

TECHNICAL REPORT

72-42-FL

**DEVELOPMENT OF RADIATION STERILIZED  
FISH ITEMS**

by

Louis J. Ronsivalli

U.S. Department of Interior,  
Fish and Wildlife Service,  
Bureau of Commercial Fisheries,  
Technological Laboratory,  
Gloucester, Massachusetts

Contract No: Project Order No. 66-88

February 1972

**UNITED STATES ARMY  
NATICK LABORATORIES  
Natick, Massachusetts 01760**



**Food Laboratory**

**FL-153**

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## FOREWORD

The availability of shelf-stable, highly acceptable seafood items for use in military feeding systems is considered a necessity. Radiation processing, or "cold" sterilization as it is frequently called, has the potentiality of yielding products that have good military utility, good storage stability, and good acceptability. Therefore, research to develop process criteria that can be used to produce irradiation sterilized seafoods is under way.

The work covered in this report was performed by U. S. Department of Interior, Fish and Wildlife Service, Bureau of Commercial Fisheries, Technological Laboratory, Gloucester, Massachusetts under Project Order No. 66-88 during the period from 14 December 1966 to 13 March 1968. It represents an investigation on the development of edible coatings for the stabilization of the physical structure of radiation sterilized fish fillets, and an investigation on the effects of variation in processing conditions and selected additives for the prevention of browning of fish fillets during storage.

Mr. L. Ronsivalli was the Project Officer and Official Investigator and Robert J. Learson the Chief Collaborator in the research work for the Bureau of Commercial Fisheries. The U. S. Army Natick Laboratories' Project Officer was Dr. Fred Heiligman and the Alternate Project Officers were Messrs. George Giddings and Gary Shults, all of the Food Laboratory.

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## ABSTRACT

Research was carried out on edible coatings to stabilize the physical structure of radiosterilized fish fillets and on additives to prevent browning of these products during room temperature storage. Ground or blended fish flesh which has binding properties upon heating was tested as a batter material to coat fillet portions prior to breading. It was found that a coating composed of 50 percent ground flesh and 50 percent methylcellulose solution (2 percent) in conjunction with a normal slow-browning breading material greatly enhanced the structural and storage stability of the fillet. This coating which seals itself during the frying process was considered highly acceptable by the taste panel and proved to be shelf stable through nine months' storage at room temperature.

Although this coating resulted in acceptable products immediately after irradiation and up to one month at room temperature storage, marked discoloration (browning) of the flesh was observed after 2-3 months' storage, resulting in an unacceptable product. Research indicates that the browning is nonenzymatic although the availability of reactants may be the result of enzymatic activity. Attempts to eliminate browning in prefried radiosterilized fillets have failed. Of all the additives tested, only bisulfite treatment affected the browning reaction. However, shelf life extension was not significant.



# DEVELOPMENT OF RADIATION STERILIZED FISH ITEMS

## INTRODUCTION

The primary problems involved in the production of acceptable radio-sterilized fish fillets concern the structural breakdown of the fillet during processing for enzyme inactivation and "browning" of the product during room temperature storage. This report covers research carried out on edible stabilizing coatings for retaining the structural integrity of the fillets and on determining the effects of heat treatment, irradiation, various additives and packaging environment on the browning of these products during room temperature storage. The effects of different treatments for proteolytic enzyme inactivation were also studied.

## EXPERIMENTAL

### Irradiation Facility and Procedures

All irradiation was carried out at the Marine Products Development Irradiator at Gloucester, Massachusetts. (1)

During this reporting period experimentation was primarily concerned with cod and haddock fillets. All products were irradiated to receive a minimum dose of 4.5 megarads (maximum = 5.9 megarads) at a temperature of  $-80^{\circ}\text{C}$ . (1)

### Structural Stability of Fillets

As reported in a previous progress report (1), extreme physical damage occurring during enzyme inactivation treatments and storage resulted in products which were unrecognizable or unusable as fish fillets. Because of this, experimentation was carried out on methods to improve the structural stability of the fillets.

To increase the uniformity of the geometry of the fillets and to facilitate handling during processing, sections were cut from the thickest portion of each fillet with a 2-1/2" diameter cookie cutter. Three portions of this size were packed in a standard No. 2 can after processing to inactivate enzymes.

Previous research has shown that an edible stabilizing coating is necessary to minimize the physical damage to the fillets occurring during the enzyme inactivation process. (1) Although commercial breading and batter coating materials eliminate physical damage during frying, these coatings absorb moisture from the fish flesh during room temperature storage, and, subsequently, the product breaks apart, becoming unusable as a fillet or portion.

The requirements for a suitable coating for radiosterilized fillets are extremely strict. The coating must 1) cover the product completely, 2) seal itself during heat treatment, 3) remain stable during enzyme inactivation, 4) adhere to the fillet surface, 5) remain intact during room temperature storage and subsequent handling (reheating and serving), and 6) have good quality characteristics (flavor and odor). Various materials were tested for suitability. These were as follows:

1. Methylcellulose in various concentrations was used by itself as a batter material and also as an additive to commercial starch batter materials.

2. Myvacet (acetylated Monoglyceride) was tried as a dip coating. Since this material must be kept hot (82°C) during use, attempts were made to carry out the coating process and the enzyme inactivation simultaneously.

3. Edible shellac, a coating material commonly used for candies was tried as a dip and as a sprayed coating in a variety of concentrations.

