

AN ABSTRACT OF THE THESIS OF

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Title: EFFECT OF WASHING ON THE QUALITY, DESIRABILITY
AND STABILITY OF DRUM-DRIED MACHINE SEPARATED
ROCKFISH FLESH

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The effect of washing machine separated rockfish flesh on composition, quality and desirability was investigated.

Washing machine separated rockfish flesh resulted in a 36.88% loss of solids. The greatest reductions were found in the ash (80.32%) and lipid (65.13%) levels in the washed flesh. Most of the sarcoplasmic proteins were lost during washing and only 77.29% of the protein (N X 6.25) was recovered.

Washing the machine separated fish flesh did not appreciably alter the amino acid composition. The content of essential amino acids of the drum-dried treatments was similar to casein and this was reflected by the high protein efficiency ratios compared to the casein reference. The mineral composition of the unwashed machine separated flesh was similar to that of the fillet. Washing the machine

separated flesh greatly altered the mineral composition in the flesh. The levels of calcium, magnesium, barium, strontium, boron, and manganese were not appreciably altered by washing. Phosphorus, potassium, and sodium levels were reduced while the iron, copper, zinc and chromium levels were increased in the washed flesh.

The rapid formation of trimethylamine (TMA) in the drum-dried rockfish fillet and unwashed machine separated flesh treatments may be related to the development of off-odors and off-flavors formed during storage at 30°C. The over-all desirability of bland wafers consisting of 10% drum-dried washed flesh increased slightly during storage but was not appreciably higher than the wafers containing the drum-dried fillet and unwashed fish flesh. Over-all desirability may not be directly linked to the formation of trimethylamine (TMA) since washing drastically reduced the level of trimethylamine oxide (TMAO) in the washed flesh and the subsequent formation of trimethylamine (TMA) during storage at 30°C.

The much higher levels of malonaldehyde found in the drum-dried washed machine separated rockfish flesh may implicate the oxidation of the lipids as being responsible for the lower over-all desirability of the washed flesh. The rapid formation of malonaldehyde may also indicate that naturally occurring antioxidants are leached from the flesh during the washing process.

Effect of Washing on the Quality, Desirability and Stability
of Drum-dried Machine Separated Rockfish Flesh

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EFFECT OF WASHING ON THE QUALITY, DESIRABILITY
AND STABILITY OF DRUM-DRIED MACHINE
SEPARATED ROCKFISH FLESH

INTRODUCTION

Some species of fish are caught which are not fully utilized due to some specific undesirable characteristics (31, 34, 38, 49, 52). Miyauchi (40) pointed out the problem associated with black rockfish (Sebastes sp.). The fresh fillet has acceptable taste, but when stored in the frozen state, rancidity and toughening develop rapidly in the dark muscle.

Attempts have been made to use fish muscle in the minced form, although it has been reported to have poor palatability and mouth feel (12, 40, 49). Teeny and Miyauchi (52), have tried to improve the quality of the minced blocks made from rockfish by using sugar and monosodium glutamate to enhance the flavor; salt and sodium tripolyphosphate to serve as a binder, and antioxidants to inhibit rancidity. An extension in the storage time was achieved with this modified block.

Other workers have tried to blend the minced rockfish with other species in an attempt to improve its quality. Babbitt (13) suggested a combination of shad and rockfish. The acceptance and shelf-life stability of the minced rockfish was improved by mixing it

with shrimp (14). Steinberg (49) stated that rockfish could be reduced to a smooth, white paste with low fat and high protein which can be used for making spreads with different flavors.

Miyauchi et al. (41) have tried washing the minced rockfish used for the modified, minced blocks. They found that the washed modified blocks were better in color and had a significantly higher flavor score than the unwashed modified blocks. The washing also gave an improvement in the odor and the storage life.

Considering what has been reported above, this investigation was carried out to determine whether washing machine separate rockfish flesh would improve:

1. The quality of drum-dried material as determined by color, proximate composition and over-all desirability.
2. The nutritional value of the drum-dried material as determined by protein efficiency ratio (PER), amino acid and mineral composition.
3. The storage stability of the drum-dried material determined by sensory evaluation, amine content, trimethylamine oxide (TMAO) and trimethylamine (TMA), 2-thiobarbituric acid content (TBA No.), and microbial load.

REVIEW OF LITERATURE

Machine Separated Minced Fish Flesh

The demand for good quality protein to feed the world's population cannot be over-emphasized. Seafoods are known to be an excellent source of protein and polyunsaturated lipids (31). In order to meet the demand, the total utilization of undeveloped fisheries and industrial fisheries as sources of food, has been suggested by several workers.

Miyauchi and Steinberg (38) pointed out the waste of potential edible protein encountered by present fishing techniques and conditions. Furthermore, there are no reserve stocks for human food. To overcome the problem of total utilization of fisheries resources, machines for removing edible muscle from bone and skin have been introduced (31).

The most commonly used machine consists of a rubber or plastic belt driven tightly against the outside of a rotating perforated stainless steel or cast iron drum. The perforations of the drum are of uniform size and different sizes are available ranging from three to seven millimeters in diameter. The belt and the drum move in the same direction, but at different speeds, producing a shearing action. The muscle which is torn off, passes through the

perforations into the drum, while the skin and bones are scrapped off the outside of the drum. The separated flesh is removed from inside the drum. Several types of these meat/bone separators are now available, and by adjusting the belt and the drum, the yield of the flesh can be varied with differing quality (15, 38).

The yield of edible flesh has been found to be high for dressed samples of several species. Miyauchi and Steinberg (38) using a wide variety of Pacific Ocean fish obtained a yield of 37-60% of the whole weight of the fish compared to a 25-30% yield by filleting operations. Crawford et al. (21) have obtained yields of 40.4-54.4% whole weight of Dover and English sole. They have also found that planking the fish before introduction into the deboner reduces the quantity of skin in the separated flesh. For black rockfish species, Miyauchi et al. (41) have obtained yields of 47.4% and 46.4% at normal pressure. The machines have also been used to recover significant amounts of flesh from fish frames produced during filleting operations (12, 20, 32, 38).

King and Carver (31) have suggested that species which are under-utilized as a result of unsuitability for filleting because of the size and anatomy are suited for mechanical processing. Steinberg (49) has emphasized the great flexibility of processing the minced flesh obtained.

In the production of "kamaboko", a Japanese elastic fish cake

made from ground fish muscle, starch is used as a thickening agent to improve succulence and texture. Sugar, salt, and monosodium glutamate are used for flavoring (45). For fish sausage, starch is also used as well as salt and spices for flavor. Sodium or potassium nitrite is used for coloring tuna and whale meat. Chemical preservatives, such as 5-nitro-2-furfural semi-carbazone, Nitro-furyl-acrylamine and sorbic acid are also employed.

Miyauchi et al. (39) reported the use of additives such as glucose, tripolyphosphates, and sucrose or sorbitol to maintain the white color of Surimi, a semi-processed wet fish protein. Teeny and Miyauchi (52) have used sugar and monosodium glutamate to enhance the flavor of fish blocks produced from machine separated black rockfish. Salt and sodium tripolyphosphate were also used to partially solubilize the muscle protein and to act as a binder. Antioxidants, butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) were also incorporated to inhibit rancidity in these modified fish blocks.

It has also been suggested that the minced fish gives opportunity for development of products which are unidentifiable as to particular species and will appeal to the customer on the basis of their attributes (31).

Quality of the Minced Fish Flesh

Despite the high yield of the minced flesh as compared to the fillet, the material has a number of associated problems. The flesh is visibly colored with blood pigments and clots have been found in some cases (32, 35). The texture, which has been described as a granular mouth feel, has been attributed to the disruption of the cell during machine separation and the straining process (12, 35, 46). Fishy flavors and odors, nerve tissue, fibers, pieces of skin and membrane have also been noted (32, 46).

Babbitt et al. (12, 21) found that the muscle from English sole frames was discolored and distinctly off-flavored. This correlated with the low over-all acceptance. The problems were attributed to substances which were pressed from the backbone, the high level of lipid and the pieces of skin that were not completely removed. The discoloration has been found to increase with the tension exerted by the belt (17).

