Progress on Projects, February 1953

REFRIGERATION: Preparation of a Manual on the Refrigeration of Fish: In order to provide certain information to include in the manual, a survey was made of refrigerated retail display cases in approximately 100 supermarkets from Washington, D. C., to Boston, Massachusetts. The following observations were made: (1) availability of frozen fishery products was generally adequate; (2) sales effort towards marketing frozen fish was quite variable; (3) holding temperatures in frozen-food cabinets were generally higher than they should be to protect the quality of the frozen fish. Work on the manual had been delayed due to the illness and subsequent death of the technologist assigned to the project. Another technologist has been transferred from the Boston Technological laboratory to College Park to continue the work. (College Park)

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Freezing Fish at Sea, Defrosting, Filleting, and Refreezing the Fillets: Vessel Operation: VESSEL: Specifications for bids were prepared for the annual overhauling of and for certain alterations on the research trawler Delaware. Alterations on the Delaware will include the installation of a new ventilating system in the freezing-machinery room and the installation of a new basket-type brine freezer. The new brine freezer has been designed so that loading and unloading operations can be carried out on deck. The new arrangement will also allow fuller use of available frozen storage space. The capacity of the brine freezer has been increased so as to permit continuous freezing of fish during normal fishing operations.

LABORATORY: To obtain further information on salt penetration into fish, two samples of round scrod haddock were immersed in cold (5° F.) sodium-chloride brine (25 percent) for 7 days. Weight losses in the two samples during the immersion period were 5.3 percent and 6.4 percent. The fish were extremely desiccated, the surfaces were soft and rubbery, and the tail sections were flexible. The surface areas and tail sections apparently were not frozen even at 5° F. due to the penetration of salt from the cold brine into the meat of the fish. Cross-section cuts of the fish revealed that the center area was frozen, but the outer area--3/16-inch from the surface--was unfrozen. This unfrozen area was highly discolored. The samples were inedible due to saltiness and off-flavors. Further tests will be made to determine the maximum time scrod haddock can be held in cold brine before excessive salt penetration occurs. Present recommended practices of removing the fish from the brine immediately after they are frozen result in negligible penetration of salt. (Boston)

* * * * *

BYPRODUCTS: Vitamin Content and Nutritive Value of Fishery Byproducts: Analyses were made of samples of anchovy, sardine, and mackerel scrap, and whale-loin meal for riboflavin, niacin, and vitamin B_{12} content. The results are as follows:

Sample	Sample Number	Method of Drying	Moisture	Oil	Vitamin Content Moisture-and-Oil-Free Basis		
					Riboflavin	Niacin	Vitamin B_{12}
			Percent	Percent	... Micrograms Per Gram ...		
Anchovy scrap	1	Steam-tube dryer	9.90	8.86	-	-	0.24
	2		3.06	7.61	4.7	41	0.14
Sardine scrap	1	Steam-tube dryer	3.46	6.67	5.6	65	0.26
Mackerel scrap	1	Steam-tube dryer	9.71	7.32	5.5	62	0.40
	2		9.03	6.65	5.6	62	0.41
	3		10.38	6.90	6.8	71	0.49
Whale-loin meal	1	Air dryer (at 160° F. for 10 to 12 hours)	7.26	15.97	10.9	117	0.081
	2		8.82	15.73	10.6	124	0.078

Development of a Dried Product from Condensed Menhaden Solubles or Stickwater: After a delay by the manufacturer, the experimental drum dryer was received at the College Park Technological Laboratory. During the month the equipment was installed and certain preliminary experiments were carried out.

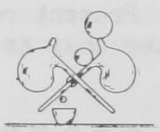
* * * * *

ANALYSIS AND COMPOSITION: Composition and Cold-Storage Life of Fresh-water Fish: The proximate composition of six additional individual samples of lake trout was determined. The fish were caught in June 1952 in Lake Superior.

