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A CHEMICAL STUDY OF THE OIL OF SALMON IN COLD STORAGE.

Author

H. N. BROCKLESBY

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The investigation to be reported in this manuscript is one which was undertaken in connection with the general study of the chnages which occur in fish when stored for short and long periods in cold storage. , It is of a preliminary character only.

Certain visible evidences of chemical change make themselves apparent on fatty fish which are kept for any length of time in a frozen condition. The most common phenomenon is the appearance of a reddish-brown exudation known to the trade as "rust" and commonly ascribed to the oxidation of the oil of the fish. This "rusting" of stored fish is a source of continual loss to the trade. It was with particular reference to this condition, that the chemical examination of the oils of several salmon, stored under different conditions and for varying lengths of time, was carried out.

The work of Smith (1) and of Perlzweig and Geis (2) showed that the fats of the fish that they examined (fluke and flounder) underwent no hydrolysis even when stored for a period of two years. Clark and Almy (3) reported on the chemical composition of some twenty fresh fish on different species. The most striking fact about their analyses of the oils of these fish was that they found the acid value to be a quantity varying inversely as the oil content of the fish. Forinstance, they found the acid value of fresh bonita to be 28.6 and that of Spanish mackerel 2.6.

In this regard, they say, "This seems to point to the presence, in the tissues of fish in a more or less constant proportion, regardless, of the fat content, of an acid substance soluble in the fat solvent, or carried out with the fat". It might be stated at once, that no such values have been obtained for fresh fish during the present investigation. Clark and Almy (4) also reported a chemical study of the composition of fish stored for short and long periods in the frozen condition. In regard to the changes of the oil of the weakfish and of the bluefish (with which they worked), they concluded that the acid values of the samples increased and the iodine samples decreased with the length of time in storage. They again found high acid values for the fresh samples. Their data showed that there was considerable variation in the oils of the different fish of the same species. One specimen showed a higher iodine value after being in storage for ten months than that of a fresh speciment. In the case of the bluefish examined, fish that were in storage for four, five, eight, twelve and sixteen months respectively all had higher iodine values than one of the two fresh specimen examined. It seems to the present writer that no definite conclusions can be drawn from the analysis of so few fish, in view of the great variation which occurs in the character of the oils of fish of the same species.

In the present investigation, salmon were used. With one exception, they were all of the genus Oncorhynchus, the exception being some steelhead trout which belong to the closely related genus Salmo. The salmon used were as follows: redspring, <u>Oncorhynchus tschawytscha,</u> cohoe, <u>Oncorhynchus kisutch,</u> chum <u>Oncorynchus keta,</u> humpback, Oncorhynchus gorbuscha; the steelhead trout examined was the Salmo gairdneri.

Preparation of the samples of oil.

Each fish was skinned and the dark muscle removed from the great pink muscle. The oil was extracted from the great pink muscle separately from that of the skin and dark muscle. The extraction was carried out by cooking the finely chopped-up material at a pressure of fifteen pounds for about fifteen minutes and then immediately pressing the hot mass in a Carver hydraulic press at a pressure of from three to four thousand pounds. The resulting mixture was allowed to separate and the oil skimmed off; the latter was then dehydrated with anhydrous sodium sulphate and filtered.

Methods of analysis.

From the preliminary investigation in which a sample of salmon oil was oxidized slowly by means of a current of air, it was found that the iodine value, the saponification value, refractive index and the percentage of ether-insoluble brominated glycerides were changed to the greatest extent and it was thus possible to follow very closely the gradual oxidation of the oil. The determination of the insoluble brominated glycerides was only made in the fish examined at the commencement of the work (Experiment 1). To determi[ne] very slight oxidations, the insoluble bromides were not found reliable. With the brominating reagents tried in this investigation, it was not possible to control the bromination with sufficient accuracy to be of any value in detecting small changes in the degree of unsaturation.

However in Experiment 1, where this determination was made, the method of Bailey and Johnson (5) was followed.

The iodine values, saponification values and refractive indices were determined according to the methods suggested by the Committee on the Examination of Oils, Fats, and Waxes of the American Chemical Society. The method used for determining the acid value was Davies (6) modification, using ether and neutral barium chloride. The refractive indices were determined with a Bausch & Lomb refractometer with prisms maintained at 25° Centigrade.

