooking (Hickey *et al.* 1982). However, a uniform spray treatment with sorbates requires the installation of somewhat sophisticated, and relatively expensive, machinery in the processing line. Small tortilla manufacturers, particularly those in developing countries, operate with limited capital and are unable to make such large investments. Hence, the conventional method of incorporating an antifungal agent into the product would be more desirable.

Recently, Islam (1982) demonstrated that the dimethyl ester of fumaric acid can effectively prevent mold growth in bread. Islam *et al.* (1982) also found that dimethyl fumarate can inhibit the growth of aflatoxin producing mold, *Aspergillus flavus* in poultry feed. Huhtanen (1983) demonstrated antibotulinal activity of dimethyl fumarate in cans of comminuted nitrite-free bacon.

The main objective of this study was to assess the preservative effect of dimethyl fumarate in tortilla and compare it with that of a commonly used preservative, calcium propionate.

MATERIALS AND METHODS

Nine batches of tortilla were prepared using commercially available lime-treated corn flour, Maza Harina (Quaker Oats Co., Chicago, IL). The corn flour was mixed with water in the proportion of 165 g flour to 200 ml water. The first batch was a control with no preservative. The next four batches contained 0.05, 0.1, 0.2 and 0.4 g of calcium propionate per 100 g corn flour respectively. The remaining four batches contained corresponding amounts of reagent grade dimethyl fumarate obtained from Eastman Kodak Co. (Rochester, NY). The additives were incorporated into the tortilla dough by first mixing them with water. Round shaped doughs weighing about 35 g each were pressed into flat discs of 12.5 cm in diameter and about 2 mm

thickness using a tortilla press. They were cooked on an electric pan at 193.3°C for about 1.5 min per side plus an additional 15 s on the first side. They were cooled on racks, packaged individually in polyethylene bags and stored at room temperature which ranged from 22-23°C. Three tortillas from each batch were observed daily at a specific time by the authors for visible signs of mold growth. Shelf-life of each tortilla was calculated based on the number of days it maintained a mold-free appearance.

Three tortillas from each batch were used for determination of moisture and water activity (A_w). Moisture was determined by oven drying method of AOAC (1975). Water activity was determined by a Beckman SINA hygrometer (Beckman Instruments Co., Cedar Grove, NJ).

RESULTS AND DISCUSSION

The moisture content of the tortillas ranged from 47.6 to 50.6%. This range is somewhat higher than the figures reported by Rubio (1974a) and Johnson *et al.* (1980). Neither calcium propionate nor dimethyl fumarate had any influence on the moisture content of the tortillas. Water activity of tortilla samples ranged from 0.97-0.98 indicating highly favorable environment for microbial growth (Jay 1978). Again the level of calcium propionate or dimethyl fumarate had no effect on the water activity of the tortillas.

The mold-free shelf-lives of tortillas containing calcium propionate and dimethyl fumarate are shown in Table 1. Tortillas in the control batch had

Table 1. Mean Shelf-life of Tortillas Containing Various Levels of Calcium Propionate or Dimethyl Fumarate During Storage at Room Temperature[1]

PRESERVATIVE	LEVEL (g/100 g Flour)	SHELF-LIFE (Days)[2]
Control	0.00	1.3 ± 0.6a[3]
Calcium Propionate	0.05	1.7 ± 0.6a
	0.10	2.0 ± 0.0ab
	0.20	3.7 ± 0.6bc
	0.40	4.7 ± 0.6c
Dimethyl Fumarate	0.05	2.0 ± 0.0ab
	0.10	3.7 ± 0.6bc
	0.20	7.7 ± 0.6d
	0.40	10.6 ± 1.5e

[1]Average of 3 tortillas
[2]Based on number of days required for the appearance of moldy spot
[3]Means not followed by a common letter are significantly ($P<0.01$) different according to Duncan's multiple range test

average shelf-life of only 1.3 days. Average shelf-lives of tortillas containing calcium propionate ranged from 1.7 to 4.7 days depending on the concentration used. Dimethyl fumarate at 0.2 and 0.4% levels maintained the tortillas mold-free for significantly ($P < 0.01$) longer time than calcium propionate. The highest average shelf-life obtained by 0.4% calcium propionate was 4.7 days while this level of dimethyl fumarate gave a shelf-life of 10.6 days. It should be emphasized that the average shelf-life assessment in this study was based on only the absence of visible signs of mold growth. No attempt was made to obtain any mold or bacteria count. In general, the shelf-lives obtained in this study are considerably lower than those reported by Pelaez and Karel (1980). The higher moisture content and water activity in this study are likely to be the most probable causes of the large discrepancy in shelf-life.

