# Canadian Technical Report of Fisheries and Aquatic Sciences No. 1206

# NUTRITION AND MARINE SURVIVAL OF CHINOOK SALMON (Oncorhynchus tshawytscha).

I. POTENTIAL ROLE OF SMOLT BODY COMPOSITION (ROBERTSON CREEK HATCHERY 1979 BROOD).

December 1983

bу

M. Dianne Plotnikoff $^1$ , David A. Higgs $^1$ , Jack R. Markert $^1$ , Bakhshish S. Dosanjh $^1$ , Jack R. McBride $^1$  and J. Thomas Buckley $^2$ 

With the assistance of: Karl A. Petersen and Gerry R. Cook

<sup>1</sup> Fisheries Research Branch Dept. of Fisheries and Oceans West Vancouver Laboratory 4160 Marine Drive West Vancouver, B.C. V7V 1N6 Canada

Dept. of Biochemistry and Microbiology University of Victoria Victoria, B.C. V8W 2Y2 Canada

(c)Minister of Supply and Services Canada 1983
Cat. No. Fs 97-6/1206 ISSN 0706-6457

#### ABSTRACT

Plotnikoff, M.D., D.A. Higgs, J.R. Markert, B.S. Dosanjh, J.R. McBride, and J.T. Buckley. 1983. Nutrition and marine survival of chinook salmon (Oncorhynchus tshawytscha). I. Potential role of smolt body composition (Robertson Creek Hatchery 1979 brood). Can. Tech. Rep. Fish. Aquat. Sci. No. 1206: jv + 20 p.

Juvenile chinook salmon with different levels of body lipid and protein were differentially tagged and released from Robertson Creek Hatchery at the end of the freshwater rearing stage to determine whether body composition plays a role in ocean survival. Differences in body composition were created by feeding triplicate groups of approximately 8,000 fish either an experimental dry diet with one of three levels of protein and lipid or Oregon Moist Pellets (OMP), the standard hatchery diet. Lipid levels in the dry diets were 8, 13 or 17% of dry matter and protein to lipid ratios varied between 3.0 and 6.8. OMP had 17% lipid on a dry matter basis with a protein to lipid ratio of 2.8. Inclusion of the OMP diet permitted evaluation of fish performance when fed either a dry or semi-moist (OMP) diet.

The different dietary lipid levels or protein to lipid ratios had pronounced effects on growth, food, protein and energy utilization, and on body proximate and lipid composition. Growth, food, protein and energy utilization were best for fish fed diets with highest lipid content. Body protein and lipid contents were respectively directly and inversely related to dietary protein to lipid ratio. The fatty acid composition of total body neutral lipid mirrored that of dietary lipids whereas changes were less pronounced in phospholipid. Percent returns of two and three-year old fish to the hatchery were low in all cases and did not permit any definite conclusions regarding the influence of smolt body composition on ocean survival. Food category did not influence fish performance.

Key words: diet, body composition, marine survival, salmon.

#### RÉSUMÉ

Plotnifoff, M. D., D. A. Higgs, J. R. Markert, B. S. Dosanjh, J. R. McBride, and J. T. Buckley. 1983. Nutrition and marine survival of chinook salmon (Oncorhynchus tshawytscha). 1. Potential role of smolt body composition (Robertson Creek Hatchery 1979 brood). Can. Tech. Rep. Fish. Aquat. Sci. No. 1206: iv + 20 p.

Nous avons apposé des étiquettes distinctes à des saumons quinnats junéniles, aux niveaux de lipides et de protéines différents, et les avons relâchés de la piscifacture du ruisseau Robertson à la fin de la phase d'élevage en eau douce, afin de déterminer si la composition des tissus corporels jouait un rôle dans la survie en milieu océanique. Nous avons obtenu ces variations dans la composition corporelle en nourrissant des groupes triples composés d'environ 8000 individus, soit avec une nourriture sèche expérimentale contenant un des trois niveaux de protéines et lipides ou avec des boulettes humides Oregon (Oregon Moist Pellets), le régime habituel des piscifactures. Les taux de lipide dans la nourriture sèche s'élevaient à 8, 13 et 17 % de matières sèches et les rapports protéines/lipides variaient entre 3.0 et 6.8. En fonction de l'état sec, les boulettes humides contenaient 17% de lipides et le rapport protéines/lipides était de 2.8. L'utilisation de nourriture humide a permis l'évaluation du rendement du poisson nourri soit d'aliments secs ou mi-humides.

Les différents niveaux de lipides dans les aliments, ou les rapports protéines/lipides, ont eu une incidence élevée sur la croissance, l'utilisation de la nourriture, des protéines et de l'énergie et sur la composition immédiate et lipidique corporelles. La croissance et l'utilisation de la nourriture, des protéines et de l'énergie ont été supérieures chez les poissons nourris d'aliments contenant le plus de lipides. Les teneurs en protéines et en lipides corporels ont respectivement été directement et inversement reliés au rapport protéines/lipides des aliments. La quantité d'acides gras contenus dans les lipides neutres corporels a reflété celle des lipides alimentaires tandis que, chez les phospholipides, la variation a été moins prononcée. Le pourcentage des poissons de deux et de trois ans qui sont retournés à la piscifacture a été faible dans tous les cas et n'a pas permis la formulation de conclusions précises pour ce qui est de l'influence de la composition corporelle des saumoneaux sur la survie en milieu océanique. Les variétés d'aliments n'ont pas influé sur le rendement du poisson.

