Nutrient Requirements of Coldwater Fishes (1981)

Pages
71

Size
8.5 x 11

ISBN
0309031877

Subcommittee on Coldwater Fish Nutrition; Committee on Animal Nutrition; Board on Agriculture and Renewable Resources; Commission on Natural Resources; National Research Council

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OF
DOMESTIC
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NATIONAL ACADEMY PRESS

Washington, D.C. 1981

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This report has been reviewed by a group other than the authors according to procedures approved by a Report Review Committee consisting of members of the National Academy of Sciences, the National Academy of Engineering, and the Institute of Medicine.

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This study was supported by the Agricultural Research Service, U.S. Department of Agriculture, by the Bureau of Veterinary Medicine, Food and Drug Administration of the U.S. Department of Health and Human Services, and by Agriculture Canada.

Library of Congress Cataloging in Publication Data
Main entry under title:

Nutrient requirements of coldwater fishes.

(Nutrient requirements of domestic animals; no. 16)
Bibliography: p.
1. Fishes—Feeding and feeds 2. Salmon—Feeding and feeds. 3. Trout—Feeding and feeds. I. National Research Council (U.S.). Subcommittee on Coldwater Fish Nutrition. II. Series.
SF95.N32 no. 16 [SH156] 636.08'52s 81-18701
ISBN 0-309-03187-7 [639.3'11] AACR2

Available from

NATIONAL ACADEMY PRESS
2101 Constitution Avenue, N.W.
Washington, D.C. 20418

Printed in the United States of America

PREFACE

This report contains information on the nutritional requirements of coldwater (< 20°C) fishes. It was developed and expanded to update the 1973 publication *Nutrient Requirements of Trout, Salmon, and Catfish.* Catfish (warmwater fish) and coolwater species were excluded from this publication. Most of our present knowledge pertains to the euryhaline salmonid. Whenever appropriate data on other coldwater species were available, these have been included. When available, specific requirement recommendations, both qualitative and quantitative, have been made for growth and maintenance. Recommendations for nutrients required for reproduction were not made; obviously, more research is needed on this critical life stage.

Many of the valuable fisheries, both sport and commercial, have been overexploited. Harvesting of greater amounts of food from the seas and inland waters does not appear promising because the costs of equipment and effort are far greater than prospective fishery yields. Furthermore, the supply and demand situation for desirable fish is rapidly changing, and production by fish farming or aquaculture is more feasible. Fishes are among the most efficient of animals for converting feed energy into human food.

A combination of factors including nutrition, genetics, and disease control is principally responsible for the recent phenomenal growth in animal agriculture. Fish farming, like other forms of animal agriculture, is a product of man's ingenuity and an endeavor to which scientific advances and agricultural principles can be applied. It differs as much from the lake, sea, and river fisheries as animal husbandry differs from prehistoric hunting. However, fish culture (farming) has not kept pace with other animal agriculture. Principles regulating agricultural production, namely level of demand and technological development, should apply equally well to fish production. Demand for fishes is increasing and will stimulate technology.

Concentrated research on nutrition, feeds, and feeding has been limited to only a few coldwater species. Little definitive work on fundamental nutritional requirements of many freshwater or estuarine and marine species has been reported.

Two other publications in the "Nutrient Requirements of Domestic Animals" series deal with fishes. A complementary 1977 publication, *Nutrient Requirements of Warmwater Fishes*, covers catfish and other economically important species. A section devoted to nutrition and feeding of fishes used in medical and biological research appears in the 1978 publication entitled *Nutrient Requirements of Laboratory Animals.*

The subcommittee acknowledges the efforts of the International Feedstuffs Institute, Utah State University, in establishing and maintaining the data bank that supplied feed composition data for the subcommittee's consideration. The assistance of the Committee on Animal Nutrition's Subcommittee on Feed Composition is also acknowledged with appreciation.

The subcommittee is indebted to Philip Ross, Executive Secretary, and Selma P. Baron, Staff Officer, of the Board on Agriculture and Renewable Resources for their assistance in the production of this report; to the members of the Committee on Animal Nutrition; to Neville P. Clarke, reviewer for the Board on Agriculture and Renewable Resources, and C. P. Idyll, reviewer for the Commission on Natural Resources, for their comments and suggestions; and to Harry K. Dupree, George Post, John Spinelli, and John C. Wekell, who prepared critical reviews for the advice and guidance of the subcommittee.

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INTRODUCTION

The quantitative nutritional requirements of coldwater fishes are not all known. The subcommittee has reviewed and evaluated the available information and defined requirements where possible. When the information available was sparse or when published results did not agree, the subcommittee has estimated requirements and these estimates are so indicated. The requirements listed should be considered as minimum requirements and do not include a margin of safety. Recommendations are based upon the assumption that diets contain about 90 percent of dry matter.

Species, genetic strain, sex, and sexual maturity affect growth rate, feed conversion, and carcass composition and, therefore, the nutrient requirements. The required dietary level will also be influenced by:

(1) feed intake;
(2) energy density of the diet;
(3) level and interaction of nutrients in the diet;
(4) availability of nutrients to the animal;
(5) presence and level of feed additives;
(6) temperature, flow rate, and chemical composition of water;
(7) infectious disease, clinical or subclinical;
(8) presence of toxins, enzyme inhibitors, and microorganisms; and
(9) expected level of performance and carcass composition.

Ingredient quantity and composition of "open formula" feed formulations are available to the interested public. The most widely used coldwater fish diets are "closed formulas," the quantitative ingredient composition of which is proprietary information of the diet manufacturers. Least-cost linear programming, a recent innovation in fish feed formulation, is being used by most commercial fish feed manufacturers to reduce costs.

The Committee on Animal Nutrition's Subcommittee on Coldwater Fish Nutrition has revised the report with the practicing nutritionist, students of fish nutrition, and practical fish farmer in mind. Separate sections have been developed for each major nutrient class. In addition, sections have been developed on adventitious toxins, formulating diets and feeding practices, and feed processing and storage. Tables on composition of feedstuffs complete the report. Because of the wide variation in diet components, recommendations on open formula and semipurified diets are not presented.

NUTRIENT REQUIREMENTS

PROTEIN AND AMINO ACIDS

Proteins are complex, organic compounds composed of many amino acids linked together through peptide bonds and cross-linked between chains by sulfhydryl bonds, hydrogen bonds, and van der Waals forces. There is greater diversity of chemical composition in proteins than in any other group of biologically active compounds. Proteins in the various animal and plant cells confer on these tissues their biological specificity.

Proteins can be classified as *simple proteins*, when hydrolysis yields only the amino acids and occasional small carbohydrate compounds; *conjugated proteins* are simple proteins combined with some nonprotein material in the body; and *derived proteins* are those derived from simple or conjugated proteins by physical or chemical means (West and van Bruggen, 1963; Lehninger, 1975).

Ingested proteins are first split into smaller fragments by pepsin in the stomach, then by trypsin or chymotrypsin from the pancreas in the mid gut. These peptides are further reduced by carboxypeptidase or amino peptidase, which hydrolyze one amino acid at a time beginning at the free carboxyl end or at the free amino end of the polypeptide chain, respectively. The free amino acids released are absorbed through the gastrointestinal walls into the blood stream and are resynthesized into new tissue proteins or are catabolized for energy into other cellular metabolites (West and van Bruggen, 1963; Weissbach and Ochoa, 1967; Halver, 1972; Lehninger, 1975).

The nitrogen content of most proteins found in animal, nut, oilseed, and grain tissue is about 16 percent; therefore, protein content is commonly expressed as nitrogen content × 6.25. Each feedstuff has a characteristic nitrogen content determined by its amino acid profile, and may vary from 12 percent to as high as 20 percent nitrogen with 6.25 × nitrogen as an average estimate.

Amino Acids

Amino acids are the building blocks of proteins, with about 23 amino acids having been isolated from natural sources. These occur as L-stereoisomers in nature, but chemical synthesis results in isomeric mixtures of D- and L-forms, with the former being generally inactive. Ten of these are indispensable for fish, i.e., fish are incapable of synthesizing these particular amino acids and must therefore obtain them from the diet (Halver, 1957; Halver *et al.*, 1957; Weissbach and Ochoa, 1967; Aoe *et al.*, 1970; Halver, 1972; Mertz, 1972; National Research Council, 1973; Lucas-Lenard and Beres, 1974). The same 10 amino acids have been classified as indispensable for each finfish species studied (Cowey, 1979).

Salmon and trout fed diets devoid of any of the indispensable amino acids—arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, or valine—failed to grow (Figure 1) (Halver, 1957; Halver *et al.*, 1957; Mertz, 1972; National Research Council, 1973). Tryptophan deficiency induced scoliosis and lordosis in salmon and trout (Halver, 1972; National Research Council, 1973). Fish fed diets devoid of other L-amino acids grew as well as fish receiving all 18 amino acids tested (Figure 2). The nitrogen component in the test diets was made up of 18 L-amino acids in the pattern found in whole egg protein (Halver, 1957). All fish on test recovered rapidly when the missing amino acid was replaced in the diet. The slope of the growth curve of the recovery group was identical with that of fish receiving the complete amino acid test diet.

Dispensable amino acids tested were alanine, aspartic acid, cystine, glutamic acid, glycine, proline, serine, and tyrosine. These amino acids were found nonessential for the growth of salmon, trout, and other fishes (Halver, 1957; Halver *et al.*, 1957; Halver, 1972; National Research Council, 1973). When uniformly ^{14}C-labeled glucose was fed to several species of fishes, it was incorporated into these same dispensable amino acids (Cowey, 1979).

Quantitatve studies of the requirements of the 10 indispensable amino acids used a casein–gelatin mixture supplemented with crystalline L-amino acids. The test diet had an amino acid pattern of whole chicken egg protein. Similar experiments conducted with plaice, turbot, and red sea bream likewise demonstrated poor growth when an indispensable amino acid was absent from the diet (Aoe *et al.*, 1970; Nose *et al.*, 1974; National Research Council, 1973, 1977; Cowey, 1979).

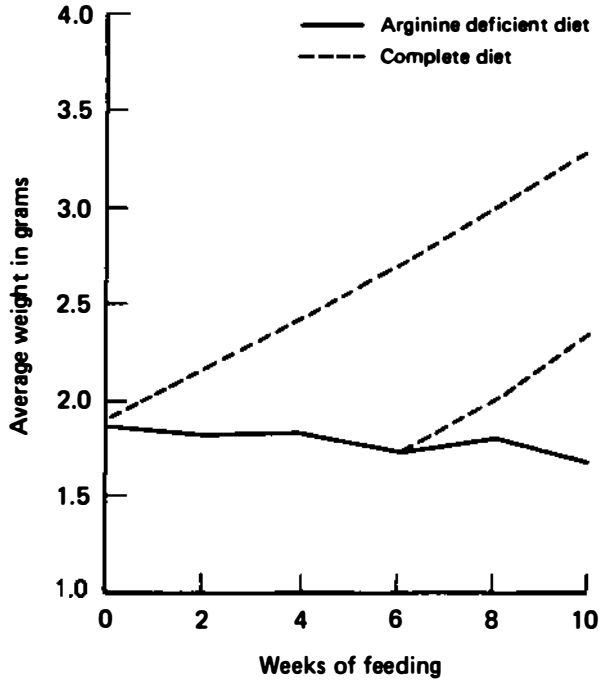

FIGURE 1 Growth of a typical indispensable amino acid-deficient group of chinook salmon. The deficient group was divided after 6 weeks on the deficient diet and the missing amino acid was replaced in one of the two sublots (Halver *et al.*, 1957).

Gross Protein Requirements

Gross protein requirements have been determined for salmon, trout, and flatfish (DeLong *et al.*, 1958; Cowey and Sargent, 1972; Cowey *et al.*, 1972; Cowey *et al.*, 1974). The simulated avian egg protein component of the test diets contained an excess of indispensable amino acids. These diets were made approximately isocaloric by adjusting the total of protein plus digestible carbohydrate components to a fixed amount as the protein diet treatments were varied over the ranges tested. Tests in feeding fry, fingerling, and yearling fishes show that gross protein requirements, as a percent of diet, are highest in initial feeding fry and decrease as size increases (Ogino and Saito, 1970; Nose and Arai, 1972; Satia, 1974; Garling and Wilson, 1976). For maximum growth, fry must ingest a diet in which nearly half of the digestible ingredients consist of protein containing minimal amounts of the 10 required amino acids. The requirement is decreased to about 40 percent of the diet for salmon and trout at 6–8 weeks, and to about 35 percent of the diet for yearling salmonids raised at standard environmental temperature (SET) (DeLong *et al.*, 1958) (Figures 3 and 4).

Some feeding trials with salmon have indicated direct relationships between changes in the protein requirements of young fishes and changes in water temperature. Chinook salmon, *Oncorhynchus tshawytscha*, in 7°C water require about 40 percent protein with the amino acid composition of whole egg for maximum growth; the same fish in 15°C water require about 50 percent protein (DeLong *et al.*, 1958). In contrast, rainbow trout fed practical diets showed no dif-

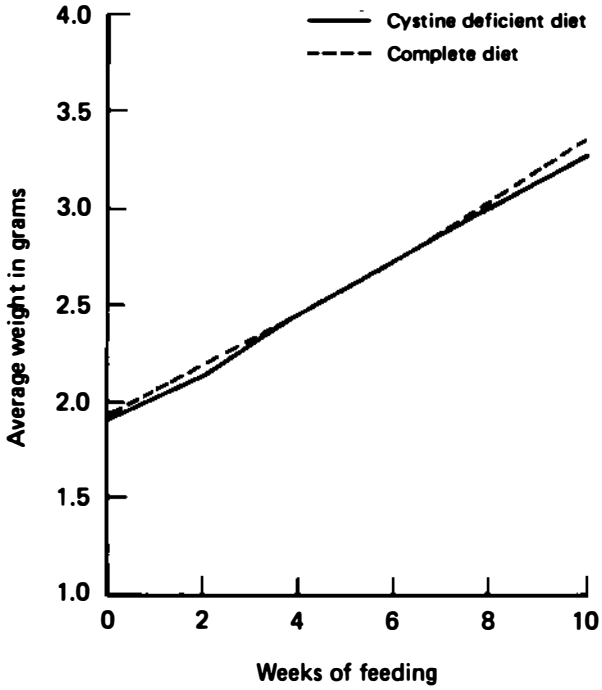

FIGURE 2 Comparison of growth rates of chinook salmon fed diets with and without a dispensable amino acid (Halver *et al.*, 1957).

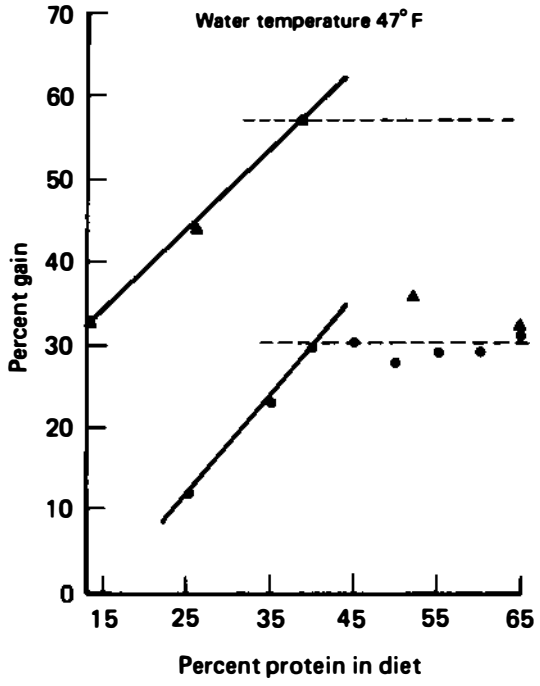

FIGURE 3 Protein requirement at 47°F (8°C). Top curve: initial individual average weight of chinook salmon, 1.5 g. Bottom curve: initial individual average weight of chinook salmon, 5.6 g (DeLong *et al.*, 1958).

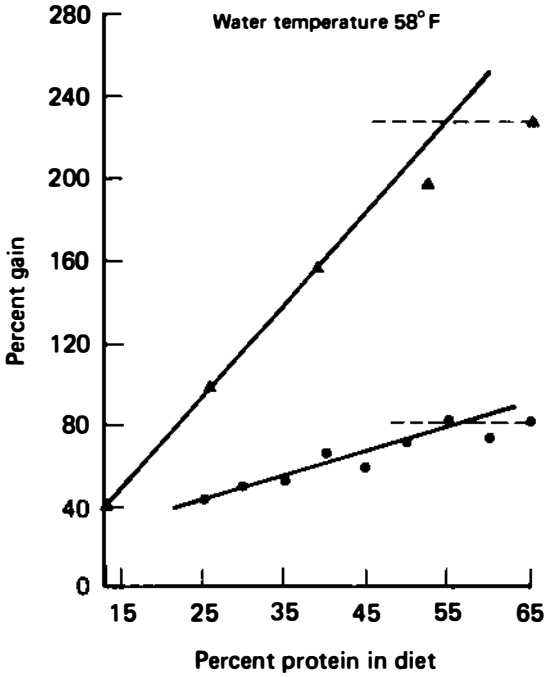

FIGURE 4 Protein requirement at 58°F (15°C). Top curve: initial individual average weight of chinook salmon, 2.6 g. Bottom curve: initial individual average weight of chinook salmon, 5.8 g (DeLong *et al.*, 1958).

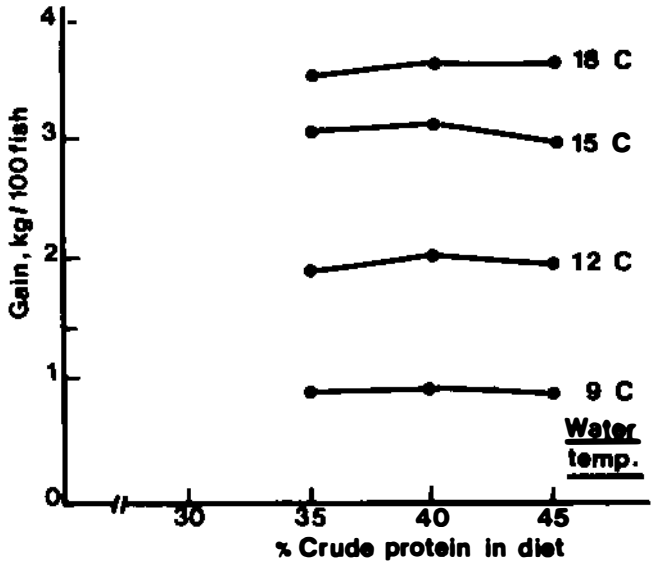

FIGURE 5 Effect of water temperature and protein level on growth of rainbow trout. Herring fish meal was reduced to lower the protein content while keeping metabolizable energy and fat contents constant. Average initial body weight was 2 g per fish, and the experimental period was 16 weeks (from Slinger *et al.*, 1977).

ferences in weight at protein levels of 35, 40, and 45 percent at temperatures of 9, 12, 15, and 18°C in one experiment (Slinger *et al.*, 1977), or in another experiment with temperatures of 9, 15, and 18°C (Cho and Slinger, 1978), even though there were distinct temperature effects (Figures 5 and 6). The greater absolute need of the trout for protein at the higher temperatures appeared to be satisfied through increased consumption of the lower-protein diets. Salmon, trout, and flatfish can use more protein than required for maximum growth because of efficiency in eliminating nitrogenous wastes in the form of soluble ammonia compounds through the gill tissue directly into the water environment (Cowey and Sargent, 1972; Cowey *et al.*, 1972, 1974). This system for eliminating nitrogen is more efficient than that used by birds and mammals to eliminate nitrogen. These animals require energy to synthesize urea, uric acid, or other nitrogen compounds, which are excreted through the kidney tissue and concentrated in urine (West and Van Bruggen, 1963; Cowey and Sargent, 1972; Lehninger, 1975). Excess protein in the diet of fishes can be used to spare digestible carbohydrate and fat as long as the protein requirement for maximum growth is met (Lee and Putnam, 1973; Rumsey, 1973; Sabaut and Luquet, 1973; Kaushik, 1977).

Zeitoun *et al.* (1973, 1974) showed that the protein requirement of rainbow trout, *Salmo gairdneri*, or coho salmon, *Oncorhynchus kisutch*, reared in water of 20‰ (parts per thousand) salinity is about the same as the requirement in fresh water. No data are available for the protein requirement of these species in full strength seawater (35‰).

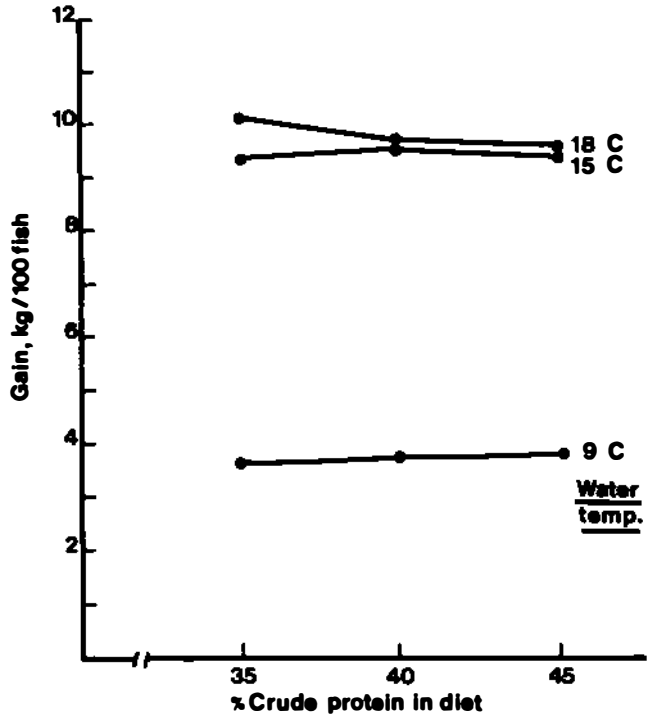

FIGURE 6 Effect of water temperature and protein level on growth of rainbow trout. As the level of protein was increased, both the levels of fish meal and soybean meal were increased to maintain the same ratios. The levels of crude fat and crude fiber were maintained constant at 11.5 and 4.0 percent, respectively. Average initial body weight was 3.45 g per fish, and the experimental period was 24 weeks (from Cho and Slinger, 1978).

Zeitoun *et al.* (1976) introduced studies on the concept of *economic protein requirement* of fishes. They suggested that the parabolic curve relating growth to dietary protein level does not reflect the practically insignificant differences in percentage gain below and beyond the maximum point. The minimum and maximum levels of intake occur over the range in which the animal is able to adapt to the level of nutrient supplied without substantial changes in metabolic processes. They determined economic protein requirement from the growth curve. A value of 440 g of protein per kilogram of diet as the economic requirement for rainbow trout was calculated.

Similar calculations can be made for the balanced gross protein requirement for other coldwater fishes and should result in more economical formulations from the feedstuffs available for coldwater fish diets.

Quantitative Amino Acid Requirements

Quantitative requirements of salmonids for the 10 indispensable amino acids were determined by feeding linear increments of 1 amino acid at a time in a test diet containing an amino acid profile identical with whole egg protein except for the amino acid tested (Mertz, 1972; National Research Council, 1973). Replicate groups of fish were fed the diet treatments until measurable differences appeared in the growth of test lots. A graph of growth response relative to dietary concentration of the amino acid under test indicated the requirement for maximum growth under those specific test conditions. Diets were designed to contain protein at or slightly below the optimum protein requirement for that species and test condition to assure maximum utilization of the limiting amino acid. A comparison of the requirements for the 10 indispensable amino acids of different species is shown in Table 2.

Test diets of proteins relatively deficient in a given essential amino acid are now being used. Kaushik (1977) used combinations of fish meal and zein in test diets to define the requirement of rainbow trout for arginine. Andrews and Page (1974) used diets containing different relative amounts of casein and gelatin and showed that an increase in the level of protein-bound arginine from 11 to 17 g/kg resulted in a significant increase in the growth of channel catfish. Rumsey and Ketola (1975) confirmed this principle in rainbow trout and Atlantic salmon (*Salmo salar*).

Kaushik (1977) determined the arginine requirement of rainbow trout from a conventional dose–response (growth) curve. He also measured the tissue (blood and muscle) levels of free arginine in groups of trout given increasing amounts of dietary arginine. After the dietary requirement of the trout for arginine had been met, any further increase in arginine intake led to an increase in the concentration of free arginine in blood and muscle. Good agreement was obtained between the two methods (Cowey, 1979).

The data in Table 2 suggest that differences exist among fish species in requirements for certain amino acids. Formulating the protein component of practical diets for those species whose amino acid requirements are not yet known is difficult. A possible solution is to use the highest level of each amino acid required by any of those species for which data are available (Mertz, 1972; Halver, 1976). Further quantitative data on the amino acid requirements of fishes are needed, especially for those fishes actually or potentially useful as farm animals.

Probably the most conservative technique is to estimate the quantitative amino acid requirements by calculating the portion of each indispensable amino acid present in the protein component of the diet. Examination of these estimates indicates a remarkable similarity of indispensable amino acid requirements between species (Table 2) (Mertz, 1972; Halver, 1976).

Availability of Amino Acids in Feed Proteins Cowey *et al.* (1974) compared the known amino acid requirements of chinook salmon with the amino acids provided by each of five proteins at a level of 500 g/kg diet. They showed that these proteins apparently supplied a considerable excess of essential amino acids. A similar comparison was made by Rumsey (1973). Besides a relative deficiency of methionine and phenylalanine, he noted that many plant proteins (seed meals, glutens) are deficient in lysine.

Phenylalanine can be spared by tyrosine (Mertz, 1972; National Research Council, 1973; Robinson *et al.*, 1980). It is not chemically modified nor rendered unavailable by the harsh conditions feedstuff proteins undergo during processing.

Lysine is a basic amino acid. It contains two amino groups. The ϵ-amino group must be free and reactive, or lysine will not be biologically available (Carpenter and Ellinger, 1955). During processing, the ϵ-amino group of lysine may react with nonprotein molecules present in the feedstuff to form additional compounds that render the lysine biologically unavailable (Cowey and Sargent, 1972). Methods for measuring "available" lysine in proteins correlate well with the biological value of these proteins in tests on birds and mammals and could prove useful in evaluating the lysine content of fish diets (Cowey, 1979).

Methionine can be spared by cystine. Measurement of methionine in feed proteins is difficult, as the amino acid is subject to oxidation during processing. Fishes may be able to reduce some of the oxidized methionine and thus recover some of the methionine oxidized (Cowey, 1979).

Methionine in proteins may be measured using iodoplatinate reagent before and after reduction with titanium trichloride, to give values for both methionine and the sulfoxide in the original protein (Njaa, 1977). Ellinger and Duncan (1976) have described a method to measure methionine by cyanogen bromide cleavage. Microbiological assay of methionine in feed proteins is a valuable tool, although there is the danger that oxides of methionine may differ in their activity in microorganisms and may misrepresent biological values of available methionine to fishes (Cowey *et al.*, 1974).

Supplementation of Proteins with Amino Acids Fishes appear to utilize free amino acids at various degrees of efficiency. Aoe *et al.* (1970) found that young carp (*Cyprinus carpio*) were unable to grow on diets in which the protein component (casein and gelatin) was replaced by a mixture of amino acids similar in amino acid profile. A trypsin hydrolysate of casein

was equally ineffective in supporting sustained growth. Nose (1971) showed that a diet containing free amino acids as the protein component, when adjusted to pH 6.5–6.7, allowed some growth in young carp. This growth was less than that on a comparable casein diet under the same experimental conditions.

Salmonids are able to utilize free amino acids for growth in an amino acid test diet (Halver, 1957). Nose (1971) showed that a zein–gelatin diet supplemented with lysine and tryptophan was superior to an unsupplemented zein–gelatin diet for rainbow trout.