EXPERIMENT 1.

In this experiment fish were chosen at random from the storage rooms of the Canadian Fish and Cold Storage at Prince Rupert. The object was to get samples of fish in all stages from the strictly fresh frozen to the badly rusted condition. As it was necessary to take those samples that were available, it was not possible to obtain a complete set of fresh to rusted fish on one specie. The history of these fish other than the length of time in cold storage and the fact that they were air frozen in a sharp freezer, was not known. The condition of the fish at the time of sampling, was noted and the two samples of oil were extracted from each specimen, in the manner already outlined. The acid value, iodine value, insoluble bromides and saponification value were determined for the majority of these samples. In one case (Number 18/8), not enough oil was obtained for complete analysis, The refractometer was available only at the end of this experiment.

Each sample of oil was tested for the presence of organic peroxides with alcoholic potassium iodide and starch but negative results were obtained in each case. The biochemical reaction of Vintilescu and Popescu (7) also gave negative results. The results of the analysis of this lot of fish are given in Table 1.

EXPERIMENT 2.

In this experiment fresh redspring salmon were used. Three of these fish were analysed whilst they were in the fresh condition. One of the three was fresh frozen, the other two simply being chilled The remainder of the fish used in this experiment were air frozen in a sharp freezer at -20° Fahrenheit. One half of them were glazed, the other half left unglazed. They were placed in four cold storage rooms of the Canadian Fish and Cold Storage Company, each room being kept at a different temperature and having a different rate of temperature change. In each of these four rooms, both glazed and unglazed samples were stored. The samples were analysed for the most part, after seven and twelve months respectively. In one case where the fish were kept in a sharp freezer, they were analysed at the end of three, six and twelve months respectively. The temperature of these four rooms were recorded each six hours for the whole twelve months. From this data, the degree of temperature fluctuation was calculated. The lowest and highest temperature of storage has also been included. The results of the analysis of this set of fish will be found recorded in Table II. In this experiment the acid values, iodine values and saponification values only have been determined. Each sample was tested for traces of peroxides by the two methods mentioned previously and in each case, negative results were obtained.

EXPERIMENT 3.

In this experiment fresh humpback salmon and fresh redspring were utilized. They were all stored in the same storage room for the same length of time. Each sample was air frozen and glazed. Half, however, were frozen in the round and the other half eviscerated before freezing. Samples of each were kept in sealed tin boxes containing an atmosphere of oxygen, carbon dioxide and ammonia respectively. Controls of the same lot of fish were also kept in the ordinary atmosphere of storage rooms. In the case of the humpback salmon not enough oil was obtained for complete analysis. The acid value and refractive index alone were determined on these samples. The results of the analysis of these samples will be found in Table III.

DISCUSSION.

An inspection of the tables will show that the oils from the fresh fish examined in this investigation were neutral as indicated by their acid values; this is in contra distinction to the findings of Clark and Almy (loc. cit.), who claim that the acid values of the oils of fresh fish may vary from 2.2 to 28.6. The discrepancy probably arises in the methods of preparing the samples. Clark and Almy extracted the vacuum-dried flesh with ether and the probability is that the "ether-soluble acidic substance" (sarcolactic acid?) is more soluble in hot water than in oil and thus does not appear in the samples prepared in the manner described in this paper.

Out of forty-four samples examined, forty specimen showed a higher acid value of the oil from the pink muscle than that of the oil from the skin and dark muscle. Of the other four, three of the samples were from humpback salmon which contain very little oil. The pink muscle of sample 18/8 was divided into two portions, the first being that part of the muscle lying within one inch of the backbone and the second the remainder of the pink muscle from one inch of the backbone to the dark muscle and skin. The acid value of the skin and dark muscle oil, outside pink muscle and inside pink muscle oil were 8.82, 13.98 and 14.88 respectively. A similar division was made in sample 12/8 when 8.77, 1084 and 14.26 were the acid values which were obtained from the dark muscle and skin oil, outer pink muscle and inner pink muscle oils respectively. It it therefore apparent that the hydrolysis of the oil proceeds more rapidly in the interior of the fish than it does towards the outside.