Dimethyl fumarate exhibited strong inhibitory effect on the mold growth on tortillas. Besides its normal antifungal property (Gershan and Shanks 1978) in tortilla, dimethyl fumarate seems to have an added advantage over calcium propionate. Since tortilla is made from lime-treated corn, the pH of tortilla is usually about 7 or higher. The antifungal activity of calcium propionate is highly dependent upon the pH; the lower the pH, the higher the activity. This is because the undissociated form of propionic acid is higher at lower pH as explained in detail in a review article by Sauer (1977). On the other hand, dimethyl fumarate, being an ester, does not dissociate under conditions normally encountered in food systems. Similar to the esters of p-hydroxybenzoic acid, its activity is expected to be pH-independent (Heimann 1980). Thus, dimethyl fumarate provides considerable flexibility over calcium propionate for use in a product such as tortilla. Similar, but less, advantage may be expected over potassium sorbate which is also used for tortilla preservation. However, it should be mentioned that potassium sorbate is more expensive than calcium propionate.

Dimethyl fumarate is not an approved food additive. According to the Registry of Toxic Effects of Chemical Substances (DHHS-NIOSH 1979) the oral LD_{50} in rats is 2240 mg/kg for dimethyl fumarate compared with 5160 mg/kg for calcium propionate and 3000 mg/kg for sodium chloride. Based on these comparisons, dimethyl fumarate may have low toxicity. Detailed toxicological studies have to be carried out before it can be recommended for use in food.

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COMBINED EFFECTS OF ELECTRICAL STIMULATION AND METHODS OF MEAT PRESERVATION UPON THE SURVIVAL OF BACTERIA[1]

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Received for Publication February 29, 1984
Accepted for Publication June 28, 1984

ABSTRACT

The synergistic effect caused by electrical stimulation and methods of meat processing on microbial numbers in meat was investigated. Electrical stimulation in all experiments reduced initial bacterial numbers and lowered tissue pH. Under the conditions of this research, no synergistic effect could be proven for electrical stimulation and 3% salt or 3% salt and 200 ppm of nitrite or for storage at -21°C. However, a synergistic effect was observed for electrical stimulation and heating at 60°C. This reduction in thermoresistance caused by electrical stimulation did not appear to be caused by the lowering of pH of the heating media.

INTRODUCTION

Bacterial action is one of the most important factors affecting the shelf-life and wholesomeness of meat and meat products. Therefore, any new procedure which may even slightly alter the number or characteristics of bacteria in meat should be investigated. One such process which has received considerable attention in recent years is electrical stimulation.

[1]Salaries and research support provided by State and Federal Funds appropriated to The Ohio Agricultural Research and Development Center, The Ohio State University. Journal Article No. 218-82.
Submitted to 29th European Meeting of Meat Research Workers in Parma, Italia, August 28, 1983

Journal of Food Processing and Preservation 8 (1984) 47–56

Post-mortem electrical stimulation of carcasses accelerates the onset of rigor mortis allowing easy, early hot-boning and rapid chilling of primal cuts without the toughening caused by cold shortening (Asghar and Henrickson 1982). Electrical stimulation can also speed up the fabrication of carcasses, reduce refrigeration requirements, lower energy usage, reduce cooler space needs (Kastner 1977) and improve the organoleptic properties of meat (Savel *et al.* 1982; McKeith *et al.* 1982; Asghar and Henrickson 1982).

Available information on the effect of electrical stimulation upon meat microbiology is very inconsistent. Corte *et al.* (1980) found higher initial superficial contamination of meat from stimulated and hot-boned carcasses than from traditionally chilled carcasses. Jeremiah and Martin (1980), Kotula (1980) and Taylor *et al.* (1980) failed to identify any differences in the bacterial counts from electrically stimulated and unstimulated meat initially or after storage. However, a number of research reports have shown a statistically significant reduction in total plate counts as a result of electrical stimulation (Mrigadat *et al.* 1980; Ockerman and Szczawinski 1983; Raccach and Henrickson 1978, 1980; Lin *et al.* 1984). Lin *et al.* (1984) also reported that electrical stimulation caused a reduction in the number of microorganisms in inoculated media.