Several investigators have demonstrated the potential of supplementing amino acid-deficient proteins with limiting amino acids in diets for salmonids. Rumsey and Ketola (1975) supplemented casein with six amino acids and obtained feed conversion ratios with Atlantic salmon similar to those obtained when isolated fish protein was fed as the dietary protein source. They showed that soybean meal supplemented with five or more amino acids was a superior protein source for rainbow trout to soybean meal alone. Additions of methionine or lysine did not improve the value of soybean meal (Rumsey and Ketola, 1975). The amino acid profile of the isolated fish protein used may approximate the amino acid requirement of rainbow trout (Cowey, 1979).

Dabrowska and Wojno (1977) showed that diets containing fish meal, meat and bone meal, yeast, and soybean meal were improved by supplementation with cystine (10 g/kg) and tryptophan (5 g/kg). Tiews *et al.* (1976) showed that fish meal can be replaced in diets for rainbow trout by a mixture of poultry by-product meal and feather meal if 17 g lysine HCl per kilogram, 4.8 g DL-methionine per kilogram, and 1.44 g DL-tryptophan per kilogram are added. More research is needed to define the utilization of free or bound amino acids as supplements for other species of coldwater fishes.

LIPIDS

Introduction

Coldwater fishes use lipids for energy, for cellular structure, and for maintenance of the integrity of biomembranes. Membrane fluidity is regulated in part by the fatty acid composition of the phospholipids that control such processes as cellular transport and the activities of membrane-associated enzymes. The degree of fatty acid unsaturation in fish tissue increases when environmental temperature is lowered, thereby maintaining membrane fluidity to allow normal cellular functions (Slinger *et al.*, 1977; Leger *et al.*, 1977b). Large amounts of polyunsaturated lipids are found in the natural diets of coldwater and marine fishes.

Lipid requirements for coldwater fishes are not adequately described. A wide range of opinions exists on both the qualitative and quantitative fat needs and types of fats that are most suitable (Phillips *et al.*, 1952; Phillips and Podoliak, 1957; Castell *et al.*, 1972a,b,c; Sinnhuber *et al.*, 1972; Kay *et al.*, 1976; Takeuchi *et al.*, 1978a,b,c, 1979; Cowey and Sargent, 1979; Yu and Sinnhuber, 1979). Several studies have shown that lipids can be used effectively as an energy source (Brown and Tappel, 1959; Nicolaides and Woodall, 1962; Cowey and Sargent, 1979; Takeuchi *et al.*, 1978a,b,c). Pelleted diets can be produced more economically by increasing the level of fat relative to an acceptable level of protein. Protein in excess of that required for growth and maintenance is used for energy and can be replaced with higher levels of fat in diets.

Dietary Lipid Levels

The optimal lipid intake for hatchery-reared coldwater fishes is essentially similar to that for wild fishes (Cowey and Sargent, 1979). Data from several experiments with several species suggest that not less than 10 percent and not more than 20 percent of lipid can be added to fish diets with excellent results (Lee and Putnam, 1973; Adron *et al.*, 1976; Yone *et al.*, 1971; Stickney and Andrews, 1972; Takeuchi *et al.*, 1978a). Higher levels of lipid may not cause metabolic defects but do alter carcass composition by deposition of excess lipid (Phillips *et al.*, 1952; Davis, 1953; Higashi *et al.*, 1964; Satia, 1974; Watanabe, 1977; Takeuchi *et al.*, 1978a). Watanabe (1977), Reinitz *et al.* (1978), and Takeuchi *et al.* (1978a) found that whole body lipids increased as dietary lipid levels increased and that as the percentage of fat in a diet increased the percentage of body protein decreased.

Protein and Lipid Content in Diets

Several studies have shown that providing adequate energy with dietary lipids can minimize the use of more costly protein as an energy source (Ringrose, 1971; Lee and Putnam, 1973; Watanabe, 1977; Reinitz *et al.*, 1978; Takeuchi *et al.*, 1978a). Takeuchi *et al.* (1978a) fed diets containing 5–20 percent lipid with protein levels ranging from 16 to 48 percent. Weight gain and feed conversion improved at each protein level as the lipid levels increased, reaching a maximum in the 35 percent protein diets with 15–20 percent lipid. The higher the lipid content, or the lower the protein content in the diets, the higher the proportion of protein retained. Thus protein was more efficiently utilized at increased levels of dietary lipids (Takeuchi *et al.*, 1978a). More than 10 percent of dietary lipid is necessary in order to increase the efficiency of protein utilization. Takeuchi *et al.* (1978a) suggest that the optimum ratio in diets of rainbow trout is 35 percent protein to 15–20 percent lipid.

Animal Fats and Marine Oils

Results from several experiments demonstrate that when animal fats, such as lard (Atherton, 1975; Yu *et al.*, 1977a,b), beef tallow (Holub *et al.*, 1976; Takeuchi *et al.*, 1978b), and yellow grease (Hardy *et al.*, 1980) are combined with marine fish or vegetable oils, the energy density of the diet can be increased without creating adverse effects on efficiency of diet utilization, fish growth, or survival. Since marine fish oils are generally rich in $\omega 3$ and low in $\omega 6$ fatty acids, these are most suitable for use as the lipid source in formulated coldwater fish diets.

Fatty Acids

There are four common families of polyunsaturated fatty acids in fishes. The exact structure of an unsaturated fatty acid is given by three numbers (1) the number of carbon atoms in the chain, (2) the number of double bonds, and (3) the omega (ω) number, which indicates the number of carbon atoms from the terminal methyl group to the carbon atom of the first double bond. The omega system designates those unsaturated fatty acids belonging to each series, such as the ω3 series or the ω6 series. The chemical name, family name, and chemical designation of four families are shown in Table 3.

The position of double bonds in the fatty acid carbon chain denotes both the physical characteristics and nutritional value. Coldwater fishes contain polyunsaturated fatty acids of the ω3 series. The major polyunsaturated fatty acids of fishes are 20:5ω3 and 22:6ω3, whereas 18:2ω6 and 20:4ω6 are the major polyunsaturates of terrestrial animals.

Essential Fatty Acids The fatty acid composition of coldwater fishes is determined by both the fatty acids supplied by the diet and fatty acids derived biosynthetically. The general conclusion from available data is that coldwater fishes have limited ability to synthesize fatty acids of the ω3 and ω6 series (Lee *et al.*, 1967; Castell *et al.*, 1972a,b,c; Yu and Sinnhuber, 1972; Sinnhuber *et al.*, 1972; Watanabe *et al.*, 1974). Adequate sources of ω3 and ω6 must accordingly be supplied in the diet.

Fish nutrition studies have established that ω3 fatty acids are essential for the maintenance of good health and promotion of rapid growth in rainbow trout (Lee *et al.*, 1967; Castell *et al.*, 1972a,c; Yu and Sinnhuber, 1972; Watanabe *et al.*, 1974). Similar work showed a necessity for dietary ω3 fatty acids in the red sea bream (Fujii *et al.*, 1976) and turbot (Leger *et al.*, 1977a).

Different levels of the triglycerides, trilinolenin (ω3), and trilinolein (ω6) in diets containing 10 percent lipid reduced growth in rainbow trout fed a diet (1) deficient in ω3 fatty acid, (2) high in ω6 and low in ω3 fatty acids, and (3) high in both ω3 and ω6 fatty acids (Yu and Sinnhuber, 1975).

An optimum level of unsaturated fatty acids is necessary for maximum growth and the requirement for ω3 fatty acids may be species specific (Yu and Sinnhuber, 1979). Rapid growth was observed in trout fed diets with high levels of ω3 fatty acids and a high ω3:ω6 ratio (Yu and Sinnhuber, 1975). The concentration of total ω3 and ω6 fatty acids in the fish body lipids reflected the dietary levels of 18:3ω3 and 18:2ω6.

Yu and Sinnhuber (1979) reported an optimum dietary level of ω3 fatty acid for coho salmon (*Oncorhynchus kisutch*) in the range of 1 to 2.5 percent. Concentrations of dietary ω6 fatty acids above 1 percent depressed growth. Extremely low and high levels of ω3 fatty acids will inhibit growth. The most desirable total polyunsaturated fatty acids (ω3 + ω6) in the salmon diets appeared to be approximately 2.5 percent.

Poor growth, low feed efficiency, high mortality, and swollen pale livers were reported in chum salmon (*Oncorhynchus keta*) fed a diet deficient in essential fatty acids. Weight gain and feed efficiency were maximized by supplementing the diet with both 1 percent 18:2ω6 and 1 percent 18:3ω3 or a mixture of 20:5ω3 and 22:6ω3 at the 0.5 and 1.0 percent level [i.e., ω3 highly unsaturated fatty acids (ω3HUFA)] (Takeuchi *et al.*, 1979). The requirement of chum salmon for linoleic and linolenic acids was approximately 1 percent, or 0.5–1.0 percent for ω3HUFA of the diet.

Some studies have described essential fatty acid deficiency signs in fishes (Nicolaides and Woodall, 1962; Castell *et al.*, 1972b,c; Sinnhuber *et al.*, 1972). These deficiency signs in trout are: (1) poor growth; (2) elevated tissue levels of ω9 fatty acids (particularly 20:3ω9); (3) necrosis of the caudal fin; (4) fatty, pale liver; (5) dermal depigmentation; (6) increased muscle water content; (7) syncope accentuated by stress; (8) increased mitochondrial swelling; (9) increased respiration rate of liver homogenates; (10) heart myopathy; and (11) lowered hemoglobin level (Sinnhuber, 1969).

The ω3 fatty acid series can prevent or remedy all the signs listed except dermal depigmentation. Addition of ω6 fatty acids (linoleic) to the diet of trout will aid in preventing some of the above signs, such as poor growth, fatty livers, and high tissue levels of ω9 fatty acids. However, linoleic acid has little effect on the other signs and appears to aggravate the syncope and heart myopathy.

Autoxidation and Antioxidants

Autoxidation of lipids is one of the most deleterious changes that may occur in fish diets. It is most likely to occur during manufacture, during storage of ingredients, and in finished feed. Oxidized fat in a diet may have adverse effects due to destruction of vitamin E and vitamin A and other essential nutrients, or the development of new compounds with toxic properties (Hung *et al.*, 1980). Free radicals resulting from peroxidation react with fat intermediates and form polymerized compounds that alter metabolism and nutritive values (Watanabe and Hashimoto, 1968). Chinook salmon fingerlings fed rancid fish meals developed signs that included dark coloration, anemia, lethargy, brown-yellow pigmented livers, abnormal kidneys, and gill clubbing (Smith, 1968).

Diets should be protected by adequate levels of antioxidants, such as ethoxyquin, butylated hydroxyanisole (BHA), and butylated hydroxytoluene (BHT) (Rumsey, 1980). A level of 100 mg/kg (0.01 percent) of these antioxidants is adequate but must not exceed 0.02 percent of a finished ration. Adequate amounts of vitamin E in the tissues must be present to retard *in vivo* fat peroxidation (Sinnhuber *et al.*, 1968; Watanabe *et al.*, 1967; Murai and Andrews, 1974).

CARBOHYDRATES

Carbohydrate provides the least expensive dietary energy source for coldwater fishes. Hexoses are of major nutritional significance and all fish studied have some ability to utilize carbohydrate as an energy source (Cowey and Sargent, 1972). Whereas carbohydrate provides the starting point for many biochemical syntheses, little may be found in natural diets. Little is present in the body, and fishes can grow on diets devoid of carbohydrate (Cowey and Sargent, 1979). However, low levels of dietary carbohydrate may lead to in-

sufficient body reserves of carbohydrates. Any energy deficit must be replaced by the more expensive protein and fat intermediates.

Carbohydrate Function

Carbohydrate may function as (1) an immediate energy source (Phillips *et al.*, 1966, 1967; Buhler and Halver, 1961), (2) a quick energy reserve stored as glycogen in the liver and muscle (Phillips *et al.*, 1948; Wendt, 1964), or (3) a long-term energy reserve when converted to fat in the body. Glycogen energy reserves prevent delayed fish mortality after stocking (Wendt, 1964). Carbohydrates also improve pellet binding and lower cost of diets.

Carbohydrate Digesting Enzymes

Carbohydrate-digesting enzymes in fishes are produced in the pyloric caeca, pancreas, and intestinal mucosa. Salmonids produce maltases, sucrases, lactases, amylases, and other enzymes for the digestion of carbohydrates (Phillips *et al.*, 1948). Enzymes are released into the pyloric caeca and anterior intestinal tract to hydrolyze disaccharides and starches into absorbable monosaccharides (Phillips *et al.*, 1948; Kitamikado and Tachino, 1961). Simple sugars follow the same Embden–Meyerhof and pentose phosphate intermediary pathways of carbohydrate metabolism as in other animals (McCartney, 1971; Cowey and Sargent, 1972).

Carbohydrate Utilization One of the first studies on utilization of carbohydrates by salmonids indicated a limited usefulness in brook trout diets (Phillips *et al.*, 1948). Higher carbohydrate levels resulted in low growth, high-glycogen livers, and increased mortality. Buhler and Halver (1961) found that chinook salmon tolerated relatively high levels of dietary carbohydrate without the development of abnormal signs. They suggested that the earlier results of Phillips *et al.* (1948) were caused by vitamin deficiency and dietary imbalance rather than by an inability to utilize carbohydrate. Others have fed salmonids diets containing higher levels of carbohydrates without adverse effects (DeLong *et al.*, 1958; McLaren *et al.*, 1947; Bergot, 1979a,b,c).

Buhler and Halver (1961) reported that salmon can tolerate either 48 percent dextrin or 20 percent sugars of low molecular weight in their diets. Bergot (1979a,b,c) found that the digestible carbohydrate can be raised from 15 to 30 percent without an adverse effect on growth. A 45 percent protein diet with 30 percent glucose provided the best weight gain, feed conversion, and protein efficiency, whereas a level of 30 percent glucose in a 30 percent protein diet had a negative effect on growth and feed efficiency (Bergot, 1979a,b). A similar trend was evident in a study on sucrose utilization at protein levels of 35 and 55 percent (Luquet *et al.*, 1975). Rainbow trout growth and feed conversion were greater on diets containing 17 and 25 percent of their metabolizable energy as carbohydrate than on a diet containing 38 percent (Edwards *et al.*, 1977). Studies with rainbow trout (Ringrose, 1971; Lee and Putnam, 1973), plaice (Cowey *et al.*, 1975), catfish (Page and Andrews, 1973; Garling and Wilson, 1977), and yellowtail (Takeda *et al.*, 1975) indicate that starch or dextrin up to a level of 25 percent of the diet was effective as an energy source. The 25 percent level was as effective as fat on an isocaloric basis (Cowey and Sargent, 1979).

Sparing Action of Carbohydrate The protein-sparing action of carbohydrate in fish diets has been reported for salmonids (Ringrose, 1971; Buhler and Halver, 1961; Lee and Putnam, 1973; Pieper and Pfeffer, 1979; Bergot, 1979a,b) and in other species, such as plaice (Cowey *et al.*, 1975), turbot (Adron *et al.*, 1976), catfish (Tiemeier *et al.*, 1965), and sea bass (Alliot *et al.*, 1979).

Buhler and Halver (1961) fed a series of diets to chinook salmon with a constant protein content (37.5 percent) but with increasing amounts of dextrin (0–48 percent) substituted for the inert α-cellulose. Weight gains increased with dietary dextrin concentration up to a level of 20 percent. Further increase in dextrin content had little effect on weight gain. Protein efficiency ratio (PER) was highest on diets containing 20–30 percent dextrin, indicating that with these diets less protein was used for energy purposes. Similar results in other studies have established the sparing action of carbohydrate on protein use for energy. It appears that sufficient carbohydrate in the diet obviates the need for gluconeogenesis.

Liver Size and Glycogen Levels An observed increase in liver size and glycogen level in proportion to the level of digestible carbohydrate in salmonid diets has been reported by several authors (Phillips *et al.*, 1948, 1966; Buhler and Halver, 1961; Lee and Putnam, 1973; Austreng *et al.*, 1977; Pieper and Pfeffer, 1979; Bergot, 1979a,b). Similar effects have been reported in red sea bream (Furuichi and Yone, 1971), plaice (Cowey *et al.*, 1975), and yellowtail (Shimeno *et al.*, 1979). DeLong *et al.* (1958) found no extensive liver damage in diets containing up to 61 percent dextrin. However, their results may have been explained by the presence of adequate vitamins and a relatively high water temperature at which the fish had an elevated energy requirement.

DIGESTIBILITY AND ABSORPTION

Digestibility of a feedstuff is normally defined in terms of energy availability. Terminology for digestibility and absorption can be found in National Research Council (1981).

Digestible energy (DE) = intake of energy in feed (IE) minus energy in feces (FE)

$$DE = IE - FE$$

Metabolizable energy (ME) = DE minus energy in the urine (UE) and gill wastes (ZE)

$$ME = DE - (UE + ZE)$$

Recovered energy (RE) = ME minus total heat production

$$RE = ME - HE$$

Both digestible energy and metabolizable energy values are used to express feed values and energy requirements. Metabolizable energy is preferable to digestible energy, because, in fishes, it accounts for energy losses from the gills and urine. Metabolizable energy is used for stating the energy requirements of several, if not most, animal species, and is currently used by the National Research Council (1981).

Digestibility values have been determined for many feedstuffs for mammals and birds. With present knowledge, however, extrapolation of these values to fishes is tenuous. Fish diets should be based on data obtained with the same species of fish.

Direct and Indirect Methods

Methods for the determination of digestibility coefficients have typically involved either a direct or an indirect quantitative measurement of the amount of nutrient ingested and subsequently egested.

Direct Method The earliest methods were direct and involved laborious and time-consuming water filtration techniques that required collection, measurement, and analysis of all egesta and excretions (Migita *et al.*, 1937; Tunison *et al.*, 1942). A modern extension of the direct method has been developed by Smith (1971, 1976) and Smith *et al.* (1980). A modified metabolism chamber permits separate and quantitative collection of feces, urine, and gill excretions. The procedures followed are essentially the same as for digestion trials with other animals. Fishes are fed a measured amount and the excretions are collected, measured, and analyzed. A 3-day preliminary period and a 4-day collection are normally used.

This method has the advantage of permitting the determination of digestibility coefficients and metabolizable energy (ME), as well as carbon and nitrogen balances. However, metabolism chambers have been criticized because the fishes are confined, force fed, and may be under some stress that reduces their ability to digest and utilize feed.

Indirect Methods An indirect method successfully applied to terrestrial animals has been used with several species of fishes (Nose, 1960a,b; Hastings, 1969; Smith and Lovell, 1971, 1973; Windell, 1978b). It is based on the assumption that the amount (0.5–1.0 percent) of chromic oxide (Cr_2O_3) in the feed and feces remains constant over the experimental time period. Its advantage over the direct method is elimination of the need for quantitative collection of all wastes. Digestion coefficients can be calculated for crude protein, lipid, carbohydrates, energy, and total dry matter by performing appropriate chemical analyses for the nutrients in the feed consumed and in the feces. Data for ash have been reported, but are of little significance in evaluating digestibility (Maynard and Loosli, 1969).

Nutrient digestibility of dry matter, protein, lipid, and carbohydrate is estimated according to the following formula (see Table 4):

$$\text{Percent Nutrient Digestibility} = 100 - 100 \times \frac{(\%\,Cr_2O_3 \text{food})}{(\%\,Cr_2O_3 \text{feces})} \times \frac{(\%\text{ nutrient feces})}{(\%\text{ nutrient food})}.$$

The major disadvantages of this method are the variety of techniques used to collect fecal samples with inherent nutrient leaching errors and the inability to compute reliable metabolizable energy values. ME values from the collected data cannot be calculated without making assumptions and applying correction factors for the energy value of gill and urinary excretions.

Water Temperature, Fish Size, and Digestibility

Windell *et al.* (1978a) found no effect of water temperature or body size (except 18.6 g fish at 7°) on the dry matter and nutrient digestibility of a pelleted diet fed to rainbow trout. Digestibilities of total dry matter, protein, and fat were similar at 7, 11, and 15°C water temperatures and at body sizes of 18.6, 207.1, and 585.7 g. Cho and Slinger (1979) determined the digestibility of the proximate components of a reference diet (CRT-73) fed to rainbow trout maintained at 9, 12, 15, and 18°C. No differences in digestibility were observed between 0 and 15°C, whereas values for dry matter, crude protein, crude fat, and energy were 2.0 to 3.5 percent higher at 18°C. It may be concluded that digestibility of nutrients is similar over the range of temperatures used in normal aquaculture practices.

Meal Size and Digestibility

Elliot (1976) fed brown trout, *Salmo trutta*, meals ranging from 10 to 100 percent of maximum consumption. Absorption efficiency decreased with an increasing level of energy intake. Other workers also have found decreased absorption efficiency as meal size was increased (Kinne, 1960; Pandian, 1967; Solomon and Brafield, 1972; Windell *et al.*, 1978a).

True and Apparent Digestibility

The determination of nutrient digestiblity involves measurement of the amounts of nutrients consumed and voided in the feces. Fecal matter contains compounds other than those coming from feed. In the course of feed movement through the digestive tract, feed residue accumulates substances originating in the body. These endogenous substances include digestive enzymes and other secretions, epithelial cells abraded from the alimentary canal, and other materials of metabolic origin. While apparent digestibility determination treats feed and metabolic (endogenous) excretions collectively, true digestibility partitions the undigested fraction into those of feed and metabolic origin.

Endogenous or metabolic fecal losses by rainbow trout have been studied by Nose (1967) and Foltz (1978). Nose (1967) fed trout a nitrogen-free diet and measured endogenous nitrogen loss. Analysis of the fecal output revealed 0.5 g endogenous nitrogen per 100 g of diet. Using the value of 6.25 to convert endogenous nitrogen to crude protein, fecal matter contained endogenous protein equivalent to 3.1 percent of the amount fed. Foltz (1978) investigated the effect of temperature and reported that endogenous protein increased from 3.1 percent of the meal at 7°C to 8.4 percent at 19°C for all feeding levels.

While such a distinction between true and apparent digest-

ibility is desirable in certain experimental work, it does not have important significance in feeding practice. Thus reported digestibility coefficients commonly represent apparent digestibility.

Protein Digestibility Hastings (1969) summarized rainbow trout digestibility values for a large number of proteins or materials containing proteins (Table 4). Inaba *et al.* (1963) examined the digestibility of protein (mainly from white fish meal) by rainbow trout at dietary levels of between 24 and 45 percent. Protein digestibility was uniformly high, ranging between 85 and 94 percent at all dietary levels. Austreng and Refstie (1979) reported that apparent protein digestibility increased when protein level was increased in diets fed to different families of rainbow trout.

The digestibility of protein within the limits of practical diets was not greatly affected by other dietary ingredients except in the presence of high levels of carbohydrate. Dietary levels of lipid, up to 30 percent of the diet, did not affect the digestibility of either casein or white fish meal (Inaba *et al.*, 1963).

Fat Digestibility Early studies of fat digestibility were limited to the determination of digestibility of fat present in individual ingredients (Windell *et al.*, 1974; Cho and Slinger, 1979; Austreng, 1978) or in diets (Cho *et al.*, 1974; Austreng, 1979). Most feedstuffs contain less than 15 percent fat, of which 85–95 percent is digested and absorbed. Since only small amounts of fat remain in the feces, digestibility estimates can be highly variable (Smith *et al.*, 1980).

Phillips and Brockway (1956, 1959) reported an average of 85 percent digestibility for fats fed to rainbow trout. Austreng *et al.* (1979) confirmed this value for fats with a low melting point, such as soybean meal, cod liver oil, and crude capelin oil. Fat and fatty acid digestibility was essentially the same at different water temperatures of 3°C and 11°C, and Austreng (1979) suggested that fatty acid composition rather than melting point may be the main factor governing fat digestibility. Austreng *et al.* (1979) found that digestibility of individual fatty acids decreased with increasing chain length up to C_{18}. A further increase in chain length up to C_{22} resulted in increased digestibility. Unsaturated fatty acids showed higher digestibility than saturated fatty acids of the same chain length. These results agree with those obtained with several species, including pigs, rats, chickens, and mink.

Carbohydrate Digestibility The ability of brook trout, rainbow trout, and chinook salmon to digest different carbohydrates has been examined (Phillips *et al.*, 1948; Buhler and Halver, 1961; Singh and Nose, 1967; McCartney, 1971). Singh and Nose (1967) reported greater than 95 percent digestibility for glucose, sucrose, and lactose, and digestibility remained nearly constant regardless of the level in the diet. The discrepancy between the digestibility values reported for lactose by Phillips *et al.* (1948) (60 percent) and Singh and Nose (1967) (96.4 percent) may have been a result of different methodology (Tables 1 and 2). At a dietary level of 20 percent, dextrin and potato alpha-starch digestibility was 77 and 69.2 percent, respectively. At a 60 percent dietary level, the respective digestibilities were 45.5 and 26.1 percent. Digestibility of raw starch was low and decreased with increasing starch content in the diet (Phillips *et al.*, 1948; Singh and Nose, 1967; Spannhof and Kühne, 1977; Kitamikado *et al.*, 1964b). Dextrin and cooked starch fed to trout were better digested than raw starch (Smith, 1976; Smith *et al.*, 1980). Falge *et al.* (1978) proposed that the low digestibility of starch was related to low enzyme activity. Consumption of a high-starch diet by trout caused a depression in the activity of amylase. McCartney (1971) showed that the type and level of simple hexose fed to trout influenced activity of enzymes involved in carbohydrate metabolism.

Kitamikado *et al.* (1964a) determined the digestibility of protein in four diets containing 10, 20, 40, and 60 percent potato starch added to 90, 80, 60, and 40 percent whitefish meal, respectively. Digestibility of the protein fraction decreased from a high of 81 percent in the diet containing 10 percent starch (63.6 percent protein) to 32 percent in the diet containing 20 percent starch (56.5 percent protein).

Inaba *et al.* (1963), Kitamikado *et al.* (1964b), Nose (1967), Page and Andrews (1973), and Austreng *et al.* (1977) reported decreased protein digestibility in diets with high carbohydrate and a low level of protein. Rychly and Spannhof (1979) also reported a lowered protein digestibility with a high carbohydrate level and low-protein diet. Austreng *et al.* (1977) showed that utilization of metabolizable energy and protein progressively decreased in diets containing carbohydrates ranging from 17 to 38 percent of the metabolizable energy. Although crude protein and crude fat showed 90 percent digestibilities in the CRT-73 reference diet, the starch component averaged only 34 percent digestibility (Cho and Slinger, 1979).

Ash Digestibility Fish meal produced from the scraps remaining after fillet removal has higher levels of ash relative to the protein content than in meal from whole fish. Higher ash levels in fish meals have significant negative effects on the total dry matter digestibility of dry diets (Nose and Mamiya, 1963). These workers related flatfish meal ash content fed to rainbow trout to protein digestibility using the chromic oxide technique. Protein digestibility coefficients (79 to 95 percent) were negatively correlated ($r = -0.829$) with the ash content (14 to 21 percent of the meal), but were positively correlated ($r = 0.984$) with protein content (Figure 7, A and B). Gulley (1980) reported similar results after feeding Atlantic menhaden meal containing 18.8 percent ash to a control group of rainbow trout at total levels of 21.5, 23.8, and 26.3 percent ash. Protein, lipid, and dry matter digestibilities were negatively correlated with ash content. An interaction between ash content and nutrient digestibility was observed. The effect was not nutrient-specific because both lipid and protein digestibility were affected.

Table 4 contains a summary of digestion coefficients for protein, fat, carbohydrate, total dry matter, energy, and metabolizable energy values of diet ingredients determined by direct and indirect methods. The metabolizable energy values of some of the materials may vary from those previously published (Smith, 1976; Smith *et al.*, 1980).

