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DEVELOPMENT OF RATIONS FOR THE ENHANCED SURVIVAL OF SALMON

Annual Report 1988

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Development of Rations for the
Enhanced Survival of Salmon

Annual Report
1988

by

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ABSTRACT

The nutritional quality of feed plays an important role in determining the health and "fitness" of smolts. Commercial fish meal, the major source of protein in salmon rations, may be reduced in quality from poor drying techniques during manufacture. Dietary stress in the hatchery may result. This investigation tests the hypothesis that protein quality of fish rations can influence the survival of smolts and the ultimate return of adults. The test involves a comparison between performances of coho and chinook salmon reared on rations containing very high quality protein derived from vacuum dried meals and those of fish reared on commercial rations, with commercial fish meal as a source of protein. Survival and return of several brood years of test and control fish are used to measure the influence of ration on survival.

Rearing and release of tagged fish to date include 1982, 1983, 1984 and 1985 broods of coho salmon; the 1983 and 1984 broods of fall chinook (tule stock) salmon; and the 1985 and 1986 broods of fall chinook (upriver bright stock) salmon. This report includes recovery data from these marked fish collected through September 1988.

Recovery data of the 1982, 1983, and 1984 broods of coho salmon (Sandy stock) suggested an improved survival for fish supplied test rations. Recovery rates varied significantly ($P < 0.05$) by brood year and ration treatment. The interaction of ration and brood year was not significant. Recovery data (as of September 1988) showed a somewhat (but not significant) improved survival for the 1983-brood fall chinook salmon (tule stock) reared on test ration. Control rations produced better survival for the 1984 brood fall chinook salmon. This latter result was due to the use of an unpalatable test ration for three months prior to release. The fish oil supplement used was highly susceptible to

autooxidation, could not be stabilized with antioxidants, and quickly became rancid.

INTRODUCTION

The natural habitat for the spawning and rearing of salmon in the Columbia River systems has been reduced by hydroelectric development and other encroachments. Artificial production of salmon in hatcheries has become a critical link in the restoration of stocks.

Time of release, natural abundance of food, fish size, and the health or "fitness" play important roles in determining survival of hatchery-reared smolts and their ultimate return as adult fish. It is believed that nutritional quality determines the health and/or "fitness" of smolts. Ration regimes containing high quality components in uniform and fine-free pellets produce efficient fish growth and minimize loss of nutrients, resulting in fish that are less susceptible to disease and of more uniform size at release. Smolts produced by these high quality feeds are thought to migrate rapidly to the sea and successfully adapt to salt water.

Quality in fish feed is determined in large part by its protein complement. Protein is the major food component in fish rations. The most successful fish rations rely on large quantities of fish protein in the form of fish meal. Plant sources of protein, such as soybean and cottonseed meal, are tolerated to a certain extent, but an excessive replacement of fish protein with plant protein results in a reduction in feed consumption, conversion, and/or weight gain. This reduction creates a dietary stress that affects smolt "fitness."

Commercial fish meal supplies used to formulate successful rations are declining in availability and quality. Industrial round (whole) fish that, in the past, formed the raw material base for high quality meal is no longer available because of cost and/or regulation dictating its use for human food.

Carcass waste is replacing round fish as a raw material. Resulting meals have a lower protein content and an elevated mineral level because of the removal of muscle tissue for human food. The majority of meals are produced by high-temperature direct-flame dryers. Excessive heating damages the proteins and initiates lipid-protein interactions. Both of these effects reduce the biological value of fish proteins.

Meals and fish protein concentrates produced from round fish and/or fish-processing waste using processes with low temperatures and reduced pressures yield protein of optimum quality. These gentle drying and concentration procedures coupled with the use of fat antioxidants limit heat damage to proteins and lipids, and markedly reduce lipid-protein interactions. Ration regimes that incorporate these sources of protein are more costly, but additional feed costs may be offset by greater survival of smolts and increased return of adult fish. Efficiency of hatchery production would thus be improved.