These inconsistent microbial results would suggest that electrical stimulation causes a slight decrease in the microflora of meat but this decrease is often not large enough to be of practical significance under all experimental conditions.

Several hypotheses on the mechanism by which electrical stimulation can affect the bacteria in meat are possible. They include the hypothesis that a fast reduction in the muscle pH value may retard microbial growth (Kotula 1980, 1981; Mrigadat *et al.* 1980), that electrical stimulation impairs the metabolism of bacterial cells (Raccach and Henrickson 1980) or that electrical stimulation has adverse effects on the meat as a growth medium (Riley *et al.* 1980). Bacteria may also be destroyed by electrical stimulation initiating the release of some proteolytic enzymes from the meat tissue (Dutson *et al.* 1980; Sorinmade *et al.* 1978), by changing meat Eh (the oxidation reduction potential) or by generating free radicals in the stimulated tissue (Mrigadat 1980).

Ockerman and Szczawinski (1983) found that bacterial cells are destroyed directly during the process of electrical stimulation. They also suggested that it is less probable that bacterial cells are retarded only by a drop in pH or by proteolytic enzymes; however, free radicals or other unknown factors acting during electrical stimulation may be responsible for the observed results.

Regardless of the mechanism of destruction, it seems possible that sublethal damage of bacteria may take place during electrical stimulation. This

damage can increase the sensitivity of microorganisms to other injurious agents such as the presence of salt and nitrite or the effect of low or high temperatures which could be important from a practical processing standpoint.

The purpose of this research was to investigate if electrical stimulation and other fundamental processes applied in meat technology (salting, curing, freezing) will cause synergistic effects on the growth of microorganisms.

MATERIALS AND METHODS

Experimental Design

Three experiments were conducted to evaluate the effect of electrical stimulation on bacteria.

In the first experiment, samples were collected from the necks *(M. sternocephalicus)* of cattle immediately after slaughter, inoculated by dipping in a suspension of bacteria and subjected to electrical stimulation. Control samples were not stimulated after inoculation. Next, both stimulated and control samples were aseptically ground through a 3.2 mm grinder plate and subdivided into three 50 g portions. The first portion was stored without any additives. The second portion was mixed with 3% salt (NaCl). To the third portion 3% salt and 200 mg of nitrite ($NaNO_2$)/kg were added. All samples were stored under aerobic conditions in petri dishes at 0-2°C for 14 days. Aerobic plate count (APC) and pH were determined immediately after grinding (before the addition of salt and nitrite) and after 7 to 14 days of storage. This procedure was repeated 6 times.

In the second experiment, inoculated control as well as inoculated and stimulated samples of meat were homogenized in distilled water, using a ratio of 1 part meat to 9 parts water. The meat slurry was poured into 12 sterile test tubes (2.5 ml into each tube), capped with rubber stoppers and stored at -21°C for 21 days. The measurement of pH was conducted immediately after homogenization. APC was determined after 0, 7, 14 and 21 days of storage (three repetitions for each time period). This procedure was repeated three times.

In the third experiment, the samples of meat slurry were prepared in the same way as in the second experiment. After determination of pH, the test tubes with meat slurry from control and stimulated meat were heated in the water bath at 60°C for 0, 5, 10 and 15 min. Immediately after heating, the samples were cooled in ice water and APC was determined (three repetitions for each heating time). This experiment was repeated three times.

Collection of Samples

The muscles *(M. sterno-cephalicus)* used in these experiments were collected from both sides of the beef carcasses immediately after bleeding and divided into 50 ± 10 g samples. Samples from 9 animals were used in this study.