Mots-clés: alimentation, composition corporelle, survie en milieu océanique, saumon

#### INTRODUCTION

One of the major goals of the Salmonid Enhancement Program in British Columbia (B.C.) is to restore salmon stocks to historic levels. Major emphasis has been placed on the use of hatcheries to rear and then release smolts in order to enhance stocks. Optimization of fish culture techniques so that juveniles have the best chance of hatchery and ocean survival will yield considerable economic benefits to the commercial and recreational fishery.

In B.C., no thorough study has been undertaken to compare the returns of wild versus cultured salmon. Preliminary information, however, indicates that the returns of wild and cultured coho salmon are about equivalent, whereas those of cultured chinook are considerably less than their wild counterparts (T. Perry, pers. comm.).

In addition to the difference in return rates, other differences between wild and cultured salmonids exist (Blaxter 1975; Buckley and Groves 1979). Biochemically, hatchery fish have lower protein and ash content and higher lipid content than wild fish (Phillips et al. 1957; Wood et al. 1969; Ludwig 1980). This results largely from differences in the composition of artificial and natural foods, ration level, and the level of physical activity. The variation in level and composition of body lipid between cultured and wild salmonids is likely to be of most significance for ocean survival because protein and ash content do not differ on a moisture-lipid free basis (Ludwig 1980).

Two viewpoints exist concerning the influence of body lipid content on smolt survival. The first suggests that those cultured fish which can most rapidly transform their body composition to the wild profile have the best chance for survival (Wood et al. 1960). By contrast, the second considers that the high lipid content or enhanced energy reserves in cultured fish may be advantageous during transition from artificial to natural food (Burrows 1969; Reinitz and Hitzel 1980).

With the two opposing viewpoints in mind, the long-term goal of this work is to assess whether body composition, and specifically body lipid content, can influence ocean survival of chinook smolts released from Robertson Creek Hatchery near Port Alberni, B.C. Fish of different body compositions were produced by varying the protein to lipid ratio in an experimental dry diet. Further, some groups received OMP the standard diet used for culturing chinook in B.C. hatcheries. All fish were differentially tagged and then simultaneously released to the ocean at the end of the freshwater rearing stage. Final assessment of the possible involvement of body composition in chinook marine survival will be possible only after 1985 owing to the long marine residence of this species.

The use of two different types of diets also permitted as a second goal, a comparison of chinook performance, that is growth, protein utilization, osmoregulatory ability and state of health when fed either dry (< 10% moisture) or semi-moist ( $\sim$  30% moisture) foods of the same lipid content.

#### MATERIALS AND METHODS

#### A. CULTURE CONDITIONS:

On April 2, 1980, chinook salmon fry at Robertson Creek Hatchery were taken directly from Heath trays and distributed 5 kilograms at a time into each of 12-three meter diameter fibreglas tanks. Each tank finally contained approximately 8,000 fish whose initial mean wet weight ranged from 0.60-0.64 g. The water supply from Great Central Lake to each tank ranged in temperature from  $8.0-15.2\,^{\circ}\text{C}$  (Fig. 1) and was adjusted from 45-76 1/min depending upon fish biomass. A natural photoperiod was in effect throughout the study.

#### B. DIET ADMINISTRATION AND PREPARATION:

Three groups were randomly assigned to each of four dietary treatments. These were OMP and the West Vancouver Laboratory dry diet No. 30 (WV30) (Table 1) supplemented with either 0, 5.6, or 9.3% herring oil. Total dietary lipid and protein contents expressed as a percentage of dry matter ranged between 8 to 17 and 48 to 55, respectively. Protein to lipid ratios in the diets varied between 2.8 and 6.8 (Table 2).

The diets were commercially prepared by Moore-Clarke Co., La Conner, Washington, U.S.A. However, the WV30 mash was pelleted, crumbled, screened and sprayed with marine oil at the West Vancouver Laboratory using methods previously described (Higgs et al. 1979). All diets were kept frozen until fed. Fish in each tank were fed to satiation 5 to 7 times a day and food particle size was adjusted according to fish size (Fowler and Burrows 1971). Records of food intake and mortalities were maintained daily.

#### C. GROWTH MEASUREMENT AND TAGGING:

The initial weight of fish in each tank was determined by lot weighing two groups of fifty fish into a beaker of water. Thereafter, sixty fish were randomly removed from each tank at 21-day intervals for measurement of individual weights and fork lengths (Higgs et al. 1982).

To evaluate ocean survival, all fish in each treatment group, except those too small for tagging (1.7-2.5% of the population), were marked by removing the adipose fin and differentially tagged by inserting a coded wire tag into the snout. All fish were then simultaneously released into the ocean on June 3, 1980.

### D. ANALYTICAL TECHNIQUES:

Fish from each treatment group were sampled for the following analyses using either methods referred to below or those previously described by Higgs et al. (1979, 1982).

- 1. Proximate composition Initially, 25 fish from each dietary replicate were sampled on April 23, 1980. Final samples of 30 fish per dietary replicate were taken on May 29, 1980 and subsequently subdivided into six groups of 5 fish each for analysis. All samples were stored in heat-sealed pouches at -40°C until analyses were performed.
- 2. Fatty Acid analysis At the end of the study, 3 fish were sampled from each of two dietary replicates. Lipid was extracted and separated by the methods described by Clarke et al. (1982).
- 3. Fish health and histological examination Before release, 20 fish per dietary treatment were randomly collected and examined externally and internally for state of health. Additionally, 10 fish per dietary treatment were examined for any abnormalities in the histological structure of selected tissues and organs.
- 4. Data Analysis Data were subjected to various types of Analyses of Variance (ANOVA, see individual tables) and when appropriate to Duncan's Multiple Range test or Scheffé's test (growth rates only) with P = 0.05 to detect significant differences between treatment means. The standard error of the mean was derived by using the appropriate error mean square from the ANOVA as the estimate of sample variance.