The basic hypothesis of this investigation is that protein quality of the rations can influence the survival of smolts and the return of adult salmon to the Columbia River basin. The general approach to test this hypothesis involves the rearing of coho and chinook salmon on rations containing a high quality fish protein complement. Fish reared on hatchery rations with commercial fish meals as a source of protein were used as controls. Coded-wire tagging experiments were conducted on replicate brood years of test and control fish to determine the influence of ration protein on survival. Beginning with the 1985 brood, fall chinook (upriver bright stock) salmon were assessed for physiological changes associated with smoltification and correlated with ration type and smolt "fitness." These physiological changes are discussed in other reports.

Project rearing and release of tagged fish to date include 1982-, 1983-, 1984-, and 1985-brood coho salmon, the 1983-, and 1984-brood fall chinook salmon

(tule stock) and the 1985- and 1986-brood fall chinook salmon (upriver bright stock). This report includes preliminary recovery data on these release groups collected through September 1988.

METHODS AND MATERIALS

General Project Operation

This project combines the facilities and expertise of the Seafoods Laboratory of the Department of Food Science and Technology, Oregon State University, and the Oregon Department of Fisheries and Wildlife (ODFW). ODFW carried out fish husbandry tasks involved in feeding trials at their Sandy and Bonneville hatcheries and conducted coded-wire tagging of experimental and control groups. The task of ration component acquisition and/or production and test ration production and characterization were carried out at the Seafoods Laboratory. The Department of Fisheries and Wildlife, Oregon State University, and ODFW examined physiological changes due to diet during the course of parr-smolt transformation in the fall chinook.

Formulation and Production of Test Feeds

The hatchery supply of rations (Oregon Moist Pellets) served as a control ration for both coho and fall chinook salmon. This included, when applicable, Biomist Starter Ration and the OP-4 and OP-2 formulations of the Oregon pellet feed. Coho salmon were supplied with two test rations deriving their major protein complement from vacuum-dried carcasses of salmon collected from hatcheries and from vacuum-dried round Pacific hake. A single test ration containing vacuum-dried salmon meal as the major protein source was supplied to fall chinook. The major protein complements provided by both vacuum-dried salmon and hake were supplemented by hydrolyzed and vacuum-concentrated bone-free fish derived from round hatchery salmon carcasses.

Ration Component Production and Acquisition

Advanced Hydrolyzing Systems, Inc. of Astoria, OR, in cooperation with the Seafoods Laboratory, produced high quality vacuum dried meal, using the facilities, power, and steam of the Seafoods Laboratory. Concentrated hydrolysates were produced in the company's own facilities. Salmon carcasses were provided by the Oregon Department of Fish and Wildlife. Hake and groundfish carcass waste were purchased on the open market.

Fish meals were prepared by placing coarse ground fish into a steam jacketed chamber equipped with a stirring-scraping device, and subjecting the meal to a vacuum of 25-27 inches of Hg. Product temperature was maintained at 101-105 °F except for pasteurization, when the temperature of the moist feed was raised to 180 °F for 5.0 minutes. All vacuum dried meals, if not used immediately for ration preparation, were sacked and held frozen <) °F.

Concentrated fish hydrolysates were prepared by exposing coarse ground fish to a temperature of approximately 140 °F with mechanical agitation until sufficient liquefaction was achieved to allow screen removal of bones. The temperature of the liquefied material was raised to 180 °F to achieve pasteurization and then concentrated in vacuum with scraped surface heat transfer equipment to approximately 50% solids. Concentrates were sacked or boxed, cooled and then frozen and stored at <0 °F.

Remaining components required for ration preparation were purchased from commercial firms that either produce moist pelletized fish rations or provide components to the fish feed industry. All purchased components met specifications for the Oregon pellet feed.