Preparation of General Inoculum and Inoculation of Meat

A cotton-tipped swab moistened in 0.5% solution of Bacto-Peptone was used to remove bacteria from a meat cutting table that had previously been used for 3 hr in handling beef carcasses. Five swabs representing 5 different 25 cm^2 areas of the table for a total of 125 cm^2 were placed into 200 ml of peptone diluent. After shaking, 20 ml of the diluent was poured into 180 ml of nutrient broth and incubated for 3 days at 25°C. This culture of bacteria was poured into test tubes and stored at -25°C. After frozen storage and before each of the experiments, the bacteria were allowed to multiply in the nutrient broth for 3 days at 25°C and diluted with a 0.5% solution of Bacto-Peptone to obtain a suspension containing approximately 5×10^8 bacteria/ml (determined by OD plotted against an APC curve). This high level of bacterial inoculation was used so that if electrical stimulation did influence bacterial numbers it would be easier to observe. Samples of meat were inoculated by dipping 3 times for 1 s each into this suspension and were stimulated immediately after inoculation.

Electrical Stimulation

Electrical stimulation was accomplished within 30 min post-mortem. The samples were stimulated in sterile petri dishes with 21 mA (60 Hz) current, 42 V for 4 min, with thirty 2-s duration shocks per min. Before and during electrical stimulation, both control and stimulated samples were held at ambient temperature (ca. 20°C). The sample was transferred from the petri dish with the aid of sterile distilled water to avoid loss of inoculum and/or blood or moisture.

Bacterial Enumeration and pH Measurement

The samples of meat (approximately 50 g and 7 × 4 × 2 cm) were homogenized in distilled water, using a ratio of 1 part meat to 9 parts water, by using a Stomacher Lab-Blender 400. The pH of the slurry was measured using a Beckman pH meter (Expandomatic SS-2). From another portion of the meat slurry, appropriate dilutions were prepared with a 0.5% solution of Bacto-Peptone and plated using Tryptone Glucose Extract Agar (Difco). Plates were incubated at 25°C and colonies were counted after 4 days of incubation (Ockerman 1980).

Statistical Analysis

The microbial counts per gram were transformed to logarithmns to the base 10. Calculations and analyses were conducted on the transformed data.

Statistical analyses of data for pH and microbial counts were carried out using the General Linear Models and Correlation Procedures supplied through the Statistical Analysis System (SAS 1979).

RESULTS AND DISCUSSION

Combined Effects of Electrical Stimulation, Salting (3% NaC1) and Curing (3% NaCl and 200 mg/kg $NaNO_2$) on Behavior of Microflora in Ground Beef Stored at 0-2°C

As shown in Table 1, electrical stimulation caused an initial (0 day, each salting and/or curing group) decrease in APC of meat but this reduction was not large enough to be statistically significant. APC of stimulated meat was also slightly lower (significant 1/3 of the time) in all treatment groups after 7 and 14 days of storage except for meat without salt and nitrite after 14 days of storage. However, significant differences between control and electrically stimulated meats were found only for samples without any additives (NaCl 0%, $NaNO_2$ 0%) after 7 days ($P < 0.01$) and for meat with salt NaCl 3%, $NaNO_2$ 0%) after 14 days of storage ($P < 0.05$).

Table 1. Effects of electrical stimulation, salting and curing on pH and log of APC of inoculated beef stored at 0-2°C

TRAIT	TIME OF STORAGE (days)	NaCl 0% $NaNO_2$ 0% Control	NaCl 0% $NaNO_2$ 0% Stimulated	NaCl 3% $NaNO_2$ 0% Control	NaCl 3% $NaNO_2$ 0% Stimulated	NaCl 3% $NaNO_2$ 0.02% Control	NaCl 3% $NaNO_2$ 0.02% Stimulated
pH[e]	0	6.11^{ax}	5.99^{ax}	6.11^{ax}	5.99^{ax}	6.11^{ax}	5.99^{ax}
	7	5.63^{ay}	5.55^{ay}	5.97^{bx}	5.92^{bx}	5.97^{bx}	5.94^{bx}
	14	5.96^{ax}	5.89^{ax}	6.07^{ax}	6.02^{ax}	6.11^{ax}	6.08^{ax}
APC[f]	0	4.67^{ax}	4.45^{ax}	4.67^{ax}	4.45^{ax}	4.67^{ax}	4.45^{ax}
	7	6.11^{ay}	5.76^{by}	4.56^{cx}	4.32^{cdx}	4.30^{cdy}	4.04^{dy}
	14	9.03^{az}	9.06^{az}	6.30^{by}	5.99^{cy}	4.19^{dy}	4.01^{dy}

n=6

^{a,b,c,d}Means within the same row bearing different superscripts are different at P<0.05