## RESULTS AND DISCUSSION

I. INFLUENCE OF DIETARY LIPID LEVEL OR PROTEIN TO LIPID RATIO ON GROWTH, FOOD, PROTEIN AND ENERGY UTILIZATION, STATE OF HEALTH AND OSMOREGULATORY PERFORMANCE.

Our original intent was to produce fish of equal size at release as this factor as well as time of release are known to influence return rate of coho (Bilton 1982) and chinook (Fowler 1980) salmon. However, those groups fed OMP or WV30 with 17% lipid were significantly larger at the time of release than those fed WV30 with 8 or 13% lipid. The faster rate of growth of groups fed the two former diets stemmed from improved food and protein utilization and in one instance from enhanced appetite and food conversion (Tables 3 and 4). The improved food conversion of fish fed OMP or WV30 with 17% lipid likely reflected the increased metabolizable energy contents of these diets relative to those of diets with reduced lipid content (Table 2).

Moreover, enhanced protein utilization in groups fed high lipid content diets or diets with reduced protein to lipid or calorie ratios probably was a consequence of diminished use of protein to satisfy a portion of daily caloric demands. Therefore, the provision of optimum levels of protein and energy is necessary to achieve maximum growth. Tables 3 and 4 show that diets with protein to lipid ratios of 2.78 to 2.99 promoted better growth and food, protein and energy conversion in chinook than diets with protein to lipid ratios of 4.71 to 6.70.

Within the groups fed the dry diets, it was noted that appetite bore an inverse relationship to dietary metabolizable energy content (Tables 2 and 4). Those fish fed WV30 with 8% lipid had significantly greater food consumption than groups fed WV30 with high lipid content. This supports the concept that fish eat to satisfy caloric demands (Lee and Putnam 1973).

Tables 3 and 4, illustrate that the difference in moisture content between WV30 with 17% lipid (8.7% moisture) versus OMP (28.6% moisture) did not significantly influence chinook performance while in freshwater. Thus equal growth in chinook smolts can be achieved by feeding a dry or moist feed provided that the available dietary nutrient levels are similar.

Condition factors of the fish did not bear any consistent relationship to dietary lipid content or to protein to lipid ratio (Table 3). Groups fed WV30 with 17% lipid had highest growth in weight relative to length whereas those fed WV30 with 13% lipid or OMP had the lowest.

Overall mortalities were 3.5% of the initial number of fish in each dietary treatment or less. Moreover, no incidence of infectious disease or evidence of pathology with regard to the histological structure of the thyroid liver, kidney, gill and alimentary tract was found in any group. Although the mean hematocrits of the groups fed WV30, irrespective of lipid content, were significantly higher than those of fish fed OMP, this was probably of no biological consequence since they exceeded 40% in all groups.

At the end of the study, representative numbers of fish from each treatment group were subjected to a seawater challenge test. Mean plasma sodium titres ranged from 190.4 to 201.7 meq/l. Because no significant differences due to dietary treatment were found, we conclude that all groups had similar osmoregulatory abilities before seawater entry.

# II. INFLUENCE OF DIETARY LIPID CONTENT ON BODY PROXIMATE COMPOSITION AND LIPID COMPOSITION

Dietary lipid content had the greatest influence on body composition which supports the findings of Buckley and Groves (1979). Each increase in dietary lipid level resulted in a significant elevation in the amount of body lipid and a concomitant significant decrease in the amount of body protein. Body moisture and ash contents were noted to be inversely related to dietary lipid content (Table 5). Moreover, the values for percent body protein and ash did not differ when they were expressed on a moisture-lipid-free basis. Histological findings confirmed that dietary lipid levels influenced body

lipid content. Hepatic and omentum lipid deposition increased in fish fed diets with elevated lipid levels. Dietary moisture, on the other hand, exerted no influence on carcass proximate composition, when dietary lipid content was the same.

The level of lipid in the diet influenced not only body lipid deposition, but also the fatty acid profile of both body and dietary lipids (Tables 6 and 9). Fatty acids of the linolenic series (ω3 fatty acids) increased whereas those of the linoleic (ω6 fatty acids) and oleic (ω9 fatty acids) series decreased with each stepwise increment in the lipid content of WV30 (Tables 6 and 9). Not all of the individual fatty acids of a particular series in the diets showed similar trends. Specifically, levels of linolenic acid (18:3ω3) decreased, and percentages of 18:4ω3, 20:1ω9, 20:4ω6 and 22:1ω9 increased or showed no clear trends. Percentages of individual fatty acids in OMP were generally slightly above or between levels found in WV30 with low and high lipid content. The notable exceptions were 20:1ω9 and 22:1ω9 which were present in much higher percentages in OMP than in WV30 regardless of lipid level.

Dietary fatty acid composition had more effect on the whole body neutral lipid composition than it did on phospholipid and total body lipid composition (Tables 7, 8 and 9, Fig. 2). Generally, percentages of individual fatty acids in neutral lipid showed similar trends to those described above for the diets. In phospholipid, fatty acid percentages were similar in all treatment groups except for  $18:1\omega 9$ ,  $18:2\omega 6$  and  $20:4\omega 6$  which were inversely related to the lipid content of WV30. These trends in fatty acids resulted in similar  $\omega 6/\omega 3$  fatty acid ratios in phospholipid but in neutral lipid  $\omega 6/\omega 3$  ratios ranged from 0.31 (WV30 with 17% lipid) to 1.01 (WV30 with 8% lipid). Other studies have shown that the composition of the body lipid is directly related to the character of the dietary lipid (Buckley and Groves 1979; Mugrditchian et al. 1981; Reinitz and Yu 1981).