Composition of Edible Portion of Lake Trout (Cristivomer namaycush)

Sample Number	Length	Weight of Eviscerated Fish	Fillet Yield from the Eviscerated Fish	Proximate Composition of Edible Portion			
				Moisture	Fat	Protein	Ash
	Centi-meters	Grams	Percent	Percent	Percent	Percent	Percent
7	70	2745	63.7	72.4	11.65	17.4	0.97
8	61	1955	59.3	71.0	12.68	18.3	1.13
9	60	1535	59.1	75.1	6.91	18.6	1.03
10	56	1255	58.0	75.7	5.36	18.8	1.08
11	54	930	55.9	77.6	2.76	19.6	0.94
12	54	960	56.1	80.3	1.93	18.4	0.84

(Seattle)



TECHNICAL NOTE NO. 25--AMINO-ACID CONTENT OF SALMON ROE

ABSTRACT

THE "ESSENTIAL" AMINO-ACID CONTENT OF ROE FROM THE FIVE SPECIES OF SALMON HAVE BEEN DETERMINED. ANALYSES OF EIGHT INDIVIDUAL ROE FROM PINK SALMON OF SIMILAR AND MODERATE MATURITY SHOWED NO STATISTICALLY SIGNIFICANT VARIATION IN AMINO-ACID CONTENT. THERE WAS AN INDICATION THAT THE ESSENTIAL AMINO-ACID CONTENT OF KING SALMON ROE SHOWED AN INCREASING TREND WITH MATURITY. THERE WAS NO APPARENT DIFFERENCE IN THE AMINO-ACID CONTENT OF ROE FROM THE FIVE SPECIES OF SALMON WHICH WERE OF SIMILAR (MODERATE) MATURITY. THE AVERAGE AMINO-ACID CONTENT OF MATURE ROE FROM THE FIVE SPECIES OF SALMON EXPRESSED AS A PERCENTAGE OF PROTEIN WAS: ARGININE 7.3; HISTIDINE 2.6; ISOLEUCINE 7.4; LEUCINE 9.9; LYSINE 8.8; METHIONINE 3.0; PHENYLALANINE 4.8; THREONINE 5.7; TRYPTOPHANE 0.9; AND VALINE 7.2.

INTRODUCTION

Studies on the utilization of Alaska salmon-cannery waste have indicated that salmon eggs offer considerable promise for the development of valuable industrial products (Jones, Carrigan, and Dassow 1948), besides serving as an excellent source of feed for hatchery fish (Robinson, Palmer, and Burrows 1949). Little is known, however, about the composition of the protein of salmon roe. Since most of the present uses for salmon eggs depend on their high protein content, 23 to 29 percent (Jones, Carrigan, and Dassow 1948), a study of the amino-acid content of this material was considered desirable.

The amino-acid content of salmon roe was studied by considering possible variation in composition due to species, maturity, and individual variation. Preliminary studies, using the microbiological assay technique, have been limited to determinations of the "essential" amino acids (arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophane, and valine).

EXPERIMENTAL PROCEDURES

Source and Preparation of Samples: The roe from king (Oncorhynchus tschawytscha), chum (O. keta), red (O. nerka), coho (O. kisutch), and pink (O. gorbuscha) salmon, of varying degrees of sexual maturity, were obtained in a random manner from local sources. The skeins were sorted as to relative maturity, sealed in cans, and frozen at -20° F. The frozen samples were stored at 0° F. until needed for analysis.

Crude protein samples were prepared as follows: The eggs were thawed then blended with acetone in an electric blender. The samples were covered with additional acetone and permitted to stand overnight. They were then subjected to continuous Soxhlet extraction with acetone for 20 hours. The residue was dried in a vacuum oven (29 inches of mercury vacuum) at 40° C. for one hour, and finely ground. The dry and fat-free samples were stored in airtight containers.

The degree of development or maturity of the salmon eggs was estimated by means of the arbitrary scale suggested by Davidson and Shostrom (1936). The scale consists of arbitrary values ranging from 1 to 4 in relation to the size and condition of the eggs as they are found either in ovaries or loose in the body cavity of the salmon. Eggs that were found to be small and compact in the ovaries were given a classification of 1. Those that were greatly increased in size and had been shed from the ovaries into the body cavity were given a classification of 4. Those found in intermediate stages of development were graded 2 or 3.