[e]Standard error of least square means 0.1030

[f]Standard error of least square means 0.1033

^{x,y,z}Means for the same item (pH or APC) within the same column bearing different superscripts are different at P<0.05

The overall analysis of variance indicates that electrical stimulation as well as treatment (additives) and storage time influenced significantly ($P < 0.01$) the bacterial counts. However, the interactions of stimulation × treatment, stimulation × storage time and stimulation × treatment × storage time were not statistically significant indicating that NaCl or NaCl and $NaNO_2$ added to the meat had the same retarding influence on bacteria from electrically stimulated or unstimulated meat.

Electrical stimulation also caused a slight decrease in pH for all treatment groups but observed differences were not statistically significant (Table 1). The overall analysis of variance indicates that pH was significantly ($P < 0.01$) influenced only by treatment (additives) and storage time.

The obtained results generally confirmed previously conducted work (Ockerman and Szczawinski 1983) in this laboratory. However, initial differences in the APC and pH between control and stimulated meat had been slightly greater and statistically significant when unground beef cuts had been used for samples in the previous experiments.

In prior studies, the analysis of variance of APC indicated nonsignificant statistical interactions between electrical stimulation and levels of salt and nitrite in the culture media suggesting the same effect for NaCl and $NaNO_2$ on bacteria from control and stimulated beef tissue (Ockerman and Szczawinski 1980). The results of the present research endorsed former observations though the experimental conditions in both works were entirely different.

Although a slight reduction (in some cases nonsignificant) in the microflora caused by electrical stimulation could be seen initially in this experiment and also after 7 and 14 days of storage of meats subjected to salting (NaCl 3%, $NaNO_2$ 0%) and curing (NaCl 3%, $NaNO_2$ 0.02%), most research workers consider differences in this range as being unimportant in commercial production of meat (Kotula 1981).

Effect of Electrical Stimulation on the Survival of Bacteria During Storage at -21°C

As shown in Table 2, the initial difference (0.26 reduction due to stimulation) in APC between samples from control and stimulated meat (significant at $P < 0.01$) remained almost identical for the 21 days of storage at −21°C.

The analysis of variance indicated that electrical stimulation as well as the time of frozen storage and individual animals or handling of these animals affected significantly ($P < 0.01$) the number of bacteria in the meat slurry.

A nonsignificant interaction for frozen storage time × stimulation indicates that the responses of bacteria, from control and stimulated meat, to

Table 2. Effect of electrical stimulation on the survival of bacteria in meat slurry during storage at -21°C

Time of storage (days)	Log of aerobic plate counts		
	Control	Stimulated	Difference[a]
0	5.94	5.68	0.26**
7	5.77	5.51	0.26**
14	5.68	5.45	0.23**
21	5.68	5.42	0.26**

[a] Standard error of least square means 0.0145

n = 9

** Significant difference at P<0.01

low temperature are approximately the same and that a synergistic effect did not occur under these experimental conditions.

Very little information is available on the effect of freezing upon bacteria in stimulated meat. Corte *et al.* (1980) reported that after 3 months of storage at -20°C thawed cuts from electrically stimulated and hot boned sides showed a tendency to be more contaminated than the control. Mrigadat *et al.* (1980) discussing the results obtained by Raccach and Hendrickson (1980) suggested that any injurious effect from electrical stimulation on the bacterial cells may have been magnified by freezing. The present study indicated a reduction in the number of microorganisms due to electrical stimulation but did not confirm these suggestions of a potential synergistic effect.

Effect of Electrical Stimulation on the Thermoresistance of Bacteria During Heating at 60°C

As shown in Table 3, an initial statistically significant ($P < 0.01$) difference in APC between control and stimulated samples increased systematically with time of heating. This decrease in bacterial number at 0 time for stimulated tissue again indicates the effect of stimulation on microorganisms and the increase with heating time suggests a synergistic effect.