Both the quantity of lipid reserves and the fatty acid composition of fish body and membrane lipids are thought to influence ocean survival. Adaptation to increased salinity or to colder temperatures increases requirements for fatty acids of the  $\omega 3$  series (Castell 1979). A high degree of unsaturation in membrane phospholipids, especially with  $\omega 3$  fatty acids may facilitate successful seawater transfer (Cowey and Sargent 1977). Whether the differences in the fatty acid composition of total body neutral lipid observed between groups exerted any influence on ocean survival of chinook smolts in this study remains an area for future investigation.

#### III. INFLUENCE OF DIETARY TREATMENT ON MARINE SURVIVAL

It is not possible at this time to draw any definite conclusions regarding the influence of diet treatment on chinook marine survival. Percent returns of two and three-year-old chinook to the hatchery were low in all groups (Table 10) and information on returns of chinook to the commercial and recreational fisheries is presently in preliminary form.

Many factors are believed to influence the ocean survival of

released juvenile Pacific salmon. Hopefully, it will be possible to clearly differentiate the effects of body composition from those of size at time of seawater entry, stamina or osmoregulatory ability. It is conceivable that the absence or presence of an estuary and the paucity or abundance of natural food organisms will have considerable influence on the preferred body composition for maximum survival in the wild. Indeed, both viewpoints previously expressed regarding the ideal body composition of smolts for ocean survival could prove to be correct depending upon the circumstances.

Additionally, the presence of toxic or anti-nutritional factors in the diet may have an indirect influence on survival. There has been some evidence that dietary anti-oxidants used to stabilize lipid sources can alter either hepatic carcinogen activation or detoxification mechanisms (Eisele et al. 1982). The biological consequences of this are unknown but as lipids were incorporated proportionately into the diets, the antioxidant effects would likely be most severe in fish fed high lipid content diets. Future work should address this possibility.

#### ACKNOWLEDGMENTS

We would sincerely like to thank Gary Hoskins and staff, and Craig Clarke and John Blackburn of the Pacific Biological Station for conducting fish health examinations and seawater challenge tests, Jim Bjerring of the University of British Columbia Computing Centre for conducting statistical analyses of the data, John Davidson of the Environmental Protection Service - Department of Fisheries and Oceans Laboratory Services for performing mineral analyses, A. J. Castledine for assistance in fatty acid analyses, and Morva Young for typing the manuscript.

#### REFERENCES

- Bilton, H. T., D. F. Alderdice, and J. T. Schnute. 1982. Influence of time and size at release of juvenile coho salmon (Oncorhynchus kisutch) on returns at maturity. Can. J. Fish. Aquat. Sci. 39: 426-447.
- Blaxter, J. H. S. 1975. Reared and wild fish--how do they compare? 10th European Symposium on Marine Biology, Ostend, Belgium, Sept. 17-23, 1975. Vol. 1: 11-26.
- Brett, J. R. and T. D. D. Groves. 1979. Physiological energetics. In: W. S. Hoar, D. J. Randall, and J. R. Brett (eds), Fish Physiology, Vol. 8, Academic Press, New York, San Francisco and London, pp. 279-352.

- Buckley, J. T. and T. D. D. Groves. 1979. Influence of feed on the body composition of finfish. In: J. E. Halver and K. Tiews (eds), Finfish nutrition and fishfeed technology. Vol. 2, Heenemann Verlagsgesellschaft mbH., Berlin.
- Burrows, Roger E. 1969. The influence of fingerling quality on adult salmon survivals. Trans. Amer. Fish. Soc. 98: No. 4, 777-784.
- Castell, J. D. 1979. Review of lipid requirements of finfish. In: J. E. Halver and K. Tiews (eds), Finfish nutrition and fishfeed technology. Vol. 1, Heenemann Verlagsgesellschaft mbH., Berlin, pp. 59-84.
- Castledine, A. J. and J. T. Buckley. 1980. Distribution and mobility of ω3 fatty acids in rainbow trout fed varying levels and types of dietary lipid. J. Nutr. 110: 675-685.
- Clarke, W. C., D. A. Higgs, J. R. Markert, J. E. Shelbourn, and A. J. Castledine. 1982. Effect of varying dietary protein: lipid ratios on growth and body composition of coho salmon fry (Oncorhynchus kisutch) reared at different temperatures. Can. Data Rep. Fish. Aquat. Sci. No. 373: 18 p.
- Cowey, C. B. and J. R. Sargent. 1977. Lipid nutrition in fish. Comp. Biochem. Physiol. 57B: 296-273.
- Eisele, T. A., J. E. Nixon, N. E. Pawlowski, and R. O. Sinnhuber. 1982. Comparative effects of dietary antioxidants on the hepatic mixed function oxidase system of rainbow trout. Federation Proc. 41: 358.
- Fowler, L. G. and R. E. Burrows. 1971. The Abernathy salmon diet. Prog. Fish-Cult. 33: 67-75.
- Fowler, L. G. and J. L. Banks. 1980. Survival rates of three sizes of hatchery reared fall chinook salmon. Technology Transfer Series No. 80-1 April, 1980.
- Fowler, L. G., R. E. Burrows, B. D. Combs, and J. L. Banks. 1980. The effect of size and time of release of fall chinook salmon fingerlings on adult survival. Abernathy Salmon Cultural Development Center, Technology Transfer Series No. 80-3, pp. 13.
- Higgs, D. A., J. R. Markert, D. W. MacQuarrie, J. R. McBride, B. S. Dosanjh, C Nichols, and G. Hoskins. 1979. Development of practical diets for coho salmon, (Oncorhynchus kisutch), using poultry by-product meal, feather meal, soybean meal and rapeseed meal as major protein sources. In: J. E. Halver and K. Tiews (eds), Finfish nutrition and fishfeed technology. Vol. 2, Heenemann Verlagsgesellschaft mbH., Berlin, pp.191-218.
- Higgs, D. A., J. R. McBride, J. R. Markert, B. S. Dosanjh, M. D. Plotnikoff, and W. C. Clarke. 1982. Evaluation of Tower and Candle rapeseed (canola meal and Bronowski rapeseed protein concentrate as protein supplements in practical dry diets for juvenile chinook salmon (Oncorhynchus tshawytscha). Aquaculture 29: 1-31.

