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PRESERVATION OF ENZYME INACTIVATED
LAMB SLICES EFFECTS OF FREEZE DESICCATION
AND LOW DOSE IRRADIATION

B. Y. K. Rao, et al

Army Natick Laboratories
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Dehydration	10		6	10		
Irradiation Dosage	10		6	10		
Low	0					
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Flexible Food Packaging				9		
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PRESERVATION OF ENZYME INACTIVATED LAMB SLICES - EFFECTS OF
FREEZE DESICCATION AND LOW DOSE IRRADIATION

by

B. Y. K. Rao, G. W. Shultz and E. Wierbicki

Project 1G762713A033

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Food Laboratory
U. S. ARMY NATICK LABORATORIES
Natick, Massachusetts 01760

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FOREWORD

This report describes the work that was performed to preserve lamb meat by a combination process of freeze-desiccation (partially dehydrated) and low dose irradiation. The objective of this combined process was to develop a shelf-stable product that is capable of storage up to 90 days at ambient temperatures.

The combined process reduced the microbial count in the samples irradiated at lower pasteurizing doses. However, at the higher pasteurizing doses in the range of 1 Mrad, no difference between samples of partially dehydrated and non-dehydrated slices were found. Additional studies are not warranted because of the time and expense of the freeze-desiccation process.

The work covered by this report was performed under Project 1G762713A033, Radiation Preservation of Foods. These investigations were performed by Mr. Gary Shults and Dr. E. Wierbicki, Food Laboratory, US Army Natick Laboratories, and Mr. B.Y.K. Rao, Bhabha Atomic Research Center, P&T Div., Bombay, India. Mr. Rao was a visiting scientist at the US Army Natick Laboratories on a research fellowship sponsored by the International Atomic Energy Agency.

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ABSTRACT

The purpose of the present study was to develop a shelf stable product, having satisfactory texture and flavor characteristics, by combination treatments of heat, freeze-desiccation (partial dehydration) and low dose radiation.

The following conclusions were drawn from the ambient temperature (21 to 25°C) storage studies conducted over a period of ninety days on the enzyme inactivated, freeze-desiccated lamb slices, vacuum packed in flexible pouches and irradiation with 0.25, 0.5 and 1.0 Mrad doses at 0 to 4°C.

1. A dose of 0.25 Mrad was not found to be adequate for the ambient temperature storage of flexible packed, enzyme inactivated, non-freeze desiccated lamb samples.
2. A dose of 0.5 and 1.0 Mrad is optimum for the ambient temperature storage of enzyme inactivated, partially dehydrated lamb slices and a dose of 1.0 Mrad for the non-dehydrated, enzyme inactivated lamb slices.
3. At an irradiation dose of 1.0 Mrad, the non-dehydrated lamb slices scored slightly higher than the partially dehydrated lamb slices in technological panel evaluations for the sensory characteristics and preference.

INTRODUCTION

Radiation sterilization of foods has been successfully employed to produce meats and meat products suitable for storage at ambient temperatures (21 to 25°C) (Heiligman 1965), Wierbicki, et al 1970). However, at the higher doses necessary to insure sterility, deleterious effects on the sensory characteristics are found. In order to minimize these effects on the odor and flavor characteristics, the entire radiation process has to be carried out at temperatures in the range -35° to -40°C.

Low dose radiation at refrigerated temperature in combination with dehydration has been shown to be effective in extending the shelf life of enzyme inactivated meats stored at ambient temperatures up to 4 months (Kumta and Sreenivasan, 1970, Gore et al 1970). Agarwal et al (1972) reported the stability of pre-cooked dehydro-irradiated lamb and chicken stored at ambient temperatures over a ninety day storage period. Additionally, it was reported that doses in the range of 0.5 to 1.0 Mrads, in combination with partial dehydration, were adequate to produce a shelf stable item. However, the conventional dehydration methods, such as freeze drying and air drying, appeared to affect the preference of the partially dehydrated items due to adverse changes in color and textural characteristics.

This study was made to obtain a stable product having satisfactory texture and flavor attributes by using a combination of heat, partial dehydration and low dose irradiation.