Test Ration Formulation and Production Protocol

Test rations were formulated to contain 28 lbs of protein derived from meal and 7.7 lbs from concentrated hydrolyzed fish for each 100 lb of ration. Water

and wheat germ meal were added to yield rations with 76% solids (24% moisture). Herring oil was added in amounts needed to provide a fat:protein caloric ration of 0.95 (protein = 4.0 kcal/g, fat = 9.0 kcal/g). A computer controlled the percentage of vacuum dried meal and concentrated hydrolyzed fish used for each batch of ration. The formulation of test and control rations is listed in Appendix II. Ration dry components (vacuum dried fish meal, wheat germ meal, dried whey product, spray dried blood, mineral and vitamin premixes, and sodium bentonite) were mixed in 600-1000 lb batches and hammer-milled to achieve a fine particle size. Milled dry mix was sacked in 50 lb units and held frozen at 0 to -30 °F if not immediately used to prepare rations.

Milled dry mix was mechanically mixed with remaining "moist" components (antioxidant-stabilized herring oil, choline chloride, concentrated hydrolyzed fish, and water) in 150-250 lb batches. The thoroughly mixed components were then mechanically extruded into pellets of the desired length and diameter, screened to remove fines, sacked into 40 lb (1/32-inch pellets only) or 50 lb units, and immediately frozen at -30 °F.

Ration Composition Control

The proximate analysis (moisture, ash, protein and fat content) of test and control rations was determined to assure composition and for computation of dry weight and protein consumption and conversion. The entire hatchery supply of control ration was sampled by pellet size and, if possible, by production date. Test rations were sampled during production. At least two samples were selected for analysis from each 150-250 lb batch. The mean for all samples derived from each production lot prepared from the same dry mix formulation was used as the composition of a particular lot of pelletized ration. The mean composition of each pellet size of control ration was used to compute dry weight and protein consumption and conversion.

Husbandry Protocol

Coho Salmon (Sandy Hatchery): Coho salmon (Oncorhynchus kisutch) (Sandy stock) were reared in 20 x 80 x 4 ft. (variable depth) raceways with a volume of 4,290 cu. ft. (32,089 gal.) at a maximum water depth of 3.5 ft. Raceways were supplied with 228 to 396 gpm/pond of Cedar Creek water that varied from 38 to 59°F (three year monthly mean range) (Appendix I). The lowest flow rates occurred during the summer, and the highest during the spring before release of smolts. The hatchery had north- and south-facing banks of ten ponds each with a separate head box for each bank. The north head box was constructed so that only a single pass of water will go into each pond. The south head box was equipped with a pipe and pump system that was used to recirculate water into the head box (along with the normal creek water). This system was used only during the summer and early fall when the water flow in the creek was too low to meet the needs of the hatchery. Under normal circumstances, the pump is used only three months during the year.

Groups of 600,000 to 650,000 unfed fry were placed in each pond during late March or early April at about 1,100 fish/lb (0.4 g/fish). Fish were supplied starter ration and progressed through the pellet size guide for salmon recommended by the Oregon Department of Fish and Wildlife for moist pelletized feeds:

Pellet size (in.)	Fish size	
	fish/lb	g/fish
Starter	1,000-700	0.4- 0.6
1/32	700-500	0.6- 0.9
3/64	500-250	0.9- 1.8
1/16	250-150	1.8- 3.0
3/32	150- 50	3.0- 9.1
1/8	50- 13	9.1-34.9

Fish at 195 to 212 fish/lb (2.1 to 2.4 g/fish) were randomly distributed in 10-lb lots into six ponds on April 30 to June 11. Final numbers were 54,000 to 60,000 fish per pond. Control rations and two test rations were randomly assigned to provide duplicate ponds for each ration type.

Control rations and two test rations were supplied to fish for about 10 months from June to release on April 30. Each ration (in recommended pellet sizes) was fed by hand to replicate ponds of fish at the feeding frequencies listed as follows:

Fish size (fish/lb)	Feeding frequency (times/day)
1,200-800	8-10
800-500	6
500-250	4
250-150	3
150- 15	1-2

Control fish were supplied feed according to a feeding guide which scheduled fish to be 15 fish/lb (30.24 g) at liberation. Fish supplied test rations were fed at a rate less than the feeding rate guide to achieve equal size at liberation.