There seems to be no clear-cut relationship between the degree of hydrolysis and the temperature at which the samples were stored, the amount of temperature fluctuation or the glazed or unglazed condition of the fish. In general, the longer a fish is in storage the higher the acid value, but so many exceptions occur that it is quite evident that the rates at which the oil hydrolyzes in different fish stored under similar conditions might be totally different. Whether or not the history of a fish from the time that it is caught to the time that it is frozen has any effect on the subsequent rate of the hydrolysis of the oil is a problem that is now being worked on.

In the case of fish stored in the different gases, those kept in ammonia gave by far the lowest acid values. Apparently the ammonia had penetrated the flesh of the fish, neutralizing the fatty acids. These samples were also the most rusted of the series. In this regard, it is interesting to note the results of some experiments now being carried out. Whilst determining the oxygen uptake of some salmon oil, it became necessary in one case to keep the atmosphere above the oil strongly ammoniacal. It was found that the oils kept in such an atmosphere turned a rusty brown colour. Whether or not there is any connection between the two phenomena, has not yet been decided.

The statement[(3)] that the amount of rusting can be estimated by determining the acid value of the oil was not substantiated in the present work. For example, samples 3/8 and 4/8 had acid values of 79.7, 40.1 and 3.38, 4.25 respectively, yet the two fish were equally rusted. In many cases fish that had not shown any signs of rusting had higher acid values than those in which rusting had been very marked.

Not a great deal of information could be obtained from the iodine values. There was so much variation between fish of the same specie that it was difficult to draw any conclusions from values obtained from different specimens even though they were stored under identical conditions. In general, the degree of unsaturation of the oil from the inner pink muscle seemed to be higher than that of the oil from the dark muscle and skin. This was true of fresh as well as stored fish so that oxidation during storage could not be the controlling factor.

That the oil of fish in cold storage undergo perceptible oxidation is neither proven nor disproven by the data presented here. Certainly, when one considers how quickly the oil oxidizes when a sample is exposed to the air and how easily this oxidation can be detected by the fall in iodine value or by sensitive peroxide tests, it is surprising that the iodine value of the oils from fish that have been stored for a long period do not show a more definite decrease. It is even more remarkable that not a single sample gave a positive test for a peroxide.

No rancidity was noted in any samples. Even the samples of oil which had the highest acid value and which came from some of the worst-looking fish were not rancid when judged organoleptically. Since rancidity is caused in part by oxidation and since there is no chemical test at present more sensitive in detecting rancidity than the organs of taste and smell, it must be concluded that the oxidation which took place in the oils of these fish during storage must have been very slight. The writer has obtained some evidence that the oils from certain stored fish have very short induction periods when subjected to oxygen absorption tests. These oils did not give any reaction when tested for peroxides. The induction period is not lengthened by subjecting the oil to vacuum treatment and it must be concluded that the oxygen absorbed is in the "mol ide" condition. This phase of the problem is now under investigation.

The saponification values and refractive indices of the oils also corroborated the evidence that little oxidation had taken place.

Samples of fresh oil when exposed to the air of the laboratory for three or four days, sometimes show an increase of the saponification value from the normal value of about 190 to a value of 200 and over whilst refractive indices increase their value in the same length of time from 8 to 15 units in the fourth decimal place. No such abnormal values were found for any of the oils examined.

Whilst the results described in this paper are not in themselves conclusive, they indicate very cloarly along what lines further investigation should proceed. The following points should be investigated further. (1) Is the unsaturated character of the oil of fish of the same specie a function of the age, spawning stage, or type of food of the salmon. (2) How does the methods of handling the fish from the time that it is placed in cold storage affect its subsequent keeping qualities whilst in cold storage. (3) Are the keeping qualities of a fish dependent upon its oil content. These problems are now being worked on and it is hoped to report on the progress in due time.

NNOTE.

Since the above manuscript was written the author has had further opportunity of examining the oils from the pink and dark muscle of fresh Red Spring Salmon. In no case was a specimen more than six hours out of the water before the oil was removed from the flesh. The results of the analyses of these oils are given in Table 4.