4. Ground or blended fish flesh which has binding properties was also used to coat fillets prior to breading. This material was tested in a number of different formulations. Various dilutions of the fish slurry, by itself or in combination with methylcellulose solutions, starch batter material, and gelatin were all tested as batters for fried breaded products.

The same procedure was used to test the various coating formulations. The portions of fillet were first dipped in the batter materials and rolled in a commercial "slow-browning" breading. Three portions were then heated in a microwave oven for 30 seconds and deep-fat fried for 60 seconds. The portions were vacuum packed in No. 2 C-enamel cans using CO<sub>2</sub> as a flush gas. Finally, the cans were cooled to -80°C and irradiated. Some cans were stored at about 1°C, and some were stored at room temperature (about 22°C).

To determine the best coating material the products were evaluated by a trained panel immediately after irradiation and, if acceptable, on a monthly basis during room temperature storage. In all cases visual examinations of the product were carried out upon removal from the can to determine the condition of the fillet prior to serving. Samples stored at room temperature were toxin tested at the U. S. Army Natick Laboratories prior to taste testing.

#### Discoloration of Radiosterilized Fillets

As previously reported (1) marked discoloration was observed in radiosterilized fillets after 3-6 months of storage at room temperature, a slight yellowing becoming visible during the first month, progressing to a deep brown after 4 to 5 months of storage.

A number of different additives were tested as possible browning inhibitors. Bisulfite, which is commonly used as a browning inhibitor in other food products (2), was tested first. Haddock fillets were dipped in a sodium bisulfite solution (2.5% sodium bisulfite in 1% ascorbic acid solution) for various times prior to processing. Treated fillets were processed both as coated and uncoated portions. The coated portions were preheated in a microwave oven and fried as described above, and the uncoated products were canned (vacuum + CO<sub>2</sub> flush) and steam, retorted to an internal temperature of 77°C for enzyme inactivation. In other experiments designed to eliminate any questions regarding penetration of the salt, the bisulfite solution was added directly to ground fish flesh which was canned and retorted prior to irradiation. These products were evaluated visually immediately after irradiation and on a monthly basis during room temperature storage.

Because of visible damage to the fillet occurring during the treatment of fillets with the "sulfiting" solution (presumably acid attack), experiments were carried out using a sodium bicarbonate solution as a neutralizing dip. Both coated and uncoated portions were prepared using this sequence of treatments prior to enzyme inactivation and irradiation.

As reported by Furia (3), butyl hydroxy anisole (BHA) ethylenediaminetetracetic acid (EDTA) and alum all have browning inhibiting properties. To determine the effects of these additives on the discoloration of radiosterilized fillets, experiments were carried out involving the use of various combinations of these additives both as a dip for fillet portions and as a direct additive to ground flesh. All these products were evaluated visually immediately after irradiation and on a monthly basis during room temperature storage.

A series of experiments was carried out to determine the effects of air, irradiation and heat treatment on the browning reaction in both whole fillets and ground flesh.

To determine whether or not a more complete elimination of air from the package would have an effect on discoloration, the fish fillets were processed as follows:

Cut portions of the fish fillets were coated with a fish slurry-methocel batter material, rolled in breading, microwave-treated for 30 seconds and fried for 60 seconds at 204°C prior to packaging and irradiation. Experiments using BHA (butyl hydroxy anisole) as a dip prior to the coating process, and CO<sub>2</sub> as a flush gas were carried out. Samples were prepared using various dip times in BHA and multiple flushes with CO<sub>2</sub>. Products were visually evaluated immediately after irradiation and on a monthly basis at room temperature storage.

It was observed that there were some differences in the "browning" rate among products heated to different internal temperatures during

enzyme inactivation. To determine the effects of heat treatment on the browning reaction rate a number of experiments were carried out using different methods of heating prior to irradiation.

1. Fillet portions were coated (fish slurry + methocel + breading) microwave-oven treated for 30 seconds and fried for either 40 seconds, 60 seconds, or 90 seconds prior to irradiation. These heating procedures resulted in internal temperatures of 55°C, 77°C, or 93°C respectively in the fillets. The products were canned (vacuum + CO<sub>2</sub> flush) and irradiated (4.5 Mrads at -80°C) and stored at room temperature.

2. Whole fillets were microwave treated to internal temperatures of 24, 49, or 77°C prior to irradiation. These products were vacuum-packed (CO<sub>2</sub> flush) in cans, irradiated at -80°C and stored at room temperature.

3. Whole fillets and ground fish flesh were retorted in cans to various internal temperatures (49°, 77°, or 121°C) prior to irradiation. Half of these samples was vacuum-packed and half was air-packed to determine the effects of oxygen on the samples. Nonirradiated controls were prepared for samples heated to 77°C or 121°C.

All of these products were evaluated visually on a monthly basis during room temperature storage. Reflectance measurements were made with a MEECO differential colorimeter on each sample in order to follow the rate of browning during storage and the reflectance of the samples is reported as the Munsel Value. (4)

Experiments designed to test the possibility of extracting fish flesh with water to remove reactants were initiated. Whole fillets as well as ground flesh were extracted for 30 minutes in both warm (38°C) and cold (8°C) tap water. The leached flesh was vacuum-packed in cans (CO<sub>2</sub> flush) irradiated at -80°C (4.5 Mrads) and placed at room temperature storage.