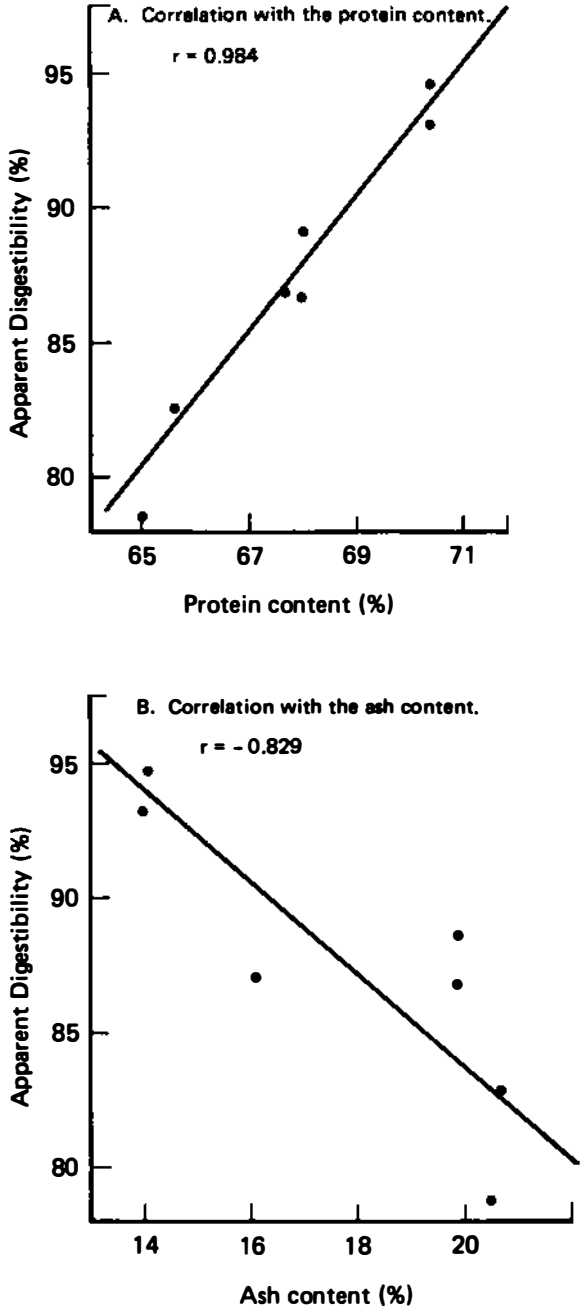

FIGURE 7 Correlation of the apparent digestibility of protein to the protein content or ash content of the fish meal (Nose and Mamiya, 1963).

NUTRITIONAL ENERGETICS

Intake of dietary energy is one of the major requirements for all animal production systems. Diets made from several carefully chosen feedstuffs will be adequate in the amounts of protein, although coldwater fishes with a simple intestine need special attention to provide protein of adequate quality, as well as certain lipid, mineral, and vitamin components. These nutrients are less likely to limit the gross efficiency of fish production than is the level of dietary energy intake.

The gross energy of ingested feed is divided into several metabolic pathways. A graphical illustration of food energy utilization with accepted abbreviations of energy metabolism terms (National Research Council, 1981) is shown in Figure 8. The magnitude of losses depends on the material fed and the level of feeding. It is desirable to know the magnitude of losses in digestion and metabolism when evaluating feeds for production purposes. Methods have been developed to measure the metabolizable energy (ME) of diets and dietary ingredients for trout and salmon (Smith, 1971; Smith and Rumsey, 1976). Figure 8 shows that this measurement takes into account the losses in digestion and excreted wastes. Not all of the ME is available for growth. Requirements for basal metabolism, voluntary activity, and heat of nutrient metabolism must be satisfied first. The portion of the ME not available for growth appears as heat. An estimation of the amount of heat produced under various feeding conditions, body sizes, and water temperatures, coupled with data on ME values of feeds and feed ingredients, is essential for the nutritional and economic evaluation of feed materials for fishes.

Energy Value of Feeds and Feedstuffs for Fish Gross energy (E), digestible energy (DE), metabolizable energy (ME), net energy (NE), and physiological fuel values (PFV) are different measures of food energy that have been used in fish culture. DE and ME are used to express feed values and energy requirements of fishes. There are not enough data available to ascertain whether DE values alone are sufficient or if the extra work associated with determining ME is justified. Theoretically, ME should be superior to DE, since, in fish, it accounts for energy losses from protein via urine and gills. ME is used for stating the energy requirements of several animal species (National Research Council, 1981). Different methodologies, i.e., total collection or use of indigestible matter, have been developed for derivation of these measurements, but more comparative data are needed to determine which method is most accurate. The aquatic environment makes quantitative feeding and separation and collection of metabolic waste difficult. The samples analyzed must be representative of the feed ingested and the wastes excreted. Energy studies with fish present several unique considerations. Usually the animals are small (<500 g). This mandates using special techniques to separate the excretions from large amounts of water, as well as microtechniques for analysis of small amounts of fecal and urinary wastes. Care must be taken to avoid mixing uneaten food with fecal material and to prevent excessive dilution and leaching by water. Teleost fish excrete ammonia as the primary nitrogen excretion product of protein metabolism (Hillaby and Randall, 1979). Most of the ammonia is excreted down a concentration gradient from blood to water, via the gills, with less than 2 percent excreted by the kidney (Goldstein and Forster, 1970). Therefore, quantitative data on both urinary and branchial excretions must be obtained in order to determine metabolizable energy measurements for fish.

Body temperature of fishes is not constant, but is dependent upon the temperature of the aquatic environment. Tem-

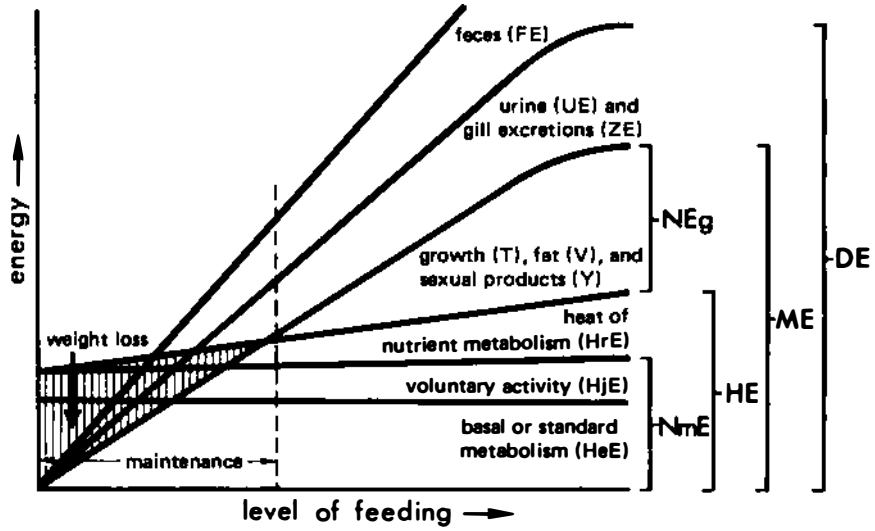

FIGURE 8 Distribution of food intake gross energy (IE) in a growing fish at various levels of feeding:

$$IE = FE + UE + ZE + HE + NE_g;$$

$$DE = IE - FE;$$

$$ME = IE - (FE + UE + ZE);$$

$$HE = H_rE + H_jE + H_eE.$$

perature affects the ability of fishes to digest and absorb nutrients (Brocksen and Bugge, 1974; Davies, 1964).

Phillips (1969) assigned "available caloric values" expressed in calories to protein, fat, and carbohydrate based on average digestion coefficients and heats of combustion of the components in a feed. While these physiological fuel values (PFV) should approximate ME values, they do not because of general assumptions made. The kilocalories for protein are corrected for energy loss in the urine, assuming a 90 percent digestibility, based on the determination that 7.9 kcal of energy were lost for each gram of nitrogen in the urine of human subjects consuming mixed diets (Atwater and Rosa, 1899) [*viz.*, if 1 g of nitrogen in the urine represents the breakdown of 6.25 g of digestible protein, there would be 7.9/6.25 = 1.25 kcal lost per gram of digested protein.] The 1.25 value can vary according to the factor used to convert nitrogen to protein, which can vary from 5.18 to 6.38, the assumed variable digestion coefficients for protein that were derived from both plant and animal sources and the chemical procedure used to determine nitrogen. There is little evidence that the constant 1.25 that is deducted from the gross energy of protein is applicable to fishes. It is tenuous to apply to fishes those data that were obtained with studies in humans, other mammals, or even birds because of energy required to produce the different nitrogen excretion compounds. Use of the factor 1.25 based on the composition of urine of urea-excreting humans fed a "mixed diet" is invalid for computing the PFV of protein for fishes [e.g., 5.65 kcal (GE of protein) − 1.25 kcal (unavailable for energy) × 0.9 (percent digestibility) = 3.9 kcal (available per gram of protein)]. Fishes excrete a very dilute urine in terms of nitrogenous compounds, and protein nitrogen is excreted primarily through the gills (Goldstein and Forster, 1970; Hillaby and Randall, 1979).

Phillips (1969) considered that the digestibility value for fat "is a compromise value between the digestibility of hard and soft fats" in salmonid diets [*viz.*, 9.45 kcal (GE of fat) × 0.85 (percent digestibility) = 8.0 kcal (available per gram of fat)]. Definition of hard versus soft fat as well as levels used were not given.

Physiological fuel values for carbohydrate are based on the assumption that most of the carbohydrate in trout diets is similar in form to raw corn starch that is about 40 percent digested [4.10 kcal (GE of starch) × 0.4 (percent digestibility) = 1.6 kcal (available per gram of carbohydrate)]. Smith (1971) and Buhler and Halver (1961) showed that digestibility of carbohydrate is dependent on the complexity of the molecule. Apparent digestion coefficients can range from 86 percent for glucose to 2.8 percent for alpha-cellulose. Hydrolysis, which occurs when starch is exposed to heat and moisture, can make starch more digestible. The type or nature of carbohydrate in feed and amount of heat processing must be known before a reasonable estimate of the energy value can be made. The PFV of available caloric values (Phillips, 1969, 1970) commonly used (4, 8, and 1.6 kcal/g of protein, fat, and carbohydrate) are inadequate to use in rigorous scientific investigation regarding the bioenergetics of feed utilization by fish.

Fish Energy Metabolism Differences in energy metabolism by fish and by mammals and birds that affect energy requirements are: the low basal energy needs (fish do not need to expend energy to maintain body temperature); a low energy cost of locomotion and voluntary activity (fish have no need for large antigravitation muscles in their aquatic environment and a streamlined fish moving through water represents one of nature's most efficient modes of locomotion); and the low energy cost for protein catabolism and waste nitrogen excretion (ammonia is the principal end product of protein catabolism as contrasted with urea and uric acid excreted by terrestial homeotherms). Fishes excrete ammonia principally through the gills and expend little or no energy in the formation, concentration, and excretion processes (Smith *et al.*, 1978a,b).

Body Temperature Regulation Freshwater fishes cannot maintain an internal body temperature more than a fraction of a degree different from the environment and have no presently known physiological means to conserve heat. Most species of fishes, however, have a preferred temperature and will seek water near that temperature. The high surface-to-vol-

ume ratio of these relatively small animals, the flow of blood through the gills, separated from the water by only two layers of cells, and the high thermal conductivity of water are some of the responsibile factors. Some species of fishes can live and grow in Arctic and Antarctic waters where body temperatures are at or below freezing temperature of fresh water, while other species thrive in hot springs with body temperature as high as 40°C.

Some difference of opinion exists regarding the effects of temperature on metabolic rate and increase in heat production following consumption of food (heat increment [HI]). Much of the difference probably can be attributed to methodology used, i.e., direct versus indirect calorimetry. Smith *et al.* (1978a) studied the net energy maintenance requirement of salmonids at different temperatures using direct calorimetry. Their work indicated a straight-line relationship from 3° to 18°C for four species of salmonids. The relationship between metabolic rate as measured by heat production (HP) and water temperature (T) of fasting rainbow trout is described by the equation $HP = 0.59 + 0.0525T$. Cho and Slinger (1979) employed indirect calorimetry using an open-circuit indirect respirometer with an automated data processor (Cho *et al.*, 1976) to study maintenance energy requirements and heat increment of feeding. Their maintenance energy requirement values at 7.5 and 15.0°C were 4.54 and 15.29 kcal/kg/24 h, respectively, which are consistent with those derived using the equation ($HP = 57.6\ W^{0.63}$) of Smith *et al.* (1978a). Cho *et al.* (1976) found heat increment to be 11 and 18 percent of the estimated ME when diets containing 40 and 60 percent protein, respectively, were fed to rainbow trout. Smith *et al.* (1978b) found that less than 5 percent of the ME is lost as HI in fish. It is difficult to account for the above-mentioned differences. Aside from differences in body weight, shifts occur in metabolic pathways and substrates oxidized at different temperature (Kanungo and Prosser, 1959; Hochachka and Hayes, 1962). These shifts would change the respiratory quotient (RQ) and thermal equivalent and explain the departure of the oxygen-consumption curve from the heat-production curve. Furthermore, the factor for converting oxygen consumption to heat production depends upon the material being oxidized and the nature of the waste products being excreted (Braefield and Solomon, 1972). Heat increment associated with food consumption is highly variable and depends, in part, upon the manner in which nutrients are utilized (Brody, 1945). Protein, for example, has a lower heat increment when used for tissue synthesis than when used to supply energy.

Energy for Voluntary Activity The metabolism of fish is difficult to measure under basal conditions. Experimentally, it is almost impossible to separate the energy expended for voluntary activity from that of basal metabolism. The more that a "motionless" condition is imposed upon a fish, the more it struggles to free itself. The terms "standard" and "routine" have been used to describe metabolism of fishes (National Research Council, 1973). Routine metabolism is the metabolic rate of undisturbed, acclimated fish that are allowed normal voluntary movement in relatively still water. Standard metabolism is an attempt to describe basal conditions by subtracting from routine metabolism the calculated value of activity. The activity factor is usually determined by forcing fish to swim at different rates and extrapolating to zero activity. Energy expended for voluntary activity is dependent on water movement, availability of feed, social/hierarchial competition, and sexual activity. A primary objective of fish-farming operations should be to provide conditions that will minimize fish activity and thereby increase the portion of dietary energy available for growth. However, some modicum of exercise might be in order if the fish are to be used for stocking.

Energy for Waste Excretion The primary waste product of protein catabolism in all animals is ammonia, which possesses no biological energy value and must be rapidly excreted or converted to less toxic compounds. Terrestrial homeotherms convert ammonia to urea and uric acid. This is energetically very costly, and as much as 30 percent of the gross energy of protein can be lost as heat increment (HI) (Brody, 1945). More than 98 percent of the ammonia excreted by teleost fish exits via the gills, with little or no energy expenditure (Goldstein and Forster, 1970; Hillaby and Randall, 1979). Less energy is lost in the excreted product, since ammonia has less energy per unit nitrogen (0.79 kcal/g) than does urea (5.4 kcal/g) or uric acid (8.2 kcal/g). Additionally, the fish, by virtue of the gills, does not have to concentrate or excrete these metabolic end products. Metabolism of protein by fish results in more of the gross energy left in the net energy fraction.

Fish Calorimetric Methods

The heat produced by an animal can be measured in a calorimeter (direct method) or it can be calculated by measuring oxygen consumption and carbon dioxide excretion and applying appropriate thermal equivalents (indirect method). In the absence of anaerobic metabolism and other unusual biochemical reactions for which the thermal equivalents are unknown, the two methods should give equal results. Both methods are equally simple in principle, but not in practice.

Historically, indirect methods have been used to study metabolic rates in fishes wherein the energy requirements are determined indirectly by measuring oxygen consumption (Brett and Groves, 1979). The measurement of O_2 consumption alone implies that metabolism is entirely aerobic (energy is derived from aerobic sources). Fishes probably do not satisfy all their energy needs through aerobic processes (Kutty, 1968). There is ample reason to question indirect methods that measure only oxygen consumption and use oxycalorific coefficients to convert oxygen consumption to heat. Values obtained with mammals have been extrapolated directly to fish. Brett (1973) proposed an oxycalorific equivalent of 4.8 kcal/liter O_2 for fishes. Brett and Groves (1979) adopted a new value of 4.63 kcal/liter O_2 based on lipid and protein rather than lipid and carbohydrate being the principal energy sources in carnivorous fish. Most times the respiratory quotient (RQ) is not determined. Indirect calorimetry, employing theoretical oxycalorific equivalents, yields equivocal data for fish energetics.

Brett and Groves (1979) suggest that direct calorimetry is not an appropriate technique for fishes because of problems of measuring heat liberation in water with its large specific

heat and the relatively low metabolic rate of ectotherms. Smith *et al.* (1978a,b) found, with a highly sensitive calorimeter, that heat produced by the chemical reactions within the bodies of fishes is rapidly transferred to water, and specific changes in metabolic rate can be detected immediately. Smith (1976) demonstrated a maximum variation of less than 1 percent in repeated calorimeter runs. The problems of heat loss by vaporization and radiation, which complicate direct calorimetry with land animals, do not occur with fishes. An adiabatic calorimeter, with its inherent stability and accuracy, appears to be an accurate instrument for use in fish energetic studies.

It has been shown that overall energy efficiency of fishes is higher than for most animals because of the relatively low maintenance requirement (Smith *et al.*, 1978a,b). Protein efficiency in terms of nitrogen retained/nitrogen eaten is lower because fishes tend to use protein as an energy source. The 24-h energy cost for maintenance of trout at 15°C has been measured as 58 kcal $w^{0.63}$ (Smith *et al.*, 1978a) as compared with 70 kcal $w^{0.73}$ for mammals and 83 kcal $w^{0.75}$ for avian species (Brody, 1945; Kleiber, 1975).

VITAMINS

Vitamins are organic compounds that are required in minute amounts for normal growth, reproduction, health, and general maintenance of fish metabolism. Eleven water-soluble vitamins and four fat-soluble vitamins are known to be required by coldwater fishes. Compared with most mammals and birds, the gastrointestinal tract of fishes does not contain a typical pattern of established microorganisms (Margolis, 1953). Accordingly it cannot be assumed that the fish obtains any appreciable quantities of vitamins from microbial synthesis in the intestine. Enzymic transformation of the amino acid, tryptophan, into the vitamin, niacin, is very inefficient in trout and salmon (Poston and DiLorenzo, 1973; Poston and Combs, 1979). Therefore, niacin and probably most other vitamins essential in metabolism must be included in salmonid feeds. Intensive production of fishes in water low in extraneous natural feed amplifies the importance of fulfilling the dietary requirements for the vitamins discussed and listed in Table 7. Vitamins are grouped into fat-soluble vitamins A, D, E, and K; the water-soluble B vitamins; and L-ascorbic acid, choline, and myo-inositol.

Fat-Soluble Vitamins

Vitamin A Each clinical sign of deficiency of vitamin A (Kitamura *et al.*, 1967b) described in Table 8 is nonspecific, because these can result from a lack of other nutrients. The signs are reliable, considered collectively, in conjunction with levels of liver vitamin A, hematology, and histopathology (Poston *et al.*, 1977). A dietary concentration of 2,500 to 5,000 international units (IU) of vitamin A per kilogram of diet is adequate for young trout (Kitamura *et al.*, 1967b). One IU of vitamin A is defined as *0.30* μg all-*trans* retinol.

Liver oils and meals from marine fish, and synthetically produced vitamin A, can provide vitamin A in fish diets. The efficacy of β-carotene in meeting the vitamin A requirement of trout and salmon apparently is dependent upon water temperature. Coldwater fishes utilize precursors of vitamin A at 12.4°C to 14°C, but do not at 9°C (Poston *et al.*, 1977). The activity of β-carotene 15,15'-dioxygenase, which oxidizes β-carotene to retinal in the intestinal mucosa (Gronowska-Senger and Wolf, 1970), may be restricted at cold temperatures. The biopotency of β-carotene has not been determined for trout and salmon at their standard environmental temperatures (SET) of 15°C and 10°C (National Research Council, 1973), respectively.

Vitamin A is essential for normal structure and function of eye and gill, through its involvement in the metabolism of mucopolysaccharides and visual pigments, and for general maintenance of epithelial tissues of various physiological systems.

Feeding massive levels (2.2 million IU) of vitamin A (retinyl palmitate) per kilogram of diet to trout at 8.3°C reduced growth and hematocrit, or packed cell volume (PCV) and caused necrosis of the caudal fin (Poston *et al.*, 1966). Feeding similar levels (up to 2.5 million IU) of retinyl palmitate at 12.4°C also reduced body fat and liver size of trout (Poston, 1971a). A high intake of dietary protein (Poston and Livingston, 1971) or methionine (Eckhert and Kemmerer, 1974) by young trout reversed or ameliorated some of the toxic effects of excess vitamin A in fish fed a low level of protein.

Vitamin D Vitamin D_3 (cholecalciferol) is at least three times as effective as vitamin D_2 (ergocalciferol) in satisfying the requirement for vitamin D in trout (Barnett *et al.*, 1979). One IU of vitamin D is defined as the biological activity of 0.025 μg of cholecalciferol. Cholecalciferol is the precursor of the hormone, 1,25-dihydroxy D_3, which is the biologically active form responsible for its functions of facilitating absorption and utilization of calcium and phosphorus. The vitamin is first converted to 25-hydroxy D_3 by the liver and then is further hydroxylated in the kidney to 1,25-dihydroxy D_3. The requirement for vitamin D_3 by fingerling trout fed a semipurified diet at 15°C is between 1,600 and 2,400 IU/kg of diet.

Signs of vitamin D deficiency include slow growth, an impairment of calcium homeostasis manifested by clinical signs of tetany of the white skeletal muscles and ultrastructural changes in the white muscle fibers of the expaxial musculature (George *et al.*, 1979), and an increase in plasma triiodothyronine (Leatherland *et al.*, 1980b). No hypocalcemia or changes in bone ash were detected (Barnett *et al.*, 1979). Trout responded favorably to supplemental 25-hydroxy D_3 and 1,25-dihydroxy D_3, but quantitative requirements have not been determined for these two substances.

Fingerling trout fed 3.75 million IU of vitamin D_3 per kilogram of semipurified diet for 40 weeks had hypercalcemia and increased PCV but no difference in rate of growth or survival (Poston, 1969b).

Vitamin E Activity of vitamin E is present in several tocopherols. Each tocopherol has a different biological activity with *d*-α-tocopherol having highest activity. One IU of vitamin E is defined as the biological activity of 1 mg of *dl*-α-tocopheryl acetate. Vitamin E acts as a lipid-soluble antioxidant and is required to protect phospholipid-containing

biological membranes, such as the erythrocyte plasma membrane and subcellular membranes of mitochondria and microsomes. Susceptibility of these membranes to oxidative degradation and ultimately to disruption of cell function varies directly with the level of polyunsaturated fatty acids in the phospholipids. The dietary requirement for vitamin E depends upon the level of polyunsaturated fatty acids in the diet and in the fish tissues, as well as the level of other antioxidants and selenium that, as a component of glutathione peroxidase, also protects against peroxidative degradation.

Vitamin E acetate at 30 IU per kilogram of diet, in the presence of selenium, is adequate for salmonid diets (Woodall and LaRoche, 1964; Hung *et al.*, 1979, 1980). Higher dietary levels of the more labile forms of vitamin E, such as the alcohol, may be required to prevent deficiency signs in fish fed other diets.

Signs of vitamin E deficiency in trout and salmon, either with or without selenium, are numerous (Poston *et al.*, 1976). These include impaired erythropoiesis, indicated by many immature, irregularly shaped erythrocytes (poikilocytosis) of various sizes (anisocytosis), fragility and fragmentation of erythrocytes, extreme anemia, marked susceptibility to stress of handling, high mortality, yellow-to-brown serous fluid in the body cavity (ascites), and increased content of body water (exudative diathesis). Both dietary vitamin E and selenium are needed to prevent dystrophic skeletal muscle lesions, consisting of enlarged muscle fiber bundles with loss of definition of the degenerating bundles.

Massive levels of vitamin E (5,000 mg of *dl*-α-tocopherol per kilogram of diet) caused reduced PCV values in trout (Poston and Livingston, 1969).

Vitamin K Trout require vitamin K for the synthesis of plasma proteins (prothrombin and thromboplastin), which are essential in blood coagulation (Poston, 1964). No effect on growth or survival has been observed. Typical intestinal vitamin K synthesizing microflora have not been described in fishes (Margolis, 1953), but dietary sulfaguanidine, as well as coldwater temperatures, caused prolonged blood coagulation times and low PCV values in trout (Poston, 1964).

Vitamin K compounds are 2-methyl-1,4-naphthoquinones and are involved in electron transport and oxidative phosphorylation reactions of cellular metabolism (Brodie, 1961). Investigations by Suttie (1980) demonstrated that vitamin K is a required cofactor for a microsomal enzyme system that carboxylates glutamyl residues of precursors of blood-clotting proteins to γ-carboxylglutamic acid residues in biologically active proteins. One-half to 1 mg of menaquinone equivalent of menadione sodium bisulfite (MSB) or menadione pyrimidinol (MPB) per kilogram of semipurified diet is sufficient to maintain normal coagulation and PCV of fingerling trout blood (Poston, 1976a). Massive levels (2,400 mg/kg diet) of MSB evidently do not adversely affect growth, survival, blood coagulation, or PCV values of young trout (Poston, 1971b).

Water-Soluble Vitamin Complex

Nutrients are often classified quantitatively as micronutrients when required at less than 100 mg/kg of complete diet and as macronutrients when needed at 100 or more mg/kg. Eight vitamins of the vitamin B complex are micronutrients, whereas choline, inositol, and vitamin C are macronutrients.

Vitamin C Conclusive evidence that salmonids require dietary L-ascorbic acid was provided by Kitamura *et al.* (1965) and Halver *et al.* (1969). The dietary requirement for ascorbic acid by trout and salmon apparently depends upon several factors, including fish size, growth rate, other dietary components, and stress conditions under which fish may be reared. Young, rapidly growing fish require the highest amounts of L-ascorbic acid. Halver *et al.* (1969) reported that rainbow trout and coho salmon weighing less than 1 g require 100 and 50 mg of L-ascorbic acid per kilogram of diet, respectively, for optimal growth. Hilton *et al.* (1978) reported, however, that not more than 40 mg of ascorbic acid per kilogram of diet is required by rainbow trout weighing about 7 g. Deterioration of dietary vitamin C due to processing and storage losses necessitates addition of excess vitamin C in trout and salmon diets (Hilton *et al.*, 1977). Environmental contaminants, such as toxaphene, in dietary ingredients increase the apparent vitamin C requirements of fish (Mehrle and Mayer, 1975; Mayer *et al.*, 1978).

L-ascorbic acid acts as a reducing agent and works synergistically with vitamin E and selenium to combat peroxidation of tissues (Poston and Combs, 1980b). It is primarily involved in the formation of procollagen and is necessary for the biosynthesis of collagen and cartilage, and in tissue membrane synthesis and repair. Hilton *et al.* (1978) reported that ascorbic acid also functions in iron metabolism of rainbow trout. Many cellular hydroxylases require vitamin C for maximum activity.

Signs of vitamin C deficiency include structural deformities, scoliosis, lordosis, and abnormal support cartilage of the eye, gill, and fins, and internal hemorrhaging usually preceded by nonspecific signs such as anorexia and lethargy (Hilton *et al.*, 1978), ascites and hemorrhagic exophthalmia (Poston, 1967), low serum triiodothyronine (T3) levels (Leatherland *et al.*, 1980a,b), high levels of plasma triglycerides and cholesterol (John *et al.*, 1979), and anemia in advanced stages of deprivation. Hilton *et al.* (1978) suggested that a liver ascorbate concentration of below 20 μg per gram indicated a dietary deficiency. Halver (1972), however, recommended use of anterior kidney ascorbate concentration for the clinical assessment of vitamin C status of fish.