- Lee, D. J. and G. B. Putnam. 1973. The response of rainbow trout to varying protein/energy ratios in a test diet. J. Nutr. 103: 916-922.
- Ludwig, B. W. 1980. A morphological and biochemical comparison of artificially and naturally-reared salmonids. M.Sc. Thesis, Univ. British Columbia, Vancouver, B. C., pp. 130 mimeographed.
- Mugrditchian, Doris D., Ronald W. Hardy, and Wayne T. Iwaoka. 1981. Linseed oil and animal fat as alternative lipid sources in dry diets for chinook salmon (Oncorhynchus tshawytscha). Aquaculture 25: 161-172.
- Phillips, Jr., A. M., D. R. Brockway, F. E. Lovelace, and H. A. Podoliak. 1957 A chemical comparison of hatchery and wild brook trout. Prog. Fish-Cult. 19: 19-25.
- Reinitz, Gary L., L. E. Orme, C. A. Lemm, and F. N. Hitzel. 1978. Influence of varying lipid concentrations with two protein concentrations in diets for rainbow trout (Salmo gairdneri). Trans. Am. Fish Soc. 167(5): 751-754.
- Reinitz, Gary and Frances Hitzel. 1980. Formulation of practical diets for rainbow trout based on desired performance and body composition.

  Aquaculture 19: 243-252.
- Reinitz, Gary L. and T. C. Yu. 1981. Effects of dietary lipids on growth and fatty acid composition of rainbow trout (Salmo gairdneri). Aquaculture 22: 359-366.
- Wood, E. M., W. T. Yasutake, J. E. Halver, and A.N. Woodall. 1960. Chemical and histological studies of wild and hatchery salmon in fresh water. Trans. Amer. Fish. Soc. 89(3): 301-307.

TABLE 1. Composition of West Van30 diet fed to juvenile chinook salmon at Robertson Creek Hatchery.

Ingredients	g/kg dry diet
Herring meal (crude protein = 66.8%)	510.0
Canola meal (crude protein = 39.8%)	164.3
Blood flour (crude protein = 90.1%)	50.3
Shrimp meal (crude protein = 48.2%)	42.9
Wheat millrun	90.2
Vitamin supplement <sup>1</sup>	40.0
Mineral supplement <sup>2</sup>	20.0
Herring oil - antioxidant <sup>3</sup> , <sup>4</sup>	57.0
Permapell (lignin-sulphonate binder)	20.0
DL-methionine	5.3

 $<sup>^1</sup>$  The vitamin supplement supplied the following levels of nutrients/kg of dry diet: vitamin A acetate 7958 IU; vitamin D $_3$  526 IU; DL-alpha-tocopheryl acetate 842 IU; vitamin B $_{12}$  0.02 mg; riboflavin 63.2 mg; niacin 666 mg; D-calcium pantothenate 94.8 mg; menadione sodium bisulphite complex 84.6 mg; folic acid 10.5 mg; pyridoxine HCl 42.8 mg; thiamine mononitrate 48.2 mg; biotin 1.07 mg; inositol 475 mg; 50% choline chloride 3717 mg; ascorbic acid 1146 mg.

 $<sup>^2</sup>$  The trace mineral supplement provided the following levels of minerals (mg/kg dry diet): Mg (as MgSO4) 856; Mn (as MnSO4.H2O) 40.9; Zn (as ZnSO4.7H2O) 11.7; Co (as CoCl2.6H2O) 0.86; Cu (as CuSO4.5H2O) 8.1; I (as KI) 5.38; Na (as NaCl) 5000.

 $<sup>^3</sup>$  Stabilized with 0.3% BHA-BHT (1:1).

<sup>&</sup>lt;sup>4</sup> To increase lipid levels in test diets 0, 5.6, or 9.3% additional herring oil was sprayed onto pellets to produce WV30 with 8, 13 and 17% lipid.

TABLE 2. Proximate composition of WV30 with different levels of supplemental marine oil and of Oregon Moist Pellets (OMP).

		WV30		OMP
% lipid	8	13	17	17
Protein (% N x 6.25)	55.29	52.84	49.89	47.97
Crude lipid (Bligh-Dyer)	8.16	12.86	16.70	17.27
Crude fiber <sup>l</sup>	4.06	4.06	4.06	3.64
Nitrogen-free extract	18.72	17.29	17.12	18.78
Ash	13.77	12.95	12.23	12.34
Moisture (as fed)	10.67	9.50	8.72	28.62
Metabolizable energy (kcal/g) <sup>2</sup>	3.27	3.53	3.71	3.69
Digestible protein (mg/dig. kcal) <sup>3</sup>	152.7	134.7	121.0	117.0
Protein/lipid	6.78	4.71	2.99	2.78

 $<sup>^{\</sup>mathrm{1}}$  Crude fiber contents of the diets were estimated.

<sup>&</sup>lt;sup>2</sup> Brett and Groves (1979) coefficients used for estimating caloric value of the diets, i.e. 4.2 kcal/g crude protein, 8.0 kcal/g crude lipid and 1.6 kcal/g carbohydrate.