Assay Method: The microbiological procedure and media of Henderson and Snell (1948) were used. This method employs various lactic acid-producing bacteria with a single uniform medium. Acid production was measured by electrometric titration. Streptococcus faecalis R (American Type Culture Collection #8043) was the assay organism used for arginine, leucine, methionine, threonine, and valine; Leuconostoc mesenteroides (A.T.C.C. #8042) was used for histidine, isoleucine, lysine, and phenylalanine; and Lactobacillus plantarum (A.T.C.C. #8014) was used for tryptophane. Protein-acid hydrolysates were prepared by autoclaving 0.5 gm. of the dried protein in sealed pyrex tubes with 20 ml. of 3 N hydrochloric acid for 5 hours at 15 pounds pressure. Alkaline hydrolysates for tryptophane assays were prepared by autoclaving 0.5 gm. of the dried protein with 10 ml. of 5 N sodium hydroxide for 10 hours at 15 pounds pressure in covered, stainless steel beakers. Complete racemization of the tryptophane was assumed. The alkaline hydrolysates were acidified with excess acetic acid and extracted twice with equal volumes of ethyl ether (Neilands, Sirny, Sohljell, Strong, and Elvehjem 1949). Nitrogen determinations were carried out on triplicate aliquots of the protein-acid hydrolysates by the micro Kjeldahl method of Ma and Zuazaga (1942).

RESULTS

The first phase was a determination of the variability of the amino-acid content of egg samples from salmon of identical species and of similar physical characteristics. Eight individual skeins of eggs of moderate maturity (Classification 3) were selected from eight pink salmon and assayed separately. In each case the whole skein was utilized, including the skein membrane and adhering slime layer.

The amino-acid distribution in the eight samples of salmon eggs of similar sexual maturity examined was very similar (table 1), giving no indication of a

Table 1 - Amino-Acid Content of Salmon Egg Protein from Pink Salmon of Similar Sexual Maturity1/

Sample	Skein Number	Amino-Acid Content (In Grams per 100 Grams of Protein)2/									
		Argi-nine	Histi-dine	Isoleu-cine	Leucine	Lysine	Methio-nine	Phenyl-alanine	Threo-nine	Trypto-phane	Valine
Pink Salmon Roe	1	7.5	2.8	7.1	9.0	8.9	2.7	4.8	5.4	1.1	7.8
	2	6.8	2.8	7.0	9.0	8.8	2.7	4.7	5.2	1.0	7.9
	3	7.1	2.9	6.8	9.6	9.0	2.9	4.7	5.1	1.2	8.7
	4	7.2	2.8	6.8	9.3	8.8	3.0	4.7	5.1	1.1	8.8
	5	7.3	2.9	6.9	9.6	8.8	3.2	5.1	5.1	1.1	8.0
	6	6.9	2.8	7.1	9.5	9.1	3.3	5.2	5.0	1.1	8.4
	7	7.5	2.9	7.0	9.7	8.8	3.2	4.9	5.1	1.1	7.7
	8	7.6	2.8	6.9	9.7	8.9	3.3	5.0	5.2	1.1	7.9
	Avg.3/	7.2±0.3	2.8±0.1	7.0±0.2	9.4±0.3	8.9±0.1	3.0±0.3	4.9±0.2	5.2±0.1	1.1±0.1	8.1±0.5

1/ALL EGGS WERE MODERATELY MATURE AND OF CLASSIFICATION 3 (EGG DIAMETER, 6.0 ± 0.5 MM.).
2/PROTEIN EQUALS NITROGEN TIMES 6.25.
3/AVERAGE VALUES ARE GIVEN AS THE MEAN PLUS OR MINUS THE STANDARD DEVIATION.

statistically significant variation, (Recovery experiments, in which amino-acid standards were added to protein hydrolysates, averaged from 96 to 107 percent of the theoretical amount.)

The second phase was a determination of the essential amino-acid content of egg samples from salmon of identical species but of varying sexual maturity. Protein samples were prepared from the eggs of king salmon corresponding to the different arbitrary levels of sexual maturity described previously. For each protein sample, 25-gm. portions were cut vertically from the midsections of 6 individual skeins, combined, and homogenized with acetone. The proportional parts of the roe (egg casings, sac contents, and skein membrane when present) were not considered, since the relative amounts vary with maturity. An effective separation of these components is impossible in some cases and any error thus introduced is probably of small magnitude. A protein hydrolysate was prepared from a representative sample of each homogenate and assayed for the essential amino acids.