Choy et al. (17) working with Gulf Coast trawl fish, obtained minced flesh that was contaminated with bone fragments when the machine was operated at maximum pressure. This has been found to be the case by other workers (26, 32).

The moisture level and the fat content of the separated material have been found to be significantly higher than the intact fillet for

several species of fish (12, 20, 21, 41). The water holding capacity is also higher and the ash level usually lower (36).

The quality of the protein as measured by the protein efficiency ratio (PER) changes little with machine separation. Choy et al. (16) suggested on the basis of their experimental protein efficiency ratio (PER) determination, that there is no significant difference between the fillet and the minced flesh of the same or different species of fish when proper deboning operation is employed. They found that the net protein ratio (NPR) determined on spot croaker was high. The amino acid score (CS) was similar for the species used and not different from values of edible portions of a variety of known fish.

Babbitt et al. (11, 12) have reported a lower protein content for deboned muscle, especially from the frames. The trimethylamine (TMA) levels were found to be two to three times higher in the deboned material than in the intact fillet. The formation of malonaldehyde (TBA No.), which has been shown to be an index of rancidity in fishery products (59), was rapid in the deboned muscle. They also found that formation of dimethylamine (DMA) and formaldehyde (FA) occur more rapidly in the minced flesh than the intact fillet of Pacific hake during frozen storage.

Recent work with frozen stored minced flesh of Atlantic cod and pollock showed a rapid loss of protein solubility at -5°C (22). This was reportedly due to the presence of kidney tissue which caused the

formation of dimethylamine (DMA) and formaldehyde (FA) from trimethylamine oxide (TMAO). The decrease in extractable protein nitrogen (EPN) has been related to increased toughness in the cooked flesh and also a decline in acceptability by taste panels. This is accompanied by a loss in water holding capacity which can cause decreases in functional properties during subsequent processing. Recent work on the functional properties done on species of North Carolina fin fish indicates differences and suitability for preparing gel-type products (26).

Crawford et al. (20) have shown that the machine separated fish flesh have favorable mineral compositions.

Effect of Washing on the Minced Fish Flesh

It was demonstrated by Japanese workers in the production of "kamaboko" that washing the separated minced flesh with chilled water improved the color, odor, and elasticity of the processed product. One part of the weight of the flesh to seven parts of water is used for the washing which is carried out three to five times. The washing operation removes the blood, flesh pigments, mucus and fat in the minced flesh (45). This operation is called "bleaching" by the Japanese and it is a step in the production of Surimi, fish sausage and ham (1, 39, 50, 51).

Okada (44) stated that the repeated washing does not only remove

the soluble substances and fat from the minced fish, but also gives a high concentration of myosin responsible for the elastic nature of "kamaboko." The water soluble substances are supposed to protect the myosin from forming a network structure essential for the jelly nature of "kamaboko."

Several workers have modified this washing process in the handling of the minced flesh. King (33) found that washing the flesh as it is being separated from the skin and bones offer several advantages. It leaches a significant amount of blood before the blood clots become insoluble in water. The amount of water used was in ratio of two to three times the weight of the minced flesh obtained. It was presumed that water soluble components were washed out as well. Ten percent of the total solids was assumed lost during the washing operation. It was also found that washing improved the appearance of the minced blocks produced.

Miyauchi (40) stated that washing the minced flesh of black rockfish (Sebastes sp.) improved the quality. The washing removed water soluble constituents that catalyze the undesirable changes in flavor and appearance during frozen storage. Some lipids and lipoproteins were also removed, thus lessening the rancidity problem. This washing operation was in the ratio of five parts of ice water to one part by weight of the fish flesh, and it was carried out once. It was also found that the washed minced flesh had a longer storage life,

nine to twelve months, than the unwashed which became rancid in three months. It was concluded that washing the minced black rockfish is necessary in order to obtain a consistently higher quality product.

Kudo et al. (37) in preparing "kamaboko" from washed and unwashed minced flesh of six species of rockfish found that the "kamaboko" made from the washed flesh was better in quality. The washing improved the color, flavor and the flexibility of the "kamaboko." The chilled water used was also effective in improving the gel forming capacity. The washing removed blood, fat, small pieces of skin and any "fishy" odors.

Recent work with minced flesh by Dingle and Hines (22) showed that trimethylamine (TMA) in the washed, minced flesh is lower than the unwashed flesh and that the pH of the latter increased during frozen storage. The dimethylamine (DMA) production in the washed minced flesh was found to be less than in the unwashed material. This would be the case if the trimethylamine oxide (TMAO), normally present in the fish muscle and degraded to dimethylamine (DMA) and formaldehyde (FA), had been removed by the washing. They showed also that the loss in the extractable protein nitrogen (EPN) is greater in the unwashed minced flesh as compared to the washed and that the unremoved blood decreases the extractable protein nitrogen (EPN) in some species. Thus showing that washing is necessary to

improve the quality of the minced fish flesh.

Product Development

Several product applications have been proposed for the minced flesh, as a means of utilization, both as a supplement and in its own entity. Amano described the use of the minced flesh in making fish sausage by the Japanese (1). Advantage is taken of the loss of identity of the species of fish during the chopping and grinding operations. The product is also modified by use of food additives and spices.

The Japanese have for several years, prior to the introduction of fish sausage, been producing an elastic fish cake, "kamaboko" from a number of different species of fish (45, 51). The minced flesh which has been ground with starch, sugar, salt and monosodium glutamate (MSG) is processed in many ways including steaming, boiling and deep-fat frying. The appearance and flavor of the "kamaboko" is varied by the addition of various ingredients and the form is also modified to give different named products: "Chikuwa," "Hanpen," and "Sumaki" (51).

The production of "Surimi," a semi-processed wet fish protein from the minced flesh, led to the expansion of the "kamaboko" industry in Japan (39). The ground, washed fish flesh, is frozen into rectangular blocks after the addition of various additives. The

"Surimi" can then be made into any of the several Japanese minced fish products.

Carver and King (15) elaborated on the possible uses of the minced flesh. It was suggested for use as fish cakes consisting of 50% whiting flesh, 49% potatoes, 1% dehydrated onions, salt and pepper. The fish cakes were judged as acceptable as those prepared from cod, although the color was darker. The possibility of making frankfurters, fish sausage, croquettes, fish loaf, and jelly roll from the flesh of cod and haddock filleting wastes was mentioned. They further suggested the production of a fish protein concentrate with a low mineral content for supplementation of low cost protein foods for various feeding programs.

The color of the minced fish has been used to advantage by King and Flick (32) in the production of beefish patties. They mixed minced fish with ground beef (25-50% substitution) and seasoning in varying amounts. The sensory evaluation indicated that it was as acceptable as all-beef patties, in appearance, odor, flavor and texture. Further work along this line demonstrated that mixing beef and the colored minced fish is economically justifiable.

Also, the mixture of ground beef and minced flesh was tested in a variety of main dishes, such as lasagna, meat/potato burgers, spanish rice, American chop suey, beef stroganoff, meat balls, and meat loaf. It was found that these main dishes could be

successfully made with the mixture, particularly that made from ground fish visibly colored with pigments.

Various other substitutions have been tried. Babbitt et al. (10) incorporated 20% shrimp into minced fish and found improvement in texture, flavor and desirability. Recent work by these authors with the fish-shrimp portion made from black rockfish (Sebastes melanops) showed that increasing the amount of shrimp in the portion improved the acceptance and the shelf-life stability of the minced flesh. This was related to the decrease in the formation of malonaldehyde and peroxides (14).

Patashnik et al. (46) proposed the potential use of the machine separated minced flesh for high protein snack items. The process tested produced a bland, flavored spread base with smooth texture, appealing white color and excellent stability. It was suggested that modifications in the process could lead to production of dips or fried snack items.

Microbiological Spoilage

King (33) working with minced fish for the production of blocks for fishsticks, found that the lowest total aerobic plate counts in the minced blocks were in the order of 10^3 to 10^4 per gm. The counts were found to be dependent on the freshness of the material used and

that they were lowered by the washing process. Similar results have been obtained by other workers (12).