It will be noted that in the majority of cases the iodine value of the pink muscle oil is lower than that of the dark muscle oil. The refractive indices are also lower for the pink muscle oil than for the dark muscle oil. No great variations are shown for the saponification values. The acid values are all nil, again in contra-distinction to the results of Clarke and Almy.

References.

1. Biochem. Bull. 1913, 3, 54.
2. Biochem. Bull. 1913, 3, 69.
3. J. Biol. Chem. 1918, 33, 486.
4. J. Ind. Eng. Chem. 1920, 7, 656
5. J. Ind. Eng. Chem. 1918, 10, 999.
6. J. Soc. Chem. Ind. 1928, 47, 24T.
7. Chem. Abst. 1916, 10, 646.

Fish	Acid Value	Iodine Value	Sap Value	Ref. Ind.
1 P	0	166.8	196.1	1.4691
1D	0	112.8	196.1	1.4705
2P	0	98.5	196.8	1.4688
2D	0	110.0	194.4	1.4703
3P	0	119.3	192.6	1.4716
3D	0	118.4	195.5	1.4723
4P	0	124.1	194.4	1.4719
4D	0	132.5	196.0	1.4229
5P	0	no oil	no oil	1.4691
5D	0	1o4.5	no oil	1.4696
9P	0	104.6	no oil	1.4701
9D	0	113.8	194.8	1.4708
12P	0	105.5	no oil	1.4700
12D	0	118.3	193.8	1.4713
11P	0	133.3	192.9	1.4693
11D	0	124.2	193.4	1.4707
13P	0	127.1	191.1	1.4780
13D	0	123.7	191.1	1.4719
16P	0	91.8	no oil	1.4696
16D	0	111.4	no oil	1.4690.

TABLE I.

ef. No.	Description of Fish	Time in Storage	Condition at End of Storage	Source of Oil	Description of Oil	Acid Value	Iodine Value	Insol. Brōmidēs	Sap. Value	Ref. Index @ 25° C.
	Cohoes									
5 A/8	Fresh Frozen	2 weeks	Excellent	Pink Muscle	Clear light red.	1.83	170.1	51.1	189.6	
5 B/8				Dark Muscle and skin	Clear light yellowish-Brown	0.91	162.9	48.4	181.4	
3 A/8	Glazed Eviscerated	2 years	Badly "rusted", not badly shrunken, unmarketable	Pink Muscle	Red, semi-solid at room temp.	79.7	183.8	47.5		
3 B/3				Dark Muscle and skin	Red, depositing crystalline ppt.	40.1	169.4	35.8	191.3	
4 A/8	Glazed Eviscerated	Not known	Badly rusted. Unmarketable.	Pink Muscle	Light red, clear liquid.	3.38	167.9	49.9	186.9	
4 B/8				Dark Muscle and skin	Yellow-brown liquid.	4.25	175.4	52.4	194.8	
7 A/8	Glazed Eviscerated.	2 years	Badly rusted, shrunken, and unmarketable.	Pink Muscle	Deep pink Solid @ 23° C.	74.4	170.6	42.1	189.6	
7 B/8				Dark Muscle	Yellow brown liquid, depositing crystals.	48.1	161.1	42.4	186.9	
	Chum Salmon									
8 A/8	Glazed Eviscerated.	1 year	Good condition, no sign of rusting	Inner Pink Muscle.	Pink Liquid	14.88				
8 B/8				Outer Pink Muscle	Pink Liquid	13.98				
8 C/8				Dark Muscle and Skin	Pink Liquid					