In general, there was an indication that for king salmon roe, the essential amino-acid content showed an increasing trend with maturity (table 2). In some

Table 2 - Amino-Acid Content of Salmon Egg Protein from King Salmon of Varying Degrees of Maturity

Sample	Sample Number	Egg Classification[1]	Size of Eggs--Diameter in Millimeters	Amino-Acid Content (In Grams Per 100 Grams of Protein)[2]									
				Argi-nine	Histi-dine	Isoleu-cine	Leu-cine	Ly-sine	Methio-nine	Phenyl-alanine	Threo-nine	Trypto-phane	Va-line
King Salmon Roe	1	1	2.0 ± 0.5	6.5	2.1	5.3	7.0	6.1	2.4	4.3	5.9	0.8	4.9
	2	1	2.0 ± 0.5	7.2	2.4	6.5	8.5	7.7	2.7	5.0	5.8	0.8	6.1
	3	2	4.0 ± 0.5	7.3	2.5	7.2	9.3	8.5	2.7	5.3	5.5	0.8	6.8
	4	3	7.0 ± 0.5	8.0	2.4	7.5	9.4	9.0	3.1	4.9	5.7	0.8	7.0
	5	4	8.0 ± 0.5	7.7	2.4	7.3	9.6	8.9	3.1	4.8	5.7	0.7	7.0
	6	4	8.0 ± 0.5	7.1	2.4	7.2	9.9	8.8	3.0	4.8	5.9	0.9	7.1
"White Spring Salmon Roe"[3]	-	-	-	5.9	1.4	8.2	11.1	1.8	1.9	6.3	4.7	0.7	7.5

1/AN ARBITRARY SCALE INDICATING DEGREE OF SEXUAL MATURITY. CLASS 1 EGGS ARE VERY IMMATURE; CLASS 4 EGGS, VERY MATURE; AND CLASS 2 AND 3 INTERMEDIATE MATURITY (BETWEEN 1 AND 4).
2/PROTEIN EQUALS NITROGEN TIMES 6.25.
3/DATA OF DEAS AND TARR (1949).

cases, the values for immature eggs (Classification 1) were 30 percent lower than the mean for all values of this species.

A comparison of the values obtained for these materials with values reported by Deas and Tarr (1949) for "white spring salmon roe" shows that, with the exception of lysine, the distribution was somewhat similar. The value of 1.8 gm. of lysine per 100 gm. of protein reported for "white spring salmon roe" is unusually low for an animal protein.

For a comparison of the essential amino-acid content of salmon egg samples from the different species, protein samples from moderately mature eggs (Classification 3) from the five species of salmon were analyzed. The results (table 3)

Table 3 - Amino-Acid Content of Salmon Egg Protein from Five Species of Salmon of Similar Sexual Maturity[1]

Sample		Number of Samples Analyzed[3]	Amino-Acid Content (In Grams Per 100 Grams of Protein)[2]									
			Argi-nine	Histi-dine	Isoleu-cine	Leu-cine	Ly-sine	Methio-nine	Phenyl-alanine	Threo-nine	Trypto-phane	Va-line
Salmon Roe	King	1	8.0	2.4	7.5	9.4	9.0	3.1	4.9	5.7	0.8	7.0
	Chum	2	7.2	2.8	7.4	10.2	9.0	3.1	4.8	6.1	0.9	7.2
	Sockeye	2	7.1	2.7	7.6	10.1	8.7	3.0	4.7	5.7	0.8	6.8
	Coho	1	7.3	2.7	8.0	10.2	8.7	3.0	4.9	5.8	0.9	6.8
	Pink	2	7.2	2.8	7.3	9.8	8.9	3.0	4.9	5.6	0.9	7.4
Animal Liver[4]		-	6.5	2.6	5.6	8.4	6.3	3.2	7.3	5.8	1.5	6.2

1/ALL EGGS WERE MODERATELY MATURE AND OF CLASSIFICATION 3. EGG DIAMETERS WERE: KING, 7 MM.; CHUM, 7 MM.; SOCKEYE, 5 MM.; COHO, 6.5 MM.; AND PINK, 5.5 MM. (ALL VALUES TO ±0.5 MM.)
2/PROTEIN EQUALS NITROGEN TIMES 6.25.
3/EACH SAMPLE WAS PREPARED FROM ROE REPRESENTING SIX OR MORE FISH.
4/DATA OF BLOCK AND BOLLING (1945).

indicate no apparent difference in the essential amino-acid content of the eggs obtained from the five species of salmon of moderate and similar maturity.