The potential for bacterial contamination of the minced flesh is increased due to the handling during production. This has been pointed out by several workers and the need for good sanitary practices has been emphasized (1, 13, 45, 49). Choy et al. (17) have reported the need for critical quality control both in the pre- and post-landing operations involved in minced flesh production. The potential for bacterial contamination from human sources should be taken into consideration.

The Japanese have, for several years, worked on the bacterial deterioration of machine separated minced flesh products such as "kamaboko" and fish sausage. Kimata (27) have classified the types of deterioration associated with "kamaboko" on the basis of the external and internal spoilage. The external spoilage varies from slimy to opaque bacteria growth and this is a reflection of the constituent of the "kamaboko." The amount of starch, presence and absence of sugar, together with the humidity dictates the type of bacteria that will grow on the surface.

Aerobic species such as Micrococcus, Serratia, Flavobacterium and Achromobacter have been found on the surface of "kamaboko" that does not contain sugar and Bacillus spp. have been associated with the contamination of the refined starch used. Counts of 10,000-

50,000 spores per gm have been made and were mostly of Bacillus megatherium. There was no relationship between the counts and the grade of the starch (30).

The spoilage of the fish sausage have been found to differ from that of "kamaboko" due to the difference in the ingredients used and the nature of the product. The fish sausage spoils by softening and high counts were obtained in the softened portion (56). The spoilage has been attributed to Bacillus circulans, a facultative anaerobe. The softening was due to the disintegration of the starch granules and an increase in the fatty acid content was noted (53).

Yokoseki (57, 58) has reported the variation in the types of bacteria in the fish sausage depending on the internal temperature reached during processing. Micrococci were found in products heated to an internal temperature of 70°C and Bacillus sp. in fish sausage made with potato starch and heated to an internal temperature of 75-90°C. Few spore-formers were detected in the products reaching 75°C. There was also changes in the microbial flora of the product heated to 70°C as opposed to negligible changes in those heated above 75°C. The fish sausage containing sugar had mainly Streptococcus faecalis and Leuconostoc dextranicum. Clostridium tertium was also found in the fish sausage that had no sugar added.

MATERIALS AND METHODS

Rockfish

Black rockfish, Sebastes flavidus were obtained from the Pacific Shrimp Company, of Warrenton, Oregon, one day post catch, for the chemical analyses and the storage test.

The fish samples for the protein efficiency ratio (PER), amino acid and mineral analyses, and the preliminary desirability test consisted of different species of rockfish obtained from the New England Fish Company of Warrenton, Oregon. The species were identified as Sebastes melanops, 12.5%; Sebastes flavidus, 12.5%; Sebastes rubrivinctus, 25%; Sebastes pinniger, 25%; and Sebastes proriger, 25%.

A Yanagiya fish separator, (Yanagiya Machinery Works, Ltd., Yamaguchi Pref., Japan), was used for the preparation of the minced fish. The whole fish was washed to remove mucus and soil, and then headed and gutted by hand. The kidneys were removed and washed off together with blood. The fish was then planked and passed through the separator once at near maximum belt tension. A stainless steel drum with four millimeter diameter openings was used.

Some of the washed whole fish were filleted by hand and ground

by passing through a Triumph grinder with eight millimeter diameter openings.

The separated minced flesh was washed twice using four parts of water to one part of fish, both on a weight basis. The wash water was cooled to 36^oF with ice and mixed with the fish by stirring in a stainless steel Hobart mixer, for five minutes at low speed. The material was then centrifuged in a Philips Drucker centrifuge operated at the speed of 2,500 rpm (1,300 x g) at 4^oC for 5 minutes.

The fillet, the washed, and the unwashed flesh were dried using a Stokes drum-drier, and then ground in a Thomas mill. The fresh and the dried samples were vacuum sealed in air-tight, moisture proof, vinyl and polyester bags, and stored at -40^oF prior to chemical analysis.

The dried samples for the storage test were held in an incubator at 30^oC for the eight weeks storage period. Sensory evaluation for "off" odor, "off" flavor and over-all desirability as well as the determinations for malonaldehyde, trimethylamine oxide (TMAO), trimethylamine (TMA), and total aerobic plate count were conducted at 0, 2, 4 and 8 weeks after preparation of the samples.

Color Determination

The color of the dried material was determined using a Hunter Color and Color Difference meter. The readings were made against

a Standard Panel 31, Ivory, with the following readings:

Rd	56.4	L	75.1
a	-1.3	a	-1.3
b	23.3	b	23.1

Proximate Analysis

The analysis for moisture, protein (crude), lipid, and ash, were carried out according to the "Official Method of Analysis" of the Association of Official Agricultural Chemists (2, 3, 4, 5, 6, 7, respectively).

Sensory Evaluation

To test the drum-dried material for "off"-odor, "off"-flavor and over-all desirability, wafers were prepared according to the procedure used for the evaluation of fish protein hydrolysate concentrate FPHC (19).

The ingredients consisted of flour, shortening (Crisco) and water at 36^oF in the following proportion:

<u>Ingredient</u>	<u>Grams</u>
Flour	280
Shortening	95
Water (36 ^o F)	103

Fisher's unbleached 100% whole wheat flour replaced the plain flour used in the original method to mask the color differences in the fillet, the unwashed, and the washed dried fish samples.

Preparation of Wafers

The shortening was cut into the flour and blended for approximately 90 seconds with a hand dough blender until the shortening was of small size. Ice water was added and tossed with a fork until all the flour was moist. The dough was worked by hand and formed into a smooth ball. The dough was then placed on an unfloured bread board and lightly rolled into 18 inch squares. All the samples were rolled to the same dimension to ensure uniform thickness. The edge of the dough was lifted and placed over the rolling pin and then a sheet of dough was gently lifted and transferred to an ungreased aluminum baking sheet. The dough was cut into one and a quarter square inch pieces. Each piece was pricked twice with a fork. They were then baked at 425^oF for eight to ten minutes or until the edge started to brown slightly. All the wafers were baked approximately 24 hours prior to panel testing and stored in tightly covered porcelain dishes until tested.

Taste Test

The dried fish samples of the fillet, the washed and the

unwashed flesh were substituted for the flour at the desired level for the appropriate test. Two wafers of each sample, together with a coded control and a known reference, were served at the same time in cups coded with three digit numbers. Twenty-five judges seated in individual booths were used for each taste panel. The wafers were evaluated for off-odor, off-flavor, and over-all desirability. The off-odor and off-flavor were scored using a nine point scale ranging from nine, none to one, foul. A nine point scale was also used for the over-all desirability varying from nine, very desirable to one, extremely undesirable. The mean scores and the least significant difference calculated at the 5% level were recorded.

Preliminary Desirability Test

A taste panel was conducted to determine the level of substitution of the fish sample for the storage trials. Levels of substitution of 5% to 25% were tested at daily intervals for each of the dried samples. Also, a direct comparison of the 10%, 15% and 20% substitution of each of the dried fish samples were made respectively.

Protein Efficiency Ratio Determination

The biological evaluation of the protein quality of the dried samples of the fillet, washed and unwashed fish flesh, was determined according to the A. O. A. C. official method of analysis (9).

Amino Acid Analysis

The amino acid content of the drum-dried fish samples with the exception of tryptophane was determined according to the method of Spackman et al. (47). The determinations were carried out by the Biochemistry Department of the Oregon State University.

Mineral Analysis

The determination of the mineral content of the drum-dried fish samples was performed according to the method of Christensen et al. (18).

Amines

Trimethylamine Determination

The method of Yamagata et al. (55), for the reduction of trimethylamine oxide (TMAO) to trimethylamine (TMA) was used to determine trimethylamine oxide (TMAO).

One ml of the 5% trichloroacetic acid (TCA) extract of the fresh sample and 0.5 ml of the extract of the dried sample were transferred to screw top culture tubes with teflon cap and made up to 2 mls with distilled water. One ml of titanium chloride (TiCl_3) was then added to the solution and mixed. The test tubes were then

placed in the water bath at 80°C for 5 min. After mixing, 1 ml of distilled water was added to bring the volume to 4 ml and the level of trimethylamine (TMA) was then determined.