 $<sup>^{3}</sup>$  Calculated assuming protein 90% digestible.

mortalities (relative to initial number in each group) and hematocrits of juvenile chinook salmon fed WV30 with varying percentages of lipid or OMP. Values are means ± 2SE. TABLE 3 - Final dry weights, specific growth rates, condition factors on day 58, overall

		1.77.30		QMO	
		200		JUI	
% Lipid	∞	13	17	17	
	of the second of				
Final dry weight $(g)^1, 2$	0.94a	0.90a	1.05b	1.08 <sup>b</sup>	
	± 0.03	+ 0.03	+ 0.03	± 0.03	
Specific growth rate $(%)^3$	3.69a	3.76a	4.09b	4.19b	
	± 0.10	± 0.10	± 0.10	± 0.10	
x 1000 - fork length	o 983p	6.64a	6.92 <sup>c</sup>	6.69a,b	
i	± 0.07	± 0.07		± 0.07	
	2.6	2.8		3.5	
Hematocrit $(%)^1$	45.5b	46.2 <sup>b</sup>		40.5a	
	₹ 0.98	± 0.98		₹ 0.98	

 $^{1}$  Two-way nested ANOVA indicated P < 0.001 for weight and P < 0.05 for condition factor and hematocrit.

<sup>2</sup> Final mean wet weights of the groups were 4.2, 3.9, 4.3 and 4.5 g, respectively.

Groups  $^3$  Analysis of covariance with day and day squared as covariates indicated P < 0.001. with the same superscript form a homogeneous subset.

gross energy utilization of juvenile chinook salmon fed WV30 with varying percentages of lipid or OMP. Values given are means ± 2SE.1 TABLE 4 - Appetite, food conversion efficiency, protein efficiency ratio, protein utilization,

		WV30		OMP
% lipid	ω	13	17	17
Appetite				
(daily dry food intake (mg)/	144.0b	132.5ª	131,7ª	143.5 <sup>b</sup>
dry body wt (g))	± 3,45	± 3,45	± 3,45	± 3.45
Food conversion efficiency <sup>1</sup>	,	,		
(dry wt gain (g) x 100 ÷	28.2ª,b	26.7ª	31,6 <sup>c</sup>	29.6b,c
dry food intake (g))	± 3.24	± 3.24	± 3,24	± 3.24
Protein efficiency ratiol				
	0.51a	0.51a	$0.63^{b}$	0.62b
wt of protein consumed (g))	<b>0.0</b> ∓	<b>0.00</b> ∓	90°0 ∓	+ 0.06
Protein utilization (%)			,	
(wt of fish protein retained (g)	36.7ª	34.9ª	41.6 <sup>b</sup>	41.5 <sup>b</sup>
x 100 ÷ wt of protein consumed (g))	± 4.32	± 4,32	± 4.32	± 4.32
Gross energy utilization (%)				
(gross energy gain x 100 ÷	$29.0^{a}$	26.7a	$31.5^{c}$	29.2b,c
gross energy intake)	± 3.26	± 3.26	± 3.26	± 3.26
			recommendately contractions of the second	

 $^{\rm l}$  ANOVA (repeated measures design) indicated P < 0.05 for appetite, food conversion efficiency, and gross energy utilization, P < 0.001 for protein efficiency ratio and P < 0.01 for apparent net Groups with the same superscript form a homogeneous subset. protein utilization.

TABLE 5 - Final proximate composition of juvenile chinook salmon fed WV30 with varying percentages of lipid or OMP. Values given are for samples collected prior to release and are means  $\pm$  2 SE.  $^1$ ,  $^2$ 

		WV30		OMP
% Lipid	∞	13	17	17
Moisture (%)	78.34b	77.64ª,b	76.96a	77.38a
	± 0.20	± 0.20	± 0.20	± 0.20
Dry-weight basis				
Crude protein (%)	72.09c	68.76 <sup>b</sup>	65.01a	65.50a
	± 0.81	± 0.81	± 0.81	± 0.81
Crude lipid (%)	12.99a	16.17 <sup>b</sup>	20.16 <sup>c</sup>	19.72 <sup>c</sup>
	± 0.59	± 0.59	± 0.59	± 0.59
Ash (%)	9.72 <sup>b</sup>	9.59 <sup>b</sup>	9.31a,b	8.69
	+ 0.44	77.0 ₹	47.0 ∓	77°0 ∓
Moisture-free-lipid-free basis				
Crude protein (%)	82.87ª	82.02ª	81.45ª	81.58ª
	± 1.16	± 1,16	± 1.16	± 1.16
Crude ash (%)	11.16a	11.44a	11.65a	10.84a
	± 0.52	± 0.52	± 0.52	± 0.52

P < 0.001 for lipid and protein and P > 0.05 for protein and ash on a moisture-lipid-free basis.  $^{
m l}$  Analysis of covariance with day as the covariate indicated P < 0.05 for moisture and ash,

<sup>2</sup> Percentages of crude protein (P), crude lipid (L), and ash (A) on a dry-weight basis and of moisture (M) in fish sampled from each treatment group on April 23, 1980 were:

(L), 9.46 (A), 80.05 (M) (L), 9.97 (A), 79.96 (M) (L), 10.69 (A), 80.23 (M) (L), 8.75 (A), 80.37 (M) WV30 with 8% lipid - 71.23 (P), 14.21 (L), WV30 with 13% lipid - 69.34 (P), 15.54 (L), WV30 with 17% lipid - 67.48 (P), 18.0 (L), OMP 70.2 (P), 15.07 (L),