Fall Chinook Salmon (Bonneville Hatchery): At Bonneville Hatchery, fall chinook salmon (upriver bright stock) (*Oncorhynchus tshawytscha*) were reared in well water (49-51⁰F) in 17.5 x 75 x 3 ft. raceways (3,948 cu ft.; 29,456 gal.). Flow rate was gradually increased from 300 gpm/pond for swim-up fry to 550 gpm/pond for fingerlings. Maximum loading occurred at 6 lbs of fish/gpm at liberation.

Approximately 600,000 unfed fry of the tule stock were stocked in each pond in late December at an average size of 750 to 1,100 fish/lb (0.4 to 0.6 g/fish). About 200,000 to 400,000 unfed fry of the upriver bright stock were stocked in each pond in February or March at an average size of 980 to 1,060 fish/lb (0.4 to 0.5 g/fish). Tule stock were fed on a demand basis until release in early May. Upriver bright stock were fed at a rate designed to achieve a target release size of 13 fish/lb in mid-October. The 1986-brood fish were liberated early due to an emergency low water supply and did not reach the target size. Control and test fish were initially supplied starter rations and then progressed through the pellet size guide recommended by the Oregon Department of Fish and Wildlife for moist pelletized feeds listed above.

Pathological Assessment

Oregon Department of Fish and Wildlife pathologists responded to any increase in mortality rates that occurred. At the pathologists discretion, appropriate diagnostic tools were employed to determine the causative agent, and remedial treatments were prescribed.

Physiological Assessment

Methodology and results are reported in past reports. No further analyses were provided during the past year.

Growth Response Parameters

Fish weight, feed consumption, feed conversion and mortality information were determined at monthly intervals and reported at two to three month intervals for coho and fall chinook salmon. At liberation, fork length, weight and blood hematocrits were measured and samples of fish from each pond were collected for the determination of body composition.

Mean fish weight and length were based on the measurement of three to six

randomly selected samples (varying in weight depending on fish size) of the pond populations. Feed consumption and mortality were recorded daily. Feed conversion (feed/gain) was computed on a cumulative and period basis for interim reporting purposes and on both a wet and dry weight basis for the entire rearing period at liberation. The blood hematocrit level for each pond replicate was the mean of twelve to fourteen fish. Determinations of body composition were based upon the means of duplicate analyses of three randomly selected samples of ten fish from each replicate.

The emergency release of 1986-brood fall chinook salmon (upriver bright stock) from Bonneville hatchery precluded the above sampling schedule. One sample from each replicate pond (293-329 fish each) was obtained at release and immediately frozen. Fish were thawed, weighed, and measured. Pooled samples were used to determine body composition. Blood hematocrit levels were not determined in these groups.

Coded-Wire Tagging Experiments

Groups of 25,000 to 31,000 coho salmon in each experimental and control replicate were tagged and marked with an adipose fin clip during September or October. Coho were randomly selected for tagging by passing the entire pond of fish over a sampling table which was adjusted to select the desired percentage of fish. Tule stock fall chinook salmon were similarly tagged and marked in April. Groups of 75,000 to 80,000 fish were marked in each replicate of control and test fish. Upriver bright stock of fall chinook salmon were tagged and marked in August. Groups of about 32,000 to 47,000 fish were marked in each replicate of control and test fish. Fish were randomly selected using a procedure similar to that used for coho salmon. Tag retention in fish from each replicate was determined prior to release, except for the emergency release of 1986-brood fall chinook salmon. In these groups, tag retention numbers were

determined from frozen samples used to determine weight and length measurements.