### Enzyme Inactivation

During research on enzyme inactivation it was found that the texture of the product was enhanced greatly by reducing the heat process to a minimum. Samples of coated portions of haddock and cod fillets were enzyme inactivated using shorter heat treatments than those required for complete enzyme inactivation. (5) Using a combination microwave-frying treatment, samples were heated to internal temperatures of about 60°C prior to canning and irradiation. Other samples were prepared by deep-fat-frying to internal temperatures of about 49°C. These samples were stored at room temperature and organoleptically evaluated monthly.

A new procedure for enzyme inactivation using microwave heating was tested. In previous work complete enzyme inactivation by microwave treatment proved unfeasible because of extreme physical damage resulting in the

product. It was found that this damage could be minimized when the microwave treatment was carried out holding the product under water pressure. This method looked promising for a product like halibut steaks which, when enzyme inactivated by conventional heating, dried out to the point of being inedible as far as texture was concerned. (1) Halibut steaks treated with 0.1% BHA for 1 minute were vacuum-packed in mylar (polyethylene coated polyester) pouches and placed in a sealed plastic container filled with hot tap water (65°C). The entire package was then treated for 5 minutes in the microwave oven, resulting in internal temperatures in the order of 60°C. These samples were sealed in a second pouch and irradiated at -80°C (4.5 Mrads).

## RESULTS AND DISCUSSION

### Structural Stability of Fillets

In general both visual and organoleptic evaluations showed that ground or blended fish muscle used as a batter material for coating fillets greatly enhanced the structural and storage stability of the fillet. All the other materials tested had major drawbacks. The methocel and the methocel-starch batter mixtures functioned well during processing but were unstable during storage. The Myvacet dip, because of the high temperature involved, had a tendency to break up the fillet during the dipping process and the fillet could not be held long enough in the dip to effect enzyme inactivation. When the fillet was treated only long enough to be coated, the coating could not withstand heat processing for enzyme inactivation. The edible shellac became very brittle and cracked from the stress of freezing and was also extremely bitter tasting.

All the coating formulations using ground or blended fish functioned extremely well and were resistant to breakage during processing, storage, and subsequent handling during serving. Their adherence properties were excellent, especially when the surface of the fillet was scored with a fork or similar instrument prior to the coating process. In most cases, the flavor and texture of the coatings were judged to be excellent.

The best coatings as determined by both visual and organoleptic examination was a mixture of equal volumes of ground fish (the fish was ground for 60 seconds in a silent cutter) and 2% methocel. This coating was less brittle than the others and had better stability during room temperature storage. Added starch batter material or gelatin to this mixture appeared to harden the coating but lessened the moisture resistance of the coating during storage.

Irradiated products coated with the fish slurry-methocel batter and a slow browning breading were scored 6-7 (like slightly-like moderately) on

a 9-point hedonic scale, by an expert panel, immediately after irradiation and after 2 months of storage at room temperature. The products became unacceptable between 2 and 3 months of storage due to marked browning of the flesh, although they remained intact and stable through 9 months of storage with no visible signs of structural damage. In all cases the product could be removed from the can and handled without being damaged.

#### Discoloration of Radiosterilized Fillets

It appears that browning in stored radiosterilized fish flesh occurs mainly through the Maillard Reaction (sugar-amino acid condensation). Fish flesh has the necessary reactants, and according to the literature (6) (7), room temperature storage meets the conditions necessary for the reaction to take place.

Although the evidence is not conclusive as yet, results indicate that oxygen is not a factor in browning. The coated fillets all browned slowly after 2 to 3 months of storage at room temperature, regardless of treatment. Neither multiple flushes with CO<sub>2</sub> nor the addition of BHA to the product affected the browning rate.

Of all the additives tested, only sodium bisulfite appeared to affect the browning reaction. Products treated with bisulfite retained a more acceptable color through two months' storage at room temperature. However, after two months these products tended to yellow rather than brown and were considered unacceptable by the trained panel.

Fillets dipped in bisulfite solution for 1 minute prior to processing retained a lighter color for a longer time than did the nontreated fillets. Increased dip times did not appear to reduce the browning any further and only resulted in increasing physical damage to the fillet. The use of sodium bicarbonate as a neutralizing dip did not eliminate the acid attack on the fillet and appeared to reduce the effect of the bisulfite. Products treated with bisulfite, coated with fish slurry-methocel batter, and foiled in slow-browning breadings were scored 6.5 on the 9-point acceptance scale by an expert panel immediately after irradiation.

Bisulfite solution added directly to ground flesh (1500 ppm) prior to canning and retorting appeared to reduce browning even after three months of storage at room temperature. The panel in general considered the color of this product more acceptable than the color of similar products not treated with bisulfite. After four or five months of storage this product became unacceptable in appearance due to a yellowish discoloration. In all cases, however, the yellow color was not as objectionable as the brownish discoloration. All the other additives tested had no visible effect on the rate of browning either in coated fillets or as direct additives to ground flesh.

Neither irradiation nor the presence of air in the package appeared to have any effect on the rate of browning in any of the products. The only

factor that affected the color of the flesh to any measurable extent was the type of heat treatment given to the product. Figure 1 shows the effect of process temperature on the color of whole fillets and ground flesh retorted in cans (vacuum-packed) to various internal temperatures prior to irradiation. Figure 2 shows the effect of process temperature on the same products air-packed in cans prior to retorting. In all cases products heated to 120°C were only marginally acceptable in regards to color and did not change appreciably during storage. The same products heated to 77°C were acceptable in color initially and remained acceptable through five months of storage. The product heated to 49°C were the highest in initial acceptability but in general were considered only marginally acceptable after five months. The products receiving no heat treatment were unacceptable after one month of storage and browned rapidly thereafter. These results indicate that heating to 77°C is the optimum heat treatment and produces the most acceptable color initially with the least amount of browning during storage.