Table 3. Effect of electrical stimulation on the thermoresistance of bacteria in meat slurry during heating at 60°C

Trait		Control	Stimulated	Difference	Std. error of least sq. means
pH		6.28	6.10	0.18	
Log of APC	0 min	5.94	5.68	0.26**	0.035
	5 min	3.92	3.59	0.33**	0.035
	10 min	2.56	1.99	0.57**	0.035
	15 min	2.03	1.37	0.66**	0.035
D value[a]	(min)	3.83	3.48	0.35*	0.093
D value adjusted for pH (min)		3.81	3.50	0.31	0.106

n = 9

[a] The time required to reduce the microbial population by 90% at a specified temperature

* Significant difference at $P<0.05$

** Significant difference at $P<0.01$

A statistically significant difference ($P < 0.05$) in the mean D value (Table 3) suggests a reduced tolerance for heat by microorganisms in stimulated tissue and the significant ($P < 0.01$) interaction for stimulation × heating time found in the analysis of variance also shows that electrical stimulation decreases the thermoresistance of bacteria.

This observation could easily be explained by the sublethal damage of bacteria occurring during electrical stimulation. However, it is a well known fact that the pH of the suspending medium in which microorganisms are heated is one of the most important factors that influences thermoresistance (Banwart 1979). Therefore, it is difficult to determine whether the damage to bacteria is caused by electrical stimulation, or whether a slightly lower pH of the samples from stimulated meat (Table 3) is responsible for the synergistic effect observed in this experiment.

In order to examine this question in more detail, additional statistical analyses were conducted and the results are as follows.

The results obtained from the analysis of variance of APC "adjusted" for pH were very similar to the results obtained prior to APC adjustment indicating that the small change in pH encountered in this experiment did not affect the thermoresistance of bacteria under these conditions. A statistically nonsignificant correlation between pH and APC, as well as between

pH and D values, seems to confirm this conclusion. However, statistical analysis of the D values "adjusted" for pH shows that the difference between control and stimulated samples is not quite large enough to be significant at the 0.05 level (Table 3).

Considering all the results, it seems that the damage to bacterial cells occurring during electrical stimulation was a more important factor than pH in affecting thermoresistance in this experiment. However, a precise separation of these two causes will require further investigations.

Irrespective of the mechanism by which electrical stimulation decreases thermoresistance of bacteria, this seems to have occurred under the conditions of this experiment. It should be emphasized that the electrical stimulation in this research was conducted on a model system of small tissue samples and this research has not attempted to translate this to commercial conditions. Little information on this subject can be found in the literature.

It would seem that further studies using various types of spoilage and pathogenic microorganisms should evaluate whether the synergistic effect of electrical stimulation and heat treatment may have any importance from a commercial standpoint.

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BOOK REVIEWS

Handbook of Tropical Foods, Harvey T. Chan, Jr. Editor. 1983. Marcel Dekker, New York, New York. pp. 156. $75.00 in U.S.A., $90 in Foreign Countries.

The Editor's stated purpose of bringing together diffuse, often obscure information on a number of tropical foods is commendable. Fifteen well-qualified contributors have indeed covered the English language and some foreign literature relating to topics which range from basic staples to lesser known dietary items. Most chapter formats (10 to 70 pages) consist of: history and geography, botany, horticulture, composition and biochemistry, nutritional and quality attributes, postharvest handling, processing and preservation, and byproduct utilization.

Overall the informational balance is suitable for food scientists and technologists. A good introduction or review of the foods is given with enough background and detail to provide a general understanding of the technical processes involved, directly or through the extensive references. The book has some intriguing, primarily qualitative descriptions of unit operations relating to the various processes. In fact, expertise such as is represented by this journal readership could make important additional contributions to tropical food processing and utilization based on background information given in these chapters.

A few comments on the 15 chapters are listed: Amaranth, contrast between seed and leaf uses: Aroid Root Crops, emphasis on botany and composition: Bananas and Plantains, comprehensive coverage of traditional and industrial products, processes and utilization practices: Cassava, stresses production and processing machinery, mechanization and industrialization: Citrus Fruits, good overview of the global picture: Fermented Fish Products, fascinating treatment of unusual preservation methods: Ginger, good introduction to products and processes: Guava, brief treatment based mostly on Hawaiian developments: Macadamia Nuts, good description emphasizing chemistry and technology: Mango, comprehensive with much useful processing information: Palm Oil, well-detailed oil chemistry and comparisons between traditional and industrial practices: Papaya, brief review stressing Hawaiian experiences: Rice, comprehensive, balanced overview of this major tropical staple; Tropical Fruit Wines, brief description of winemaking practices emphasizing pineapple, papaya, passion fruit, byproduct alcohol and vinegar and Yams, thorough, well-organized review.