Analysis of Recovery Data

Tag recovery information was analyzed using a factorial design for analysis of variance. The significance of differences between treatment means was determined using least significant difference (LSD) procedures. Data were converted from percent to arcsin derivatives for analysis. All statistical comparisons were made at the significance level of $P < 0.05$ unless otherwise indicated.

RESULTS AND DISCUSSION

Project releases of coded-wire tagged fish to date include the 1982, 1983, 1984 and 1985 broods of Sandy stock coho salmon (Sandy Hatchery), the 1983 and 1984 broods of tule stock fall chinook salmon (Bonneville Hatchery) and the 1985 and 1986 broods of upriver bright stock fall chinook salmon (Bonneville Hatchery). Preliminary recovery of coded-wire tags from the 1982, 1983, 1984, and 1985 broods of coho salmon (Sandy stock) and 1983, 1984, and 1985 broods of fall chinook salmon through September of 1987 have been reported. Recoveries include those from the hatchery and from the fishery. The recoveries from the fishery may or may not have been expanded depending upon the fishery and the brood year.

Test rations, those containing vacuum dried salmon and hake meal, appear to alter the survival of coho salmon, but not in a uniform manner (Table 1). Analysis of variance of the percent of the tags recovered to date (September 88) from the 1982-, 1983-, and 1984-brood releases in a 3 x 3 factorial design showed that the recovery rates varied significantly by brood year and ration treatment. The interaction of ration and brood year was not significant. The data for the 1985-brood fish were incomplete and not included in the analysis.

Recovery rates did not vary significantly between groups for the 1982-brood coho salmon, but recovery rates of the 1983-brood fish fed salmon meal and hake meal were significantly greater than those of the control groups fed Oregon Mist Pellet. Also, recovery rates of the 1984 brood for the salmon meal treatment were statistically greater than the other two treatments. Furthermore, all of the individual recovery means differed by brood year (1982, 1983, and 1984).

Results from the recovery of coded-wire tags from the 1983 and 1984 broods (tule stock) and the 1985 brood (upriver bright stock) of fall chinook salmon are preliminary and represent data available as of September 1988 (Table 2). Due to incompleteness, the recovery data from the 1985 brood were not included in the statistical analysis. Analysis of variance (2 x 2 factorial design) of tag recovery data to date shows a significant variation with respect to ration, brood year, and ration and brood year interaction.

Recovery of tagged fish from the 1983 brood suggested some improvement in survival by fish supplied the salmon meal ration, but the difference was not significant. For the 1984 brood, the recovery rate of the control treatment was significantly greater than the test ration. This is not a surprising result. Growth of fish between mid-February, 1985, and release in May, 1985, was compromised in an intermittent manner by poor ration palatability that resulted in reduced feed consumption and conversion. Poor ration palatability was traced to one of two lots of herring oil used to prepare rations. The lot of herring oil that produced the problems was not oxidized when the ration was made (based upon chemical analysis), but contained only traces of antioxidant. Although antioxidant protection was increased to four times that normally incorporated into the ration, the ration tended to become rancid and unpalatable to the fish.

Table 1. Summary of preliminary tag recoveries from coho salmon reared at Sandy Hatchery

Brood year, diet	Tag code	Tagged fish release	Number recovered at Age ¹			Percent of release ²
			2	3	Total	
1982						
OMP						
7-29-13		25,763	4	492	496	1.93
7-29-06		26,983	3	475	478	1.77
Mean						1.85 ^a
Salmon meal						
7-29-12		25,250	4	444	448	1.77
7-29-09		26,573	8	513	521	1.96
Mean						1.87 ^a
Hake meal						
7-29-10		26,654	7	515	522	1.96
7-29-07		26,095	3	447	450	1.72
Mean						1.84 ^a
1983						
OMP						
7-30-45		25,683	20	1,915	1,935	7.53
7-31-05		26,459	53	2,047	2,100	7.94
Mean						7.74 ^b
Salmon meal						
7-30-48		26,673	66	2,401	2,467	9.25
7-31-06		25,743	68	2,229	2,297	8.92
Mean						9.09 ^c
Hake meal						
7-30-47		25,493	48	2,216	2,264	8.88
7-31-07		25,827	55	2,308	2,363	9.15
Mean						9.02 ^c
1984						
OMP						
7-37-46		27,623	11	907	918	3.32
7-36-20		27,974	6	841	847	3.04
Mean						3.18 ^d
Salmon meal						
7-37-45		28,079	9	1,104	1,113	3.96
7-36-19		27,115	10	1,023	1,033	3.80
Mean						3.88 ^e