The results of the retort processed products contrast markedly with the results obtained from products treated by means of the microwave oven (Figure 3). In all cases the latter browned very rapidly regardless of the process temperature becoming unacceptable after two months of storage at room temperature.

Coated products microwave treated and fried for various times in corn oil (190°C) were comparable to the microwave treated products with regard to the rate of browning of the flesh (Figure 4). In all cases, these products were considered unacceptable after two months' storage. The bisulfite treated samples were considered significantly more acceptable by the panel than the nontreated samples. However, the use of this additive again resulted in a yellowish color rather than a brown color. The panel's preference for the yellowed product over the browned product was most likely due to the difference in color. The reflectance values, however, were not significantly different between sulfited and nonsulfited samples (green filter).

The results indicate that ground fish flesh can be leached with water with some reduction in browning. Whole fillets leached in the same manner, however, were not significantly different from nonleached fillets after two months of storage.

#### Enzyme Inactivation

In all cases a reduced enzyme inactivation treatment increased the product acceptability markedly immediately after irradiation. Products microwave treated and fried to internal temperatures of 60°C were considered more acceptable than those products heated to 77°C. Bisulfited products fried for one minute in corn oil at 190°C (just enough to seal the coating) to internal temperature of 49°C were judged not significantly

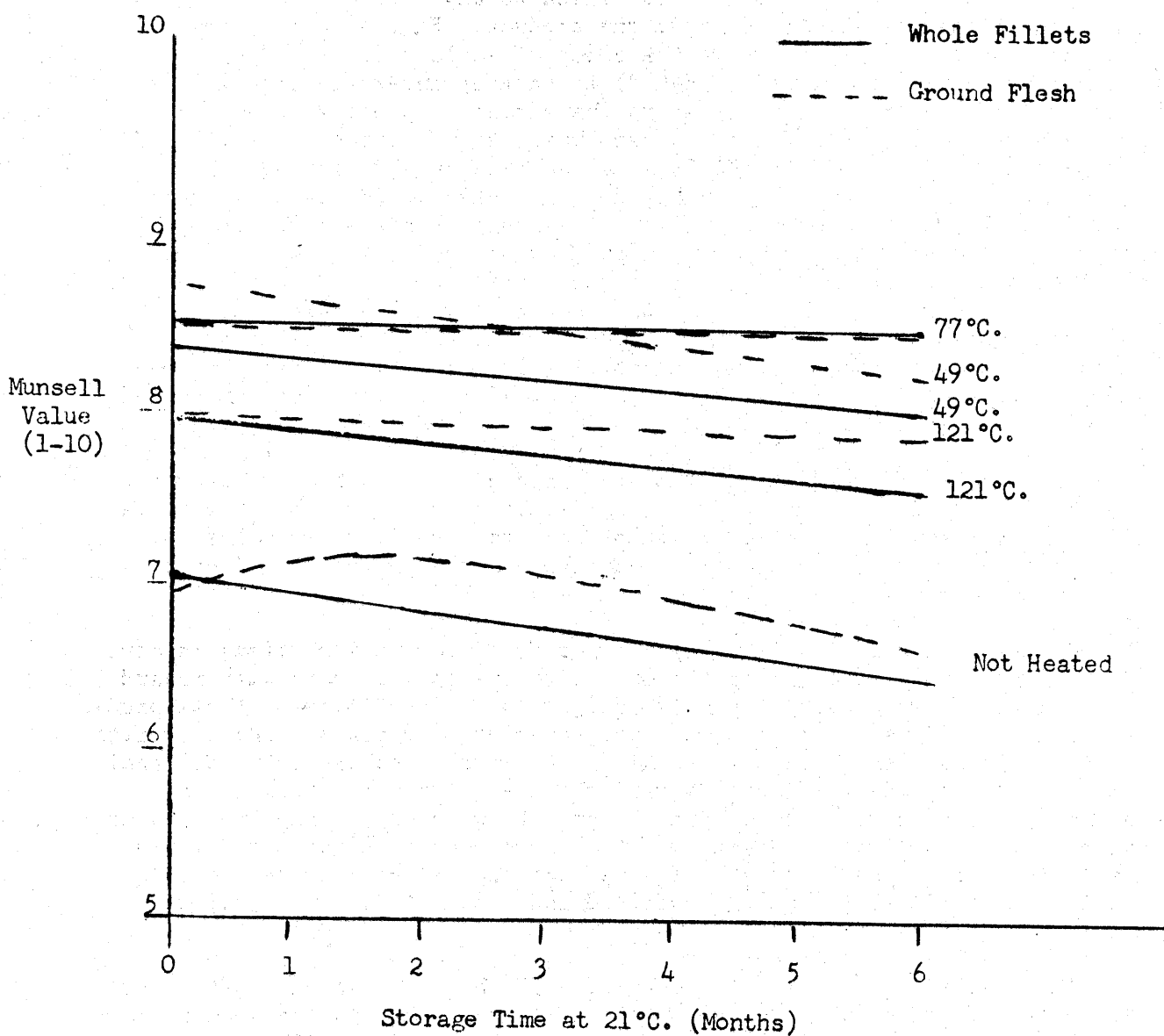


Figure 1 - The effect of room temperature storage on the color of radiosterilized whole fillets and ground flesh, vacuum packed in cans and retorted to various temperatures prior to irradiation (4.5 Mrads at  $-80^{\circ}\text{C}.$ ).