TABLE 6 - Fatty acid composition of dietary lipids.  $^{\mathrm{l}}$ 

		wv30		OMP
% lipid	8	13	17	17
Fatty acid				
14:0	3.9	3.6	4.2	5.1
16:0	17.0	15.7	15.2	15.9
16:1ω7	6.2	6.3	6.2	6.5
17:0	1.8	1.7	1.6	1.9
17:1	1.1	1.3	1.1	0.9
18:0	2.6	2.9	2.9	2.6
18:1ω9	26.7	23.1	21.8	22.7
18:2ω6	9.6	6.3	4.9	6.5
18:3ω3	2.5	2.1	1.7	1.0
18:4ω3	0.8	1.6	1.8	1.1
20:1ω9	6.4	7.2	7.3	9.8
20:4ω6	0.7	0.8	0.6	0.6
20:5ω3	5.4	7.2	7.9	5.4
22:1ω9	7.2	7.3	7.3	11.6
22 <b>:</b> 5ω3	0.5	1.2	1.5	0.7
22:6ω3	6.2	9.2	10.0	5.4
Others	2.1	3.3	4.6	2.9

 $<sup>^{\</sup>mathrm{l}}$  Fatty acids representing < 1% of total are not reported.

TABLE 7 - Percent fatty acid composition of total body neutral lipid in Robertson Creek chinook. Lipid was extracted from 3 pooled fish from each of 2 replicates.

			WV30					OMP
Fatty acid	% lipid 8		13	3	1	17		17
							1	
	1	2	1	2	I	2	7	2
14:0	3.3	2.9	3.3	3.6	3.8	3.4	5.6	5.5
16:0	15.6	13.8	15.1	14.8	13.5	14.0	14.2	14.5
$16:1\omega 7$	7.8	7.7	8.1	8.1	7.2	7.3	7.6	7.7
17:0	1.4	1.8	1.1	1.4	1.4	1.4	1.6	1.5
17:1	1.2	1.7	0.7	1.1	1.3	1,3	1.3	1.2
18:0	3.9	4.2	3.1	3.1	3.1	3.2	2.9	2.8
18:1w9	35.7	33.8	31,3	29.4	26.9	27.1	23.2	23.5
18:2w6	9.5	8.6	8.9	8.9	0.9	5.9	8.1	8.3
18:3w3	1.8	2.3	1.7	1.8	1.9	1.9	1.5	1.5
18:4m3	0.8	1.3	1.0	1.2	1.7	1.6	1.6	1.6
20:109	6.3	6.2	6.5	9.9	7.0	8.9	8.7	0.6
20:5  w3	1.4	1.9	4.0	4.0	4.8	4.7	3.1	3.0
22:109	6.4	4.5	5.4	5.4	5.5	5.6	8.7	8.9
22:5w3	0.5	0.7	1.3	1.3	1.4	1.4	6.0	0.9
22:6w3	7.7	4.0	7.8	8.6	9.6	6.7	5.2	5,3
Others	1.5	3.4	2.8	2.8	5.1	5.7	5.2	4.8

TABLE 8 - Percent fatty acid composition of total body phospholipid in Robertson Greek chinook. Lipid was extracted from 3 pooled fish from each of 2 replicates.

			WV30	0			0	OMP
Fatty acid	% lipid 8		13	3		17		17
	-	2	-	2	_	2	1	2
14:0	0.7	0.7	0.8	0.9	0.7	0.8	1,3	1.1
16:0	19.3	19.0	18.1	18.9	19.7	19.0	21.6	19.8
16:1w7	3.1	3.2	3.6	3.2	3,3	3.2	3,3	3.2
17:0	6.0	1.0	1.3	1.2	1.5	1,4	1.0	1.1
17:1	0.8	0.7	1.0	7.0	9.0	0.5	0.3	0.8
18:0	5,3	5.4	5.5	5,3	5.3	5.2	5.2	5,3
18:1w9	14.4	14.3	12.9	12.9	12.4	12.3	12.2	12.2
$18:2 \pm 0$	3,3	3.1	2.4	2.1	1.8	1.8	2.0	2.5
18:3w3	6.0	6.0	1.2	6.0	0.7	8.0	0.3	9.0
18:4m3	0.4	0.4	9.0	0.5	0.4	0.4	0.2	0.5
20:109	1.3	1.2	1.4	1.3	1.2	1.4	1.6	1.8
$20.4 \pm 0$	2.5	2.7	2.6	2.3	1.9	2.1	2.0	2.1
20:5m3	5.5	5.5	7,1	7.0	6.9	6.7	5.6	6.1
22:109	0.1	0.4	7.0	0.2	0.2	0.2	0.3	9.0
$22:5\omega 3$	1,3	1.4	1.9	1.7	1.6	1.7	1.7	1.7
22:6w3	38.3	37.9	37.3	39.5	40.4	40.4	40.1	39.3
Others	1.9	2.2	2.9	1.7	1.4	2.1	1.3	1.3
and the desired and desired and desired	Salar Control of the	Albacharder Grade Grade Grade Grade Grade	O-CO-CO-CO-CO-CO-CO-CO-CO-CO-CO-CO-CO-CO	Chapthadhe dhugardardardardarba	-Christian (heigh-Christian Christian	0-0-0-0-0-0-0-0-0-0-0-0-0-0-0-0-0-0-0-		

TABLE 9: Total percentages of w3, w6, and w9 fatty acids and w6/w3 fatty acid ratios for dietary lipid and phospholipid, neutral lipid and total body lipid.