Trimethylamine (TMA) was determined using the picric acid procedure of Dyer (22, 24) as modified by Murray and Gibbon (42, 43), where 45% potassium hydroxide (KOH) was substituted for saturated potassium carbonate (K_2CO_3). The absorbance was read at 410 nm (42, 43).

Trimethylamine Standard Preparation

Two grams of trimethylamine hydrochloric acid reagent (TMA-HCl) were weighed into a 100 ml volumetric flask. One ml of concentrated hydrochloric acid (conc. HCl) was added and the volume was made up with distilled water. Ten ml aliquots of the solution were taken and Kjeldahl nitrogen determinations were carried out. The concentration of the trimethylamine in the solution was determined using the formula:

$$\text{ml}_{H_2SO_4} \times N_{H_2SO_4} \times 1,000 \times \text{titre} = \mu\text{g TMA per ml}$$

Dilutions of the standard solution were made and the trimethylamine (TMA) concentration was calculated. The trimethylamine (TMA) determination was made on the standard dilutions and the

optical density readings recorded. A standard curve was then plotted from the values obtained.

Trimethylamine Sample Preparation

Ten grams of the fresh sample were diluted with 90 ml of 5% trichloroacetic acid (TCA) solution and 1 g of the dried sample with 99 ml of the same solution. The samples were homogenized with a Virtis blender, operated at the speed of 40 for 5 minutes, and then filtered using S and S white ribbon No. 588 (size 15) filter paper.

A 1:10 dilution of the fresh fillet and unwashed fish flesh extracts was made prior to the trimethylamine oxide (TMAO) determination.

2-Thiobarbituric Acid Method (TBA)

Malonaldehyde was determined by the 2-thiobarbituric acid procedure of Yu and Sinnhuber (59) with appropriate modifications (60). This involved the omission of the use of petroleum ether to clarify the red color produced. Instead a clear solution for spectrophotometrical analysis was obtained by centrifugation at 2000-3000 rpm. The preparation of the thiobarbituric acid reagent was simplified by using one gram of the reagent dissolved in 75 ml 0.1 N NaOH in a 100 ml volumetric flask, and made up to mark with distilled water. Five ml of the TBA solution was used in place of

the 6 ml suggested in the previous method and replaced by a buffer in the modified method. No separation of interfering substances was carried out, since Yu and Sinnhuber (60) have reported that very little interference occurs by substances such as carbohydrate in products composed entirely of fish.

The thiobarbituric acid number (TBA No.) defined as milligram of malonaldehyde per kilogram of sample, was calculated by the formula:

$$\frac{1/2 \text{ Absorbance (535 nm)} \times 46}{1.166 \times \text{Sample wt. (gm)}}$$

Microbial Analysis

A modification of the "Official Method of Analysis," of the Association of Official Agricultural Chemists for the aerobic plate count (8) was used to determine the total microbial content of the samples.

The modification involved using the 10 g of the sample and 90 ml of the diluent for the 1:10 dilution instead of the 50 g to 450 ml ratio in order to conserve samples. The subsequent dilutions were then made by using 1 ml of the previous dilution in 9 ml of the diluent. The plates were incubated at 30°C for 48 hours.

RESULTS

Yield of Rockfish

The yield of flesh from the fish separator presented in Table 1 was greater than from the filleting operation. Higher recovery was obtained using a single species of rockfish Sebastes flavidus. This higher yield of mechanical separated fish flesh confirms the results of other workers (15, 21, 31).

Table 1. Comparison of the yield of flesh from deboning and filleting operations.

Species	Treatment	Processed weight (kg)		Yield (%)
		Round	Flesh	
Black rockfish, <u>Sebastes flavidus</u>	Fillet	8.75	2.93	33.43
	Deboned	12.05	6.15	51.04
Mixed Rockfish Species ^a	Fillet	54.55	14.40	26.00
	Deboned	60.00	25.68	43.00

^aSpecies comprised of: Sebastes melanops, 12.5%; Sebastes flavidus, 12.5%; Sebastes rubrivinctus, 25%; Sebastes pinniger, 25.0%; Sebastes proriger, 25.0%.

It was found that the efficiency of the separation of the flesh from the bone and skin using the fish separator depended on the size of the fish. This is evident from the higher recovery of the single species, Sebastes flavidus, which consisted of medium size fish. There was a wide range of different sizes in the mixed species,

some of which were difficult to feed into the machine.

The flesh recovered from the fish separator contained blood clots, nerve fibers and fat tissues. Only a slight amount of skin remained in the recovered flesh. On washing, some of the fat tissue was recovered as scum on the surface of the wash water, which was visibly colored red. The latter was due to the wash water dissolving some of the blood clots.

Washing and Effect on Quality

The amount of material recovered from the washing of the mechanically separated fish flesh shown in Table 2 was greater than the original due to the increase in water content.

Table 2. Recovery of flesh from the washing operation.

	Processed weight (kg)	Recovered weight (kg)
1st wash	2.40	2.70
2nd wash	2.70	3.55

The moisture content of the washed flesh was greater than the unwashed flesh or the fillet (Table 3). King (32) has suggested means of reducing the amount of water in the material by use of screens (nylon mesh bag) and continuous centrifugation for complete recovery of solids. Also, there was a marked decrease in the ash, lipid and protein content of the washed flesh.

Table 3. Proximate composition of black rockfish, Sebastes flavidus.

Treatment	Percent ^a			
	Moisture	Ash	Lipid	Protein
Fresh fillet	78.68	1.15	4.52	19.66
Washed flesh	90.30	0.14	1.11	9.50
Unwashed flesh	77.31	1.06	4.69	18.17

^a Mean of duplicate samples.

The effect of washing on the machine separated fish flesh caused an increase in the protein content if the proximate composition of the washed flesh is compared to the unwashed flesh on a dry weight basis (Table 4). The lipid and ash levels of the washed flesh were comparatively lower than the unwashed flesh and the fillet. The values for the single rockfish species are slightly higher than the mixed species and this can be explained on the basis of the composite nature of the mixed rockfish material.

The data in Table 5 indicate that washing greatly improved the color of the machine separated flesh.

The "L" value, which is a measure of lightness, was the highest in the washed flesh and lowest in the fillet for black rockfish, Sebastes flavidus. The "a" value, which is a measure of redness, was higher for the unwashed flesh reflecting a much darker color than the washed flesh.

Table 4. Comparison of the proximate composition of drum-dried treatments of Sebastes flavidus and Sebastes species expressed on a dry weight basis.

Species	Treatment	Percent ^a		
		Ash	Lipid	Protein
Black rockfish	Fillet	4.79	12.02	83.19
<u>Sebastes flavidus</u>	Washed	1.49	8.09	90.41
	Unwashed	4.62	15.32	80.06
Mixed rockfish species ^b	Fillet	5.06	8.58	86.36
	Washed	1.67	8.43	89.89
	Unwashed	5.03	13.64	81.33

^aMean of duplicate samples.

^bSpecies comprised of: Sebastes melanops, 12.5%; Sebastes flavidus, 12.5%; Sebastes rubrivinctus, 25.0%; Sebastes pinniger, 25.0%; Sebastes proriger, 25.0%.

Table 5. Color determination of the drum-dried treatments using a Hunter Color and Color Difference Meter.

Species	Treatment	Hunter Color Values		
		L	a	b
	Ivory standard	75.10	-1.30	23.10
Black rockfish	Fillet	57.00	7.55	18.50
<u>Sebastes flavidus</u>	Washed	78.00	0.75	15.30
	Unwashed	58.00	9.20	17.65
Mixed rockfish Species	Ivory standard	75.10	-1.30	23.10
	Fillet	60.80	7.50	21.10
	Washed	73.80	1.20	17.10
	Unwashed	54.60	9.30	21.10

Little variation occurred in the Hunter values when the results for the mixed rockfish species were compared to the single rockfish species. The "a" values were slightly higher in each treatment with the exception of the fillet. The "b" value, which is a measure of yellowness, indicated that all the samples were a deeper yellow than the single species.