A detailed evaluation of the data reported in this paper will not be attempted. Corresponding amino-acid values for animal liver, a feed commonly used for hatchery fish, are presented for comparison (table 3). Any conclusions as to the quality of salmon-egg protein (based on amino-acid content) as compared with animal liver, for example, are not warranted, since the amino-acid requirements of hatchery fish have not been established. Further studies on the variability of the amino-acid content of salmon egg protein from the different species of salmon of varying sexual maturity are being carried out, and will be reported in more detail at a later date.

LITERATURE CITED

BLOCK, R. J. AND BOLLING, D.
1945. THE AMINO ACID COMPOSITION OF PROTEINS AND FOODS. SPRINGFIELD, ILL., CHARLES C. THOMAS, PUBLISHER.

DAVIDSON, F. A. AND SHOSTROM, O. E.
1936. PHYSICAL AND CHEMICAL CHANGES IN THE PINK SALMON DURING THE SPAWNING MIGRATION. INV. REP. NO. 33, VOL. 2, DEPT. OF COMMERCE, BUREAU OF FISHERIES.

DEAS, C. P. AND TARR, H. L. A.
1949. AMINO ACID COMPOSITION OF FISHERY PRODUCTS, JOURNAL OF THE FISHERIES RESEARCH BOARD OF CANADA, VOL. 7, PP. 513-21.

HENDERSON, L. M. AND SNELL, E. E.
1948. A UNIFORM MEDIUM FOR DETERMINATION OF AMINO ACIDS WITH VARIOUS MICROORGANISMS. THE JOURNAL OF BIOLOGICAL CHEMISTRY, VOL. 172, PP. 15-29.

JONES, G. I.; CARRIGAN, E. J.; AND DASSOW, J. A.
1948. UTILIZATION OF SALMON EGGS FOR PRODUCTION OF CHOLESTEROL, PROTEIN, AND INDUSTRIAL FAT. DEPT. OF COMMERCE, I.R.R.D. REPORT NO. 2, PP. 9-32.

MA, T. S. AND ZUAZAGA, G.
1942. MICRO KJELDAHL DETERMINATION OF NITROGEN. INDUSTRIAL AND ENGINEERING CHEMISTRY, ANALYTICAL EDITION, VOL. 14, PP. 280-82.

NEILANDS, J. B.; SIRNY, R. J.; SOHLJELL, F. M.; STRONG, F. M.; AND ELVEHJEM, C. A.
1949. THE NUTRITIVE VALUE OF CANNED FOODS. THE JOURNAL OF NUTRITION, VOL. 39, PP. 187-202.

ROBINSON, L. A.; PALMER, D. D.; AND BURROWS, R. E.
1949. TESTS OF HATCHERY FOODS FOR BLUEBACK SALMON. DEPT. OF INTERIOR, SPECIAL SCIENTIFIC REPORT, FISHERIES, NO. 60.

--By Harry L. Seagran, Biochemist,
Fishery Products Laboratory,
Fisheries Experimental Commission,
Ketchikan, Alaska

MAYONNAISE WITH FISH-PROTEIN STABILIZER WITHSTANDS HEAT PROCESSING

By substituting egg yolk with a fish-protein stabilizer (manufactured by a firm in Hamburg, Germany), it is possible to make mayonnaise which withstands heat processes normally used for canned foods. The experiments were made with shrimp packed in mayonnaise. (Report of the Technological Laboratory of the Danish Ministry of Fisheries, 1951.)

--World Fisheries Abstracts, vol. 3, no. 6 (Nov.-Dec. 1952), p. 1.