METHODS AND MATERIALS

Raw Material and Enzyme Inactivation

The raw material used in these experiments was USDA graded Choice leg of lamb. The lamb legs were deboned and trimmed of all surface and intramuscular fats and cut into 225 to 450 gram chunks. The lamb meat was mixed in a mechanical mixer with 0.75% NaCl and 0.4% sodium tripolyphosphate (TPP). The meat was stuffed into size 11 fibrous, regenerated cellulose casings and formed into loafs (88 x 125 mm) in stainless steel metal cages. The meat was enzyme inactivated using dry heat until an internal temperature of 71° - 76°C was obtained. The meat was cooled overnight at 5°C and then sliced into 6 mm slices.

Freeze-Desiccation

One half of the meat slices were subjected to partial dehydration using silica gel under vacuum. The silica gel was mesh size 6-16 ctr., 88 grade 0.05. The slices were frozen to -19°C , wrapped into a polyethylene net material and covered with the silica gel in a can size 603 x 700. Dehydration was carried out under a vacuum of 500 mm of mercury.

The duration of the holding time to achieve 50-60% moisture level in the lamb slices was determined by weight loss data and by a moisture balance (Table 1 and 1A). It was found that 90 minutes of freeze desiccation was sufficient for the process.

Packaging

The lamb slices were packaged in flexible pouches, 92.5 x 175 mm. The pouch consisted of three layers: medium density, polyethylene as the food contactant, aluminum foil (middle layer) and mylar (outside layer). Pouches were heat sealed under a vacuum of 650 mm Hg.

Irradiation

Both the non-dehydrated and dehydrated samples were irradiated at 0.25, 0.5 and 1.0 Mrad (+ 25%) at $-30^{\circ}\text{C} \pm 10^{\circ}\text{C}$. Dose rate of the source was 4.25×10^4 rads per minute.

Microbiological analysis

Total plate counts were carried out by standard methods. Aerobic microbial contents were determined using TSY agar medium. Anaerobic microbial content was determined using Brewers anaerobic jars.

Chemical Analysis

Samples were analyzed by AOAS procedures for moisture, fat, protein, salt, phosphate, pH and ash. Free fatty acids, peroxide and TPA values were determined for each sample. After initial determinations, free fatty acid and peroxide values were discontinued due to the low quantities of fat found in the lamb.

Organoleptic Testing

The samples were subjected to organoleptic analysis using a seven member trained technological panel. All irradiated samples used in sensory evaluations were tested to ascertain the absence of preformed toxin of *Cl. botulinum*, types A and B, by the modified method recommended by the U. S. Dept. of Health, Education and Welfare (1969). Samples were tested for color, odor, flavor, texture and preference after storage of 0, 30, 60 and 90 days at ambient temperature. The ratings were made for the sensory evaluation on a nine-point intensity scale with 1 being none and 9 being extreme in intensity. Preference ratings were made on the hedonic scale where 9 is "like extremely" and 1 being "dislike extremely". On the hedonic scale, any score 5.0 or above, neither "dislike or like" is considered to be acceptable. Statistical analysis was performed using an analysis of variance and Duncan's multiple range test (Duncan, 1955).

Samples were heated in a steam table prior to serving to the test panels.

RESULTS AND DISCUSSION

It was observed that the pouches containing the non-dehydrated, 0.25 Mrad irradiated lamb slices that were stored at ambient temperatures for four days were swollen indicating microbial activity. The head space gases were analyzed using a combined GC/MS instrument. A 2.5 ml of head space gas was removed from the pouches with a hypodermic syringe and injected on to a tris column at -100°C . The column was temperature programmed at $6^{\circ}\text{C}/\text{min}$ and the helium flow rate was 6.5 ml/min to separate compounds. The compounds were then introduced into the ion source of a TOF mass spectrometer. The compounds were identified from the mass spectra. The compounds were mainly carbon dioxide, hydrogen sulfide, methyl mercaptan, and polymethyl sulfides. The presence of these sulfur containing compounds indicated intense activity of microorganisms.