Sensory Evaluation

The preliminary taste panel evaluations of the drum-dried treatments were carried out at levels of substitution of 5% of 25%. Judgments for off-odor, off-flavor and over-all desirability are presented in Table 6. The scores for off-odor on a nine point scale were usually high, the lowest being 5.60. A significant difference ($P < .05$) was noted above the 5% level of substitution for the fillet and the washed flesh and above 10% for the unwashed flesh. The off-flavor evaluations showed a similar trend with significant differences obtained after 5% of the dried fish treatments were substituted in the bland wafers.

The over-all desirability of the wafers decreased with increasing levels of substitution for all the treatments. A significant difference in the levels of substitution was apparent after the 5% level of substitution.

Further evaluations of the effect of substituting the drum-dried

Table 6. Mean panel scores^a for the drum-dried fish treatments at increasing levels of substitution in the bland wafers.

Factor ^b	Treatment	% Substitution					
		0	5	10	15	20	25
Off-odor	Fillet	8.12 ^a	8.12 ^a	7.36 ^b	7.60 ^{ab}	6.68 ^c	7.52 ^b
	Washed	8.68 ^a	8.16 ^a	7.84 ^b	7.56 ^b	6.88 ^c	5.60 ^d
	Unwashed	8.48 ^a	7.88 ^a	7.92 ^a	6.68 ^b	6.08 ^b	6.68 ^b
Off-flavor	Fillet	8.32 ^a	7.84 ^{ab}	7.36 ^b	7.52 ^b	6.56 ^b	7.16 ^b
	Washed	8.52 ^a	7.92 ^{ab}	7.48 ^b	7.16 ^b	6.16 ^c	4.80 ^d
	Unwashed	8.52 ^a	7.72 ^b	7.80 ^b	6.88 ^c	5.52 ^e	6.20 ^d
Over-all desirability	Fillet	6.16 ^a	5.76 ^{ab}	5.12 ^b	4.88 ^b	4.32 ^b	4.36 ^b
	Washed	6.12 ^a	5.60 ^{ab}	5.20 ^b	4.52 ^b	3.84 ^{bc}	3.12 ^c
	Unwashed	6.20 ^a	5.72 ^{ab}	5.40 ^b	4.44 ^c	3.48 ^d	3.72 ^d

^an - 25

^b Nine point hedonic scale.

Mean scores in a row with the same exponent letter did not vary significantly ($p < .05$) from each other.

fish treatments in the bland wafer on off-odor, off-flavor and over-all desirability are presented in Table 7.

Each factor was directly compared at each level of substitution, 10%, 15% and 20% for the three treatments. There was a significant difference ($P < .05$) for off-odor and off-flavor at the 10% level of substitution between the fillet and the washed flesh and the control. The over-all desirability of the unwashed machine separated fish flesh was higher than the fillet and washed flesh and compared favorably with the bland wafer.

At the two higher levels of substitution, all the factors in the three treatments were significantly different from the control but not from one another. The higher preference for the unwashed flesh at the 10% level disappeared as the level of substitution was increased to 20%.

From the results of these panel evaluations, the 10% level of substitution was chosen for evaluating the stability of the dried fish treatments during the storage at 30°C.

Effect of Washing on Nutritional Value

The protein efficiency ratio (PER), which is a measure of the efficiency of utilization of absorbed nitrogen, did not vary significantly between the treatments and the casein reference (Table 8). There was a significant difference ($P < .05$) in the mean weight

Table 7. Mean panel scores^a for the drum-dried fish treatments by direct comparison at three different levels of substitution in the bland wafers.

% Substitution	Treatment	Flavor panel ^b		
		Off-odor	Off-flavor	Over-all Desirability
10	Control	8.52 ^a	8.52 ^a	6.12 ^a
	Fillet	7.28 ^b	6.96 ^b	4.40 ^b
	Washed	7.64 ^b	7.08 ^b	4.16 ^b
	Unwashed	8.20 ^{ab}	7.88 ^{ab}	5.40 ^a
15	Control	8.36 ^a	8.52 ^a	5.92 ^a
	Fillet	7.00 ^b	7.28 ^b	4.40 ^b
	Washed	7.08 ^b	6.84 ^b	4.28 ^b
	Unwashed	7.16 ^b	7.16 ^b	4.28 ^b
20	Control	8.24 ^a	8.44 ^a	5.96 ^a
	Fillet	7.04 ^b	7.04 ^b	4.56 ^b
	Washed	7.52 ^{ab}	6.64 ^b	4.12 ^b
	Unwashed	6.80 ^b	6.56 ^b	3.96 ^b

^a n = 25

^b Nine point hedonic scale.

Mean scores for each level of substitution in a column with same exponent letter did not vary significantly ($P < .05$) from each other.

Table 8. PER values for rockfish fillet, washed and unwashed machine separated rockfish flesh.

Treatment	Mean feed consumption (gm)	Mean weight gained (gm)	Protein (gm)	PER	PER ratio x 100 ^a
Casein	365.60 ± 40.04 ^a	83.70 ± 16.40 ^{ab}	32.89 ± 3.61 ^{ab}	2.53 ± 0.27 ^a	-
Fillet	342.30 ± 30.38 ^a	80.30 ± 12.77 ^b	30.81 ± 2.74 ^b	2.60 ± 0.30 ^a	103
Washed	394.30 ± 42.42 ^a	96.30 ± 15.41 ^a	35.49 ± 3.81 ^a	2.72 ± 0.36 ^a	108
Unwashed	352.30 ± 49.64 ^a	88.20 ± 21.68 ^{ab}	31.71 ± 4.71 ^{ab}	2.75 ± 0.34 ^a	109

^aPER for sample group/PER for casein reference group x 100.

Mean scores in a column with the same exponent letter did not vary significantly (P < 0.05) from each other.

gained and the protein intake of the experimental animals between the fillet and the washed flesh treatments.

It is known that the utilization of protein is related to the amino acid composition. In considering the data in Table 9, the washed flesh shows comparable levels of the essential amino acids to the fillet and the unwashed flesh. The cystine value was lower and the tyrosine value higher in the washed flesh compared to the fillet and the unwashed samples. The other amino acids, histidine, arginine, cysteine, aspartic acid, serine, glutamic acid, proline, glycine and alanine show much the same value in the three treatments with a slightly lower value of histidine for the washed flesh.

The mineral composition of the unwashed machine separated fish flesh was similar to that of the fillet (Table 10). Washing the machine separated flesh greatly altered the mineral composition in the flesh. Phosphorus, potassium, and sodium levels were reduced while the iron, copper, zinc and chromium levels were increased in the washed flesh. The levels of calcium, magnesium, barium, strontium, boron and manganese were not appreciably altered by washing.

Effect of Washing on Storage Stability

The microbial load of the washed machine separated fish flesh presented in Table 11 was higher than the unwashed flesh and the

Table 9. Amino acid composition of rockfish fillet, washed and unwashed machine separated rockfish flesh.

Amino acid	gm/100 gm protein			
	Casein	Fillet	Washed	Unwashed
*Lysine	8.1	10.3	10.2	10.0
*Histidine	3.0	3.1	2.2	2.3
*Arginine	3.9	6.8	7.1	6.7
Cysteine				
Aspartic acid		10.8	10.6	11.0
*Threonine	4.5	4.8	4.8	4.8
Serine		4.3	4.2	4.2
Glutamic acid		16.0	17.6	16.5
Proline		3.7	3.8	3.7
Glycine		4.1	3.8	4.3
Alanine		5.8	5.6	5.8
Half cystine		0.7	0.3	0.6
*Valine	7.4	5.3	5.1	5.2
*Methionine	3.3	3.5	3.5	3.6
*Isoleucine	6.4	5.1	4.7	4.7
*Leucine	10.0	8.2	8.0	8.0
Tyrosine		2.8	4.2	3.9
*Phenylalanine	5.4	4.7	4.1	4.7

* Essential amino acids.

Table 10. Mineral composition of rockfish fillet, washed and unwashed machine separated rockfish flesh.