As is common with multi-authored works, chapters are of variable quality with about 9 qualifying as average or above average. In view of the

book's price, typographical errors are detracting and more attention to original figures and less space-wasting minor tables (utilizing only 1/2 column of some pages) would be desirable. Several otherwise excellent chapters could have benefited from photos or diagrams to supplement or replace detailed descriptive narrative. Several chapters have scant references past 1979.

There are many other common and lesser know tropical foods whose treatment in such a handbook format would be a welcome professional reference. Unfortunately, such a series might be prohibitively expensive to those who need it the most, individuals, small institutions and businesses in LDCs. The book is a useful reminder of how far we have come in developing certain tropical foods and how very much remains to further exploit these and other unique, critically needed global food resources.

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Sensory Quality in Foods and Beverages: Definition, Measurement and Control. Edited by A. A. Williams and R. K. Atkin. 1983. 488 pp., 144 illus., 90 tables. ISBN 0-89573-130-4. $21.25 paperback, Ellis Horwood Series in Food Science and Technology. Distributor: Verlag Chemie International, Inc., 303 N.W. 12th Avenue, Deerfield Beach, FL 33441.

Papers presented at the University of Bristol in an April, 1982, international symposium with the book title form the contents of this volume. Although the topics of the papers range widely and cover many aspects of sensory quality, the book seems to lack the systematic approach that would make the volume useful as a textbook for university classes. However, because of the broad nature of individual contributions on the definition, measurement and control of the sensory quality of foods and beverages, technical personnel and researchers should find the book to be a useful general reference. Retrieval of information may be difficult because the index is limited to six pages, and effective use will require the user to develop a familiarity with the contents.

The first section of the book contains four papers about understanding and defining sensory quality, and this information is integrated with industrial needs and practices. The second section presents four chapters on the state of the art methodology for measurement of sensory quality, but the depth of coverage is somewhat limited. As with other later chapters, this section contains short contributions from poster session presentations that relate to the topic of the section.

Section 3 deals with the use of instrumental methods and their validation for the measurement of the sensory quality of foods. Chapters are devoted to appearance, color, texture and flavor as they may be determined by analytical instrumentation. Some of the presentations are quite well done and provide a reasonable basis for the following Section 4 which deals with applications of sensory and instrumental methods to specific products. Section 4 comprises the greatest number of papers and these address vegetables, fruit and fruit products, fish and meat, and dairy products from various quality-sensory-instrumental perspectives. The final Section 5 provides three papers on the influence of sensory quality on food choice and intake and includes an interesting paper on the topic of marketing and sensory quality.

In the preface, the authors have stated accurately that the symposium attempts to explore the state of our knowledge and understanding of sensory quality in foods and beverages, and how we interpret information about consumer preferences in relation to chemical and physical measurements pertinent to food quality. Thus, the contents of the book focus on issues and information that frequently are meaningful to individuals well-versed or very curious about the field rather than providing discussions about the usual well-used methods and procedures covered in other sensory publications.

Robert C. Lindsay
University of Wisconsin-Madison
June 26, 1984

ERRATA

The errata listed below were inadvertently printed in the *Journal of Food Processing and Preservation* Volume 7, Number 3 in the paper: Kinetics of Oxidation of Dehydrated Food at Low OxygenPressures by Louis J. Kacyn, Nutrition and Flow Control Division, Baxter Travenol Laboratories, One Baxter Parkway, Deerfield, Illinois; Israel Saguy, Department of Food Technology, Agricultural Research Organization, The Volcani Center, Bet Dagan, 50-250, Israel and Marcus Karel, Department of Nutrition and Food Science, Massachusetts Institute of Technology, Cambridge, Massachusetts.

page	line	wrong	correct
164	30	$\frac{dy}{dt} + K_B(y)(1-y)$	$\frac{dy}{dt} = K_B(y)(1-y)$ (2)
165	3	$y = y_o e^{K_B t}$	$y = y_o e^{K_B t}$ (3)