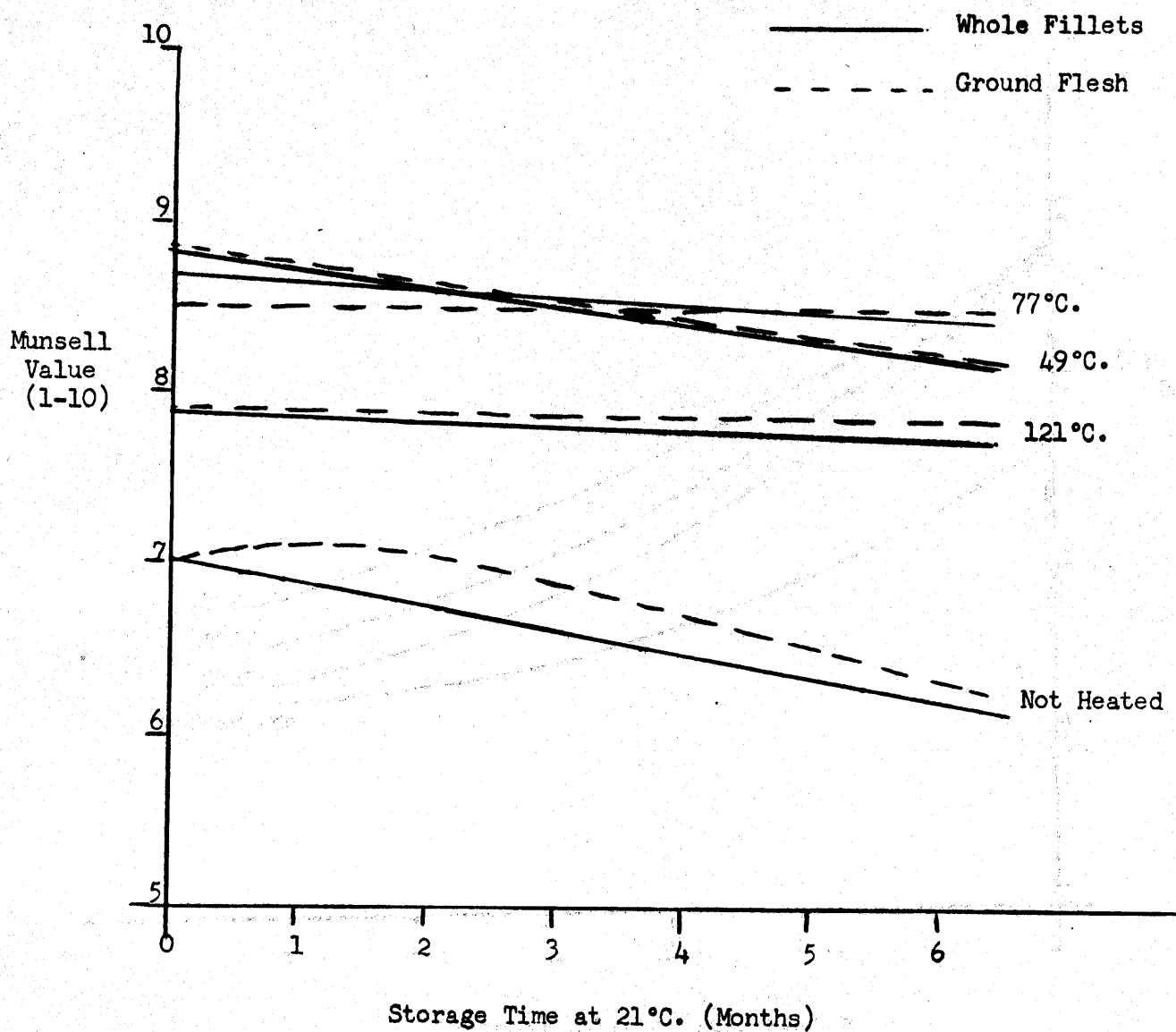


Figure 2 - The effect of room temperature storage on the color of radiosterilized whole fillets and ground flesh, air packed in cans and retorted to various temperatures prior to irradiation (4.5 Mrads at  $-80^{\circ}\text{C}.$ ).