Mineral		Treatments		
		Fillet	Washed	Unwashed
P	%	0.81	0.26	0.76
K	%	1.82	0.28	1.65
Ca	%	0.05	0.11	0.09
Mg	%	0.10	0.08	0.10
Na	%	0.18	0.06	0.28
Al	ppm	10.00	10.00	20.40
Ba	ppm	2.00	2.00	2.00
Fe	ppm	6.02	168.00	98.20
Sr	ppm	1.00	2.54	2.00
B	ppm	2.49	2.74	2.58
Cu	ppm	4.69	11.30	4.74
Zn	ppm	18.30	21.30	17.90
Mn	ppm	2.00	2.17	2.00
Cr	ppm	3.00	12.10	5.72

fillet. The number of microorganisms present in the dried fish treatments was surprisingly low. The rapid decrease in the microbial load is common for dried foods and indicates a lack of available moisture for growth.

Table 11. Total aerobic plate counts^a of the drum-dried treatments held at 30°C.

Treatment	Microorganisms/gm sample			
	Time (weeks)			
	0	2	4	8
Fillet	340	160	95	65
Washed	660	130	55	25
Unwashed	210	100	65	30

^aMean of duplicate samples.

The formation of malonaldehyde (TBA No.) which has been shown to be an index of rancidity in fishery products (59) was higher in the washed flesh compared to the unwashed flesh and the fillet (Table 12).

Table 12. 2-Thiobarbituric acid values (TBA number) of the drum-dried fish treatments stored at 30°C.

Treatment	TBA number (mg malonaldehyde/kg) ^a			
	Time (weeks)			
	0	2	4	8
Fillet	1.45	2.69	1.48	0.95
Washed	5.83	7.89	3.74	6.31
Unwashed	2.66	3.06	2.73	2.54

^aMean of duplicate samples.

The higher TBA number may be related to the presence of high levels of iron and copper in the washed flesh (Table 9).

The results of Table 13 show a drastic reduction in the trimethylamine oxide (TMAO) content of the freshly washed flesh. This reduction in TMAO content is the result of the washing process and has been reported by other workers (12, 22). The levels of TMAO in the dried samples decreased during storage which corresponded to the formation of trimethylamine (TMA).

Table 13. Amine content^a of the fresh and drum-dried fish treatments held at 30°C.

Factor	mg/gm Sample			
	Fillet	Washed	Unwashed	
TMAO	Fresh	2.44 ± 0.12	0.68 ± 0.00	2.93 ± 0.21
	Dried 0 wk	17.41 ± 0.51	0.87 ± 0.09	11.38 ± 0.09
	2 wk	11.73 ± 0.76	0.62 ± 0.00	10.71 ± 0.00
	4 wk	11.67 ± 1.41	0.00	11.99 ± 0.25
	8 wk	7.77 ± 0.00	0.00	6.28 ± 0.00
TMA	Fresh	0.01 ± 0.001	0.00	0.01 ± 0.002
	Dried 0 wk	0.39 ± 0.00	0.00	0.34 ± 0.017
	2 wk	0.91 ± 0.01	0.00	0.51 ± 0.00
	4 wk	1.13 ± 0.001	0.10 ± 0.007	1.09 ± 0.14
	8 wk	0.89 ± 0.005	0.13 ± 0.00	0.90 ± 0.005

^a Mean of duplicate samples.

The formation of TMA in the dried fillet and unwashed treatments may be related to the development of off-odors and off-flavors detected by the flavor panels (Tables 14 and 15). This is very apparent in the drum-dried unwashed machine separated flesh.

Off-flavor scores for the unwashed flesh decreased from a very high score of 8.12 to 6.56 after 8 weeks storage at 30°C. The TMA levels in the unwashed flesh increased from 0.01 to 1.09 mg/gm during the first 6 weeks of storage. The off-odor and off-flavor scores for the drum-dried washed flesh did not decrease during storage at 30°C and this was accompanied by only a slight formation of TMA.

Table 14. Taste panel evaluation^a for off-odor of the drum-dried fish treatments stored at 30°C.

Factor	Treatment	Time (weeks)			
		0	2	4	8
Off-odor	Control	7.88 ^b	8.40 ^a	8.34 ^a	8.72 ^a
	Fillet	8.24 ^{ab}	8.00 ^{ab}	7.72 ^b	7.72 ^b
	Washed	7.80 ^b	8.32 ^{ab}	7.84 ^{ab}	7.80 ^b
	Unwashed	8.56 ^a	7.88 ^b	8.04 ^{ab}	7.44 ^b

^a n = 25

Range of scores: 9, "none" to 1, "foul".

Mean scores in a column with same exponent letter did not vary significantly ($P < .05$) from each other.

The changes in off-odors and off-flavors occurring in the drum-dried fillet and unwashed flesh during storage was also reflected in the over-all desirability of the treatments (Table 16). The low over-all desirability scores given to the washed flesh may be attributed to its blandness. The freshly prepared drum-dried fillet and unwashed flesh had a higher over-all desirability than the plain wafer control. Washing the flesh resulted in a very bland drum-dried

Table 15. Taste panel evaluation^a for off-flavor of the drum-dried fish treatments stored at 30°C.

Factor	Treatment	Time (weeks)			
		0	2	4	8
Off-flavor	Control	7.20 ^b	8.28 ^a	8.36 ^a	8.76 ^a
	Fillet	7.44 ^{ab}	7.28 ^b	7.32 ^b	7.04 ^{bc}
	Washed	6.96 ^b	7.36 ^b	7.00 ^b	7.16 ^b
	Unwashed	8.12 ^a	7.32 ^b	7.40 ^b	6.56 ^c

^a n = 25

Range of scores: 9, "none" to 1, "foul".

Mean scores in a column with same exponent letter did not vary significantly ($P < .05$) from each other.

Table 16. Taste panel evaluation^a for over-all desirability of the drum-dried fish treatments stored at 30°C.

Factor	Treatment	Time (weeks)			
		0	2	4	8
Over-all desirability	Control	5.20 ^b	6.44 ^a	6.12 ^a	7.08 ^a
	Fillet	5.44 ^b	5.20 ^b	5.24 ^b	5.32 ^b
	Washed	4.84 ^b	5.00 ^b	4.80 ^b	5.24 ^b
	Unwashed	6.40 ^a	5.08 ^b	5.60 ^{ab}	5.00 ^b

^a n = 25

Range of scores: 9, "extremely desirable", to 1, "extremely undesirable".

Mean scores in a column with same exponent letter did not vary significantly ($P < .05$) from each other.

powder and may be related to the removal of minerals (Table 9) and lipids (Table 3).

DISCUSSION

The mechanical separation of the flesh from cleaned and dressed rockfish greatly increased the yield of edible flesh compared to conventional hand-filleting operations. The increased yields of edible flesh were similar to results of other workers (21, 31, 38) and indicates the potential of increasing the utilization of the total fish catch by machine separation.

A major drawback in the utilization of machine separated minced fish flesh is the lower acceptance of the flesh. Off-flavor, texture, and over-all desirability have been found to vary significantly between machine separated flesh and the intact fish fillet (12, 21). To overcome these drawbacks, Japanese fishery scientists have developed a procedure for preparing surimi, a semi-processed wet fish protein. Washing the machine separated fish flesh is an essential step for manufacturing surimi and it is responsible for the rapid increase in the production of "Kamaboko"-type products in Japan to over one million metric tons in 1970 (39).

There is limited knowledge available in the literature concerning the losses of solids during the washing process and the effect of washing on the quality of the fish flesh. From Tables 2 and 3, it can be observed that there was a 36.88% loss of solids during washing. The greatest reductions were found in the ash (80.32%) and lipid

(65.13%) levels in the washed flesh. Most of the sarcoplasmic proteins were lost during washing and only 77.29% of the protein (N X 6.25) was recovered.

Washing the machine separated fish flesh did not affect the amino acid composition (Table 9). The content of essential amino acids of the drum-dried fish treatments were similar to casein (Table 9) and this is reflected by the high protein efficiency ratios compared to the casein reference (Table 8).