Table 2 lists the results of the microbiological analysis of both dehydrated and non-dehydrated lamb slices irradiated at 0.25, 0.5, and 1.0 Mrad. The data show that at a 0.25 Mrad dose, the microbial count of the non-dehydrated sample is high in both aerobic and anaerobic population. At 0.5 and 1.0 Mrad these counts were reduced to <100 per gm. In the partially dehydrated samples, <100 per gm. were found in the samples irradiated at the 3 doses. After 30 days storage (Table 3), the 0.25 Mrad non-dehydrated samples were found to be very high in microbial counts, a further indication

that a 0.25 Mrad dose is not sufficient to retard the growth of microorganisms in the non-dehydrated lamb slices. Samples irradiated at 0.5 and 1.0 Mrad, both partial dehydrated and non-dehydrated, were found to contain less than 100 per gm. in aerobic and anaerobic counts. It was concluded that 0.5 Mrad were sufficient to retard the growth of microorganisms in the non-dehydrated lamb slices, while a dose of 0.25 Mrad was sufficient for the partially dehydrated slices.

Organoleptic results of three technological panels on the lamb slices immediately after irradiation processing are shown on Table 4. The partially-dehydrated samples (A) were rated in the acceptable range (5.0 or above) but were found less acceptable than the irradiated non-dehydrated reference sample, 4.7 Mrad at -30°C . This difference was found to be significant in the case of the dehydrated 0.5 and 1.0 Mrad samples. The most pronounced difference in the partial dehydrated samples as compared to the non-dehydrated reference sample was the discoloration. This is a result of the drying effects on the surfaces of the lamb slices. The non-dehydrated samples (B) were found more acceptable than the 4.7 Mrad reference sample. A definite dose effect can be seen in the preference ratings for these samples. As the dose increased, the product acceptability decreased from 6.7 at 0.25 Mrad to 5.5 at 4.7 Mrad. This difference in preference ratings was found to be significant at the 5% level.

A third test compared 1.0 Mrad partial-dehydrated and non-dehydrated samples against the 4.7 Mrad reference samples and frozen controls. Again the 1.0 Mrad, non-dehydrated sample scored higher than the other irradiated samples. Also, the non-dehydrated frozen control was rated higher than the partial dehydrated frozen control. However, all samples were found acceptable and statistical differences were not found for any of the ratings.

After one month's storage at 21°C , all of the pouches containing non-dehydrated slices irradiated at 0.25 and one half of pouches containing non-dehydrated slices irradiated at 0.5 Mrad were found to be swollen. It was determined that these groups were not suitable for organoleptic testing. No swollen pouches were noted in the 1.0 Mrad non-dehydrated and the partial-dehydrated irradiated samples. Results on Table 5 again show that all the samples scored in the acceptable range. No statistical differences were found in the ratings for the sensory characteristics and preference ratings of the partially dehydrated samples. Comparison of the 1.0 Mrad samples and the 4.7 Mrad reference sample yielded comparable results as the zero time storage. The 1.0 Mrad non-dehydrated sample was found more acceptable than the other two irradiated samples. Again the non-dehydrated frozen control samples scored higher than partial-dehydrated frozen control. Both frozen control samples were significantly better than the 4.7 Mrad sample.

The organoleptic data shows that the partial-dehydration affects the organoleptic characteristics of the lamb slices and lowers the acceptability. However, all samples were rated in the acceptable range. Both the partial-dehydrated and the non-dehydrated samples received higher acceptability ratings than the 4.7 Mrad reference sample, although the differences were not significant.

Table 6 tabulates the data of a duplicate experiment in which samples were tested after storage of 0, 30 and 90 days at 21°C. At zero time storage, the 0.5 Mrad sample, partially dehydrated, had a significantly higher intensity for discoloration which resulted in the sample being rated unacceptable (score 4.85). However, at 1 and 3 months storage this intensity for discoloration was not found, indicating that product variability accounted for differences. At 1 and 3 months storage, no significant differences could be found for any of the sensory characteristics and preference ratings of the samples. All samples were still rated in the acceptable range after 90 days storage.

Proximate analysis was run on all samples of dehydrated and non-dehydrated lamb slices. Free fatty acids and peroxide values could not be run because of the low fat content of the samples. TBA values were obtained but data varied and did not fall into any definite trends that could be associated with treatment given to the individual samples.