Thiamin Thiamin is a part of the coenzyme, thiamin pyrophosphate (also known as cocarboxylase), which is required for the decarboxylation of pyruvic acid and α-ketoglutaric acid in the intermediary metabolism of carbohydrates and lipids, respectively. Thiamin is also involved enzymatically in activating tissue transketolase, which is essential for the direct oxidative cellular metabolism of glucose. The level of erythrocyte transketolase activity (Brin, 1963) has been used as a sensitive, specific indicator of thiamin status in fish (Cowey *et al.*, 1975). The level of kidney transketolase activity declines in thiamin-deficient rainbow trout well in advance of appearance of external signs of thiamin deficiency such as slow growth, anorexia, hyperirritability, convulsions, instability, and loss of equilibrium (Lehmitz and Spannhof, 1977).

Trout require 1 to 10 mg of thiamin per kilogram of diet

(McLaren *et al.*, 1947), depending on other dietary ingredients, fish size, and growth rate.

Riboflavin Riboflavin functions as two flavoprotein enzymes, flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD), which serve as prosthetic groups of many enzymes involved in oxidation-reduction reactions (e.g., transport of hydrogen ions during cellular respiration). Based on growth and maximum storage of riboflavin (McLaren *et al.*, 1947), young trout (2 to 12 g) require 5 to 15 mg of riboflavin per kilogram of diet. Work by Hughes (1980) indicated, however, that 3 mg per kilogram of diet was adequate for rainbow trout. Signs of riboflavin deficiency in trout and salmon include slow growth, anorexia, inefficient conversion of feed, opaque lens and cornea of the eye (Halver, 1957; Poston *et al.*, 1977), and dark pigmentation of the body (Halver, 1953). The degree of *in vitro* stimulation (activity coefficient) of the $NADPH_2$-dependent erythrocyte glutathione reductase (EGR) activity in the presence of added FAD has been used as a sensitive enzymatic measurement of the biochemical riboflavin status in mammals (Tillotson and Sauberlich, 1971). Hughes (1980) found that an increase in the EGR activity coefficient in rainbow trout fed less than 3 mg of riboflavin per kilogram of diet preceded a measurable reduction in growth, or miscroscopically detectable cataracts.

Vitamin B_6 (Pyridoxine) Pyridoxine and vitamin B_6 are terms used to denote at least three chemically, metabolically, and functionally related substances—pyridoxine, pyridoxal, and pyridoxamine. Pyridoxal phosphate is the coenzyme involved in decarboxylating amino acids, in at least 22 transaminases (transferases), and in other aspects of protein metabolism, as well as in the metabolism of lipids (especially fatty acids) and carbohydrates. This coenzyme is essential for the synthesis of many neuroendocrine substances, such as 5-hydroxytryptamine or serotonin, from tryptophan. Consequently, signs of a deficiency of pyridoxine include general nervous disorders, epilepticlike seizures, hyperirritability, lowered resistance to handling, erratic and spiral swimming, rapid breathing, flexing of opercles, and rapid onset of rigor mortis after death, as well as other less specific signs such as anorexia, reduced growth, and poor feed conversion. Tissue pyridoxine is depleted rapidly in fish fed a pyridoxine-deficient diet. Signs of deficiency appear quickly, with high mortality usually occuring within 4 to 6 weeks. The reduction of alanine aminotransferase activity in erythrocytes and muscle (Smith *et al.*, 1974; Juerss, 1978) of pyridoxine-deficient rainbow trout, followed by an increase in the enzyme in trout fed a pyridoxine repletion diet (Juerss, 1978), indicated that the level of tissue aminotransferases can be useful in monitoring the biochemical status of pyridoxine in fish.

The dietary equipment for pyridoxine increases with level of intake of protein and rate of growth in trout (Phillips and Livingston, 1966). Young trout and salmon require at least 5 to 15 mg pyridoxine hydrochloride per kilogram of diet, depending on fish size and level of dietary protein.

Pantothenic Acid Pantothenic acid functions as part of coenzyme A (Co A), in metabolism and release of energy from all three energy-providing nutrients—carbohydrate, fat, and protein—by way of the tricarboxylic acid (TCA) cycle. Pantothenic acid, as a component of Co A, is required for the synthesis of fat. Co A is involved as an acceptor and donor of acetate groups (acetylation reactions) and is vital to all energy-requiring processes. A dietary insufficiency of pantothenic acid impairs the normal metabolism within mitochondrial-rich cells undergoing rapid mitosis and high energy expenditure. Structures such as the gill and kidney tubules are involved in osmoregulation, or active hydromineral homeostasis, and pancreatic acinar cells almost constantly synthesize enzymes essential for digestion of fats, carbohydrates, and proteins. A continual high level of energy is essential for these activities. Results of work with pantothenic acid-deficient trout (Poston and Page, 1980) showed occurrence of conglutinated megamitochondrial lesions (i.e., clumped mitochondria). These subcellular lesions, which apparently are caused by anoxia within cellular energy transfer mechanisms (Hartroft, 1964; Rouiller, 1964), initially appear as vacuoles or hyaline bodies and eventually lead to necrosis. Grossly, the signs of pantothenate deficiency are manifested as a condition called dietary gill disease (Wolf, 1945), including clubbed, exudate-covered gill lamellae, swollen opercles, fused gill filaments, abnormal swimming near the surface of the water, anorexia, poor feed conversion, loss of weight, and high mortality within 8 to 10 weeks.

Trout and salmon require 10 to 20 mg of calcium pantothenate per kilogram of diet, depending on the size of fish and composition of the diet fed (McLaren *et al.*, 1947).

Niacin Niacin, in the form of niacinamide, is an essential component of two enzymes, nicotinamide adenine dinucleotide (NAD) and nicotinamide adenine dinucleotide phosphate (NADP). Both NAD and NADP are involved as hydrogen donors and acceptors in the release of energy from all three energy-yielding nutrients—carbohydrate, fat, and protein. The vitamin is required by all living cells. It is equally active either as the nicotinic acid or as the amide.

Signs of nutritional deficiency of niacin in trout and salmon are pellagralike and include anorexia, reduced growth, poor feed conversion, a photosensitivity or sunburn, intestinal lesions, muscular weakness, and spasms and increased mortality (McLaren *et al.*, 1947). Since trypotophan is not an adequate precursor of dietary niacin for trout and salmon (Poston and Combs, 1980a), salmonids must obtain preformed niacin to meet their requirements. Based on maximum liver storage of niacin, trout require at least 95 mg of the vitamin per kilogram of diet (Phillips and Brockway, 1947). Salmon possibly require up to twice that level (Halver, 1972). The presence of phenothiazine or antimetabolites such as pyridine-3-sulfonic acid and thioacetamide may increase the requirement for dietary niacin (Rucker, 1957; Halver, 1972). However, niacin in excess of requirements inhibits growth (Poston, 1969b).

Biotin Biotin, as a component of several coenzymes, plays an important role in the addition (carboxylation) and removal (decarboxylation) of carbon dioxide in various reactions such as synthesis of fatty acids (acetyl Co A carboxylase),

oxidation of carbohydrate (pyruvate carboxylase), synthesis of niacin and microsomal protein, and formation of pancreatic amylase. Based on growth, maximum liver storage of biotin and optimum feed conversion, young trout need a minimum of 0.05 to 0.25 mg of biotin per kilogram of diet (McLaren *et al.*, 1947; Poston, 1976b; Castledine *et al.*, 1978). Most diets containing fish meal, or a combination of fish and plant proteins, probably will contain sufficient biotin.

Signs of biotin deprivation in trout and salmon include anorexia, slow growth, increased mortality, poor feed conversion (McLaren *et al.*, 1947), degeneration of gill lamellae (Castledine *et al.*, 1978) and epithelium, depressed activity of liver acetyl Co A carboxylase and pyruvate carboxylase, abnormal synthesis of fatty acids and glycogen (Poston and McCartney, 1974), degenerative acinar cells of the pancreas, and deposition of glycogen in kidney tubules (Poston and Page, 1980). As in other animals, type and level of dietary fat can affect signs of biotin deficiency in salmonids. Feeding a partially hydrogenated soybean oil obscured biotin deficiency signs in trout (Kitamura *et al.*, 1967a; Poston and McCartney, 1974).

Folacin(s) Folacin (also called pteroylglutamic acid or folic acid) is essential for synthesis of the nucleic acids, DNA and RNA, and thus is necessary for normal erythrocyte formation. As the biologically active coenzyme, formyltetrahydrofolic acid, it is required for the transfer of single-carbon units (e.g., methyl groups) within rapidly metabolizing and dividing cells. Examples of this function are the formation of the amino acid methionine from homocysteine, serine from glycine, and the synthesis of choline from ethanolamine. The requirement for folacin is from 1 to 5 mg per kilogram of diet for young trout and salmon (McLaren *et al.*, 1947).

Signs of folacin deficiency in trout and salmon include anorexia, slow growth, inefficient feed conversion, and macrocytic normochromic, megaloblastic anemia (Smith, 1968; Smith and Halver, 1969) characterized by pale gills, anisocytosis, and poikilocytosis. The erythrocytes are large with abnormally segmented and constricted nuclei. A large number of megaloblastic proerythrocytes were present in the erythropoietic tissue of the anterior kidney. Production of erythrocytes decreases with time in fish fed a folic acid-deficient diet for at least 6 weeks. Anemia disappears when fish are fed folic acid for about 8 weeks. Combined dietary deficiencies of either folic acid and *p*-aminobenzoic acid (Phillips *et al.*, 1964) or of folic acid and vitamin B_{12} (John and Mahajan, 1979) accelerated the development of a more pronounced anemia in fish.

Vitamin B_{12} Vitamin B_{12} (cyanocobalamin), the functional forms of which are referred to collectively as cobamide coenzyme, is necessary for normal growth, normal formation of blood, and healthy nervous tissue. Vitamin B_{12}, together with folic acid, is needed to provide single carbon units such as methyl groups for formation of DNA in hemopoietic tissue. Vitamin B_{12} catalyzes the formation (release) of folic acid with only one glutamic acid molecule from more complex naturally occurring folic acid containing several molecules of glutamic acid. It also is involved in the formation of folic acid coenzymes. Lack of either of these vitamins accentuates the deficiency of the other, which apparently remains in a metabolically unavailable form. Folic acid and cyanocobalamin, thus, have complementary roles in fish metabolism.

Salmon (Halver, 1957) and trout (Phillips *et al.*, 1964) fed low amounts of vitamin B_{12} showed a high variability in numbers of fragmented erythrocytes, and in hemoglobin values, with a tendency for a microcytic, hypochromic anemia. In the presence of adequate dietary folic acid, a tentative requirement of 0.002 to 0.003 mg for vitamin B_{12} per kilogram of diet has been suggested (Halver, 1957).

Choline The presence of three methyl (CH_3) groups in the choline molecule makes it a desirable methyl donor in a variety of biological reactions. Choline reacts with acetyl Co A to form the neurotransmitter acetylcholine. It is also a constituent of the fat-related substances lecithin and sphingomyelin. Although the amount required in the diet is influenced by the level of other nutrients such as methionine, folacin or vitamin B_{12}, it is essential for growth, efficient conversion of food and prevention of fatty livers in trout (McLaren *et al.*, 1947) and salmon (Halver, 1957). The minimum level required by young trout for optimal growth is no more than 1,000 mg per kilogram of purified diet (Ketola, 1976). Trout can synthesize sufficient choline from dietary methylaminoethanol and from dimethylaminoethanol but not from aminoethanol or betaine (Ketola, 1976).

Myoinositol (Inositol) Myoinositol, a biologically active cyclohexitol, is a structural component in living tissues. It occurs as a phospholipid, lipositol, in animal cells. It is present in nucleated erythrocytes. A deficiency of inositol causes anorexia, anemia, and reduced growth and inefficient feed conversion, as well as a decreased rate of gastric emptying and activity of cholinesterase and certain transaminases. Increased concentration of dietary glucose may raise the need for inositol in some fishes (Shitanda *et al.*, 1971), however, the tentative quantitative requirement for inositol is 250–300 mg per kilogram of diet for young salmon and trout (McLaren *et al.*, 1947). A nutritionally inactive form of inositol occurs in plants, especially oilseed products and grains, as a more complex, water-insoluble phytin (calcium–magnesium salt of inositol hexaphosphoric acid).

MINERALS

The mineral requirements of fishes are difficult to quantify accurately. Since most minerals are required in small quantities, it is difficult to formulate diets and maintain environments that are sufficiently free of minerals to conduct requirement or deficiency studies (Phillips, 1959). Essential minerals may be obtained from water by exchange across the gill membrane or from food by absorption across the gut (Phillips, 1959). At least part of the salmonid requirement for calcium, cobalt, iron, magnesium, potassium, sodium, zinc, and others can be obtained directly from the water (Phillips, 1959; Phillips *et al.*, 1956, 1957, 1958, 1959, 1963, 1964; Podoliak, 1970; Podoliak and Holden, 1965, 1966; Zeitoun *et*

al., 1976). Certain minerals such as chlorides, phosphates, and sulfates are more effectively obtained from feed sources (Phillips *et al.*, 1959, 1963). Mineral interactions may also affect absorption from feed or water sources (Phillips *et al.*, 1956, 1958, 1960; Phillips, 1959; Hunn and Fromm, 1966; Podoliak, 1970).

Minerals perform a wide variety of structural, biochemical, and physiological functions (Phillips and Podoliak, 1957). At least 22 minerals, 7 major minerals (calcium, phosphorus, potassium, sodium, chlorine, magnesium, and sulfur) and 15 trace elements (iron, zinc, copper, manganese, nickel, cobalt, molybdenum, selenium, chromium, iodine, fluorine, tin, silicon, vanadium, and arsenic), are essential for animal life (Underwood, 1977). Athough most of these elements are probably required by fishes, only 7 dietary minerals have been shown to be required or utilized by salmonids (Table 9). Additional trace minerals are often added to salmonid diets to assure adequate supplies of minerals (Halver and Coates, 1957; Ketola, 1975a). Mineral mixtures for natural ingredient and purified diets that should satisfy all coldwater fish requirements are given in National Research Council (1978).

Total Mineral Intake

Wolf (1951) fed rainbow trout a synthetic diet without mineral supplementation for 23 weeks. The trout grew and survived at normal rates. Abnormal signs usually associated with mineral deficiency were not observed. He concluded that the mineral requirements of the trout had been met by solutes in the hatchery water supply. However, the diets used contained 55 percent casein, which probably provided calcium, phosphorus, and many trace elements (Phillips *et al.*, 1963). A later study by Ogino and Kamizono (1974) demonstrated the need of rainbow trout for dietary mineral supplementation. Young trout were fed a synthetic diet containing graded levels of McCollum's salt mixture No. 185 (formulated for rats) with trace elements (Halver and Coates, 1957) for 50 days. Diets lacking the salt mixture caused growth retardation, high mortality, and skeletal deformities (malformed heads, scoliosis, and lordosis). The optimum dietary level of salt mixture under their experimental conditions was 4 percent based on growth and red blood cell counts.

Young coho salmon fed salt-enriched diets (1.5 to 12 percent NaCl–KCl mixture or instant ocean salts added to the 3 percent mineral supplement of the basal diet) gained less weight and utilized diets less efficiently than controls (Zaugg and McLain, 1969). However, salt-enriched diets resulted in greater survival of young coho salmon exposed to marine salinities (33‰). The growth and feed conversion of Atlantic salmon smolts were not significantly affected by dietary sodium chloride levels (Shaw *et al.*, 1975a,b).

Water salinities (10 to 20‰) have been shown to have only a minor effect on net protein utilization, protein efficiency ratios, and growth of Atlantic salmon smolts (Shaw *et al.*, 1975a), coho salmon smolts (Zeitoun *et al.*, 1973), and rainbow trout fingerlings (Zeitoun *et al.*, 1974). Free amino acid concentrations in blood and latero-dorsal muscle tissues of rainbow trout were affected by environmental salinities (Kaushik *et al.*, 1977; Kaushik and Luquet, 1979). Blood concentrations of aspartic acid, threonine, and histidine were reduced in saltwater-adapted trout, while other essential amino acids increased in relative concentrations (Kaushik and Luquet, 1979). Total α-amino nitrogen levels (Kaushik *et al.*, 1977) and free amino acid levels (Kaushik and Luquet, 1979) were significantly increased in the blood and muscle tissue of saltwater-adapted rainbow trout. Relative differences in free amino acid pools induced by salinity were not significantly affected by dietary amino acid patterns (Kaushik and Luquet, 1979).

Calcium and Phosphorus Because of their structural importance and metabolic interactions, calcium and phosphorus requirements are often considered together. Calcium phosphate is the major mineral constituent of bone. Other important functions of calcium include osmoregulation, blood clotting, nerve irritability, and as a cofactor in enzymatic reactions. Unlike calcium, phosphorus is a component of a wide variety of organic molecules.

The major portion of the calcium requirement of salmonids can be met by calcium absorption from the water (McCay *et al.*, 1936; Ogina and Takeda, 1978; Phillips *et al.*, 1960), while dietary calcium is poorly utilized (Phillips *et al.*, 1963). Dissolved calcium absorption by brook trout was only slightly affected by the calcium concentration of the water from 5 to 50 ppm (Phillips *et al.*, 1957). Other minerals (magnesium, strontium, barium, copper, and zinc) may depress calcium absorption (Podoliak, 1970). Vitamin D does not appear to be necessary for absorption of calcium from the water or diet (Barnett *et al.*, 1979). Conversely, dissolved phosphorus is poorly absorbed by trout (Phillips *et al.*, 1958), while dietary phosphorus is used to meet the majority of the salmonid requirements (Phillips *et al.*, 1960).

The calcium requirement of rainbow trout can be met by 16–20 ppm dissolved calcium (Nose and Arai, 1976). Under conditions of low water calcium, trout will increase calcium absorption from feed sources (Phillips *et al.*, 1958). Both brook and brown trout have been shown to use dietary calcium (McCay *et al.*, 1936; Podoliak and Holden, 1965).

Natural ingredient diets containing high levels of animal proteins may not require supplemental inorganic phosphorus (as phosphate) (Phillips *et al.*, 1957, 1963). Supplementation of the U.S. Fish and Wildlife Service production diet PR-11 containing 25 percent whole herring meal and 0.55–0.65 percent available phosphorus with inorganic phosphates produced no significant differences in weight gain, feed conversion, or mortalities of rainbow trout (Reinitz *et al.*, 1978). Ketola (1975a,b) demonstrated that mineral supplements are required when soybean meal is substituted for fish meal in diets fed to Atlantic salmon and rainbow trout. Atlantic salmon fed a diet containing 0.7 percent phosphorus from plant sources required a minimum inorganic phosphorus supplement of 0.6 percent of the diet for normal growth and survival. Phytin phosphates apparently are not used as efficiently as organic phosphates from animal sources by Atlantic salmon. The level of available dietary phosphorus required to maintain normal growth in rainbow trout was estimated to be 0.7–0.8 percent of the diet (Ogino and Takeda, 1978).

Studies by Phillips *et al.* (1958) indicated that phosphorus

utilization by brook trout was best at a dietary calcium:phosphorus ratio of 1:1. Later studies failed to demonstrate a significant effect of dietary calcium (Nose and Arai, 1976) or dietary calcium:phosphorus ratios (Reinitz *et al.*, 1978) on the performance of rainbow trout. Differences in response to the level of dietary minerals may have been influenced by levels of calcium and phosphorus in the water supply.

Dietary induced phosphorus deficiency signs include reduced growth, poor feed conversion, and depressed bone ash content (Ketola, 1975a). Dietary induced calcium deficiency signs have not been described. However, alterations in calcium metabolism or excretion producing renal calcinosis in rainbow trout have been described as a result of dietary deficiencies of magnesium (Cowey *et al.*, 1977) and tryptophan (Kloppel and Post, 1975). Similar renal calcium deposits have been described in rainbow trout caused by noninfectious diseases of unknown etiology (Gillespie and Evans, 1979).

Other Minerals Iodide deficiency was probably the first nutritional deficiency described for salmonids (Gaylord and Marsh, 1914). Marine and Lenhart (1910) correctly described proliferation of thyroid tissue in brook trout as simple hyperplasia (endemic goiter) instead of thyroid carcinoma. They demonstrated that complete remission of thyroid hyperplasia could be effected by adding an iodide–iodine solution to the culture water. Although iodide is actively absorbed via the gills by rainbow trout (Hunn and Fromm, 1966), dietary iodide supplementation is required when iodide levels are low in culture water. Quantitative dietary requirements have been established for premigrant chinook salmon raised in water containing low (0.2 μg/liter) concentrations of iodide (Woodall and LaRoche, 1964). Chinook salmon fingerlings (0.5 g) and parr (9 g) required 0.6 mg and 1.1 mg iodide per kilogram of dry diet, respectively. These requirements were based on iodine storage, since growth retardation and clinical deficiency signs were not observed.

Dietary magnesium deficiencies may result in renal calcinosis (Cowey, 1976; Cowey *et al.*, 1977). Rainbow trout fed magnesium-deficient diets (4 mg of magnesium per 100 g of diet) developed renal calcification at dietary calcium levels of 2.7 percent and a dietary calcium to phosphorus ratio of 1:1 (Cowey, 1976). Lower dietary calcium levels (1.4 percent) and magnesium supplementation (0.1 percent) produced trout with normal renal calcium levels (Cowey *et al.*, 1977). Trout fed diets supplemented with magnesium had weight gains twice those of trout fed no supplemental magnesium (<0.063 g of magnesium per kilogram of diet). Rainbow trout fed a 45 percent casein diet containing McCollum salt No. 185 fortified with trace elements (Halver and Coates, 1957) required a magnesium supplement of 0.06–0.07 percent of the dry diet (Ogino *et al.*, 1978). Deficiency signs included loss of appetite, decreased growth, lethargy, reduced whole body and vertebral ash, spinal curvature, and histological changes in muscle, pyloric caeca, and gill filaments. Calcium and magnesium interactions were not considered.

The dietary zinc requirement of rainbow trout has been estimated to be between 15 and 30 ppm based on growth rates (Ogino and Yang, 1978). Feeding diets extremely low in zinc (1 mg/kg of diet) produced rainbow trout with retarded growth rates, high mortality, and other deficiency signs, which included erosion of fins and skin and eye cataracts. Deficiency signs were eliminated by feeding diets containing 5 ppm zinc. Bilateral lens cataracts have also been observed in salmonids fed diets containing whitefish meal (WFM) as a substitute for herring meal. Ketola (1978, 1979) demonstrated that lens cataracts could be prevented in rainbow trout fed diets containing WFM supplemented with zinc or Na_2EDTA. Whitefish meal diets supplemented with phosphates and carbonates of calcium, sodium, and potassium increased the severity of trout lens cataracts. Since the zinc content of diets containing WFM was relatively high (60 ppm), Ketola (1979) speculated that zinc absorption or utilization from WFM may have been impaired by minerals found in excess in WFM (calcium, phosphorus, sodium, or potassium).

Information concerning the dietary requirements of other minerals is limited. Dietary selenium supplements increased growth and survival of Atlantic salmon, and both selenium and vitamin E supplements were required to prevent muscular dystrophy (Poston *et al.*, 1976). Iron supplements increased trout growth and prevented anemia (Kawatsu, 1972); however, trout may be able to satisfy this requirement by absorption of iron directly from the water (Zeitoun *et al.*, 1976). Since dietary cobalt may be required for the synthesis of vitamin B_{12} by gut bacteria, it is often added to practical diets (Halver, 1976). Salmonids may also require in feed or water sources other trace minerals known to be required by other animals.

OTHER FEED COMPONENTS

Feedstuffs are complex mixtures of many components, including nutrients and nonnutrients. Nonnutrient components are either genetically determined within the organism from which the feed ingredient was derived, or arise from outside the ingredient itself. Some of the nonnutrient components are man-made; others result from uncontrollable events. This discussion pertains primarily to the physiological significance of nonnutrient components that are genetically determined or have been incorporated for enhancement of dietary properties such as acceptance, palatability, odor, color, stability, digestibility, and bioavailability.

The physiological significance of a feed component may be evaluated on the basis of the level of intake. Accordingly, an ingredient may be classified as either physiologically inert, beneficial (i.e., serving a useful function), or potentially harmful or hazardous (Friedman and Shibko, 1969; Shibko and Friedman, 1972).

Fiber

The generic term, fiber, refers to mixtures of cellulose, hemicelluloses, lignin, pentosans, mannans, and other complex carbohydrates or polysaccharides. These components are usually indigestible unless bacterial action occurs within the digestive tract. Use of plant products in fish feeds has resulted in the introduction of varying amounts of complex carbohydrates. The possibility of using complex polysaccharides

to improve physiological utilization by providing dietary bulk has not been investigated extensively with fish. Although growth and food consumption in salmonids are inversely proportional to the molecular weight of the carbohydrate consumed, a low concentration of dietary alpha-cellulose apparently increases the efficiency of protein utilization (Buhler and Halver, 1961); a small amount of digestion and absorption of cellulose reportedly occurs in salmonids (Smith, 1971).

High dietary levels ($\geq$ 20 percent) of carbohydrates of even low molecular weight suppress the digestion of dietary protein, food conversion, and growth in trout (Kitamikado *et al.*, 1964; Singh and Nose, 1967; Luquet *et al.*, 1975; Edwards *et al.*, 1977). Logarithmic gradations of wood cellulose from 2 to 32 percent of a natural-ingredient diet linearly suppressed growth, feed conversion, and dietary metabolizable energy of trout offered increased levels of diet to compensate for dilution of nutrients (Poston, 1980). The primary adverse effect was due to dilution of nutrients by cellulose. Although dilution of nutrients caused increased frequency of voluntary feeding and total amount of feed consumed, rate of gastric emptying was accelerated and intestinal transit time was reduced (Lee and Putnam, 1973; Grove *et al.*, 1978). Use of wide ranges of concentrations of nutritionally inert cellulose and hemicellulose as dilutents in experimental fish diets (Cowey *et al.*, 1971; Lee and Putnam, 1973; Poston, 1975) has obscured the true requirements for nutrients by altering physiological responses of fishes to the nutrient.

Dietary Binding Agents

Very little experimental testing has been conducted on the use of natural and synthetic binding agents to minimize leaching of nutrients in fish diets. The addition of 2 percent sodium chloride to trout diets consisting of liver and spleen (1:1), or meat and dry meals, reduced nutrient loss and improved fish growth (Phillips, 1970). Although carboxymethylcellulose (CMC), a commercial cellulose product, is physiologically inert when included at 2 percent of trout diets (Wood *et al.*, 1954), its strong binding effect on the physical state of the diet suppresses availability of nutrients and reduces fish growth. Increased levels of dietary water reduce the binding capability of CMC in the diet. Algin products, both natural and commercial, are binding agents (Wood *et al.*, 1954). Their use in flake or extruded diets for crustaceans (New, 1976) and tropical fishes (Meyers *et al.*, 1972) suggests a potential use in diets for salmonid fry (Meyers, 1979).

Water

Information is sparse on the effect of level of dietary water on fish growth and efficiency of feed utilization by salmonids. Growth and feed/gain efficiency of trout fed a semipurified diet containing either 9.6 or 55 percent water did not differ significantly when compared on the basis of intake of dry matter or energy (Poston, 1974). Unpublished results by Smith (Poston, 1974) indicated that rainbow trout and coho salmon grew equally well with either 10 or 35 percent dietary water, but that chinook salmon grew faster when the diet contained 35 percent water.