CORRECTED.
Au.
27/227

F N P

JOURNALS AND BOOKS IN FOOD SCIENCE AND NUTRITION

Journals

JOURNAL OF FOOD SERVICE SYSTEMS, G. E. Livingston and C. M. Chang

JOURNAL OF FOOD BIOCHEMISTRY, H. O. Hultin, N. F. Haard and J. R. Whitaker

JOURNAL OF FOOD PROCESS ENGINEERING, D. R. Heldman

JOURNAL OF FOOD PROCESSING AND PRESERVATION, T. P. Labuza

JOURNAL OF FOOD QUALITY, M. P. De Figueiredo

JOURNAL OF FOOD SAFETY, M. Solberg and J. D. Rosen

JOURNAL OF TEXTURE STUDIES, M. C. Bourne and P. Sherman

JOURNAL OF NUTRITION, GROWTH AND CANCER, G. P. Tryfiates

Books

PRODUCT TESTING AND SENSORY EVALUATION OF FOODS, H. R. Moskowitz

ENVIRONMENTAL ASPECTS OF CANCER: ROLE OF MACRO AND MICRO COMPONENTS OF FOODS, E. L. Wynder *et al.*

FOOD PRODUCT DEVELOPMENT IN IMPLEMENTING DIETARY GUIDELINES, G. E. Livingston, R. J. Moshy, and C. M. Chang

SHELF-LIFE DATING OF FOODS, T. P. Labuza

RECENT ADVANCES IN OBESITY RESEARCH, VOL. III, P. Bjorntorp, M. Cairella, and A. N. Howard

RECENT ADVANCES IN OBESITY RESEARCH, VOL. II, G. A. Bray

RECENT ADVANCES IN OBESITY RESEARCH, VOL. I, A. N. Howard

ANTINUTRIENTS AND NATURAL TOXICANTS IN FOOD, R. L. Ory

UTILIZATION OF PROTEIN RESOURCES, D. W. Stanley, E. D. Murray and D. H. Lees

FOOD INDUSTRY ENERGY ALTERNATIVES, R. P. Ouellette, N. W. Lord and P. E. Cheremisinoff

VITAMIN B_6: METABOLISM AND ROLE IN GROWTH, G. P. Tryfiates

HUMAN NUTRITION, 3RD ED., F. R. Mottram

DIETARY FIBER: CURRENT DEVELOPMENTS OF IMPORTANCE TO HEALTH, K. W. Heaton

FOOD POISONING AND FOOD HYGIENE, 4TH ED., B. C. Hobbs and R. J. Gilbert

POSTHARVEST BIOLOGY AND BIOTECHNOLOGY, H. O. Hultin and M. Milner

THE SCIENCE OF MEAT AND MEAT PRODUCTS, 2ND ED., J. F. Price and B. S. Schweigert

GUIDE FOR AUTHORS

Typewritten manuscripts in triplicate should be submitted to the editorial office. The typing should be double-spaced throughout with one-inch margins on all sides.

Page one should contain: the title, which should be concise and informative; the complete name(s) of the author(s); affiliation of the author(s); a running title of 40 characters or less; and the name and mail address to whom correspondence should be sent.

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.

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Results: The results should be presented as concisely as possible. Do not use tables *and* figures for presentation of the same data.

Discussion: The discussion section should be used for the interpretation of results. The results should not be repeated.

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.

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Tables should be numbered consecutively with Arabic numerals. The title of the table should appear as below:

Table 1. Activity of potato acyl-hydrolases on neutral lipids, galactolipids, and phospholipids

Description of experimental work or explanation of symbols should go below the table proper. Type tables neatly and correctly as tables are considered art and are not typeset.

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Short notes will be published where the information is deemed sufficiently important to warrant rapid publication. The format for short papers may be similar to that for regular papers but more concisely written. Short notes may be of a less general nature and written principally for specialists in the particular area with which the manuscript is dealing. Manuscripts which do not meet the requirement of importance and necessity for rapid publication will, after notification of the author(s), be treated as regular papers. Regular papers may be very short.

Standard nomenclature as used in the scientific 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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JOURNAL OF FOOD PROCESSING AND PRESERVATION Vol. 8, No. 1

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