Washing the machine separated fish flesh increased the microbial load compared to the unwashed flesh and fillet (Table 11). Although it has been reported by other workers that washing increases the microbial load, the number of microorganisms found in the washed flesh does not justify concern since it is well below the unacceptable level in fishery products (13, 33, 45).

The rapid formation of trimethylamine (TMA) in the dried rockfish fillet and unwashed machine separated flesh treatments was unexpected and difficult to explain. The breakdown of trimethylamine oxide (TMAO) to TMA by bacterial enzymes has been shown to cause undesirable odors and flavors (23). The formation of TMA in fish held at refrigerated temperatures has been used for many years as an index of quality (24, 25, 42, 43). It has been noted that there is no TMA formed during frozen storage of rockfish (12, 22). Since the microbial load was very low in the dried fish treatments, the

formation of TMA must have been non-enzymatic. Vaisey (54) has reported that TMAO at temperatures between 22° - 24° is readily reduced by cysteine in the presence of iron or hemoglobin as a catalyst.

The formation of TMA in the drum-dried fillet and unwashed flesh treatments may be related to the development of off-odors and off-flavors formed during storage at 30°C (Tables 14 and 15). The over-all desirability of the drum-dried washed flesh increased slightly during storage but was not appreciably higher than the drum-dried fillet and unwashed flesh treatments (Table 16). It appears that the over-all desirability is not directly linked to the formation of TMA since washing drastically reduced the level of TMAO in the washed flesh and the subsequent formation of TMA during storage at 30°C.

The lower over-all desirability of the drum-dried washed flesh may be attributed to its blandness. Washing the flesh greatly reduced the levels of ash and lipids (Table 4). However, the concentration of iron and copper in the washed flesh was considerably higher than the fillet or unwashed flesh treatments and may be responsible for the rapid formation of malonaldehyde (Table 12).

The formation of malonaldehyde is a useful index as a measure of lipid oxidation or rancidity (59, 60). During lipid oxidation, various compounds are formed which can complex with other

compounds in the tissue to form off-odors and off-flavors (48). Thus, the higher levels of malonaldehyde in the drum-dried washed flesh may implicate the oxidation of the lipids as being responsible for the lower over-all desirability of the washed flesh.

The data in Table 12 might also suggest that there are antioxidants naturally present in fish flesh. The higher TBA values for the washed flesh may be the result of leaching the antioxidants from the flesh during the washing process.

SUMMARY AND CONCLUSIONS

The effect of washing of the machine separated flesh as reflected by such parameters as quality, desirability and storage stability was investigated. The study involved the comparison of the washed flesh with the unwashed flesh and the fillet. The results and conclusions may be summarized as follows:

1. Higher yields of flesh were obtained from the mechanical separation than from the filleting operation.
2. The color of the drum-dried washed flesh was superior to the drum-dried unwashed flesh.
3. The proximate composition of the washed flesh showed a lower protein content and considerably reduced ash and lipid levels compared to those of the unwashed flesh and the fillet.
4. Sensory evaluation of the three treatments incorporated in a bland wafer was comparable for off-odor, off-flavor and over-all desirability. The over-all desirability of the wafers decreased with increasing levels of substitution for all the treatments.
5. Washing the machine separated fish flesh did not appreciably alter the amino acid composition. The content of essential amino acids of the drum-dried treatments was similar to

- casein and this was reflected by the high protein efficiency ratios compared to the casein reference.
6. Washing the machine separated fish flesh increased the microbial load although the number of microorganisms found were well below acceptable levels.
 7. The mineral composition of the unwashed machine separated fish flesh was similar to that of the fillet. Washing the machine separated flesh greatly altered the mineral composition in the flesh. Phosphorus, potassium, and sodium levels were reduced while the iron, copper, zinc, and chromium levels were increased in the washed flesh. The levels of calcium, magnesium, barium, strontium, boron, and manganese were not appreciably altered by washing.
 8. The rapid formation of trimethylamine (TMA) in the drum-dried rockfish fillet and unwashed machine separated flesh treatments may be related to the development of off-odors and off-flavors formed during storage at 30^o C.
 9. The over-all desirability of the bland wafers consisting of 10% drum-dried washed flesh increased slightly during storage but was not appreciably higher than the wafers containing drum-dried fillet and unwashed fish flesh. It appears that the over-all desirability may not be directly linked to the formation of trimethylamine (TMA) since washing drastically reduced the

level of trimethylamine oxide (TMAO) in the washed flesh and the subsequent formation of TMA during storage at 30°C.

10. The much higher levels of malonaldehyde found in the drum-dried washed flesh may implicate the oxidation of the lipids as being responsible for the lower over-all desirability of the washed flesh.

BIBLIOGRAPHY

1. Amano, K., Fish Sausage processing in Japan. *Fish. New. Intl.* 1. 5:29, 1962.
2. Association of Official Agricultural Chemists, "Official methods of analysis." 10th ed. p. 345, 23.003. 1965a.
3. Association of Official Agricultural Chemists, "Official methods of analysis." 11th ed., p. 295, 18.012. 1970.
4. Association of Official Agricultural Chemists, "Official methods of analysis." 11th ed., p. 297, 18.0022. 1970.
5. Association of Official Agricultural Chemists, "Official methods of analysis." 11th ed., p. 295, 18.013. 1970.
6. Association of Official Agricultural Chemists, "Official methods of analysis." 11th ed., p. 842, 41.014. 1970.
7. Association of Official Agricultural Chemists, "Official methods of analysis." 11th ed., p. 843, 41.015. 1970.
8. Association of Official Agricultural Chemists, "Official methods of analysis." 12th ed., p. 904, 46.005, 46.0037a, 46.038. 1975.
9. Association of Official Agricultural Chemists, "Official methods of analysis." 11th ed., p. 800, 39.116, 39.167, 39.168, 39.169, 39.170. 1970.
10. Babbitt, J. K., Law, D. K. and Crawford, D. L. Acceptance of a fish shrimp portion utilizing machine separated minced fish flesh. *J. Food Science* 39:1130-1131. 1974.
11. Babbitt, J. K., Crawford, D. L. and Law, D. K. Decomposition of trimethylamine oxide and changes in protein extractability during frozen storage of minced and intact hake (Merluccius productus) muscle. *J. Agric. Food Chem.* 20 (5):1052-1054. 1972.

12. Babbitt, J. K., Crawford, D. L. and Law, D. K. Quality and utilization of minced deboned fish muscle. Second Technical Seminar on Mechanical Recovery and Utilization of Fish Flesh. June 12-13, 1974, Boston, Mass. (In Press).
13. Babbitt, J. K. Minced fish flesh research and development at Oregon State Seafood Laboratory. In: "Oak Brook Seminar, Mechanical recovery and utilization of Fish.", Ed., Martin, R. E. p. 49. National Fisheries Institute, Washington, D. C. 1972.
14. Babbitt, J. K., Law, D. K. and Crawford, D. L. Improved acceptance and shelf-life of frozen minced fish with shrimp (In Press). 1975.
15. Carver, J. H. and King, F. J. Fish scrap offers high quality protein. Food Eng. 43(1):75-76. 1971.
16. Choy, A. A., Meinke, W. W. and Mattil, K. F. Comparative nutritive value of fillets and minced fish prepared from some species of fish. Presented at the 35th annual IFT Meeting. Chicago, IL. 1975.
17. Choy, A. A., Meinke, W. W. and Mattil, K. F. Preparation and characterization of some fish flesh products from Gulf Coast trawl fish. Presented at the 35th annual IFT Meeting, Chicago, IL. 1975.
18. Christensen, R. E., Beckman, R. M. and Birdsall, J. J. Some mineral elements of commercial spices and herbs as determined by direct reading emission spectroscopy. J. A. O. A. C. 51(5):1003-1010. 1968.
19. Crawford, D. L., Law, D. K. and Babbitt, J. K. Utilization of Hake, Dogfish, and By-Products of the Fillet Industry for Protein Supplements. Completion Report. United States Dept. of Commerce, National Oceanographic and Atmospheric Administration, National Marine Fisheries Service Commercial Fisheries Research and Development Act. 1973.
20. Crawford, D. L., Law, D. K., and Babbitt, J. K. Nutritional characteristics of marine food fish carcass waste and machine separated flesh. J. Agric. Food Chem. 20(5): 1048-1051. 1972.