The microbiological analysis for the duplicate experiment are shown on Table 7. The analysis showed that all samples had <100 per gm. for the 90 days storage period. Again it was concluded that a dose of 0.25 Mrad was sufficient to retard microbial growth for up to 90 days in samples of partially dehydrated lamb slices.

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TABLE I

Holding time for freeze-desiccation of lamb slices

Time (min)	Initial ¹ Weight	Final Weight	Weight Loss	% Loss
60	191 gms	159 gms	32 gms	16
90	186	147	39	21
180	190	141	49	26
240	191	133	58	30
300	185	126	59	32

¹Total weight of 81 mm slices of lamb

TABLE 1A

Moisture content of lamb slices¹

Time (minutes)	Calculated % Moisture	Moisture ² Balance Determination
60	52	58
90	49	51
180	43	49
240	42	44
300	34	43

¹As determined by weight loss data and moisture balance.

²Determined on Ohaus Moisture Balance 175 – watts – 20 minutes.

TABLE 2

Microbiological analysis of non-dehydrated and partially dehydrated
lamb slices – 0 month storage

Sample	Aerobic Count*	Anaerobic Count*
A 0.25 Mrad	<100/gm	<100/gm
A 0.5 Mrad	<100/gm	<100/gm
A 1.0 Mrad	<100/gm	<100/gm
A Frozen Control	< 10/gm	< 10/gm
B 0.25 Mrad	8×10^4 /gm	8.5×10^5 /gm
B 0.5 Mrad	<100/gm	<100/gm
B 1.0 Mrad	<100/gm	<100/gm
B Frozen Control	<100/gm	< 10/gm

*Incubation: +21°C

A – Partially Dehydrated Samples

B – Non-Dehydrated Samples

TABLE 3

Microbiological analysis of non-dehydrated and partially
dehydrated lamb slices - 1 month storage

Sample	Aerobic Count*	Anaerobic Count*
A 0.25 Mrad	<100/gm	<100/gm
A 0.5 Mrad	<100/gm	<100/gm
A 1.0 Mrad	<100/gm	<100/gm
A Frozen Control	<100/gm	<100/gm
B 0.25 Mrad	11.2×10^6 /gm	1.1×10^7 /gm
B 0.5 Mrad	<100/gm	<100/gm
B 1.0 Mrad	<100/gm	<100/gm
B Frozen Control	<100/gm	<100/gm

*Incubation: +21°C

A - Partially Dehydrated Samples

B - Non-Dehydrated Samples

TABLE 4

Organoleptic evaluation of partially dehydrated lamb slices
 - 0 storage time
 Sensory characteristics

Samples	Discoloration	Off Odor	Irradiation Flavor	Off Flavor	Mushiness	Friability	Preference
A 0.25 Mrad	3.7	2.3	1.8	2.2	1.3	1.6	5.8
A 0.5 Mrad	3.6	2.0	1.4	3.0	1.3	1.6	5.1*
A 1.0 Mrad	2.0	1.8	1.8	2.0	1.4	1.6	5.4*
4.7 Mrad, -30°C	1.8*	1.4	1.8	1.4*	2.0	2.0	6.4
A Frozen Control	2.0*	2.0	1.8	2.6	1.6	1.8	5.80
B 0.25 Mrad	1.5	1.1	1.0	1.9	1.4	1.7	6.7
B 0.5 Mrad	1.5	1.5	1.4	1.4	1.6	1.7	6.4
B 1.0 Mrad	1.2	1.4	1.2	1.6	1.5	1.9	6.4
4.7 Mrad, -30°C	1.5	2.4	2.0	2.5	2.0	2.6	5.5*
B Control	1.1	1.0	1.0	1.4	1.2	1.6	7.9
A 1.0 Mrad	1.7	1.3	1.3	1.8	1.4	1.7	6.1
B 1.0 Mrad	1.7	1.0	1.1	1.4	1.4	2.0	6.7
4.7 Mrad, -30°C	1.7	1.7	1.8	1.8	1.7	2.2	6.0
A Control	1.6	1.3	1.0	2.0	1.3	1.6	6.4
B Control	1.4	1.3	1.0	2.0	1.4	1.8	7.0