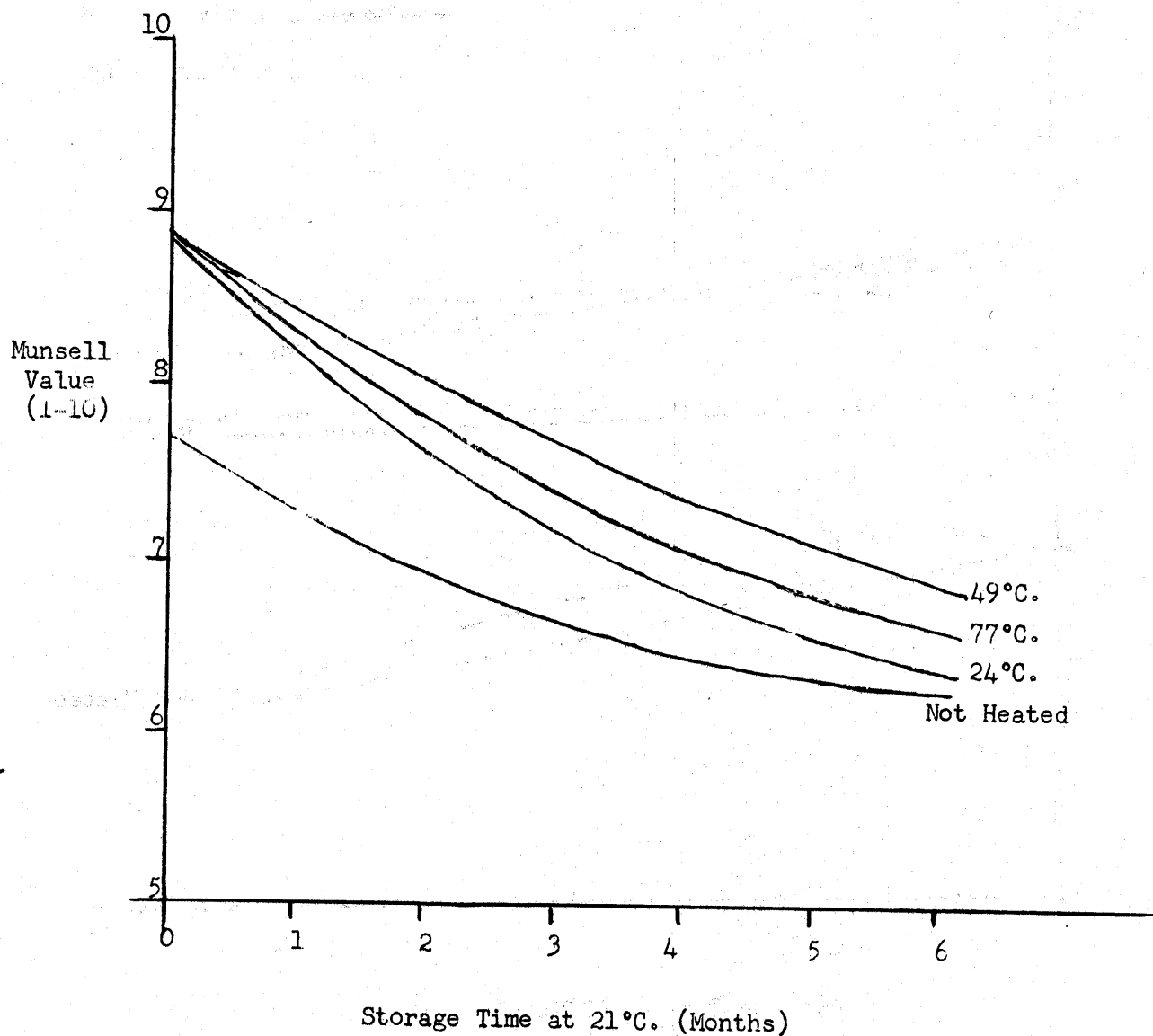


Figure 3 - The effect of room temperature storage on the color of radiosterilized fillets, microwave oven treated to various temperatures prior to canning (vacuum packed) and irradiation (4.5 Mrads at  $-80^{\circ}\text{C}.$ ).