21. Crawford, D. L., Law, D. K., and Babbitt, J. K. Yield and acceptability of machine separated flesh from some marine food fish. *J. Food Science* 37(4):551-553. 1972.
22. Dingle, J. R. and Hines, J. A. Protein instability in minced flesh from fillets and frames of several commercial Atlantic fishes during storage at -5° . *J. Fish. Res. Board Can.* 32:775-783. 1975.
23. Dyer, W. J. and Mounsey, Y. A. Amines in Fish muscles. II. Development of trimethylamine and other amines. *J. Fish. Res. Board Can.* 6:359-367. 1945.
24. Dyer, W. J. Note on the colorimetric estimation of TMA. *J. Fish Res. Board Can.* 7:576-579. 1950.
25. Dyer, W. J. Report on TMA in Fish. *J. Assoc. Off. Agric. Chem.* 42:292-294. 1959.
26. Howell, A. J., Webb, N. B., Thomas, F. B. and Rao, V. M. M. Evaluation of the functional properties of minced fish tissue. Presented at the 35th annual IFT Meeting, Chicago, IL. 1975.
27. Kimata, M. Studies on the spoilage of "Kamaboko". I. On the appearance of spoilage. *Bull. Jap. Soc. Sci. Fish.* 16:428-432. 1951.
28. Kimata, M. Studies on the spoilage of "Kamaboko". IV. On the microbiology in the spoilage (1). *Bull. Jap. Soc. Sci. Fish.* 16(12):46-48. 1951.
29. Kimata, M. Studies on the spoilage of "Kamaboko". V. On the microbiology in the spoilage (2). *Jap. Soc. Sci. Fish.* 16(12):50-57. 1951.
30. Kimata, M. and Kawai, A. Studies on the spoilage of "Kamaboko". VII. Microbiological studies of refined starch on the market (1). *Bull. Jap. Soc. Sci. Fish.* 16(12):55-58. 1951.
31. King, F. T. and Carver, J. H. How to use nearly all the ocean's food. *Com. Fish. Rev.* 32(12):12-21. 1970.

32. King, F. J. and Flick, G. J. Beefish patties. *Marine Fisheries Review* 35(7):31-33. 1973.
33. King, F. J. Improving the supply of minced blocks for the fish stick trade. A progress report. *Marine Fisheries Review* 35(8):26-32. 1973.
34. King, F. J., Carver, J. H. and Prewitt, R. Machines for recovery of fish flesh from bones. *Am. Fish Farmer* 2(11): 17-21. 1971.
35. King, F. J. Acceptability of main dishes (entrees) based on mixture of ground beef with ground fish obtained from under-used sources. *J. Milk Food Technol.* 36(10):504-508. 1973.
36. King, F. J. Characteristics, uses, and limitations of comminuted fish from different species. In "Oak Brook Seminar Mechanical Recovery and Utilization of Flesh." Ed. Martin, R. E., p. 15. National Fisheries Institute, Washington, D. C. 1972.
37. Kudo, G., Okada, M. and Miyauchi, D. Gel forming capacity of washed and unwashed flesh of some Pacific Coast species of fish. *Marine Fisheries Review.* 35(12):10-15. 1973.
38. Miyauchi, D. and Steinberg, M. Machine separation of edible flesh from fish. *Fisheries Industrial Research* 6(4):165-171. 1970.
39. Miyauchi, D., Kudo, G., and Patashnik, M. Surimi - A semi-processed wet fish protein. *Marine Fisheries Review* 35(12):7-9. 1973.
40. Miyauchi, D. Progress report on minced fish studies at National Marine Fisheries Service. In: "Oak Brook Seminar Mechanical Recovery and Utilization of Flesh." Ed. Martin, R. E., p. 35. National Fisheries Institute, Washington, D. C. 1972.
41. Miyauchi, D., Patashnik, M. and Kudo, G. Frozen storage keeping quality of minced black rockfish (Sebastes spp.) improved by cold water washing and use of fish binder. *J. Food Science* 40(3):592-594. 1975.

42. Murray, D. K. and Gibson, D. M. An investigation of the method of determining trimethylamine in fish muscle extracts by formation of its picrate salt - Part I. *J. Food Technol.* 7:35-46. 1972.
43. Murray, C. K. and Gibson, D. M. An investigation of the method of determining trimethylamine in fish muscle extracts by formation of its picrate salt - Part II. *J. Food Technol.* 7:47-51. 1972.
44. Okada, M. Effect of washing on the jelly forming ability of fish meat. *Bull. Jap. Soc. Sci. Fisheries* 30:225-261. 1964.
45. Okada, M., Miyauchi, D., and Kudo, G. "Kamaboko" - The giant among Japanese processed fishery products. *Marine Fish Rev.* 35(12):1-6. 1973.
46. Patashnik, M., Kudo, G. and Miyauchi, D. Smooth white spread from separated fish flesh forms a base for flavored dips, snack items. *Food Product Development* 7(6):82-91. 1973.
47. Spackman, D. H., Stein, W. H. and More, S. Automatic recording apparatus for use in the chromatography of Amino acid. *J. Analytical Chemistry.* 30:1190-1206. 1958.
48. Stansby, M. E. Speculations on Fishy odors and flavors. *J. Food Technol.* 16(4):28-32. 1962.
49. Steinberg, M. New opportunities through use of comminuted fish flesh. In: "Oak Brook Seminar Mechanical Recovery and Utilization of Flesh." Ed. Martin, R. E., p. 4. National Fisheries Institute, Washington, D. C. 1972.
50. Tanikawa, E. Fish sausage and ham industry in Japan. In "Advances in Food Research." Ed. Chichester, C. O., Mrak, E. M. and Stewart, G. E., 12:367, Academic Press, N. Y. 1963.
51. Tanikawa, E. A Japanese style fish meat paste. In. E. Tanikawa. *Marine products in Japan*, p. 340-372. Koseisha-Koseikaku Co., Tokyo, Japan. 1971.

52. Teeny, F. M. and Miyauchi, D. Preparation and utilization of frozen blocks of minced black rockfish muscles. *J. Milk Food Technol.* 35:414-417. 1972.
53. Uchiyama, H. and Tanaka, T. The softening deterioration of fish sausage - 1. Some chemical and microscopical aspects. *Bull. Jap. Soc. Sci. Fisheries* 24(2):148-155. 1958.
54. Vaisey, E. B. The non-enzymic reduction of trimethylamine oxide to trimethylamine, dimethylamine, and formaldehyde. *Can. J. Biochem. Physiol.* 34:1085-1089. 1956.
55. Yamagata, M., Horimota, K. and Nagaoka, C. Assessment of green tuna: determining trimethylamine oxide and its distribution in tuna muscles. *J. Food Science* 34:156-159. 1969.
56. Yokoseki, J., Uchiyama, H. and Mamizuka, T. The softening deterioration of fish sausage. II. Microbiological studies of softening deterioration. *Bull. Jap. Soc. Sci. Fisheries* 24(2):156-164. 1958.
57. Yokoseki, M. Studies on the internal spoilage of fish-jelly products - 1. Surviving microorganisms in fish-jelly products cooked at different temperatures. (In Japanese, Engl. summ.). *Bull. Jap. Soc. Sci. Fisheries* 23:539-542. 1958a.
58. _____ Studies on the internal spoilage of fish-jelly products - II. Relationship between cooking temperature and spoilage. (In Japanese, Engl. summ.). *Bull. Jap. Soc. Sci. Fisheries* 23:453-546. 1958b.
59. Yu, T. C. and Sinnhuber, R. O. 2-Thiobarbituric acid method for measurement of rancidity in fishery products. *J. Food Technol.* 11(1):104-108. 1957.
60. Yu, T. C. and Sinnhuber, R. O. Removal of interfering pigments in determining malonaldehyde by the 2-thiobarbituric acid reaction. *J. Food Technol.* 16(6):115-117. 1962.