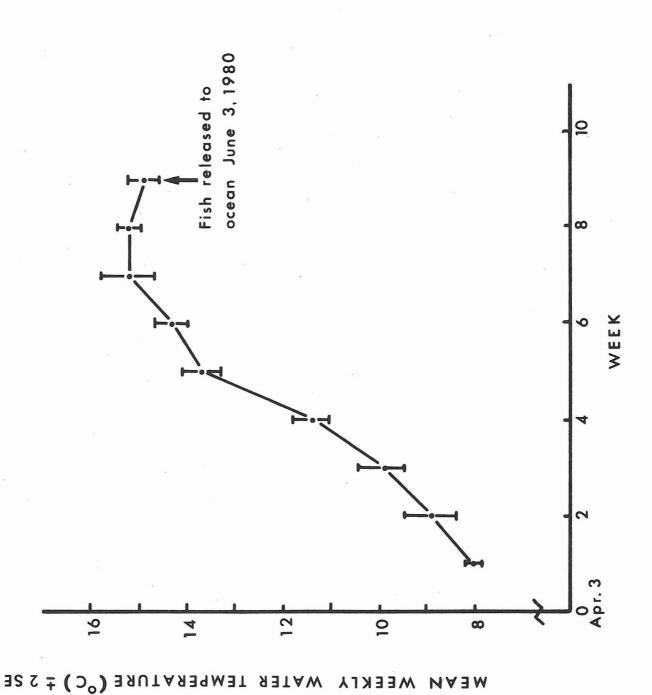
		Diet WV30	Dietary lipid 730	oid OMP		Pho WV30	Phospholipid 30	id OMP	10.0	Neu WV30	Neutral lipid WV30 CMP	pid OMP		Total body 1 WV30	body 1	lipid CMP
	% lipid 8 13	13	17	17	8	13	17	17	∞	13	17	17	∞	13	17	17
,						And the state of t			O Marie Library, printed and a				Action of the Control			
	•														10.7	
Total w3	15.4	21.3	22.9	13.6	46.3	48.9	50.0	48.1	9.6	16.4	19.3	12.3	55.9	65.3	69.3	<b>60.</b> 4
Total w6	9.6	6.3	4.9	6.5	5.8	4.7	3.8	4.3	9.7	8.9	0.9	8.2	15,5	11.5	8.6	12.5
Total w9	40.3	37.6	29,1	44.1	15.8	14.5	13.9	14.3	45.7	42.3	36.5	41.0	61.5	56.8	53,4	55.3
£m/9m		0.30	0.21	0.48	0.13	0.1	0.08	0.09	1.01	0,42	0,31	0.67	0.28	0.18	0,14	0.21

TABLE 10 - Number of two and three-year old chinook returning to Robertson Creek Hatchery (1979 brood).

Diet	% lipid	Tag code	Release wet wt (g)	Release date	No. of tagged fish released	% returns to hatchery 1981 - 1982
	00	17-02-15	4.2		20.449	700.
	13	18-02-07	3.9	June 3, 1980	20,789	. 043
	17	18-02-05	4.4	3,	20,733	.038
	17	18-02-06	4.5	3,	20,931	.052

TABLE 11 - Mortality and plasma sodium titres for representative samples of fish taken from each tank before ocean release. Fish were transferred to seawater (11°C, 30%) for 24 hr and then a blood sample was withdrawn from the caudal vessels of each fish for sodium determination. Values are means ± 2SE.

Diet	% lipid	Z	Mortality	Length (cm)	Weight (g)	Sodium mM/1
reditation to the special section of		Charles distribution distributi				
WV30	∞	18	0/22	$7.2 \pm 0.2$	$3.6 \pm 0.2$	$193.7 \pm 5.0$
WV30	13	18	0/22	$7.0 \pm 0.1$	$3.4 \pm 0.2$	$201.7 \pm 4.8$
WV30	17	18	0/21	$7.2 \pm 0.1$	$3.8 \pm 0.2$	$192.8 \pm 3.4$
OMP	17	18	0/21	$7.5 \pm 0.1$	$4.2 \pm 0.2$	190.4 ± 3.4
And any other Party and an	Contract of the Contract of th	-	Charles and the Charles of the Charles and the	elektristi om til en stande som en stande	had the statement of th	suggediskep daystartis tay tarminap attack design en



MEEKIL

Fig. 1 - Mean weekly temperature of water supplied to tanks at Robertson Creek Hatchery from time of ponding to release.

WV 30 (17 % LIPID)

LIPID)

WV 30 (13%

LIPID)

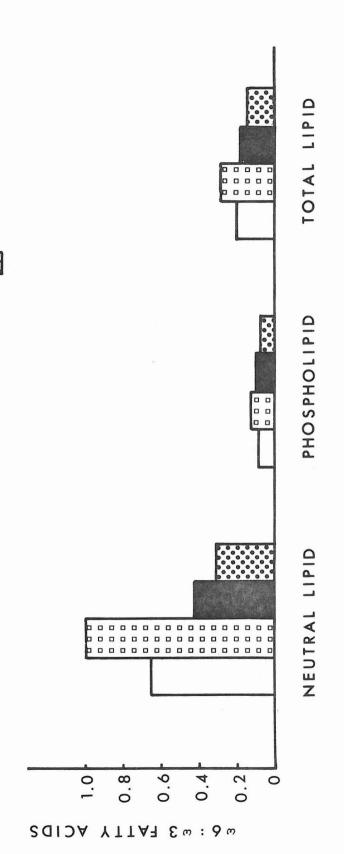
% 8 ) 0 8 %

(17% LIPID)

OMP

DIET





- Ratios of w6 to w3 fatty acids in whole body neutral lipid, phospholipid and total lipid for juvenile chinook salmon fed either WV30 with 8, 13 or 17% lipid or OMP. Fig. 2