Bromley (1980) reported that, as long as the intake of the basic nutrients by turbot was adequate, dietary water content was immaterial; quantity of feed intake was the primary factor.

Plant and Animal Pigments

Carotenoid pigments are important fat-soluble nonnutrient dietary components in the coloration of fishes and fish eggs. Carotenoid pigmentation in trout and salmon is often advantageous and desirable, both esthetically for consumer acceptance and physiologically because of reported increased reproductive efficiency (Deufel, 1965). Shrimp and daphnia and some other aquatic animals ingest plant carotenoids, including β-carotene (β,β-carotene) and xanthophylls or oxycarotenoids (e.g., lutein, β,ϵ-carotene-3,3′-diol), and convert them to such compounds as astaxanthin (3,3′dihydroxy-β,β′-carotene-4-4′-dione), which is also present, in ester form, in salmonid skin (Simpson and Kamata, 1979). Intermediate pigments such as canthaxanthin (β,β′-carotene-4,4′-dione) can be fed to salmonids, either in shrimp and crab products, and other natural ingredients (Saito and Regier, 1971) or in synthetic carotenoids (Deufel, 1965; Schmidt and Baker, 1969). Synthetic canthaxanthin, however, is not approved for addition to fish feeds in the United States. Corn gluten and alfalfa meal contain xanthophyll and impart a yellow color in trout. Paprika contains a red pigment, capsanthin, which is nontoxic, and at 2 to 10 percent of the diet produces a natural red–orange coloration in trout (Tunison *et al.*, 1944; Phillips *et al.*, 1945).

Nonnutrient Components of Oilseed Products

Oilseed products are potentially good sources of nutrients. With proper processing and supplementation with limiting amino acids (Rumsey and Ketola, 1975; Ketola, 1980), the biological and energetic values can be very high. However, the mere chemical presence of a nutrient does not guarantee adequate absorption and utilization.

Deleterious Nonnutrient Components

Minerals from plant feedstuffs, in contrast to those from animals, are generally utilized inefficiently by fishes. For example, phosphorus in plant feedstuffs is practically unavailable to salmonids (Ketola, 1975).

Phytates Soybeans, peanuts and cottonseeds, and similar oilseeds are high in phytic acid, a water-soluble organic acid of myoinositol. Phytic acid forms strong complexes (phytin) with protein, phosphorus, zinc, calcium and magnesium, and other polyvalent minerals at intestinal pH conditions of fishes and other monogastrics that have little phytase (Erdman, 1979). Phytic acid in most cereal (e.g., corn, wheat, and rice) is associated with specific parts such as endosperm, germ, and hull. Phytic acid can be selectively extracted with these components. In contrast, the phytates are found throughout the kernel of oilseeds. Phytates are resistant to heat and moisture. Their removal requires special manipulation of pH, anion exchange, and ultrafiltration (Hartman, 1979).

Protease Inhibitors Raw and underautoclaved soybeans contain a globulin protein that complexes with, and inactivates, the pancreatic and intestinal proteases, trypsin and chymotrypsin. This adverse effect accounts for about 40 percent of the growth reduction of rats fed unheated soybean meal (Liener, 1979). Feeding raw soybeans and isolated protease inhibitor itself causes, by way of a neuroendocrine negative feedback inhibition mechanism, hypertrophy of the pancreas and an accompanying increase in secretion of proteases by the pancreas of some animals (Liener, 1979). Excess secretion of pancreatic trypsin and chymotrypsin, which are rich in the sulfur-containing amino acids cystine and methionine, may result in an endogenous loss of these amino acids. Soybean protein is inherently low in both methionine and cystine.

Trout are extremely sensitive to dietary soybean protease inhibitors (Sandholm *et al.*, 1976; Combs and Poston, 1978). Autoclaving a commercial isolated soybean protein (ISP) under 1.12 kg/cm^2 of pressure for 30 min before incorporation as the sole dietary protein reduced both the *in vivo* and *in vitro* activity of dietary ISP trypsin and chymotrypsin inhibitor. This heat also caused increased growth and protein efficiency ratio (PER) in trout (Poston *et al.*, 1978). Observations with other animals (Liener, 1979) suggest that soybean protein is, in itself, highly indigestible unless denatured by heat and that this undenatured protein may bind proteases by forming an enzyme–substrate protein complex. This common mechanism between the protease inhibitors and the digestive refractoriness of unheated soybean protein possibly accounts for the adverse effects on salmonids.

Phenols Phenolic compounds in many sources of oilseed protein have a significant role in the development of undesirable flavors, odors, and antinutrients in fish diets. The phenolic compounds are of several types, and they include the hydroxylated derivatives of benzoic and cinnamic acids, coumarins, flavonoids, and the polyphenolic tannins and lignin (Sosulski, 1979). Free phenolic acids are particularly deleterious in soybean, peanut, and cottonseed products because of enzymatic oxidation to *o*-quinones and subsequent binding to the ϵ-amino group of lysine and the thioether group of methionine. The resulting products are nutritionally unavailable to monogastric animals.

Cyclopropenoid Acids Food oils, such as cottonseed oil and kapok oil, contain significant quantities of the cyclopropenoid fatty acids (CPFA)—sterculic acid and malvalic acid. Sterculic acid is a 19-carbon fatty acid with a propene ring involving carbons number 8 and 9. Malvalic acid is an 18-carbon homolog of sterculic acid. Dietary CPFA synergistically stimulate aflatoxin-induced hepatomas in trout (Lee *et al.*, 1971), possibly by altering the activity of the mixed function oxidase (MFO) system of the fish (Eisele *et al.*, 1978).

Other Toxic Components Raw linseed meal contains a water-soluble fraction that is detrimental when fed to trout and other animals. One toxic fraction is linatine, which occurs naturally as a combination in peptide linkage, between 1-amino-D-proline and glutamic acid (Liener, 1969). The toxic 1-amino-D-proline forms an *in vitro* complex with pyridoxal phosphate. Its toxic effects can be counteracted by the administration of pyridoxine. Linamarin, a cyanogenetic glucoside in linseed, is toxic to trout (Tunison and McCay, 1938). This glucoside releases acetone and hydrogen cyanide upon hydrolysis. Linamarin is heat stable and only slightly soluble in water but can be extracted satisfactorily by heating at 1.06 kg steam per square centimeter for at least 20 min (Tunison and McCay, 1938).

ADVENTITIOUS TOXINS

Toxic materials affecting feed ingredients include: accidental contaminants, factors arising from processing, microbial toxins, natural plant and animal constituents, and intentional feed additives (Wogan, 1977). Naturally occurring toxic plant and animal constituents are discussed in another section, "Other Feed Components." The discussion of the remaining potential feed-borne toxins will focus on experimentally induced or documented cases of acute and chronic effects of these toxins on salmonids.

ACCIDENTAL CONTAMINANTS

Accidental food contaminants include heavy metals, pesticides and herbicides, industrial chemicals, and other organic toxicants. These toxicants may enter feeds through agricultural practices, manipulation involved in food processing and distribution, or purely accidental circumstances (Friedman and Shibko, 1972). Most research on accidental food contaminants has resulted from interest in simulating bioaccumulation of environmental pollutants.

Heavy Metals Fishes can absorb heavy metals either directly from their environment or from feed. Availability of metals from the environment depends upon the element and its chemical form (Bryan, 1976). Certain elements in feed may not be available for uptake by fishes. Hodson *et al.* (1978) demonstrated that dietary lead (e.g., lead bound to beef liver protein) was not available to rainbow trout. Feed may play a larger role than water in providing zinc, manganese, cobalt, and iron to certain fishes (Bryan, 1976). Other metals, such as mercury, may be absorbed equally well from water or feed sources (Hartman, 1978). Because of the complex nature of absorption of metals from water and feed sources, dietary levels of heavy metals necessary to meet mineral requirements may exceed acute water-borne toxicity levels.

Mercurials are of particular interest, because they can inadvertently enter fish diets through contaminated feed ingredients. Spinelli and Mahnken (1976) demonstrated that mercury contaminated dogfish meal (2.3 ppm total mercury) could be used to replace up to 50 percent of the herring meal in Oregon Moist Pellet-type (intermediate moisture) diets fed to coho salmon without decreasing growth rates. Mercury accumulation in muscle tissues did not exceed U.S. FDA tolerance levels (0.5 ppm Hg). It was speculated that mercury accumulation in muscle was related to the content and form of mercury in the diet and to other dietary constituents. Rainbow trout are apparently unable to convert inorganic mercury to more toxic methyl mercury (Pennacchioni *et al.*, 1976) even though oral doses of mercuric chloride increased total tissue mercury burdens. Dose level and feeding schedules of ethylmercury *p*-toluene sulfonanilide influenced the concentrations of mercury in muscles (Hartman, 1978). At low dietary exposure levels (0.5 μg Hg/g of diet), mercury tissue levels were similar to the daily oral dose. Higher daily doses (0.5–25 μg/g) produced a dose related concentration of mercury in muscle, but the slope of the regression was greater than 1. Total body burden of mercury remained high after 6 months of mercury-free feeding.

Certain plants are capable of accumulating very high levels of a wide variety of minerals from soils (Friedman and Shibko, 1972). However, information on the availability of organically bound minerals and toxic reactions to dietary mineral excesses in fishes is not available.

Pesticides and Herbicides Pesticides and herbicides affect fishes mainly in areas that receive massive sprayings. Interest in bioaccumulation and bioconcentration of these contaminants has stimulated studies in the acute and chronic effects of oral doses on salmonids (Buhler *et al.*, 1969; Macek and Korn, 1970; Macek *et al.*, 1970; Mayer *et al.*, 1970, 1972; Mehrle *et al.*, 1971, 1972; Mehrle and Bloomfield, 1974; Klaverkamp *et al.*, 1976; Addison *et al.*, 1977; Hawkes and Norris, 1977; Addison and Willis, 1978). Caution should be used when extrapolating results of oral dose effects because diet quality (Mehrle *et al.*, 1977), pesticide interactions (Macek *et al.*, 1970; Mayer *et al.*, 1970), and experimental design may affect acute and chronic toxicities.

Detectable levels of organochlorine (oc) pesticides and

polychlorinated biphenyls (PCB's) have been found in commercial fish feeds (Brauhn and Schoettger, 1975; Gruger *et al.*, 1976; Parejko and Wu, 1977; Mac *et al.*, 1979). Since fish products are the principal source of the contaminants in fish feeds (Schoettger and Mehrle, 1972; Brauhn and Schoettger, 1975), it is recommended that fish oils and fish meals contain no more than 2 ppm and 0.1 ppm oc contaminants (including PCB's), respectively.

Addison (1976) reviewed the distribution, transport, and physiological significance of oc's and related compounds in aquatic organisms. Although oc's can be absorbed through the gills, feed appears to be the major source of stored oc's (Macek and Korn, 1970). Organochlorines are stored mainly in the lipids. The dynamics of oc retention and elimination are affected by oc interactions (Macek *et al.*, 1970; Mayer *et al.*, 1970). The sublethal effects of oc's are diverse, including alteration of lipid metabolism (Buhler *et al.*, 1969; Mayer *et al.*, 1970) and protein metabolism (Mehrle *et al.*, 1971, 1972; Mehrle and Bloomfield, 1974).

Industrial Chemicals and Other Organic Toxicants Polychlorinated biphenyls are a class of industrial chemicals used as plasticizers, heat transfer fluids, dielectrics, and hydraulic fluids because of their chemical unreactivity and availability in a variety of physical states. Polychlorinated biphenyls are widely distributed, persistent environmental contaminants that have been found in a variety of animal feeds above FDA-approved levels (0.2 ppm).

Leib *et al.* (1974) observed that rainbow trout fed diets containing 15 ppm PCB's (Aroclor 1254) retained stabilized relative tissue concentrations (ppm), while absolute quantities of PCB's (milligrams per fish) increased. After a 32-week feeding period, 68 percent of the PCB's fed were retained in the lipid fraction of various tissues. Trout did not eliminate the PCB's during starvation after exposure ceased.

Exposure to sublethal levels of dietary PCB's did not adversely affect growth rates of rainbow trout (Leib *et al.*, 1974) or coho salmon (Mayer *et al.*, 1977). Oral sublethal doses of PCB's administered to salmonids have been associated with liver enlargement and alteration of liver ultrastructure (Hacking *et al.*, 1977), induction of hepatic aryl hydrocarbon hydroxylase (Gruger *et al.*, 1977) and other hepatic microsomal enzymes (Sivarajah *et al.*, 1978), increased thyroid activity (Mayer *et al.*, 1977), and a reduction of plasma steroid levels (Sivarajah *et al.*, 1978). Dietary exposures of coho salmon to PCB's (Aroclor 1254) at a rate of 14.5 mg/kg body weight per day resulted in 100 percent mortality after 260 days, while lower doses ($\leq$1.45 mg/kg body weight per day) produced no observable toxicosis (Mayer *et al.*, 1977). Differences in the effects of PCB's result from the constituent polychlorinated biphenyls contained in commercial PCB mixtures.

A wide variety of other organic environmental contaminants have been administered orally to salmonids (Lotikar *et al.*, 1967; Lombardo *et al.*, 1975; Gruger *et al.*, 1976, 1977; Zitko and Hutzinger, 1976; Roubal *et al.*, 1977; Zitko, 1977; Grieco *et al.*, 1978; and others) because of interest in bioaccumulation.

TOXIC SUBSTANCES FROM FEED PROCESSING

Toxic substances can be inadvertently added to salmonid diets during processing. Processing contaminants have included: boiler water additives, hydraulic fluids, and lubricants from processing equipment; fumigants used to sterilize feeds; defoaming agents; coating materials; and toxic derivatives formed by a processing agent or procedure (Friedman and Shibko, 1972; Wogan, 1977). Unfortunately, little attention has been directed towards industrial contaminants in dietary feedstuffs and fish feeds.

MICROBIAL TOXINS

Under certain conditions, microorganisms associated with feed contamination or spoilage are capable of producing toxicoses. Feed-borne bacteria can pose toxicological hazards either through direct infection or by the production of toxic metabolites. Certain fungi can produce metabolites, termed mycotoxins, that are toxic to animals.

Food-Borne Bacterial Toxicoses Heat associated with diet processing reduces the risk of introducing feed-borne bacterial infections. Improperly stored or processed fish scraps have served as vectors of infectious bacteria. Mycobacteriosis (fish tuberculosis) in Pacific salmon was related to the practice of feeding raw fish offal to hatchery stocks (Wood and Ordal, 1958; Ross *et al.*, 1959). Infections were almost exclusively restricted to hatchery stocks (Ross *et al.*, 1959). Feeding pasteurized (30 min at 53°C) salmon viscera greatly reduced the incidence of infection (Hublou *et al.*, 1959). Transmission of the mycobacteria by transovarian passage or contact infection during artificial propagation could not be demonstrated (Ross and Johnson, 1962). Use of pasteurized salmon products in hatchery diets has essentially eliminated mycobacteriosis among Pacific salmon (Ross, 1970).

Salmonella contamination of fish feeds is possible by way of contaminated feed ingredients such as animal by-products or fish meals (Taylor and McCoy, 1969). Insects and rodents also act as *Salmonella* vectors, contaminating stored ingredients or feeds with feces (Friedman and Shibko, 1972).

Shepherd (1978) cited a recent personal communication with Christensen that reported an outbreak of botulism in trout. The trout were fed trash fish that had been stored under warm anaerobic conditions. The affected trout appeared nervous and died shortly after being fed the botulinum-toxin-contaminated trash fish. Coho salmon have also been infected by botulism apparently from the sediment of hatchery ponds at the State of Washington Elokomin Hatchery (M. Eklund and L. Peck, personal communication).

Mycotoxins A large number of mold species can produce toxic metabolites. Toxin production by molds that can contaminate fish feeds or feedstuffs requires three simultaneous conditions: (1) an environment suitable for growth, (2) a substrate suitable for growth, and (3) the physical presence of the generating organism (Mislivec, 1977). The responses of

salmonids to mycotoxicoses caused by *Aspergillus flavus* (aflatoxins) and *Fusarium* sp. (T-2 toxins) have been studied.

An epizootic of liver hepatoma occurred in farm reared rainbow trout around 1960 (Hueper and Payne, 1961; Rucker *et al.*, 1961; Wood and Larson, 1961). Wolf and Jackson (1963) demonstrated that a contaminated cottonseed meal in a commercial trout diet was responsible for the liver hepatoma. Aflatoxin B_1 was identified in toxic cottonseed meal (Engebrecht *et al.*, 1965). Rainbow trout were shown to be sensitive to aflatoxins B_1 and G (Ashley and Halver, 1961; Ashley *et al.*, 1964, 1965; Halver, 1965, 1967, 1969; Lee *et al.*, 1968, 1971; Sinnhuber *et al.*, 1968a). Levels as low as 0.4 ppb aflatoxin B_1 produced hepatocarcinoma in the Mt. Shasta strain of rainbow trout (Lee *et al.*, 1968). Because of the sensitivity of rainbow trout to aflatoxins, Sinnhuber *et al.* (1977) developed a trout mycotoxin bioassay.

The extreme sensitivity of rainbow trout to aflatoxin B_1 may be a result of liver metabolism. Aflatoxicol (R_0) is the major *in vitro* metabolic derivative of aflatoxin B_1 by rainbow trout liver (Schoenhard *et al.*, 1976). Trout mitochondrial enzymes can reconvert R_0 to aflatoxin B_1 (Loveland *et al.*, 1977), thus intensifying the aflatoxicosis. The pathology of aflatoxin induced liver carcinoma in trout has been reviewed by Scarpelli *et al.* (1963), Halver (1965, 1968, 1976), Ashley (1970), Wales (1970), Snieszko (1972), and Ghittino (1976). Brook trout (Wolf and Jackson, 1967) and coho salmon (Halver *et al.*, 1969) are apparently less sensitive to chronic aflatoxin treatments.

The effects of additional agents that may affect the hepatic carcinogenesis of aflatoxins must be considered when present in the diet. Cottonseed meal may contain several ingredients that enhance aflatoxicosis. These enhancing agents include gossypol (Sinnhuber *et al.*, 1968b), cyclopropenoid fatty acids (Lee *et al.*, 1968; Sinnhuber *et al.*, 1968b, 1974; Loveland *et al.*, 1979), and altered lipids (Sinnhuber *et al.*, 1968b, 1974; Lee *et al.*, 1971). The source and level of dietary protein can also influence the response of rainbow trout to aflatoxin B_1 (Lee *et al.*, 1978). Two common environmental contaminants have been fed in conjunction with aflatoxin B_1. The effect of dietary dieldrin on aflatoxin B_1 carcinogenesis in rainbow trout was highly variable and, consequently, was not statistically significant (Hendricks *et al.*, 1979). Polychlorinated biphenyl (Aroclor 1254) inhibited the carcinogenic effect of aflatoxin B_1 in rainbow trout (Hendricks *et al.*, 1977; Stott and Sinnhuber, 1978).

Rainbow trout fed semipurifed diets containing ammoniated aflatoxin contaminated corn for 12 months had a low incidence of hepatoma (Brekke *et al.*, 1977). Ammoniation reduced the total aflatoxin content of the contaminated corn without reducing the nutritive value of the corn. Use of an ammoniation process for detoxifying corn will require Food and Drug Administration approval.

Chronic exposure to low levels of dietary T-2 toxins failed to induce liver carcinoma in rainbow trout (Marasas *et al.*, 1969). Low doses (200 to 400 ppb) of T-2 toxins appeared to promote growth (Marasas *et al.*, 1969), but higher doses (greater than 2.5 ppm) caused a dose-dependent depression of growth and feed consumption (Coffin, 1979). Dietary T-2 toxin produced a dose-dependent anemia in trout (Coffin, 1979).

The acute oral LD_{50} of T-2 toxin for rainbow trout fingerlings has been estimated at 6.1 to 6.5 μg/kg of body weight (Marasas *et al.*, 1967; Smalley, 1973). Mature fish survived doses in excess of the LD_{50} for fingerlings (Marasas *et al.*, 1969). Both fingerling and adult rainbow trout exhibited pathological changes when fed LD_{50} levels of the toxin (Marasas *et al.*, 1967, 1969). These changes included loss of intestinal mucosa, severe edema, and exophthalmia.

INTENTIONAL FOOD ADDITIVES

Intentional food additives are chemicals added to the feed to improve nutrient value, maintain freshness, prevent disease, promote growth, increase acceptability, or aid in processing. More than 3,000 chemicals have been added to feeds for these purposes (Furia, 1972). The primary additives in animal feeds are added to increase nutrient value; however, preservatives (mainly antioxidants), growth-promoting substances, and drugs may also be added (Friedman and Shibko, 1972). Use of certain additives as growth promoters and disease prevention agents have caused adverse effects when included in fish diets.

Drugs Drugs, particularly antibiotics, have been used in small doses as growth promoters and in larger doses as disease control agents. Currently, three drugs (Table 10) are registered for fishery use (Schnick *et al.*, 1979). Oxytetracycline and sulfonamides fed to various salmonids failed to induce accelerated growth (Snieszko and Wood, 1954; Wagner, 1954; Schumacher, 1955) and sulfonamides retarded brook trout growth (Snieszko and Wood, 1954). Sulfonamides fed to cutthroat trout brood fish at the registered antibacterial dose level resulted in severe kidney histopathology and increased mortality among males (Smith *et al.*, 1973). Caution should be used in feeding medicated feeds.

Steroid Hormones Diets supplemented with steriod hormones have been fed to salmonids in attempts at sex reversal, inducement of sterility, and growth promotion. Sex reversal or inducement of sterility can postpone or minimize the need for dietary energy in gonadal maturation. Feeding 20 mg/kg of diet 17-β-estradiol to rainbow trout and Atlantic salmon for minimum periods of 30 and 21 days after first feeding, respectively, has resulted in all female stocks (Johnstone *et al.*, 1978). Conversely, feeding 1 mg/kg of diet 17-α-methyltestosterone from 2 to 25 days after hatching altered the sex ratio of rainbow trout (horai masu variants) toward an all male population (Yamazaki, 1976). Increasing the level of 17-α-methyltestosterone to 3 mg/kg of diet fed from first feeding for 90 days produced all male stocks of rainbow trout and Atlantic salmon (Johnstone *et al.*, 1978).

Fed at low levels (1.0–2.5 mg/kg of diet) over various feeding periods, 17-α-methyltestosterone has accelerated the growth and improved feeding efficiency of juvenile salmonids, including coho salmon (McBride and Fagerlund,

1973; Fagerlund and McBride, 1975; Higgs *et al.*, 1977; Yu *et al.*, 1979), chinook salmon (McBride and Fagerlund, 1973), kokanee, and rainbow trout (Yamazari, 1976). Spermatogonial development was reduced or eliminated at these dose levels. Other steroids that have promoted growth of coho salmon at low feed levels include: 11-ketotestosterone, oxymethalone (McBride and Fagerlund, 1976), testosterone (McBride and Fagerlund, 1976; Yu *et al.*, 1979), and estradiol (Yu *et al.*, 1979). Progesterone and 4-chlorotestosterone had no effect on coho salmon growth (McBride and Fagerlund, 1976). In general, steroid hormones fed to salmonids at higher levels (>10 mg/kg of diet) resulted in growth suppression, gonadal tissue degradation, and other anomalies.

Two synthetic androgens, dimethazine and norethandrolone, fed at 2.5–5.0 mg/kg of diet improved growth and feed efficiency of rainbow trout (Matty and Cheema, 1978). Diethylstilbestrol, a synthetic estrogen, has produced adverse effects on growth when fed to rainbow trout at low levels (Ghittino, 1970; Matty and Cheema, 1978) and induced hepatocarcinoma when fed at higher levels (Halver, 1965).

FORMULATING DIETS AND FEEDING PRACTICES

A number of authors have recently reviewed various aspects of formulating dry diets and feeding practices for salmonids, including Halver (1976), Higgs *et al.* (1979), Tiews *et al.* (1979), Luquet and Rumsey (1979), Spinelli *et al.* (1979), Nose (1979), Webber and Huguenin (1979), and Ghittino (1979a). Ghittino (1979b) and Solberg (1979) have reviewed studies relating to moist feed formulation. "Fish Feed Technology" was the subject of a comprehensive FAO/UNDP (1978) training course.

Numerous factors must be considered when formulating diets for coldwater fishes; some of the more important are listed below:

Nutrient Requirements. Knowledge should be available on which nutrients are required as dietary components and the quantitative needs and how they are affected by species, age, physiological functions, and water-quality parameters.

Nutrient Content of Feedstuffs. It is important to know the nutrient levels in the feedstuffs to be used along with their bioavailability. If actual analyses are unavailable, average analysis figures may be used.

Use Levels and Safety of Various Feed Ingredients. Feeding experiments have established the levels at which numerous feed ingredients may be used in combination with other commonly used ingredients. The use levels of some ingredients are restricted by the presence of heavy metals or pesticide residues or naturally occurring toxins.

Feedstuffs Market. Diets should be nutritionally and economically sound. Price and availability of feedstuffs on a given market at a specific time will influence the amount that can be used in the diet. It is possible to formulate diets in all countries of the world, but the ingredient composition will vary widely depending on local ingredient availability and economics. In some cases critical ingredients must be imported to formulate adequate diets. This applies, for example, to synthetic vitamins, amino acids, minerals, antioxidants, pellet-binding agents, antimold agents, pigmenting compounds, and other additives that form an integral part of many practical diets. Good-quality fish meals and marine oils are important ingredients in practical diets and are not produced in all countries.

Type of Processing Required. The choice of ingredients may vary, depending on whether a moist or dry pellet is desired. If a dry diet is desired, the manufacturer may wish to make steamed pellets or expanded (extruded) pellets. In the latter process there are differences in moisture, pressure, and temperature to gelatinize the starch portion so that the volume of the particles is increased due to abrupt reduction in pressure, when extruded (Robinette, 1977). Diets to be extruded must contain more starchy ingredients than are present in many diets to be steam-pelleted in order to achieve flotation. In practical diets the additional starch usually comes from vegetable protein supplements.

Stability and Palatability. Processing method affects the durability of the pellets and too great a proportion of fines may influence water quality. The composition of the diet should allow satisfactory pelleting and crumbling without undue "fines" production. Palatability and organoleptic properties of a diet are influenced by ingredient composition and nutrient balance. Some ingredients such as fish meal and fish oil are highly palatable to fish as compared with ingredients such as soybean meal; part of this difference may relate to taste and part to smell or texture (Webber and Huguenin, 1979).

Effects on Productive Performance, Feed Efficiency, and Quality of Fishes. It is possible to have marked differences in these characteristics, depending upon diet composition (Reinitz *et al.*, 1978; Cho *et al.*, 1976). The economics of the ingredient-supply situation may not always indicate the advisability of formulating diets that give maximum growth, reproductive performance, or feed efficiency; however, diets that do not approach optimum results are not likely to give maximum profitability. The composition of the fishes is determined by diet to a considerable extent, and a different composition may be required for stocking fish than for table fish (Buckley and Groves, 1979).

Effect on Water Quality. Nutrients supplied in the diet in excess of requirements are excreted into the water where they may cause eutrophication with consequent production of macrophytic plants or algae depending on the site. This is to be guarded against with all nutrients but particularly with phosphorus and protein, since this mineral and nitrogen are

usually the first and second limiting factors for plant growth in natural waters. Algal growth reduces feed consumption in fishes and decreases growth rate. It is also important to use highly digestible feed ingredients, since the undigested residues increase water pollution. Thus the balance of nutrients, the avoidance of excesses, and digestibility in fish diets assume a dimension of much greater significance in fishes than in diets for terrestrial mammals.