Table 1. Continued

Brood year, diet	Tag code	Tagged fish release	Number recovered at Age ¹			Percent of release ²
			2	3	Total	
1984 (continued)						
Hake meal						
7-36-18		27,489	6	865	871	3.16
7-36-23		27,542	7	889	896	<u>3.25</u>
	Mean					3.21 ^d
1985						
OMP						
7-44-47 R2		32,011	42	119	161	0.50
7-41-19 R2		31,475	51	116	167	<u>0.53</u>
	Mean					0.52 ^f
Salmon meal						
7-44-42 R2		30,839	52	99	151	0.48
7-41-21 R2		30,927	62	126	188	<u>0.60</u>
	Mean					0.54 ^f
Hake Meal						
7-44-41 R2		29,410	16	29	45	0.15
7-41-11 R2		28,560	31	53	84	<u>0.29</u>
	Mean					0.229

¹Includes catch and escapement data available through September 1988.

²Mean values in a column with same exponent letters did not vary significantly (\$0.05).

Table 2. Summary of preliminary tag recoveries of fall chinook salmon (tule stock) reared at Bonneville Hatchery/

Brood Year, Diet	Tag Code	Tagged Fish Release	Number Recovered at Age ¹				Percent of Release ²
			2	3	4	Total	
1983							
OMP							
7-31-20		80,348	5	9	0	14	0.02
7-31-21		80,048	6	39	0	45	0.06
Mean							0.04 ^a
Salmon Meal							
7-31-22		80,138	9	54	2	63	0.08
7-31-23		81,282	7	28	5	40	0.05
Mean							0.07 ^a
1984							
OMP							
7-33-22		78,367	314	1,851	5	2,170	2.77
7-29-20		78,962	304	1,685	4	1,993	2.52
Mean							2.65 ^b
Salmon meal							
7-33-24		80,242	148	1,323	1	1,472	1.83
7-33-23		79,750	110	1,201	4	1,315	1.64
Mean							1.73 ^c
1985							
OMP							
7-37-52		46,579	21	0	-	21	0.05
7-37-53		47,268	36	1	-	37	0.08
Mean							0.07
Salmon meal							
7-36-35		46,852	19	2	-	21	0.04
7-36-36		47,250	26	1	-	26	0.06
Mean							0.05

¹Includes catch and escapement data available through September 1988.

²Mean values in a column with same exponent letters did not vary significantly (40.05).

APPENDIX I. Monthly Water Temperatures (⁰F) at Sandy Hatchery

Mnth	1983			1984			1985		
	Max.	Min.	Av.	Max.	Min.	Av.	Max.	Min.	Av.
January	44	42.5	43.3	42	40	41	39	37	38
February	45	44	44.5	45	42.5	44	39	37	38
March	47	45	46	47	44	45.5	43	40	41.5
April	49.5	45	47	47.5	44	45.8	48	44	46
May	54.5	49.8	52.1	51	47	49	52	47	49.5
June	56	52	54	54	50	52	56	51	53.5
July	56.6	53.4	55	61	54	57.5	66	58	62
August	61	56	58.5	62	56	59	61.4	55.2	58.3
September	60	45	54	60	48	54	59	48	53.2
October	51	47	49	49	47	48	49	46	47.5
November	47	46	46.5	45	44	44.5	41	40	46.5
December	41	39	40	41	40	40.5	37	36	36.5

APPENDIX II. Ration Formulations. Components are designated as percentages of total wet weight of ration.