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. No.	Description of Fish	Time in Storage	Condition at End of Storage	Source of Oil	Description of Oil	id Value	Iodine Value	Insol. Bromides	Sap. Value	Ref. Index @ 25° C.
	<u>Steelheads</u>									
9 A/8	Glazed Evis-cerated.	At least 1 year	Badly rusted, Unmarketable.	Pink Muscle	Yellowish-brown	10.63	162 .3	36.8		1.4793
9 B/8				Dark Muscle	Yellowish-brown clear	8.22	149.4	34.5		1.4792
0 A/8	Glazed Evis-cerated	At least 1 year	Badly rusted, unmarketable.	Pink Muscle	Yellow-pink "	10.15	157.1	28.0	194.0	1.4782
0 B/8				Dark Muscle and Skin	Yellow-brown"	8.95	148.0	22.8		1.4784
	<u>Red Springs</u>									
A/9	Glazed Evis-cerated	4 months	Good condition, Glaze O.K. No rusting	Pink Muscle	Red, fairly cloudy	5.52	94.3	11.35	189.2	1.4683
B/9				Dark Muscle and Skin	Yellow-pink clear	4.18	94 .4	7.30	190.2	1.4781
A/9	Glazed Evis-cerated	4 months	Good condition. Glaze O.K. Slight signs of rusting near oval fin	Pink Muscle	Red depositing crystals.	7.05	96.5	14.05	190.3	1.4718
B/9				Dark Muscle and Skin	Yellow pink, depositing crystals	4.54	99.6		188.6	1.4716
A/9	Glazed Evis-cerated	4 months	Good condition, Glaze O.K., no signs of rusting.	Pink Muscle	Red, cloudy	8.67	133.3	21.70	190.8	1.4730
B/9	Glazed Evis-cerated			Dark Muscle and Skin	Yeloow-pink depositing crystals	5.48	137.6	28.4	190.8	1.4731

TABLE III.

No.	Description of Fish	Time in Storage	Temp. of Storage Lowest °F	Temp. of Storage Highest °F	Degree Temp. Fluctuation	Condition of Fish at End of Storage Period.	Source of Oil	Appearance of Oil	Analysis of Oil: Acid Value	Analysis of Oil: Iodine Value	Analysis of Oil: Sap. Value
	Red Spring Salmon										
A/8	Fresh Frozen gutted, not glazed.	1 day	-20°	25°	45	Excellent, perfectly fresh.	Pink Muscle	Clear Red	0	139.3	---
B/8	Fresh Chilled not frozen.						Dark Muscle	Clear Yellow pink.	0	118.7	---
A/9	Fresh Chilled not frozen.	1 day	28°	29°	1	Excellent, perfectly fresh.	Pink Muscle	Clear Red	0	108.4	---
B/9							Dark Muscle	Clear yellow pink	0	114.7	---
8	Fresh Chilled	1 day	28	28	0	Excellent	Whole Eviscerated fish	Clear Red	0	108.4	198.[illegible]
	Fresh Spring Salmon Fresh When Frozen										
/8	Eviscerated, glazed perfectly fresh.	7 months	3	12	264	Fresh rusted in interior of fish. Started to discolor along cut. Fish [illegible] firm glaze unbroken	Pink Muscle	Clear Red	11.25	---	194.7
B/8							Dark Muscle	Clear Yellow-pink.	7.65	132.8	194.1
A/8	Eviscerated, unglazed.	7 months	3	12	264	Rust commencing at nape. Fish fairly firm	Pink Muscle	Clear Red	14.2	148.2	1 85.0
B/8							Dark Muscle	Yellow Red Clear.	6.34	128.5	193.0

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Ref. No.	Description of Fish	Time in Storage	Temp. of Storage Lowest °F	Temp. of Storage Highest °F	Degree Temp. Fluctuation.	Condition of fish at End of Storage period.	Source of Oil	Appearance of Oil	Analyses of Oil. Acid Value.	Analyses of Oil. Iodine Value	Analyses of Oil. Sap Value
1 A/8	Eviscerated, unglazed.	12 months	3	12	587	Fairly good condition. Not dessicated, rusting commenced	Pink Muscle	Light red crystals precipitating.	12.48	115.2	191.9
1 B/8							Dark Muscle	Yellow pink cloudy.	9.93	111.6	198.3
2 A/8	Eviscerated, glazed.	12 months	3	12	587	Badly dessicated, but not badly rusted, discoloration commenced along napes.	Pink Muscle	Light red precipitating crystals.	12.73	116.7	192.5
B/8							Dark Muscle	Orange yellow, cloudy	7.56	116.2	196.4
A/8	Eviscerated, glazed.	7 months	6	18	671	Fish fairly firm. Napes badly rusted. Cut belly also discolored.	Pink Muscle	Clear Red	11.62		190.
7 B/8							Dark Muscle	Yellow Red Clear.	6.78	139.0	189.2
5 A/8	Eviscerated, unglazed.	7 months	6	18	671	Fish fairly firm, napes badly rusted. Body cavity discolored with rust.	Pink Muscle	Light Red	11.32	140.8	191.1
B/8							Dark Muscle and Skin	Yellow red	6.6	127.4	195.8
A/8	Eviscerated, glazed.	12 months	6	18	1,089	Fairly badly rusted; firm	Pink Muscle	Light red CLOUDY	12.42	133.8	193.8
B/8							Dark Muscle and skin	Yellow red	8.84	133.7	191.7

le II continued.