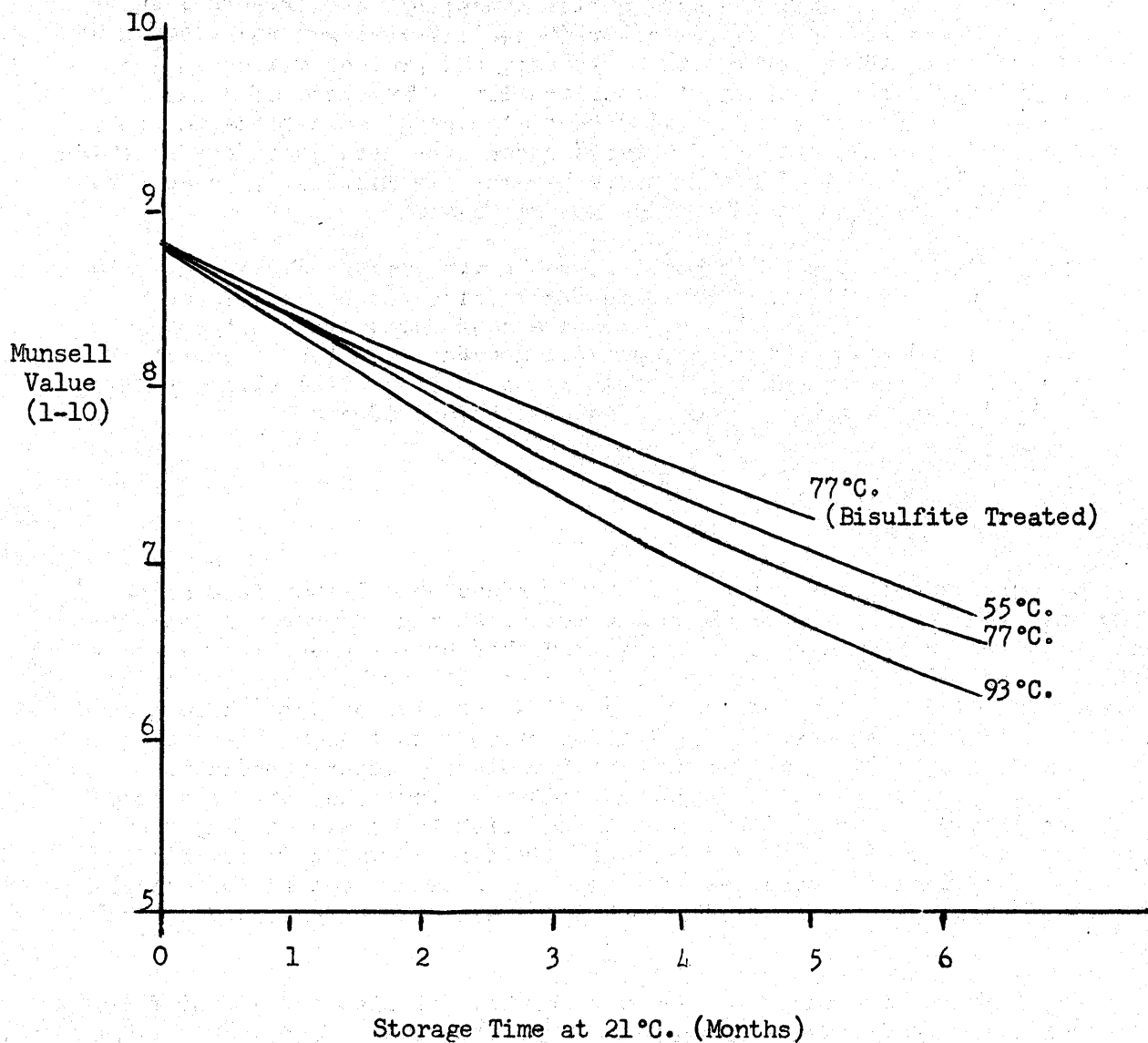


Figure 4 - The effect of room temperature storage on the color of radiosterilized fillets, microwave oven treated (30 sec.) and fried to various internal temperatures prior to canning (vacuum packed) and irradiation (4.5 Mrads at  $-80^{\circ}\text{C}.$ ).

different from the frozen control immediately after irradiation. These were the best results obtained for initial acceptability. Figure 5 shows the effect of storage on both the color (Munsell Value) and the acceptance of this product. After two months of storage the product was considered unacceptable primarily because of discoloration. The slope of Munsell Value vs. time correlates very well with the slope of acceptance vs. time. In all cases, texture, odor, and outward appearance were considered highly acceptable. The flavor of the products became only marginally acceptable because of the development of a chicken-like flavor.

The initial acceptance of halibut steaks was increased substantially when the enzyme inactivation procedure was carried out in the microwave oven under water pressure. The steaks were considered excellent as far as appearance and texture were concerned. However, a slight irradiation flavor was present reducing the overall acceptance to "like slightly-like moderately." Evaluations are continuing on these products.

#### CONCLUSIONS

Concerning the production of a highly acceptable fishery item prepared from fish in the fillet form, browning during storage represents the primary problem. During exploratory research this browning problem was only lightly considered because of structural stability problems in the fillet. Development of a structurally stable coating material prepared from ground flesh and advances in enzyme inactivation methods to improve texture resulted in a highly acceptable product immediately after irradiation. Subsequent storage at room temperature, however, indicates that decreased acceptability due to marked browning of the fish flesh was the major problem to be solved. To date, attempts to eliminate browning in prefried products have failed. Bisulfite treatment appeared to retard "browning" somewhat. However, there was no appreciable extension of acceptable shelf life.

Experiments have been carried on with whole fillets and ground flesh to determine the effects of irradiation, heat treatment, air, and various additives on the rate of browning. Results indicate that there are no significant differences between air and vacuum-packed products and between irradiated and nonirradiated products as far as browning is concerned. In some cases ground flesh did not brown as rapidly as whole fillets. The only additive that affected the browning rate was bisulfite. The color of these products, however, was still unacceptable.

The only variable that significantly affected the browning reaction to any extent during a 5-6 month storage at room temperature was the type of heat treatment and the final product temperature. Products which had been deep-fat-fried and microwave over-treated prior to canning, browned