*Significantly different (5% level) from other samples

A - Partial Dehydrated Samples

B - Non-Dehydrated Samples

TABLE 5

Organoleptic evaluation of partially dehydrated lamb slices —
1 month storage

Sample	Sensory Characteristics						
	Discoloration	Off Odor	Irrad Flavor	Off Flavor	Mushiness	Friability	Preference
A 0.25 Mrad	3.1	1.6	1.3	2.3	1.3	1.4	5.4
A 0.5 Mrad	2.4	1.8	1.6	2.0	1.4	1.7	6.2
A 1.0 Mrad	1.7	1.6	1.1	2.4	1.7	1.8	6.2
A Frozen Control	1.1	1.1	1.0	1.0	1.3	1.1	7.3
A 1.0 Mrad	1.4	1.4	1.3	2.6	1.1	1.7	5.6
B 1.0 Mrad	1.7	1.3	1.4	2.0	1.6	1.6	6.4
4.7 Mrad, -30°C	2.3	2.1	2.6	1.1	1.7	1.7	5.4
A Frozen Control	1.3	2.1	1.1	3.1	1.0	1.3	6.7*
B Frozen Control	1.1	1.1	1.0	1.4	1.1	1.3	7.1*

*Significantly different (5% level) from other samples

A — Partial-Dehydrated Samples

B — Non-Dehydrated Samples

TABLE 6

Organoleptic evaluation of partially dehydrated lamb slices

Samples	Sensory Characteristics						
	Discoloration	Off Odor	Irrad Flavor	Off Flavor	Mushiness	Friability	Preference
0 Month Storage							
A 0.25 Mrad	3.1	1.6	1.3	2.3	1.3	1.4	5.4
A 0.5 Mrad	4.0	1.7	1.7	2.1	1.3	1.4	4.8*
A 1.0 Mrad	2.4	1.8	1.6	1.8	1.4	2.3	6.1
B 1.0 Mrad	2.3	1.8	1.4	1.7	1.4	2.3	6.0
A Frozen Control	2.0	1.4	1.7	2.0	1.0	1.0	5.5
1 Month Storage							
A 0.25 Mrad	1.8	1.8	2.8	2.6	1.6	2.0	5.0
A 0.5 Mrad	1.8	2.3	3.0	2.0	1.3	1.4	5.3
A 1.0 Mrad	1.6	2.1	2.8	2.0	1.6	1.6	5.1
B 1.0 Mrad	2.0	2.3	2.5	2.1	2.3	1.7	5.3
A Frozen Control	2.0	1.4	1.7	2.1	1.3	1.7	5.5
3 Month Storage							
A 0.25 Mrad	1.4	1.9	1.3	2.0	1.0	2.0	5.6
A 0.5 Mrad	1.8	1.4	1.1	2.3	1.1	2.0	5.6
A 1.0 Mrad	1.6	1.3	1.3	1.8	1.1	2.0	5.4
B 1.0 Mrad	1.4	1.3	1.1	1.8	1.3	1.8	5.7
A Frozen Control	1.3	1.3	1.1	2.0	1.1	1.8	6.1

*Significantly different (5% level) from other samples.

A - Partially-Dehydrated Samples

B - Non-Dehydrated Samples

TABLE 7

Microbiological analysis of partially dehydrated lamb slices

Samples	Storage Time	Aerobic Count	Anaerobic Count
A 0.25 Mrad	0	<100/gm	<100/gm
	50 days	<100/gm	<100/gm
	90 days	<100/gm	<100/gm
A 0.5 Mrad	0	<100/gm	<100/gm
	50 days	<100/gm	<100/gm
	90 days	<100/gm	<100/gm
A 1.0 Mrad	0	<100/gm	<100/gm
	50 days	<100/gm	<100/gm
	90 days	<100/gm	<100/gm
B 1.0 Mrad	0	<100/gm	<100/gm
	50 days	<100/gm	<100/gm
	90 days	<100/gm	<100/gm
A Frozen Control	0	<100/gm	< 10/gm
	50 days	< 10/gm	< 10/gm
	90 days	< 10/gm	< 10/gm