STEPS IN FORMULATING A DIET

It is first necessary to decide on the levels of protein and energy required in the diet. These and other recommended nutrient allowances are suggested in earlier chapters. Protein nutrition is really amino acid nutrition; thus protein supplements should be chosen to satisfy the essential amino acid requirements. The most efficient diets will provide the essential amino acid requirements with as low a level of protein as possible for the least cost. Each protein supplement has a different amino acid profile, and they should be combined in a manner such that each complements the amino acid profile of the others. It is sometimes economical to supplement practical diets with DL-methionine or L-lysine when these are limiting (Rumsey and Ketola, 1975; Ketola, 1979a,b). All the amino acids may be provided in crystalline form in semipurified diets for specific purposes, but only methionine, lysine, and arginine have practical economic application in commercial diets.

Protein supplements such as high-quality fish meal, soybean meal, corn gluten meal, cottonseed meal, and various animal by-products such as meat meal, poultry by-product meal, hydrolyzed feather meal, and ring-dried or spray-dried blood meal all may be used in fish feeds. Fish meals made from fish carcasses after removal of the fillets are high in ash and low in fat and protein and should be used only in limited quantities (Ketola, 1979c). Many other protein supplements may be used, depending upon economics and availability in a given area. High-quality fish meal is the preferred source of protein for natural-ingredient diets, but low supply, high cost, and contamination of the product with toxicants such as heavy metals and pesticides has resulted in much research in an attempt to reduce dietary levels of fish meal (Cho *et al.*, 1974; Gropp *et al.*, 1976; Slinger and Cho, 1978b; Tiews *et al.*, 1979; Spinnelli *et al.*, 1979).

For semipurified test diets, casein and gelatin, with some supplemental methionine, will satisfy the protein needs, although other protein sources such as torula yeast are used in low-selenium diets.

After deciding on the protein and energy levels desired and the protein supplements to use, one must then consider the levels and types of oil or fat, the level and type of carbohydrate, and the vitamins, minerals, and levels and types of unidentified factor sources that are needed to round out the diet. This information is found in earlier chapters.

In formulation of diets without the availability of a computer, a worksheet will facilitate calculation of the nutrient content of the diet. The worksheet should include columns for ingredient name, International Feed Number, and percent of each macroingredient in the diet, together with the levels of dry matter and all critical nutrients. It then becomes easy to calculate the nutrient content of the macroingredient portion of the diet and to formulate suitable vitamin and trace mineral premixes to satisfy the requirements for the diet.

The use of the computer and linear programming techniques has simplified the difficult task of selecting the correct combination of ingredients to produce the most efficient diets at least cost for domestic mammalian species. The same techniques are being used in fish feed formulation and will increase in popularity and value as more is learned concerning nutrient requirements, as well as ingredient-use levels and specifications for fish diets. It must be remembered that the value of a diet, whatever the method of formulation, is only as good as the information and quality of ingredients that are used.

Synthetic vitamins are usually the most economical. Vitamin supplements of various potencies may be used to supply the requirements. Generally, the higher-potency products are more economical but require greater technical skill in mixing. Stability and biological availability must be considered in purchasing vitamin supplements. Fish meal and fish oil are often significant sources of vitamins A, D, and E, but it is not safe to assume the presence of certain levels of these vitamins in such products. It is safer to provide the total requirements with synthetic stable sources of these vitamins in the absence of analytical data on the fish meals and oils.

Natural ingredients such as dried brewer's yeast, distiller's dried solubles, and dried whey, among others, are good sources of readily available B vitamins. The fact that they are in conjugated form as they occur in nature makes them less likely to be leached into the water. One or more of these ingredients should be included to provide B vitamins, particularly in starter feeds. The possibility that there may be as yet unidentified growth factors required by salmonids is another reason for including some of these natural sources of vitamins, especially in diets for young fish and brood stock.

The trace minerals employed should be dry and free-flowing. Technieal-grade minerals are more economical than chemically pure grades; the former should be used in commercial diets, while the latter are recommended in semipurified test diets. The sulfate, chloride, and carbonate salts of the trace minerals are more biologically available than the oxides, but even the latter may prove satisfactory, depending upon relative costs.

Hilton *et al.* (1977) showed that ascorbic acid is very unstable in practical and test diets. Factors that may affect the stability of ascorbic acid are moisture content of the diet, processing method, and storage conditions. Leaching losses of ascorbic acid and other water-soluble vitamins can also be significant (Slinger *et al.*, 1979). Supplemental levels in excess of the recommended requirements for certain vitamins are therefore necessary to compensate for such potential losses. The diets that are probably at greatest risk from leaching losses are starter diets, because of their larger surface area. The problem is also exacerbated by low water temperatures in which the fish are relatively inactive, so that the small feed particles remain in the water for longer periods than at higher temperatures.

The presence of 10–30 percent wheat middlings in some

practical diets absorbs oil and acts as a pellet-binding agent. This ingredient contains only 16–17 percent protein, which is of a relatively high order of digestibility; on the other hand the digestibility of the remaining dry matter is low (Cho and Slinger, 1979) and the use of such levels of middlings can only be justified on the basis of its fat absorption and pellet-binding capabilities. Wheat gluten meal, fish solubles, and dried whey are often used in fish diets and act as sources of nutrients as well as pellet-binding agents. Other materials, such as lignosol or bentonite, are used as pellet-binding agents but supply no nutrients.

FEED QUALITY

Fish meals and fish oils are highly important in fish diets, but they tend to be of variable quality. Special attention is necessary in testing and selecting these ingredients before their use in the feed and in storing them once selected. Some of the more important quality-control measures suggested to ensure quality fish meal and fish oil are shown below. These parameters are in many cases empirical and subject to change with further research.

Quality Control of Fish Meals:

1. Should be stabilized with liquid ethoxyquin (6-ethoxy-1,2-dihydro-2,2,4-trimethylquinoline) at manufacturers plant—200 mg/kg.
2. Stored at manufacturers plant for no more than 3 months.
3. Minimum crude protein 68 percent.
4. Pepsin digestibility not less than 92.5 percent.
5. Ammonia-N less than 0.2 percent.
6. Crude fat range 8–11 percent.
7. Maximum sodium chloride 3 percent.
8. Maximum ash 15 percent.
9. Steam processed, ground finer than 0.25 mm.
10. Maximum chlorinated hydrocarbons 0.1 ppm.
11. Free from mold, not caked or heated.

Quality Control of Fish Oils:

1. Peroxide value (PV) < 10 meq peroxide per kilogram.
2. Thiobarbituric acid value (TBA) < 70 mg of malonaldehyde per kilogram.
3. Nitrogen less than 1 percent.
4. Moisture less than 1 percent.
5. Stabilized with liquid antioxidant or mixture thereof—500 mg/kg ethoxyquin.
6. Maximum chlorinated hydrocarbons 1 ppm.

A suitable fish oil when deaerated and mixed with an antioxidant by bubbling nitrogen gas will remain stable for as much as 1 year by storage under nitrogen or in air-tight containers.

Other major ingredients used in salmonid diets should be subjected to proximate analysis before being used in fish feeds. After mixing, feed samples should be taken regularly for proximate and other analyses. The samples should be labeled as to batch number and date of mixing, which should be listed on the feed tag or bag. Feed ingredients and mixed feeds should be stored in a cool, dry place; feed mixed and stored for more than 3 months should not be fed to fish.

The FAO/UNDP (1978) training course and the Feedstuffs Reference Issue (Anderson, 1980) contain additional useful information that is beyond the scope of this publication for those wishing to formulate and manufacture feeds. Included in these publications are sections on Feed Manufacturing Terminology, Feed Ingredient Definitions, Practical Fish Diets, Antioxidants in Compounded Feeds, Quality Control of Incoming Ingredients, Storage Problems of Feedstuffs, Effect of Processing on Nutritive Value of Feeds, Aflatoxin Tests, Feed Mixing Regulations, Registration of Feed, Drug Classifications, Feed Labeling, Good Manufacturing Practices, FDA Inspection Checklist for Feed Manufacturers and Mixers, Control and Prevention of Drug Cross-Contamination in Feed, and Metric System Conversions.

FEEDING PRACTICE

The most appropriate time to initiate feeding of hatchery rainbow trout is when the yolk reserves have been completely absorbed. One should keep in mind that much yolk may yet remain in the abdominal cavity after the externally visible yolk has disappeared. The onset of feeding appears to be in synchrony with the histogenesis of the oropharyngeal mucosa, notably the mucous cells and taste buds. In studies made at an ambient water temperature of 10°C, the failure of fish to feed by day 37 appeared to be due to maldevelopment of the oropharyngeal mucosa (Twongo and MacCrimmon, 1976, 1977). Feeding too soon causes pollution of the environment, which can be hazardous; however, it is desirable to feed as soon as the first fish swim up. Swimup fry should be fed in slight excess so that feed is visible on the bottom of the rearing unit (i.e., tank, trough, raceway, or jar).

Feed consumption is markedly affected by ambient temperature. Feeding activity is very low at temperatures below 5°C and gradually increases to about 18°C. Swimup fry of some salmonid species are reluctant to consume dry feed at very low temperatures, and it is sometimes necessary to start the fish on chopped liver, which may be gradually replaced by dry feed (Cho and Slinger, 1978).

Another factor affecting feed intake is the energy content of the feed, because fish, like terrestrial animals, eat to satisfy energy needs (Lee and Putnam, 1973; Page and Andrews, 1973). Feed consumption is readily reduced in polluted water, for example in the presence of heavy algal growth. Other water-quality parameters, as well as physical factors such as rate of water exchange, type of rearing facility and size, density and physiological state of the fish, will influence feed intake. It is important to keep the rearing units clean at all times (Ghittino, 1972).

FEEDING RATES

The feeding rate of salmonids is commonly expressed as a percentage of body weight fed per day (Table 11). Smaller

fishes require feed at a greater percentage of their body weight per day than do larger ones. Feeding guides also show the marked effect of ambient temperature on feed requirement. Feed as a percentage of body weight for growing fishes can vary between 0.5 to 10.0 percent, depending on numerous factors (Haskell, 1959; Buterbaugh and Willoughby, 1967). Many of the older feeding guides were based on diets containing lower energy levels than present diets, and thus the feeding levels suggested are too high.

Automatic and hand-feeding operations can be successful if the operator is experienced. Frequency of feeding is important, with swimup fry fed a small amount of feed as often as 20–24 times per day and the frequency gradually being reduced to 1 to 3 times per day as the size of the fishes increases. Some operators prefer to use a 24-hour-a-day lighting period for the first several days or until the fishes are well started on dry feed. Feed particle size, hardness, and texture, as well as placement of feed in relation to fish size, are important considerations. Very small fishes will travel only a short distance for food, but large fishes move freely about the rearing unit.

Most feeding schedules represent only approximate guides to the amount of feed that should be fed to fishes of a given weight or length. Operators should have knowledge of a satisfactory growth rate and feed efficiency for a given ambient temperature for their strain and species of fishes and follow a feeding schedule to match these standards. In the absence of such standards, one could obtain gain and feed efficiencies much below the potential of the fishes. For example, Figure 9 shows satisfactory growth curves for a strain of rainbow trout grown at 7°C, 11°C, and 15°C. More time must be spent in feeding fishes at colder ambient temperatures than at SET or above. There are times when fishes should not be fed to satiation. If the ambient temperature goes up rapidly over a period of a few days, there is real danger from suddenly increasing feed intake to the new higher appetite level. In a pond situation, a combination of days with little sunlight, so that there is no photosynthesis, coupled with increased water temperature, which has a lower oxygen-holding capacity than cooler water, plus a large biomass of fishes, calls for a reduction in feed offered even though the fishes will eat avidly. Overfeeding under such conditions could possibly deplete the dissolved oxygen level to 5 ppm or less for a prolonged period and result in excessive mortality (Ghittino, 1972).

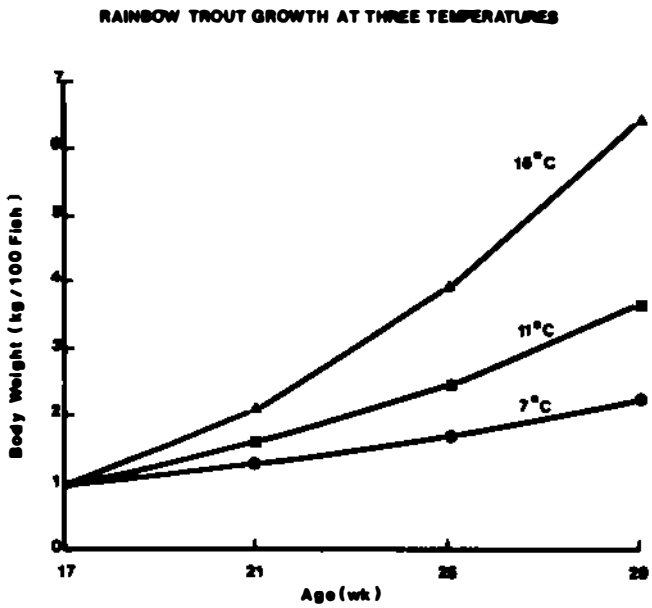

FIGURE 9 Rainbow trout growth at three temperatures (Slinger and Cho, 1978).

FEED PROCESSING AND STORAGE

Two comprehensive texts covering most aspects of feed milling, processing, and manufacturing will be of interest to readers wishing to have more details on the subjects dealt with in this chapter: Simmons (1963) and Pfost and Pickering (1976). For further information on feed processing technology and the effect of processing on feed quality and the physiology of animals, see MacBain (1968), Ott *et al.* (1973), and FAO/UNDP (1978).

Fish are more sensitive than terrestrial mammals to most chemicals, including drugs and insecticides. Cleanliness in the feed is therefore of great importance, particularly if animal feeds are being mixed with the same equipment because there is risk of cross-contamination without proper precautions.

MANUFACTURING

Salmonid feeds should be processed into pellets or granules to aid in prehension by the fish and prevent fouling of the water. Starter granules are exceedingly small in size, and therefore fine grinding of the ingredients is necessary to make a homogenous mix and for maximum digestibility. All feed ingredients should be preblended from bags or bulk stock and after thorough mixing should be pulverized in a hammer mill. For all feeds, 100 percent of the ingredients should pass through a U.S. standard sieve No. 45 (nominal sieve opening 0.354 mm).

Premix Preparation and Mixing

Premixing the microingredients is necessary to facilitate uniform distribution of vitamins, minerals, and other trace substances normally present in gram or smaller quantities. The premixes should represent a uniform mixture of one or more microingredients with a diluent of such physical properties that separation does not occur. Premixes should be systematically blended to an amount equal to about 4 percent of the total mix before being incorporated into the feed. The premixes should be introduced into a batch mixer when about one-half the ingredients that comprise the total diet have been added. The mixer should have complete clean-out features.

Steam Pelleting

Feed should be processed into pellets using dry steam to produce the proper textured pellets. Pellets must be soft enough for the fishes to prehend, yet firm enough to hold together with a minimum amount of "fines" produced in handling and transportation. "Fines" content (defined as particles passing through a 420-μ screen) should not exceed 3 percent of any feed at the manufacturing plant. It is necessary to spray most of the oil on the feed following pelleting or crumbling to produce particles of satisfactory durability and to minimize the amount of fines. The fat coating also helps prevent leaching losses from the feed. In the steam-pelleting process, steam at a pressure of about 0.5–3.5 kg/cm^2 is introduced, along with the feed in the conditioning chamber, where it is mixed with the feed to raise the temperature to 70–80°C and the moisture content to 16–17 percent. The feed remains in the conditioning chamber for about 5–6 s before being forced through the holes in the die. Dies are available with various-sized openings, and pellets may be cut to varying lengths. Satisfactory pellet sizes for salmonids are:

2.35 mm (3/32") long × 2.35 mm (3/32") diameter;
3.1 mm (1/8") long × 3.1 mm (1/8") diameter;
4.7 mm (3/16") long × 4.7 mm (3/16") diameter; and
6.25 mm (1/4") long × 6.25 mm (1/4") diameter.

Dies also vary in thickness, and the die thickness can alter the pellet durability as well as its suitability for making into crumbles. Dies 5 cm (2") in thickness with a 12.5-mm to 25-mm (1/2" to 1") release make satisfactory pellets for fish.

It is ideal if the diet ingredients permit production of a durable pellet without use of nonnutritive binding agents, such as wood pulp by-products or bentonite, which may pollute the water. Wheat middlings and wheat gluten meal, as well as dried whey and fish solubles, have good binding properties. Fine grinding of feed ingredients increases gelatinization of starch and durability of pellets. Following

pelleting the feed must be dried and cooled in order to prevent mold growth. Pelleting conditions vary with environmental factors, and the experienced mill operator will be able to make adjustments as conditions warrant.

Crumbles or granules are made by crushing pellets with a roller and then screening out the granules to the desired size. The standard granule sizes (Pfost and Headley, 1976) with the sieve numbers necessary to produce them are as follows:

Granule Size		United States and Canadian Standard Sieve	
		Opening	No.
Starter	To pass over	420 microns	40
	To pass through	595 microns	30
No. 1 Granule	To pass over	595 microns	30
	To pass through	841 microns	20
No. 2 Granule	To pass over	841 microns	20
	To pass through	1.19 mm	16
No. 3 Granule	To pass over	1.19 mm	16
	To pass through	1.68 mm	12
No. 4 Granule	To pass over	1.68 mm	12
	To pass through	2.38 mm	8
No. 5 Granule	To pass over	2.38 mm	8
	To pass through	3.36 mm	6

Starter and No. 1 and No. 2 granules should be made from 3.1-mm (1/8") or 4.7-mm (3/16") pellets. No. 3 and No. 4 granules should be produced from 4.7-mm (3/16") pellets. After sieving, the finished feed should contain not more than 15 percent oversized and/or undersized granules. The "fines" should be recirculated continuously so as to cause a minimum of alteration in feed formulation from that intended. Granules made from extruded pellets produce less "fines" than those from steam pellets, but the extrusion process is more expensive.

Pellets and granules should not be bagged or loaded for bulk delivery until cooled to within 2–3°C of ambient temperature and dried to a moisture content of 10 percent or less. Tags on bags or bulk trucks should be printed to show crumble number or pellet size, as well as diet identification and date of manufacture. Color coded tags help prevent errors in feeding.

Extrusion

The difference between steam pelleting and extrusion is that higher levels of moisture, heat, and pressure are employed in the latter procedure. The finely ground feed in the conditioning chamber is heated to 125–155°C with dry steam under a pressure of about 5–7 kg/cm^2 to raise the moisture to about 20–24 percent. The sudden reduction of pressure upon extrusion permits expansion of the water vapor, and the entrapment of air results in the production of buoyant feed particles. Feeds high in fat and animal or fish by-products are difficult to extrude, while inclusion of vegetable proteins and cereal grains provides starch, which gelatinizes with protein to give a durable water-stable pellet. Properly made extruded particles can be crushed to make crumbles with less loss of "fines" than with steam pellets (Slinger *et al.*, 1974). The starch in extruded feeds appears to be more digestible than that in steam pellets (Hilton *et al.*, 1980). Pellets smaller than 2.35 mm (3/32") in diameter will not float, so extruded pellets are usually produced as particles 4.7 mm (3/16") or larger in diameter.

Cold Pelleting

This type of processing is not commonly used in commercial feed production but is occasionally used in fish nutrition research. The steam pressure and temperature are drastically reduced in such processing; however, the moisture content may rise to 20 percent. The durability of cold pellets is poorer than that of steam or extruded pellets and therefore wastage may be increased.

Moist Pelleting

Moist pellets contain variable amounts of fresh and/or frozen, wet tissue together with some dry ingredients. The raw tissue should be pasteurized. The diet is processed in a manner similar to the cold-pelleting processing method, and the moisture content of the diet is sometimes as high as 50 percent (*v/w*). The pellets produced must be kept frozen until fed. Improper storage and handling of such diets can severely jeopardize the stability of certain vitamins and fats, as well as increase bacterial and fungal contamination. Salmonids prefer the soft moist pellets as compared with dry pellets, probably because of their soft nature and palatability. However, the disadvantages of having to keep the moist pellets refrigerated, the care needed in handling to prevent breakage, and the possible introduction of disease are factors tending to limit the use of moist pellets.

Other Feed Forms

Rumsey (1978) has described certain advantages that may be attributed to encapsulated diets, *viz.*, nutritional requirements can be met with greater precision, since there would be minimum leaching losses; water quality would be improved, minimizing BOD and other adverse effects on water quality; and shelf-life of diets would be improved and would be consistent in nutrient composition. Flaking of feed is another possibility for the future.

Pigott (1978) has described the advantages for the Dravo processing method in which dry feed forms a ball shape when sprayed with a water mist on an oscillating table. A major advantage is that much of the cost associated with maintenance of the pellet mill is eliminated. The pelleting operation does not involve the heat or pressure associated with conventional methods of pellet production. The retention of nutrients during pelleting is improved, and the final product is soft, yet hard enough to withstand normal handling procedures. The pellets do not sink as rapidly as conventional pellets, and many more sizes of pellets can be manufactured than with the present system.

FEED STORAGE

After pelleting or crumbling, the dry feed should be reduced to a moisture content of no more than 10 percent and cooled to within 2–3°C of ambient temperature before bagging or placing in bulk storage bins. Bags should be of multiwall construction with plastic liners in order to reduce moisture uptake and prevent undue peroxidation of the lipid component. Whether the feed is delivered in bags or in bulk, it should be stored in a cool, dry place. High moisture enhances the destruction of ascorbic acid, in particular, while high moisture coupled with high temperatures increase peroxidation, with consequent destruction of vitamin E and other vitamins. Poor storage conditions also result in the production of off-flavors and off-odors in the feed.

High temperature and high humidity in storage also result in mold growth. Some molds produce mycotoxins, such as aflatoxin, to which fish are much more sensitive than are terrestrial mammals. Propionic acid is sometimes added to diets at the rate of about 0.25 percent to help prevent mold growth. In general, complete fish diets should not be stored for longer than 3 months. This is particularly true for feeds stored in warm, humid periods. Feed storage spaces should be provided with satisfactory protection against rodents, and there should be good air circulation to prevent overheating.

COMPOSITION OF FEEDS

Tables 12 and 13 present the composition of some fish feed ingredients. Nutrient concentrations are organized as follows:

Table 12: Dry matter, digestible and metabolizable energy, protein, ether extract, crude fiber, minerals, and vitamins.
Table 13: Amino acids.

INTERNATIONAL NOMENCLATURE

In Tables 12 and 13 names of the feeds are based on a scheme proposed by Harris *et al.* (1980, 1981). The names are designed to give a qualitative description of each product, where such information is available and pertinent. A complete name consists of as many as six facets, separated by commas and written in linear form. The facets are as follows:

Origin (or parent material) breed or kind
Part eaten
Process(es) and treatment(s) to which the product has been subjected
Stage of maturity
Cutting or crop
Grade or quality designations

Feeds of the same origin (and the same breed or kind, if one of these is stated) are grouped into eight classes on the basis of their composition and the way they are used in formulating diets:

Code
1. Dry forages and roughages
2. Pasture, range plants, and forages fed green
3. Silages
4. Energy feeds
5. Protein supplements
6. Minerals
7. Vitamins
8. Additives

Feeds that in the dry state contain on the average more than 35 percent cell wall or 18 percent of crude fiber are classified as forages or roughages. Feeds that contain 20 percent or more of protein are classified as protein supplements. Products that contain less than 20 percent of protein and less than 35 percent cell wall or 18 percent crude fiber are classified as energy feeds.

INTERNATIONAL FEED NUMBER

Each feed name is assigned a five-digit "International Feed Number (IFN)" for identification. The numbers are assigned consecutively as new feed names are created. These numbers are particularly useful when calculating animal diets for maximum profit. The feed class number is placed in front of the international feed number when making up feed composition tables, and the entire six-digit number is entered in a column following the international feed name (Tables 12 and 13).

The following list shows how three feeds are described:

Facets and Classification of the International Feed Name	Feed No. 1	Feed No. 2	Feed No. 3
Origin (or parent material)	Fish	Soybean	Wheat
Breed or kind	herring		soft white winter
Part eaten	whole	seeds	grain
Process(es) and treatment(s) to which product has been subjected	fresh	meal solv extd	
Grade or quality designations		44% protein	
Classification; first digit in front of the International Feed Number (IFN)	(5) protein supplements	(5) protein supplements	(4) energy feeds
IFN	5-01-999	5-04-604	4-05-337

Thus, the names of the three feeds are written as follows:

No. 1: Fish, herring, whole, fresh
No. 2: Soybean, seeds, meal solv extd, 44% protein
No. 3: Wheat, soft white winter, grain

The international feed names may vary slightly in each report, because changes are made as more is known about a given feed, or the Association of American Feed Control Officials or the Canada Feed Act may change the name or definition of a feed. However, if the feed is the same, the IFN remains the same even though the name changes.

DATA

The data were primarily taken from the International Feedstuffs Institute, Utah State University data bank. The analytical data are expressed in the metric system and are on an as-fed basis. See Table 14 for weight-unit conversion factors and Table 15 for weight equivalents. Analytical data may differ in the various NRC reports because the data are updated for each report. Individual feed samples may vary widely from averages in the table. Variations are influenced by factors such as crop, variety, climate, soil, and length of storage; therefore, the values given should be used with judgment, to be related if possible, to analysis about the feed on hand for critical nutrients.

TABLES

TABLE 1 Estimated Dietary Protein Requirement of Certain Species[a]

Species	Crude Protein Level in Diet for Maximum Growth (g/kg)	References
Rainbow trout (*Salmo gairdneri*)	400–460	Satia (1974) Zeitoun *et al.* (1976) Tiews *et al.* (1976)
Carp (*Cyprinus carpio*)	380	Ogino and Saito (1970)
Catfish (*Ictalurus punctatus*)	320–360	Garling and Wilson (1976)
Chinook salmon (*Oncorhynchus tschawytscha*)	400	DeLong *et al.* (1958)
Eel (*Anguilla japonica*)	445	Nose and Arai (1972)
Plaice (*Pleuronectes platessa*)	500	Cowey *et al.* (1972)
Gilthead bream (*Chrysophrys aurata*)	400	Sabaut and Luquet (1973)
Grass carp (*Ctenopharyngodon idella*)	410–430	Dabrowska (1977)
Brycon sp.	356	Saint-Paul (1977)

[a]Adapted from Cowey (1979) and Mertz (1972).

TABLE 2 Comparative Amino Acid Requirements of Several Species[a]

Amino Acid	Eel Fingerling	Carp Fry	Channel Catfish	Chinook Salmon Fingerling	Chick	Young Pig	Rat
Arginine	3.9(1.7/42)	4.3(1.65/38.5)		6.0(2.4/40)	6.1(1.1/18)	1.5(0.2/13)	1.0(0.2/19)
Histidine	1.9(0.8/42)		1.5(0.37/24)	1.8(0.7/40)	1.7(0.3/18)	1.5(0.2/13)	2.1(0.4/19)
Isoleucine	3.6(1.5/42)	2.6(1.0/38/5)	2.6(0.62/24)	2.2(0.9/41)	4.4(0.8/18)	4.6(0.6/13)	3.9(0.5/13)
Leucine	4.1(1.7/42)	3.9(1.5/38.5)	3.5(0.84/24)	3.9(1.6/41)	6.7(1.2/18)	4.6(0.6/13)	4.5(0.9/19)
Lysine	4.8(2.0/42)		5.1(1.23/24)	5.0(2.0/40)	6.1(1.1/18)	4.7(0.65/13)	5.4(1.0/19)
Methionine[b]	4.5(2.1/42)[c]	3.1(1.2/38.5)	2.3(0.56/24)	4.0(1.6/40)	4.4(0.8/18)	3.0(0.6/20)	3.0(0.6/20)
Phenylalanine[d]			5.0(1.2/24)	5.1(2.1/41)	7.2(1.3/18)	3.6(0.45/13)	5.3(0.9/17)
Theonine	3.6(1.5/42)		2.0(0.53/24)	2.2(0.9/40)	3.3(0.6/18)	3.0(0.4/13)	3.1(0.2/19)
Tryptophan	1.0(0.4/42)		0.5(0.12/24)	0.5(0.2/40)	1.1(0.2/18)	0.8(0.2/25)	1.0(0.2/19)
Valine	3.6(1.5/42)		3.0(0.71/24)	3.2(1.3/40)	4.4(0.8/18)	3.1(0.4/13)	3.1(0.4/13)

[a]Expressed as percent of protein in parentheses, the numerators are requirements as percent of diet and the denominators are percent total protein in the diet. Data for chinook salmon, chick, pig, and rat are cited from Mertz (1972); data for eel and carp are from information of Nose and Arai (1972) and Nose *et al.* (1974); data for catfish from Wilson *et al.* (1978, 1980).
[b]In absence of cystine.
[c]Methionine plus cystine.
[d]In the absence of tyrosine.