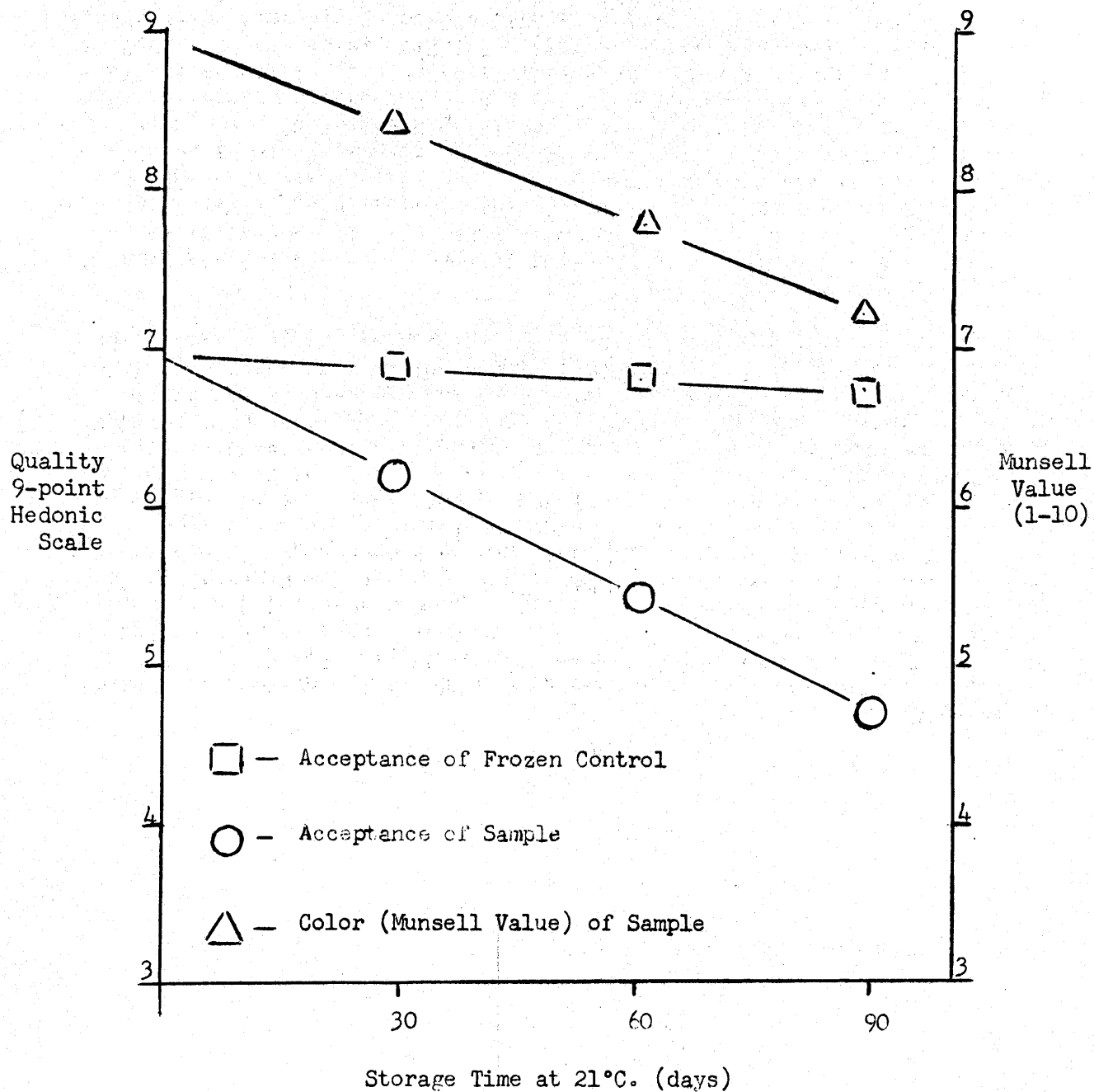


Figure 5 - The effect of room temperature storage on the panel acceptance and the color of prefried (bisulfited) radiosterilized fillet sections.

much more than products retorted in cans to the same process temperature. The only products considered acceptable in color after six months' storage at room temperature were those retorted in cans to an internal temperature of 77°C, the minimum temperature needed for enzyme inactivation. Samples retorted to 121°C for fifteen minutes were less acceptable as were samples retorted to 49°C. The results indicate that the reaction involved is non-enzymatic (Maillard condensation). However, the effect of heat treatment on browning suggests that the availability of reactants (ribose, ribose 1-phosphate, etc.) may involve enzymes. (7) The differences in browning rates among various types of heat treatment may indicate a time-temperature effect as well as a possible environmental effect. In general it appears that a more basic study of the browning reaction is necessary before a solution to this problem can be found.

Enzyme inactivation procedures attaining product temperatures lower than those reported for complete proteolytic enzyme inactivation in general resulted in less drying of the product and a much more acceptable texture. Little or no proteolysis was observed in any of these products. However, as reported above, browning resulted in unacceptable quality after two or three months of storage. Products stored up to 9 months at room temperature showed no visible signs of autolytic spoilage outside of browning. Microwave heating in a sealed container filled with water to exert pressure on the product shows promise as a method for enzyme inactivating such products as steaks or scallops. Product temperatures in the order of 60°C can be attained very quickly (five minutes) and uniformly with a minimum of physical damage to the product. Whether or not microwave treatment under water pressure will be significantly different from normal microwave treatment with regard to browning of the product remains to be seen.

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13. ABSTRACT Research was carried out on edible coatings to stabilize the physical structure of radiosterilized fish fillets and on additives to prevent browning of these products during room temperature storage. Ground or blended fish flesh which has binding properties upon heating was tested as a batter material to coat fillet portions prior to breading. It was found that a coating composed of 50 percent ground flesh and 50 percent methylcellulose solution (2 percent) in conjunction with a normal slow-browning breading material greatly enhanced the structural and storage stability of the fillet. This coating which seals itself during the frying process was considered highly acceptable by the taste panel and proved to be shelf stable through nine months' storage at room temperature.  Although this coating resulted in acceptable products immediately after irradiation and up to one month at room temperature storage, marked discoloration (browning) of the flesh was observed after 2-3 months' storage, resulting in an unacceptable product. Research indicates that the browning is nonenzymatic although the availability of reactants may be the result of enzymatic activity. Attempts to eliminate browning in prefried radiosterilized fillets have failed. Of all the additives tested, only bisulfite treatment affected the browning reaction. However, shelf life extension was not significant.			

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