. No.	Description of Fish	Time in Storage	Temp. of Storage Lowest F°	Temp. of Storage Highest F°	Degrees Temp. Fluctuation	Condition of Fish at End of Storage Period.	Source of Oil.	Appearance Of Oil.	Analysis of Oil: Acid Value.	Analysis of Oil: Iodine Value.	Analysis of Oil: Sap. Value
6 A/8	Eviscerated Unglazed	12 months	6	18	1,.89	Fairly firm. Rusting not so bad as in the glazed sample.	Pink Muscle	Red Clear	14.81	----	194.2
B/8							Dark Muscle and Skin	Yellow Red Clear	7.03	112.6	-----
A/8	Glazed Eviscerated	3 months	-20	14	1,110	Good condition, firm, no sign of rusting.	Pink	Clear Red	10.19	125.6	187.4
B/8							Dark	Clear Red	5.66	128.8	190.4
A/8	Unglazed Eviscerated	3 months	-20	14	1,110	Good condition, firm, no signs of rusting.	Pink	Clear Red	7.66	125.8	187.0
B/8							Dark	Yellow [illegible] clear.	3.69	121.6	191.1
A/8	Glazed Eviscerated	6 months	-20	30	3,407	Fair condition, Rust started at cut portions.	Inner pink	Clear Red	14.26	131.7	189.4
B/8							Outer pink	Clear Red	10.84	131.0	189.4
C/8							Dark	Yellow Pink Clear.	8.37	[illegible]	189.3
A/8	Unglazed Eviscerated	6 months	-20	30	3.407	Fair condition, rusting at cut surface more advanced than in Sample 12. [illegible]	Inner Pink	Light Red clear.	21.4	129.6	187.7
B/8							Outer Pink	Light Red clear.	14.12	126.5	186.4
C/8							Dark	Pink Yellow clear.	8.99	122.1	189.9

Ref. No.	Description of Fish	Time in Storage	Temp of Storage Lowest F°	Temp of Storage Highest F°	Degrees Temp Fluctuation.	Condition of Fish at End of Storage Period.	Source of Oil	Appearance of Oil	Analysis of Oil. Acid Value	Analysis of Oil. Iodine Value.	Analysis of Oil. Sap. Value.
5 A/8	Glazed Eviscerated	12 months	-20	30	7,275	Badly distorted, dessicated, glaze nearly off, badly rusted.	Pink	Red Cloudy	14.27	125.9	192.4
5 B/8							Dark	Yellow pink.	11.33	123.9	193.4
A/ 8	Unglazed Eviscerated	12 months	-20	30	7,275	More dessicated than sample 34, very badly rusted at cut surfaces.	Pink	Red Cloudy	14.49	126.9	192 0
B/8							Dark	Yellow pink, Cloudy	11.33	123.9	193.4
	Glazed Eviscerated	6 months	3	11	484	Fairly good condition, slight rust on cut surfaces. Rust slight under glaze on backs.	Pink	Light red crystals.	9.57	142.7	189.2
							Dark.	Yellow pink clear line	6.39	119.9	186 8
	Unglazed Eviscerated	12 months	3	11	484	Rust commenced on cut surface, dessicated, no rust along back.	Pink	Red Clear	10.22	122.8	182.8
							Dark	Yellow clear	6.77	126.3	189.9
7 A	Glazed Eviscerated	12 months	3	13	886		Pink	Red crystalling	14.38	119.1.	189.2
							Dark	Yellow clear.	12.62	114.1	194.1