Component	Control Ration	Test Rations	
		Hake Meal	Salmon Meal
Fish meal	28.0 (min) ¹¹	40.0-48.4 ¹²	37.7-41.4 ¹²
Cottonseed meal ¹	15.0	0.0	0.0
Dried whey product ²	5.0	2.0	2.0
Wheat germ meal ³	Remainder	Remainder	Remainder
Corn distillers solubles ⁴	4.0	0.0	0.0
Trace mineral premix ⁵	0.1	0.1	0.1
Vitamin premix ⁶	1.5	1.5	1.5
Spray dried blood meal ⁷	0.0	2.0	2.0
Sodium bentonite	0.0	2.0	2.0
Concentrate hydrolyzed fish ⁸	0.0	19.7-22.3	19.7-22.3
Choline chloride ⁹	0.5	0.5	0.5
Pasteurized wet fish ¹⁰	30.0	0.0	0.0
Fish oil	6.0-6.75 ¹³	1.8-7.5 ¹⁴	7.7-10.6 ¹⁴
Water	0.0	8.1-12.6	8.1-10.5
Total	100.0	100.0	100.0

¹Preprocessed, solvent extracted, min. 48% protein, max. 0.055% free gossypol.

²Min. 12% protein, max. 6% moisture, max. 10% ash, max. 3% salt

³Min. 23% protein and 7% fat

⁴May contain up to 30% "grains" in place of solubles

⁵Gm/lb: Zn (ZnSO₄), 34.0; Mn, 34.00 (MnSO₄) 9.10; Fe, Cu (CuSO₄) 0.70; I (ethylenediamine dihydroiodide), 4.54; diluted to 1.00 lb with cereal product

⁶Mg/lb: d-biotin 18.0; vitamin B 535.0 (pyridoxine HCl, 650 mg); B₁₂, 1.8; vitamin C (ascorbic acid), 27,000.0 vitamin E (water-dispersable alpha tocopheryl acetate), 15,200.0; folacin (folic acid), 385.0; Myo-inositol (not as phytate salt), 4000.0; vitamin K, 180.0 (menadione sodium bisulfite complex, 545 mg); niacin, 5700.0; d-pantothenic acid, 3200.0 (d-calcium pantothenate, 3478 mg or d,l-calcium pantothenate, 6957 mg); riboflavin, 1600.0; thiamine, 715.0 (thiamine mononitrate, 778 mg); dilute to 1.0 lb with cereal product

⁷Spray dried Whole blood

⁸Concentrated bone-free hydrolysate of salmon carcasses, groundfish carcass waste and whole Pacific hake

⁹Liquid, 70%

APPENDIX II (Continued)

¹⁰**Two or more of the following, with none exceeding 50% of the combination;**
(1) Salmon or tuna viscera (no heads or gills, with livers); (2) whole herring;
(3) bottom fish (whole or fillet scrap); (4) dogfish; (5) whole hake; and (6)
whole salmon. Approved enzymes used to aid digestion.

"Herring meal (min. 67.5% protein) used at no less than 50% of the fish meal in each batch. Anchovy (min. 65% protein), capelin (min. 67% protein), or hake (min. 67% protein) meals may be used as the remainder. Level to supply not less than 21.5% fish meal protein; max. 5% NaCl; 8-12% fat; max 17% ash.

¹²**Vacuum dried**

¹³**Herring, salmon, menhaden, dogfish (not more than 3%), or refined tuna oil; stabilized with 0.4% BHA-BHT (1:1); free fatty acids not more than 3%; BHA-BHT must be added at the time of reprocessing if reprocessed oil is used. Special condition when using hake as a wet fish: add 0.5% oil for every 10% hake in total ration.**

¹⁴**Herring oil; stabilized with 0.02% BHA-BHT (1:1); free fatty acids not more than 3%**