TABLE 3 Chemical Name, Family Name, and Omega (ω) Prefix of Four Families of Fatty Acids (Adapted from Cowey and Sargent, 1972)

Chemical Name	Family Name	Chemical Designation[a]
Hexadecenoic acid	Palmitoleic (ω7 series)	16:1ω7
		18:1ω7
Octadecenoic acid	Oleic (ω9 series)	18:1ω9
		20:1ω9
Octadecodienoic acid	Linoleic (ω6 series)	18:2ω6
		18:3ω6
		20:3ω6
		20:4ω6
		22:4ω6
Octadecotrienoic acid	Linolenic (ω3 series)	18:3ω3
		20:5ω3
		22:5ω3
		22:6ω3

[a]x:$y\omega z$, where x indicates the number of carbons in a fatty acid, y indicates the number of double bonds, and ωz indicates the number of carbon atoms from the terminal methyl group to the carbon atom of the first double bond.

TABLE 4 Digestibility Coefficients, Metabolizable Energy Values, and Digestible Energy Values for Rainbow Trout (*Salmo gairdneri*). All Values Are on a Dry Basis.

Feed Name	International Feed Number	Digestion Coefficient					Metabolizable Energy (kcal/kg)	Digestible Energy (kcal/kg)	Method	References
		Protein (%)	Fat (%)	Cho (%)	Energy (%)	Dry Matter				
Alfalfa meal, 17% protein (50% of diet)	1-00-023	61.0	—	—	45.0	—	1,383	1,934	MC[a]	15[d]
Alfalfa meal	1-00-023	—	70.7	—	—	—	—	—	MC	3
Alfalfa meal	1-00-023	22.2	—	—	20.0	—	513	859	MC	14
Animal blood, spray dehy (25%)	5-00-381	69.1	—	—	69.8	—	3,105	2,514	MC	15
(50%)	5-00-381	86.0	—	—	83.5	—	4,004	4,612	MC	15
Animal blood, spray dehy (50%)	5-00-380									
Animal blood, dehy grnd (50%)	5-00-380	32.4	—	—	44.8	—	2,441	2,514	MC	15
Animal blood, ring dehy (50%)	—	89.4	—	—	89.0	—	4,483	5,086	MC	15
(25%)	—	87.0	—	—	76.7	—	3,935	4,383	MC	15
Animal, carcass, dehy grnd	5-00-385	68.8	—	—	71.5	—	3,000	3,379	MC	15
Animal, carcass, dehy grnd	5-00-385	75.3	—	—	71.7	—	2,665	3,051	MC	15
Beef liver, frozen	—	94.3	—	—	—	—	—	—	CO	6
Beef liver	—	88.1	—	—	—	—	—	—	CO	7
Beef spleen, frozen	—	94.0	—	—	—	—	—	—	CO	6
Blood flour, spray dried	5-00-381	65.2	—	—	65.8	—	3,440	3,406	MC	14
Capelin oil	—	—	84.8	—	—	—	—	—	CO[b]	1
Cattle, whey, dehy	4-01-182	79.8	—	—	75.2	—	2,384	2,700	MC	15
whole, sweet	4-01-182	53.0	—	—	79.6	—	2,559	2,936	MC	15
low lactose	4-01-186	63.0	—	—	77.8	—	2,279	2,638	MC	15
Cattle, whey	—	—	65.6	—	—	—	—	—	CO	16
Cattle, whey low lactose, dried	4-01-186	—	91.8	—	—	—	—	—	CO	3
Cattle, whey dried	4-01-182	79.7	—	—	75.5	—	2,530	2,711	MC	14
Cod liver oil	—	—	87.2	—	—	—	—	—	CF	1
Corn, distillers solubles, dehy (25% of diet)	5-02-844	71.9	—	58.6	—	—	2,283	2,436	MC	15
Corn gluten feed	5-02-903	—	89.7	—	—	—	—	—	CO	3
Corn, gluten meal (60%)	5-09-318	—	92.5	—	—	—	—	—	CO	3
Corn, gluten meal (60% protein)										
sample 1 (50%)	5-09-318	82.4	—	—	72.0	—	3,554	4,035	MC	15
sample 2	5-09-318	86.8	—	—	83.3	—	4,149	4,668	MC	15
wet milled	5-02-900	79.8	—	—	70.9	—	3,297	3,712	MC	15
	5-09-318	83.3	—	—	87.3	—	4,435	4,682	MC	15
Casein	—	79.0	—	—	—	—	—	—	D[c]	12
Casein	—	94.3	—	—	—	—	—	—	—	6
Casein: gelatin 1:1	—	95.0	—	—	—	—	—	—	D	13
Casein, gelatin, glucose 3.5, 1.5, 5.0	—	—	—	79.3 (glucose)	—	84.1	—	—	D	13
Casein, gelatin, dextrin 3.5, 1.5, 5.0	—	—	—	77.4 (dextrin)	—	81.6	3,030	—	D	13
Casein, gelatin, raw cornstarch 3.5, 1.5	—	—	—	24.0 (starch)	60.1	—	720	—	D	13
Casein, gelatin, cooked cornstarch 3.5, 1.5	—	—	—	51.6 (starch)	70.1	—	2,140	—	D	13

TABLE 4 *Continued*

Feed Name	International Feed Number	Digestion Coefficient					Metabolizable Energy (kcal/kg)	Digestible Energy (kcal/kg)	Method	References
		Protein (%)	Fat (%)	Cho (%)	Energy (%)	Dry Matter				
Casein, gelatin cellulose flour 3.5, 1.5, 5.0	—	—	—	13.7	36.6	—	280	—	D	13
Casein, potato starch, olive oil										
85:15:00	—	97.0	—	—	—	—	—	—	—	7
76.5:13.5:10	—	97.0	—	—	—	—	—	—	—	7
68.0:12:20	—	96.0	—	—	—	—	—	—	—	7
59.5:10.5:30	—	96.0	—	—	—	—	—	—	—	7
Cotton, seeds, solv extd wo hulls										
sample 1 (50%)	5-07-874	76.1	—	—	64.0	—	2,468	2,660	MC	15
sample 2	5-07-874	78.2	—	—	63.3	—	2,453	2,928	MC	15
sample 3	5-07-874	75.7	—	—	58.2	—	2,255	2,692	MC	15
with some hulls	5-07-872	80.3	—	—	56.4	—	2,272	2,609	MC	15
Cottonseed flour	—	—	—	83.5	67.0	—	2,658	3,113	MC	15
Cottonseed meal no. 1	—	—	—	84.0	—	—	—	—	D	12
Cottonseed meal no. 2	—	—	—	72.7	—	—	—	—	D	12
Cottonseed meal, wheat flour 90:10	—	—	—	75.0	—	—	—	—	CO	4
Crab, whole dehy	—	71.9	—	—	85.1	—	3,214	3,878	MC	15
Dextrin, white	—	—	—	100	—	—	—	—	CO	3
Experimental diets										
Fat 15, protein 40	—	—	70.0	—	—	—	—	—	CO	2
Fat 15, protein 60	—	—	85.0	—	—	—	—	—	CO	2
Fat 25, protein 40	—	—	78.0	—	—	—	—	—	CO	2
Experimental diets										
1. Basal ref. diet, protein 47%, fat 11.8%, crude fiber 3.1%	—	—	89.1	—	—	—	—	—	CO	2
2. Protein 47.7%, fat 10.3%, crude fiber 2.3%	—	—	93.0	—	—	—	—	—	CO	2
3. Protein 45.8%, fat 10.5%, crude fiber 3.1%	—	—	89.8	—	—	—	—	—	CO	2
Experimental diets with:										
1. 10.8% fat	—	—	83.3	—	—	—	—	—	CO	1
2. 13.7% fat	—	—	84.9	—	—	—	—	—	CO	1
3. 16.6% fat	—	—	88.7	—	—	—	—	—	CO	1
4. 20.3% fat	—	—	85.1	—	—	—	—	—	CO	1
Feather meal no. 1	—	52.4	—	—	—	—	—	—	D	12
Feather meal no. 2	—	70.5	—	—	—	—	—	—	D	12
Feather meal, poultry hydroly	5-03-795	—	68.0	—	—	—	—	—	CO	2
Fish meal, anchovy	5-01-985	83.5	—	—	91.3	—	4,328	4,570	MC	14
Fish meal, anchovy	—	67.5 74.6	—	—	—	—	—	—	CO	11
Fish meal, anchovy, wheat flour, dextrin 70:20:10	—	75.0	—	—	—	—	—	—	CO	9
Fish, mech extd grnd										
anchovy	5-01-985	85.2	—	—	91.3	—	4,020	4,570	MC	15
herring	5-02-000	86.7	—	—	91.5	—	4,133	4,717	MC	15
salmon	5-02-012	82.5	—	—	80.0	—	3,570	4,019	MC	15
white	5-02-025	80.5	—	—	84.0	—	2,974	3,490	MC	15
Fish meal, herring	5-02-000	—	97.0	—	—	—	—	—	CO	3
Fish meal, herring	5-02-000	—	93.7	—	—	—	—	—	CO	16

TABLE 4 *Continued*

Feed Name	International Feed Number	Digestion Coefficient					Metabolizable Energy (kcal/kg)	Digestible Energy (kcal/kg)	Method	References
		Protein (%)	Fat (%)	Cho (%)	Energy (%)	Dry Matter				
Fish meal, herring 50%, rough fish 50%	—	—	90.9	—	—	—	—	—	CO	16
Fish meal, flatfish	—	79.0 95.0	—	—	—	—	—	—	CO	10
Fish meal, flatfish	—	92.3	—	—	—	—	—	—	CO	11
Fish meal, herring, starch 70:30	—	73.2	—	—	—	—	—	—	CO	9
Fish meal, herring, wheat flour, dextrin 70:20:10	—	77.0	—	—	—	—	—	—	CO	9
Fish solubles, dehy	—	69.0	—	—	78.7	—	3,345	3,684	MC	15
Flax, seeds, solv extd grnd	5-02-048	76.7	—	—	71.1	—	2,934	3,262	MC	15
Linseed meal	5-02-045	76.8	—	—	71.1	—	3,128	3,268	MC	14
Liver, spleen 1:1	—	86.3 91.7	—	—	—	—	—	—	—	14
Mysis, freeze dried, grnd	—	89.6	—	—	80.3	—	4,265	4,635	MC	15
Meat meal	5-00-388	70.3	—	—	71.7	—	3,237	3,389	MC	14
Meat (sperm whale), wheat flour 90:10	—	76.0	—	—	—	—	—	—	CO	4
Meat (fin whale), wheat flour 90:10	—	65.0	—	—	—	—	—	—	CO	4
Pancreas, glandular meat	—	91.3	—	—	84.8	—	4,218	4,775	MC	15
Poultry, viscera, dehy grnd	5-03-798	70.1	—	—	66.2	—	2,754	3,758	MC	15
Poultry by-prod meal	5-03-798	74.7	—	—	71.5	—	2,982	3,719	MC	14
Pork spleen	—	85.3 91.0	—	—	—	—	—	—	—	14
Pilchard, starch 70:30	—	77.7	—	—	—	—	—	—	CO	9
Pilchard, wheat flour dextrin 70:20:10	—	78.0	—	—	—	—	—	—	CO	9
Pollock, starch	—	79.5	—	—	—	—	—	—	CO	9
Pollock, wheat flour dextrin 70:20:10	—	80.0	—	—	—	—	—	—	CO	9
Rape, seeds, solv extd grnd	5-03-871	76.4	—	—	65.2	—	2,711	2,994	MC	15
Rape, seeds, solv dehul	5-04-612	—	61.4	—	—	—	—	—	CO	3
Redfish	—	62 68	—	—	—	—	—	—	CO	8
Salmon meal	—	80	—	—	—	—	—	—	D	12
Saury, starch	—	75.4	—	—	—	—	—	—	CO	11
Saury, wheat flour dextrin 70:20:10	—	75.0	—	—	—	—	—	—	CO	9
Soybean, CPM, "Jetsploder"										
176°C	—	76.7	—	—	75.8	—	3,926	4,248	MC	15
194°C	—	74.9	—	—	72.3	—	3,722	4,035	MC	15
204°C	—	73.0	—	—	76.1	—	3,963	4,292	MC	15
204°C	—	76.0	—	—	81.4	—	4,220	4,590	MC	15
Soybean, seeds wo hulls, dry roasted, full fat										
127°C, 10 min	—	40.3	—	—	51.3	—	2,564	2,905	MC	15
175°C, 10 min	—	69.9	—	—	71.7	—	3,641	4,083	MC	15
232°C, 8 min	—	77.5	—	—	74.4	—	3,928	4,338	MC	15
232°C, 10 min	—	71.8	—	—	70.0	—	3,761	4,113	MC	15
232°C, 5 min	—	75.2	—	—	75.5	—	3,895	4,282	MC	15
204°C, 12 min	—	73.8	—	—	72.1	—	3,839	4,188	MC	15
204°C, 10 min	—	78.2	—	—	77.3	—	4,033	4,349	MC	15
204°C, 8 min	—	74.1	—	—	70.8	—	3,872	4,121	MC	15

TABLE 4 *Continued*

Feed Name	International Feed Number	Digestion Coefficient					Metabolizable Energy (kcal/kg)	Digestible Energy (kcal/kg)	Method	References
		Protein (%)	Fat (%)	Cho (%)	Energy (%)	Dry Matter				
Starch, raw 5:1	—	—	—	65–70	—	—	—	—	CO	8
Starch, raw	—	—	—	0	—	—	—	—	CO	3
Starch, autocl	4-05-205	—	—	0	—	—	—	—	CO	3
in wheat middlings, raw	4-05-205	—	—	0	—	—	—	—	CO	3
in wheat middlings, autocl	4-05-205	—	—	62	—	—	—	—	CO	3
in wheat middlings, toasted	4-05-205	—	—	27	—	—	—	—	CO	3
in soybean meal (49%)	5-04-612	—	—	54	—	—	—	—	CO	3
in corn gluten-meal	5-09-318	—	—	62	—	—	—	—	CO	3
204°C, 5 min	—	65.6	—	—	80.9	—	3,784	4,593	MC	15
Soybean, seeds, wo hulls, microwave, full fat										
4 min	—	43.0	—	—	55.9	—	2,983	3,243	MC	15
5 min	—	46.0	—	—	71.5	—	3,803	4,131	MC	15
Soybean, seeds wo hulls, solv extd grnd commercially avail.										
sample 1	5-04-612	74.7	—	—	65.7	—	2,566	2,981	MC	15
sample 2	5-04-612	82.2	—	—	68.9	—	2,885	3,260	MC	15
sample 3	5-04-612	84.9	—	—	76.3	—	3,071	3,539	MC	15
sample 4	5-04-612	77.0	—	—	72.7	—	3,267	3,421	MC	15
Soybean, seeds wo hulls, full fat, steam cooked										
1.05 kg/cm^2, 10 min	—	74.7	—	—	68.8	—	3,510	3,917	MC	15
0.70 kg/cm^2, 10 min	—	79.5	—	—	74.7	—	3,929	4,232	MC	15
0.35 kg/cm^2, 5 min	—	37.1	—	—	42.1	—	2,074	2,395	MC	15
0.35 kg/cm^2, 15 min	—	79.5	—	—	63.4	—	3,248	3,565	MC	15
Soybean textured vegetable protein	—	85.3	—	—	84.7	—	3,601	3,942	MC	15
Soybean meal	—	70.5	—	—	—	—	—	—	CO	6
Soybean meal, wheat flour 90:10	—	81 87	—	—	—	—	—	—	CO	4
Soybean oil	—	89.2	—	—	—	—	—	—	CO	1
Sugar										
in raw wheat middlings	4-05-205	—	—	88	—	—	—	—	CO	3
in autocl wheat middlings	4-05-205	—	—	57	—	—	—	—	CO	3
in toasted wheat middlings	4-05-205	—	—	87	—	—	—	—	CO	3
in soybean meal	5-04-612	—	—	100	—	—	—	—	CO	3
in cerelose	—	—	—	100	—	—	—	—	CO	18
Wheat, flour by-prod										
stand middlings	4-05-205	85.0	—	—	39.6	—	1,509	1,804	MC	15
stand middlings (50%)	4-05-205	75.9	—	—	37.1	—	1,247	1,690	MC	15
middlings, finely grnd	4-05-205	67.7	—	—	52.8	—	2,237	2,442	MC	15
wheat middlings	4-05-199	65.3	—	—	39.6	—	1,509	1,804	MC	15
Wheat, germ, grnd	5-05-218	76.8	—	—	60.4	—	2,759	3,034	MC	15
Wheat, hard, clears	—	69.8	—	—	43.4	—	1,587	1,894	MC	15
Wheat middlings	4-05-205	89.1	—	—	—	—	—	—	CO	1
Wheat middlings	4-05-205	—	20.3	—	—	—	—	—	CO	16
Whitefish	5-02-25	80.5	—	—	84.0	—	3,289	3,490	MC	14

TABLE 4 *Continued*

Feed Name	International Feed Number	Digestion Coefficient: Protein (%)	Fat (%)	Cho (%)	Energy (%)	Dry Matter	Metabolizable Energy (kcal/kg)	Digestible Energy (kcal/kg)	Method	References
Whitefish, wheat flour										
90:10	—	88	—	22	—	—	—	—	CO	5
Whitefish	—	80.6	—	—	—	—	—	—	CO	6
Whitefish, potato starch										
90:10	—	81	—	—	—	—	—	—	CO	7
80:20	—	82	—	—	—	—	—	—	CO	7
60:40	—	78	—	—	—	—	—	—	CO	7
40:60	—	74	—	—	—	—	—	—	CO	7
Yeast, brewers, dehy grnd										
sample 1 (25%)	7-05-527	85.0	—	—	82.9	—	2,922	3,787	MC	15
sample 2 (50%)	7-05-527	71.2	—	—	63.1	—	2,465	—	MC	15
sample 3 (25%)	7-05-527	80.3	—	—	72.7	—	3,374	3,321	MC	15
sample 4 (50%)	7-05-527	77.1	—	—	84.2	—	3,881	—	MC	15
Yeast, dehy, grnd torula	7-05-527	82.3	—	—	73.0	—	3,362	3,678	MC	15
Yeast, brewers, dried	7-05-527	60.4	—	—	60.6	—	2,556	2,713	MC	14
Yeast, brewers, dried	—	80.5	—	—	—	—	—	—	D	12

[a] MC = metabolism chamber.
[b] CO = chromic oxide.
[c] D = direct.
[d] For reference 15, if no value is given, ingredient was 100 percent of diet fed.

NOTE: *References*

1. Austreng *et al.*, 1979
2. Cho *et al.*, 1974
3. Cho and Slinger, 1979
4. Inaba *et al.*, 1962
5. Inaba *et al.*, 1963
6. Kitamikado *et al.*, 1964a
7. Kitamikado *et al.*, 1964b
8. Nose, 1960a
9. Nose, 1967
10. Nose and Mamiya, 1963
11. Nose and Toyama, 1966
12. Shanks, 1964
13. Smith, 1971
14. Smith, 1976
15. Smith *et al.*, 1980
16. Windell *et al.*, 1974

TABLE 5 Digestibility of Encapsuled Carbohydrates by Brook Trout (Phillips *et al.*, 1948; Phillips and Brockway, 1956)

Carbohydrate	Absorption (%)
Glucose	99
Maltose	93
Sucrose	73
Lactose	60
Cooked starch (corn)	47
Raw starch (corn)	38

TABLE 6 Digestibility of Carbohydrate in a Dry Diet by Rainbow Trout (Singh and Nose, 1967)

	Carbohydrate Level in Diet (%)				
Carbohydrate	20	30	40	50	60
Glucose	99.3	99.0	99.0	99.6	99.5
Sucrose	99.5	98.8	99.1	99.2	98.8
Lactose	94.4	95.3	97.4	97.2	96.4
Dextrin	77.2	74.8	60.0	50.1	45.5
Potato α-starch	69.2	65.3	52.7	38.2	26.1

TABLE 7 Vitamin Requirements for Coldwater Fishes

Vitamin	Requirement[a] (mg/kg body weight per day)	Recommended (mg/kg dry diet)
Fat-soluble		
A	75 IU	2,500 IU
D	72 IU	2,400 IU
E	1 IU	30 IU
K	0.1	10
Water-soluble		
Ascorbic acid	3–5	100
Thiamin	0.15–0.20	10
Riboflavin	0.5–1.0	20
Pyridoxine	0.2–0.4	10
Pantothenic acid	1	40
Niacin (nicotinic acid)	3–6	150
Biotin	0.05	1
Folacin (folic acid)	0.20	5
B_{12}	0.0006	0.02
Choline	30–50	3,000
Myoinositol (inositol)	18–20	400

[a] Based on young fish.

TABLE 8 Signs of Nutrient Deficiencies

Nutrient	Deficiency	
	Clinical	Subclinical
Fat-soluble vitamins		
A	Impaired growth	Corneal thinning and expansion
	Exophthalmos, eye lens displacement	Degeneration of retina
	Edema, ascites	
	Depigmentation	
D	Poor growth	Impaired calcium homeostasis
	Tetany of white skeletal muscle	
E	Reduced survival and growth	Immature erythrocytes, i.e., poikilocytosis
	Anemia	
	Ascites	Variable sized erythrocytes, i.e., anisocytosis
		Erythrocyte fragility and fragmentation
		Nutritional muscular dystrophy
		Elevated body water, i.e., exudative diathesis
		Lipid peroxidation
K	Prolonged blood clotting	Reduced hematocrit
	Anemia	
Water-soluble vitamins		
Thiamin	Poor growth, anorexia	Low transketolase activity in erythrocytes and kidney
	Hyperirritability	
	Convulsions	
	Loss of equilibrium	
Riboflavin	Impaired growth	Lens cataract
	Anorexia	Adhesion of lens and cornea
		Impaired activity of erythrocyte glutathione reductase
Pyridoxine	Poor growth	Reduced muscle and erythrocyte amino transferases
	Anorexia	
	Epileptiform convulsions	
	Hyperirritability	
	Lowered resistance to handling	
	Erratic, spiral swimming	
	Rapid breathing and gasping	
	Flexing of opercles	
	Rapid onset of rigor mortis	
Pantothenic acid	Poor growth and survival	Atrophied pancreatic acinar cells
	Anorexia	Vacuoles and hyaline bodies in kidney tubules
	Clubbed, exudate-covered gills	
Niacin	Poor growth and feed conversion	Colon lesions
	Anorexia, skin lesions	
	Anemia	
Biotin	Reduced growth and feed conversion	Reduced liver acetyl CoA carboxylase and pyruvate carboxylase
	Increased mortality	Altered fatty acid synthesis
	Degeneration of gill lamellae; skin lesions, i.e., blue slime	Lipid infiltration of liver
		Degeneration of pancreatic acinar cells
		Glycogen storage in kidney tubules
Folic acid	Slow growth	Large, immature, segmented erythrocytes
	Anorexia	
	Poor feed conversion	
	Anemia, pale gills	
B_{12}	Anemia	Small erythrocytes
		Low hemoglobin
Choline	Impaired growth	Fatty livers
	Poor feed conversion	

TABLE 8 *Continued*

Nutrient	Deficiency	
	Clinical	Subclinical
Inositol	Anorexia, reduced growth Anemia	Reduced cholinesterase
C	Anorexia, reduced growth Lordosis, scoliosis Lethargy Hemorrhagic exophthalmia Ascites, anemia Intramuscular hemorrhage	Reduced concentrations of ascorbic acid in liver and anterior kidney Abnormal histology of support cartilage in eye, gill, and fin Reduced serum thyroid hormone (T3) Elevated plasma cholesterol and triglycerides

TABLE 9 Qualitative Requirements (R) and Utilization (U) of Dietary Minerals by Salmonids (Adapted from Ketola, 1977)

Species	Ca	P	Fe	I	Se	Mg	Zn	Ref.
Salmo gairdneri	—	R[a]	—	—	—	R	R	1–3
Salmo salar	—	R[a]	—	—	R[b]	—	—	4–5
Salmo trutta	U	—	—	—	—	—	—	6
Salvelinus fontinalis	U	—	R	R	—	—	—	7–9
Oncorhynchus tshawytscha	—	—	—	U	—	—	—	10

[a] As phosphate. [b] As selenite.

NOTE: *References*

1. Ogino and Takeda, 1978
2. Cowey, 1976
3. Ogino and Yang, 1978
4. Ketola, 1975a
5. Poston *et al.*, 1976
6. Podoliak and Holden, 1965
7. McCay *et al.*, 1936
8. Kawatsu, 1972
9. Marine and Lenhart, 1910
10. Woodall and LaRoche, 1964

TABLE 10 Drugs Registered for Fishery Use (Schnick *et al.*, 1979)

Drug	Fishery Use	Comments
Furanace	Antibacterial drug for myxobacteria 0.05–0.1 ppm (mg/liter) for an indefinite period; 1.0 ppm (mg/liter) for 5–10 min	Nonfood fish use only
Sulfamerazine	Antibacterial against furunculosis 10 g/100 lb (22 g/100 kg) for 14 days in feed	Food fish use in salmonids
Terramycin (oxytetracycline)	Antibacterial against *Aeromonas* and *Pseudomonas* 2.5–3.75 g/100 lb (5.5–8.25 g/100 kg) of fish per day for 10 days in feed	Food fish use

TABLE 11 Fish Feeding Guide[a]

Number Fish per Kilogram	Crumble and Pellet Size	Water Temperature, °C									
		6	7	8	9	10	11	12	13	14	15
		Percent body weight per day									
2,600	#1	2.9	3.4	3.7	3.9	4.6	4.8	5.2	5.8	6.0	6.4
1,300	#1	2.8	3.3	3.6	3.8	4.4	4.7	4.9	5.6	5.9	6.1
700	#2	2.7	3.0	3.3	3.6	4.1	4.5	4.8	5.1	5.6	5.8
400	#2	2.6	2.8	3.0	3.2	3.9	4.0	4.6	4.9	5.0	5.1
200	#3	2.3	2.6	2.8	3.0	3.6	3.8	4.3	4.5	4.6	4.7
130	#3–4	2.1	2.3	2.5	2.8	3.3	3.6	3.7	3.9	4.0	4.1
90	#4	1.9	2.0	2.1	2.4	2.7	2.9	3.0	3.2	3.6	3.8
40	3/32" (2.4 mm)	1.6	1.7	1.8	1.9	2.0	2.1	2.4	2.6	3.0	3.2
30	3/32" (2.4 mm)	1.5	1.6	1.7	1.8	1.8	1.9	2.0	2.2	2.8	2.9
20	1/8" (3.4 mm)	1.3	1.4	1.5	1.6	1.7	1.8	1.9	2.1	2.4	2.5
15	1/8" (3.4 mm)	1.2	1.3	1.4	1.5	1.6	1.7	1.8	2.0	2.3	2.4
10	3/16" (4.8 mm)	1.1	1.2	1.3	1.4	1.5	1.6	1.7	1.8	1.9	2.0
5	3/16" (4.8 mm)	1.0	1.1	1.2	1.3	1.4	1.5	1.6	1.7	1.8	1.9
2	1/4" (6.4 mm)	0.8	0.9	1.0	1.0	1.1	1.1	1.2	1.3	1.5	1.6

[a] Feeding rates based on a single strain of rainbow trout fed dry diets containing about 3,000 kcal digestible energy per kilogram (J. W. Hilton and S. J. Slinger, University of Guelph, personal communication, 1979).