T A B L E III

No.	Description of Fish	Time in Storage	Conditions of Storage.	Condition of Fish at End of Storage	Source of Oil	Description of Oil.	Acid Values	Iodine Values	Saponification	Ref. Index
	Fresh Humpback Salmon		Temp. 8-15° F.							
A/9	Frozen in the round	6 months	Ordinary atmosphere of storage room (containing a trace of ammonia)	3 Fish, Excellent condition, no rusting or dessication.	Pink Muscle	Pink, clear	14.06			1,4790
B/9					Dark Muscle and skin	Yellow pink, clear.	14.71			1,4785
A/9	Eviscerated before freezing.	6 months	Ordinary atmosphere of storage room (containing a trace of ammonia)	2 Fish. Fairly good condition. Slight rust on cut portion	Pink Muscle	Pink, clear	11.80			1,4796
B/9					Dark Muscle and skin	Yellow pink, clear	13.55			1,4770
0 A/9	Frozen in the round	6 months	Stored in sealed container in atmosphere of Oxygen.	2 Fish. No rusting or dessication.	Pink Muscle	red, clear	14.76			1,4743
0 B/9					Dark Muscle	Yellow pink, clear.	13.62			1,4737
1A/9	Eviscerated before freezing.	6 months	Stored in sealed container in atmosphere of oxygen.	2 Fish. Slight rusting on cut portion	Pink Muscle	Red, clear	[illegible].51			1.4769
B/9					Dark Muscle	Yellow pink clear	15.99			1,4773

ab. No.	Description of Fish	Time in Storage	Conditions of Storage.	Condition of Fish at End of Storage.	Source of Oil	Description of Oil	Analysis of Oil: Acid Value	Iodine Value	Saponification	Ref. Index
6 A/9	Frozen in the round	6 months	Stored in sealed container in atmosphere of ammonia.	2 Fish. Rusting fairly well advanced	Pink Muscle	Pink, clear	1.91			1,4791
6 B/9					Dark Muscle	Yellow, clear.	1.51			1.4796
5 A/9	Eviscerated before freezing.	6 months	Stored in sealed cont[illegible] in atmosphere of ammonia	Rusting fairly well advanced especially around cut portions.	Pink Muscle	Pink, clear	5.66			1.4788
5 B/9					Dark Muscle	Yellow, clear	----			1.4790
4 A/9	Frozen in the round.	6 months	Stored in sealed container in atmosphere of carbon dioxide.	Very slight signs of rusting, fairly good condition.	Pink Muscle	Yellow pink clear	16.66			1.4800
4 B/9					Dark Muscle	Yellow clear.	14.37			1.4770
5 A/9	Eviscerated before freezing.	6 months	Stored in sealed container in atmosphere of carbon dioxide	Very slight signs of rusting, fairly good condition	Pink Muscle	Yellow pink, clear.	15.81			1.4777
					Dark Muscle	Yellow, clear	15.37			1.4770
A/9	Fresh Hack Spring Eviscerated before freezing.	6 months	Ordinary atmosphere of storage room (containing trace of ammonia.	Rusting started but not advanced.	Pink Muscle	Red, clear	10.30	138.2	188.9	1.4736
B/9					Dark Muscle	Yellow pink, clear.	7.79	136.6	188.3	1.4735

No.	Description of Fish	Time in Storage	Condition of Storage	Condition of Fish at End of Storage	Source of Oil	Description of Oil	Analysis of Oil: Acid Value	Iodine Value	Saponification	Ref. Index
A/9	Eviscerated before freezing.	6 months	Tank #1. Stored in sealed container in an atmosphere of oxygen.	No rusting, very good condition.	Pink Muscle	Red, clear	9.92	136.5	188.4	1.4724
B/9					Dark Muscle	Yellow, clear.	8.44	123.2	188.4	1.4731
A/9	Eviscerated before freezing	6 months	Stored in sealed container in atmosphere of ammonia.	Rusting advanced	Pink Muscle	Deep red, clear	5.12	124.2	185.1	1.4742
B/9					Dark Muscle	Yellow pink clear	4.42	117.1	------	1,4748
A/9	Eviscerated before freezing	6 months	Stored in sealed container in an atmosphere of carbon dioxide.	Very slight signs of rust. Very good condition.	Pink Muscle	Light red, clear	11.14	133.4	194.7	
B/9					Dark Muscle	Light orange, clear.	7.92	148.8	191.2	