TABLE 12 Composition of Some Common Fish Feeds, Excluding Amino Acids; Data on an As-Fed Basis

Line No.	Feed Name	International Feed Number[a]	Dry Matter (%)	ME (kcal/kg)	Protein (%)	Ether Extract (%)	Crude Fiber (%)	Minerals: Calcium (%)	Minerals: Phosphorus (%)	Minerals: Potassium (%)	Minerals: Chlorine (%)	Minerals: Magnesium (%)
01	Alfalfa, meal dehy, 17% protein	1-00-023	92.	1272.	17.3	2.7	24.0	1.40	.23	2.39	.47	.29
02	Blood, meal spray dehy (Blood flour)	5-00-381	93.	2600.	85.6	1.3	1.0	.48	.24	.09	.25	.22
03	Brewers grains, dehy	5-02-141	92.	2392.	27.1	6.6	13.2	.30	.51	.08	.15	.15
04	Casein, dehy	5-01-162	91.	3913.	84.0	.6	.2	.61	.82	.01	—	.01
05	Chicken, egg white, dehy	5-01-209	—	—	45.0	41.0	—	.21	.68	.49	—	.05
06	Corn, dent yellow, distillers grains with solubles, dehy	5-28-236	92.	—	27.1	9.4	9.1	.14	.65	.40	.17	.16
07	Corn, dent yellow, distillers solubles, dehy	5-28-237	93.	2100.	27.6	8.6	4.6	.33	1.27	1.67	.26	.60
08	Corn, dent yellow, gluten, meal dehy, 41% protein	5-12-354	91.	3000.	42.1	2.3	3.8	.13	.40	.03	.11	.05
09	Corn, dent yellow, gluten, meal dehy, 60% protein	5-28-242	90.	3335.	60.7	2.2	2.0	.07	.48	.19	.09	.08
10	Corn, dent yellow, gluten with bran (Corn gluten feed)	5-28-243	90.	—	23.0	2.2	8.7	.33	.74	.57	.22	.33
11	Corn, dent yellow, grain	4-02-935	89.	1500.	9.6	3.8	2.6	.03	.26	.33	.04	.12
12	Corn, dent yellow, grits by-product (Hominy feed)	4-03-011	90.	—	10.4	6.9	6.0	.05	.52	.59	.05	.24
13	Cotton, seeds, meal mech extd (Whole pressed cottonseed)	5-01-609	93.	—	37.9	5.0	13.3	.20	.90	1.26	.02	.53
14	Cotton, seeds, meal solv extd	5-01-619	92.	2084.	41.8	1.8	10.9	.17	1.17	1.19	.04	.41
15	Crab, process residue, meal (Crab meal)	5-01-663	92.	2957.	32.1	2.0	10.7	14.56	1.59	.45	1.51	.94
16	Fish, solubles, condensed	5-01-969	50.	1673.	32.7	5.6	.5	.22	.59	1.61	2.70	.03
17	Fish, anchovy, meal mech extd	5-01-985	92.	3698.	65.5	4.1	1.0	3.75	2.49	.72	1.00	.25
18	Fish, herring, meal mech extd	5-02-000	92.	3802.	72.0	8.4	.7	2.20	1.68	1.08	.99	.15
19	Fish, menhaden, meal mech extd	5-02-009	92.	—	61.1	9.6	.9	5.18	2.89	.70	.55	.14
20	Fish, salmon, meal mech extd	5-02-012	93.	3320.	61.1	11.4	.3	5.47	3.46	—	—	—
21	Fish, tuna, meal mech extd	5-02-023	93.	—	59.0	6.9	.8	7.86	4.21	.72	1.01	.23
22	Fish, white, meal mech extd	5-02-025	91.	2736.	62.2	4.6	.7	7.31	3.81	.83	.50	.18
23	Gelatin, process residue (Gelatin by-products)	5-14-503	90.	4050.	87.6	.0	.2	.49	.06	—	—	.05
24	Livers, meal	5-00-389	92.	—	66.0	15.7	1.4	.56	1.26	—	—	—
25	Meat, meal rendered	5-00-385	94.	—	51.4	9.1	2.7	8.85	4.44	.57	1.19	.27
26	Meat, with bone, meal rendered	5-00-388	93.	2663.	50.4	9.7	2.2	10.30	5.10	1.33	.74	1.02
27	Milk, skimmed dehy (Cattle)	5-01-175	94.	2913.	33.7	.8	.2	1.28	1.02	1.59	.90	.12
28	Molasses and syrup, beet, sugar, molasses, more than 48% invert sugar more than 79.5 degrees brix	4-00-668	78.	—	6.6	.2	—	.13	.03	4.72	1.28	.23
29	Molasses and syrup, sugarcane, molasses, more than 46% invert sugar more than 79.5 degrees brix (Black strap)	4-04-696	75.	—	4.4	.1	—	.75	.08	2.86	2.31	.32
30	Oats, groats	4-03-331	90.	—	15.8	6.2	2.5	.08	.43	.35	.08	.11
31	Pea, seeds	5-03-600	89.	—	22.5	1.2	6.1	.14	.39	1.01	.06	.13
32	Peanut, kernels, meal mech extd (Peanut meal)	5-03-649	93.	—	48.1	5.8	6.9	.19	.57	1.16	.03	.29
33	Peanut, kernels, meal solv extd (Peanut meal)	5-03-650	92.	—	48.1	1.3	9.9	.26	.62	1.13	.03	.15
34	Poultry, by-product, meal rendered (Viscera with feet with heads)	5-03-798	93.	2561.	58.7	13.1	2.3	3.51	1.83	.39	.54	.18
35	Poultry, feathers, hydrolyzed	5-03-795	93.	2880.	84.9	2.9	1.4	.25	.66	.28	.28	.20
36	Rape, seeds, meal solv extd	5-03-871	91.	2467.	37.0	1.7	12.0	.61	.95	1.24	.10	.55
37	Rice, bran with germs, meal solv extd	4-03-930	91.	—	13.5	1.5	12.9	.11	1.37	1.49	.07	.95
38	Rice, grain, polished and broken (Brewers rice)	4-03-932	89.	—	7.6	.7	.6	.03	.27	.13	.08	.11

	Minerals							Vitamins										
Line No.	Sodium (%)	Sulfur (%)	Copper (mg/kg)	Iron (mg/kg)	Manganese (mg/kg)	Selenium (mg/kg)	Zinc (mg/kg)	Biotin (mg/kg)	Choline (mg/kg)	Folacin (mg/kg)	Niacin (mg/kg)	Pantothenic Acid (mg/kg)	Pyridoxine (mg/kg)	Riboflavin (mg/kg)	Thiamin (mg/kg)	Vitamin B_{12} (μg/kg)	Vitamin E (mg/kg)	Vitamin K (mg/kg)
01	.10	.22	9.75	405.	31.0	.33	19.	.33	1370.	4.37	37.	29.8	7.10	12.9	3.4	.004	111.3	9.0
02	.39	.34	8.20	2784.	6.4	—	306.	.28	600.	.37	22.	3.3	4.45	2.9	.3	12.3	—	—
03	.21	.30	21.17	245.	37.2	.7	27.	.63	1617.	7.12	43.	8.2	.68	1.4	.6	—	26.5	—
04	.01	—	3.80	14.	4.3	.13	27.	.04	208.	.47	1.	2.7	.42	1.5	.4	—	—	—
05	.52	—	—	—	—	—	54.3	—	—	.18	2.46	63.8	—	11.72	3.09	.1	—	—
06	.53	.31	53.19	237.	22.7	.39	80.	.78	2574.	.88	73.	14.0	5.00	9.1	2.9	—	39.9	—
07	.23	.37	82.71	566.	73.9	.33	85.	1.66	4778.	1.29	124.	23.3	8.85	21.1	6.7	2.9	45.8	—
08	.10	.50	28.16	400.	7.3	1.0	20.0	.15	330.	.20	52.	11.3	7.99	2.0	.2	—	42.0	—
09	.06	.65	25.84	282.	6.6	.83	31.	.19	352.	.25	60.	3.5	6.90	2.0	.2	—	23.6	—
10	.94	.21	47.10	424.	23.1	.27	65.	.33	1515.	.27	71.	13.6	13.35	2.2	2.0	—	12.1	—
11	.03	.11	3.53	27.	4.8	.07	13.	.07	502.	.31	25.	5.9	4.69	1.2	3.4	—	22.5	.22
12	.08	.03	13.61	67.	14.5	.10	3.0	.13	1155.	.31	47.	8.2	10.96	2.1	8.1	—	—	—
13	.04	.24	21.3	139.	22.1	.9	57.0	.6	2753.	2.7	38.0	7.7	5.3	3.1	9.7	—	15.0	—
14	.04	.3	18.0	110.	23.	.06	70.	.1	2770.	1.1	45.	12.	7.	5.	3.3	—	15.0	—
15	.88	.25	32.73	4356.	133.0	—	—	.07	2011.	.11	45.	6.5	6.63	6.1	.4	438.3	—	—
16	2.34	.12	46.08	223.	13.3	1.97	44.	.14	3389.	.22	175.	35.5	12.14	12.6	5.0	504.9	—	—
17	.88	.77	9.07	218.	11.0	1.35	105.	.20	3709.	.16	82.	10.0	4.64	7.5	.5	214.2	4.5	—
18	.60	.46	5.96	125.	5.5	1.98	132.	.48	5286.	.34	85.	16.8	4.77	10.1	.4	429.1	22.1	2.2
19	.39	.45	10.68	480.	33.7	2.19	148.	.18	3112.	.15	55.	8.6	4.66	4.8	.6	122.1	12.0	—
20	—	—	11.92	179.	7.9	1.78	—	—	2783.	—	25.	6.9	—	5.8	.9	—	—	—
21	.74	.68	10.31	355.	8.4	4.30	211.	.20	2994.	—	144.	7.7	—	6.8	1.5	300.1	5.6	—
22	.78	.48	5.90	181.	12.4	1.62	90.	.08	3099.	.35	59.	9.9	5.92	9.1	1.7	89.5	8.9	—
23	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
24	—	—	89.28	630.	8.8	—	—	.02	11359.	5.56	205.	29.1	—	36.2	.2	500.9	—	—
25	1.29	.47	9.70	440.	9.6	.44	80.	.12	2041.	.39	56.	6.1	2.75	5.2	.2	67.1	.9	—
26	.72	.25	1.53	684.	13.3	.26	89.	.10	2044.	.37	49.	4.1	8.74	4.5	.2	107.5	.9	—
27	.46	.32	11.66	9.	2.1	.12	38.	.33	1390.	.62	11.	36.3	4.25	19.3	3.7	50.8	9.0	—
28	1.15	.46	16.76	68.	4.5	—	14.	.7	826.	—	41.	4.5	—	2.3	—	—	4.0	—
29	.16	.35	59.17	186.	42.0	—	22.	.69	754.	.11	37.	37.5	4.22	2.8	.9	—	5.4	—
30	.05	.20	6.02	73.	27.8	—	.1	.2	1132.	.51	10.	13.8	1.05	1.2	6.5	—	14.8	—
31	.04	—	—	50.	—	—	29.	.2	589.	.29	32.	18.7	1.47	1.8	4.6	—	3.0	—
32	.21	.27	15.27	156.	25.9	.288	21.	.33	1900.	.66	172.	46.0	7.37	8.1	6.1	—	2.4	—
33	.07	.30	15.25	142.	26.8	—	20.	.33	1948.	.65	173.	46.6	6.38	9.0	5.7	3.0	—	—
34	.82	.52	14.12	442.	11.0	.78	121.	.09	6029.	.51	47.	11.1	4.41	10.5	.2	301.2	2.2	—
35	.69	1.47	6.4	74.	12.5	.82	68.	.04	895.	.22	21.	8.9	2.98	2.0	.1	83.3	—	—
36	.09	1.14	.7	180.	43.	.97	66.	—	6625.	—	147.	8.0	7.24	5.8	1.6	—	19.1	—
37	.10	.18	13.01	187.	232.2	—	30.	.42	1128.	2.21	284.	23.0	29.12	2.9	22.6	—	60.8	—
38	.07	.04	—	—	18.0	.27	17.	.08	877.	.2	23.	3.3	—	.4	1.4	—	14.5	—

TABLE 12 *Continued*

Line No.	Feed Name	International Feed Number[a]	Dry Matter (%)	ME (kcal/kg)	Protein (%)	Ether Extract (%)	Crude Fiber (%)	Minerals: Calcium (%)	Minerals: Phosphorus (%)	Minerals: Potassium (%)	Minerals: Chlorine (%)	Minerals: Magnesium (%)
39	Rice, polishings	4-03-943	90.	—	12.1	12.5	3.2	.05	1.33	1.14	.11	.78
40	Safflower, seeds, meal solv extd	5-04-110	92.	—	23.4	1.4	30.0	.34	.75	.76	.03	.35
41	Sesame, seeds, meal mech extd	5-04-220	93.	—	45.5	6.9	5.7	2.01	1.36	1.25	.07	.46
42	Shrimp, process residue, meal	5-04-226	90.	—	39.9	3.9	14.1	9.73	1.84	.83	1.04	.54
43	Sorghum, milo, grain	4-04-444	89.	—	10.2	2.8	2.2	.03	.28	.34	.08	.16
44	Soybean, protein concentrate, more than 70% protein	5-08-038	92.	—	84.3	.5	.1	.11	.68	.17	.02	.02
45	Soybean, seeds, heat processed 177°C	5-04-597	90.	3722.	38.0	18.0	5.0	.25	.59	1.70	.03	.21
46	Soybean, seeds, meal mech extd	5-04-600	90.	—	42.9	4.8	5.9	.26	.61	1.79	.07	.25
47	Soybean, seeds, meal solv extd	5-04-604	90.	—	44.8	1.2	5.8	.30	.63	1.97	.04	.27
48	Soybean, seeds without hulls, meal solv extd	5-04-612	90.	2610.	49.7	.9	3.4	.26	.63	2.07	.04	.28
49	Sunflower, common, seeds without hulls, meal solv extd	5-04-739	93.	—	46.3	2.9	11.4	.41	.91	1.06	.10	.71
50	Wheat, bran	4-05-190	89.	—	15.2	3.9	10.0	.11	1.22	1.38	.05	.53
51	Wheat, flour by-product, less than 7% fiber (Wheat shorts)	4-05-201	88.	—	16.5	4.6	6.8	.09	.81	.93	.07	.25
52	Wheat, flour by-product, less than 9.5% fiber (Middlings)	4-05-205	89.	1226.	16.4	4.3	7.3	.11	.88	1.00	.04	.36
53	Wheat, gluten	5-05-221	91.	—	79.0	.8	.4	.06	.23	.02	—	.04
54	Wheat, hard red winter, grain	4-05-268	88.	—	12.7	1.6	2.5	.04	.38	.43	.05	.11
55	Wheat, soft red winter, grain	4-05-294	88.	—	11.5	1.6	2.2	.04	.38	.41	.07	.10
56	Whey, dehy (Cattle)	4-01-182	93.	2160.	13.3	.7	.2	.86	.76	1.15	.07	.13
57	Whey, low lactose, dehy (Dried whey product) (Cattle)	4-01-186	93.	2119.	16.7	1.0	.2	1.59	1.05	2.95	1.03	.21
58	Yeast, brewers, dehy	7-05-527	93.	2717	43.8	.8	2.9	.12	1.40	1.67	0.7	.25
59	Yeast, torula, dehy	7-05-534	93.	3093.	49.1	1.6	2.3	.50	1.59	1.90	.02	.17

[a]The first digit is the feed class, coded as follows: (1) forages and roughages; (2) pasture, range plants, and forages fed green; (3) silages; (4) energy feeds; (5) protein supplements; (6) minerals; (7) vitamins; and (8) additives.

	Minerals							Vitamins										
Line No.	Sodium (%)	Sulfur (%)	Copper (mg/kg)	Iron (mg/kg)	Manganese (mg/kg)	Selenium (mg/kg)	Zinc (mg/kg)	Biotin (mg/kg)	Choline (mg/kg)	Folacin (mg/kg)	Niacin (mg/kg)	Pantothenic Acid (mg/kg)	Pyridoxine (mg/kg)	Riboflavin (mg/kg)	Thiamin (mg/kg)	Vitamin B_{12} (μg/kg)	Vitamin E (mg/kg)	Vitamin K (mg/kg)
39	.10	.17	3.32	160.	12.4	—	26.	.62	1249.	.2	506.	46.4	27.89	1.8	20.0	—	90.3	—
40	.05	.13	9.94	495.	18.3	—	41.	1.43	820.	.45	11.	33.9	—	2.3	2.8	—	.9	—
41	.04	.33	—	93.	47.8	—	100.	.34	1535.	—	19.	6.0	12.46	3.4	2.8	—	—	—
42	1.57	—	—	105.	29.8	—	28.	—	5498.	—	—	—	—	4.0	—	—	—	—
43	.02	.11	11.03	50.	17.3	.09	22.	.57	643.	.21	37.	11.1	4.01	1.1	4.2	—	12.2	—
44	.07	.70	14.09	137.	5.5	.14	34.	—	2.	—	5.	3.5	—	.7	.3	—	—	.02
45	.28	.22	15.8	80.	29.8	.11	54.	.29	2420.	3.52	22.	15.6	10.8	2.6	11.0	—	40.0	—
46	.27	.33	21.72	157.	31.3	.10	60.	.33	2623.	6.39	31.	14.3	—	3.4	3.9	—	6.5	—
47	.34	.43	22.80	119.	29.0	.30	43.	.32	2614.	.65	28.	16.3	5.98	2.9	5.6	—	2.5	—
48	.28	.44	20.15	130.	37.2	.10	55.	.32	2753.	.74	22.	14.8	4.93	2.9	3.1	—	2.4	—
49	.22	—	3.50	31.	19.0	—	—	1.45	4120.	—	268.	40.8	13.75	3.9	3.1	—	11.2	—
50	.04	.22	12.66	114.	110.6	.38	114.	.29	1596.	1.42	238.	29.7	8.55	4.1	7.0	—	18.2	—
51	.02	.20	11.63	73.	117.0	.43	110.	.1	1813.	1.66	107.	22.3	7.21	4.2	19.1	—	54.3	—
52	.17	.17	19.32	83.	111.7	.74	103.	.24	1252.	.96	91.	17.3	8.03	2.0	13.8	—	22.7	—
53	.06	.96	11.67	60.	18.3	3.78	39.	.00	581.	.75	75.	5.8	2.28	.7	.9	73.6	34.3	—
54	.02	.13	4.75	31.	29.0	.40	38.	.11	1041.	.39	54.	9.8	3.00	1.4	4.2	—	11.0	—
55	.01	.11	6.12	27.	31.8	.04	42.	.04	929.	.41	52.	9.6	3.21	1.5	4.5	—	15.6	—
56	.65	1.04	46.56	169.	5.9	.08	3.	.35	1793.	.85	11.	46.3	3.35	27.5	4.0	18.9	.2	—
57	1.44	1.07	7.01	245.	8.0	.05	8.	.50	3859.	.74	18.	75.0	4.91	48.6	5.0	35.4	—	—
58	.07	.42	33.01	109.	5.7	.91	38.	1.01	3949.	9.62	450.	110.7	37.14	35.6	92.3	1.1	2.2	—
59	.04	.55	13.44	118.	8.4	1.00	93.	1.37	3005.	24.20	489.	93.8	36.27	44.4	6.2	4.0	—	—

TABLE 13 Amino Acid Composition of Some Common Fish Feeds; Data on an As-Fed Basis

Line No.	Feed Name	International Feed Number[a]	Protein (%)	Arginine (%)	Histidine (%)	Isoleucine (%)	Leucine (%)	Lysine (%)	Methonine (%)	Cystine (%)	Phenylalanine (%)	Tyrosine (%)	Threonine (%)	Tryptophan (%)	Value (%)
01	Alfalfa, meal dehy, 17% protein	1-00-023	17.3	.77	.33	.81	1.28	.85	.27	.29	.80	.54	.71	.34	.88
02	Blood, meal spray dehy (Blood flour)	5-00-381	85.6	3.57	5.14	.90	10.91	7.40	.87	.72	5.85	2.24	3.62	1.04	7.48
03	Brewers grains, dehy	5-02-141	27.1	1.27	.52	1.54	2.49	.88	.46	.35	1.44	1.20	.93	.37	1.61
04	Casein, dehy	5-01-162	84.0	3.49	2.59	5.72	8.80	7.14	2.81	.31	4.81	4.90	3.91	1.08	6.71
05	Chicken, egg white, dehy	5-01-209	45.0	2.93	1.1	2.86	4.02	3.09	1.48	1.09	2.58	1.91	2.25	.73	3.3
06	Corn, dent yellow, distillers grains with solubles, dehy	5-28-236	27.1	.96	.64	1.39	2.23	.70	.50	.29	1.51	.70	.93	.17	1.50
07	Corn, dent yellow, distillers solubles, dehy	5-28-237	27.6	.97	.68	1.33	2.36	.91	.56	.45	1.49	.87	1.02	.24	1.55
08	Corn, dent yellow, gluten, meal dehy, 41% protein	5-12-354	42.1	1.38	.96	2.2	5.99	.78	1.06	.68	2.30	1.0	1.40	.21	1.80
09	Corn, dent yellow, gluten, meal dehy, 60% protein	5-28-242	60.7	2.08	1.40	2.54	10.23	1.01	1.78	.99	4.02	3.19	2.22	.30	3.09
10	Corn, dent yellow, gluten with bran (Corn gluten feed)	5-28-243	23.0	.78	.61	.88	2.20	.64	.37	.44	.81	.72	.78	.15	1.10
11	Corn, dent yellow, grain	4-02-935	9.6	.43	.26	.35	1.21	.25	.17	.22	.48	.38	.35	.08	.44
12	Corn, dent yellow, grits by-product (Hominy feed)	4-03-011	10.4	.47	.19	.39	.85	.38	.16	.15	.33	.50	.39	.11	.49
13	Cotton, seeds, meal mech extd (Whole pressed cottonseed)	5-01-609	37.9	4.48	.91	1.65	2.53	1.91	.73	.64	2.50	1.35	1.57	.66	2.08
14	Cotton, seeds, meal solv extd	5-01-619	41.8	4.28	1.01	1.23	2.22	1.71	.50	.57	2.05	1.04	1.24	.48	1.68
15	Crab, process residue, meal (Crab meal)	5-01-663	32.1	1.66	.49	1.17	1.54	1.38	.53	.24	1.16	1.17	1.00	.29	1.47
16	Fish, solubles, condensed	5-01-969	32.7	1.63	1.43	1.03	1.86	1.86	.71	.27	1.02	.44	.87	.34	1.22
17	Fish, anchovy, meal mech extd	5-01-985	65.5	3.77	1.61	3.10	4.99	5.04	1.99	.60	2.78	2.24	2.76	.75	3.50
18	Fish, herring, meal mech extd	5-02-000	72.0	4.62	1.65	3.13	5.19	5.36	2.08	.74	2.71	2.20	2.90	.77	4.30
19	Fish, menhaden, meal mech extd	5-02-009	61.1	3.75	1.45	2.88	4.48	4.72	1.75	.56	2.46	1.94	2.50	.65	3.22
20	Fish, salmon, meal mech extd	5-02-012	61.1	5.20	—	—	—	7.60	1.60	.70	—	—	—	.50	—
21	Fish, tuna, meal mech extd	5-02-023	59.0	3.43	1.75	2.45	3.79	4.22	1.47	.47	2.15	1.69	2.31	.57	2.77
22	Fish, white, meal mech extd	5-02-025	62.2	4.02	1.34	2.72	4.36	4.53	1.68	.75	2.28	1.83	2.57	.67	3.02
23	Gelatin, process residue (Gelatin by-products)	5-14-503	87.6	6.97	.76	1.38	2.91	3.55	.73	.13	1.79	.52	1.76	.05	2.09
24	Livers, meal	5-00-389	66.0	4.04	1.48	3.10	5.31	5.21	1.22	.94	2.92	1.70	2.49	.69	4.15
25	Meat, meal rendered	5-00-385	51.4	3.60	.96	1.75	3.19	3.23	.70	.65	1.81	.96	1.64	.34	2.52
26	Meat, with bone, meal rendered	5-00-388	50.4	3.49	.96	1.64	3.06	2.90	.65	.50	1.70	.79	1.65	.30	2.45
27	Milk, skimmed dehy (Cattle)	5-01-175	33.7	1.15	.86	2.18	3.32	2.53	.90	.45	1.56	1.14	1.56	.43	2.28
28	Molasses and syrup, beet, sugar, molasses, more than 48% invert sugar more than 79.5 degrees brix	4-00-668	6.6	—	—	—	—	—	—	—	—	—	—		—

29	Molasses and syrup, sugarcane, molasses, more than 46% invert sugar more than 79.5 degrees brix (Black strap)	4-04-696	4.4	—	—	—	—	—	—	—	—	—	—	—
30	Oats, groats	4-03-331	15.8	.86	.25	.55	1.04	.53	.20	.20	.67	.57	.45	.19	.76
31	Pea, seeds	5-03-600	22.5	1.39	.65	1.14	1.78	1.54	.28	.19	1.25	—	.93	.22	1.25
32	Peanut, kernels, meal mech extd (Peanut meal)	5-03-649	48.1	5.06	1.08	1.69	3.02	1.50	.49	.75	2.34	1.66	1.24	.47	2.08
33	Peanut, kernels, meal solv extd (Peanut meal)	5-03-650	48.1	4.55	.95	1.76	2.70	1.77	.42	.73	2.04	1.51	1.16	.48	1.88
34	Poultry, by-product, meal rendered (Viscera with feet with heads)	5-03-798	58.7	3.77	1.01	2.38	4.00	2.89	1.06	.92	1.84	.94	1.94	.46	2.86
35	Poultry, feathers, hydrolyzed	5-03-795	84.9	7.05	.99	4.06	6.94	2.32	.55	3.24	3.05	2.32	3.97	.52	6.48
36	Rape, seeds, meal solv extd	5-03-871	37.0	2.06	.99	1.35	2.50	1.98	.71	.30	1.41	.79	1.56	.43	1.79
37	Rice, bran with germs, meal solv extd	4-03-930	13.5	.85	.29	.45	.81	.54	.21	.07	.48	.72	.45	.21	.65
38	Rice, grain, polished and broken (Brewers rice)	4-03-932	7.6	.49	.18	.33	.68	.27	.12	.08	.39	.41	.24	.10	.47
39	Rice, polishings	4-03-943	12.1	.51	.17	.35	.70	.52	.20	.13	.38	.42	.34	.10	.72
40	Safflower, seeds, meal solv extd	5-04-110	23.4	1.95	—	.28	—	.72	.34	.36	—	—	.51	.27	—
41	Sesame, seeds, meal mech extd	5-04-220	45.5	4.55	1.07	1.96	3.20	1.27	1.37	.59	2.14	1.87	1.60	.71	2.33
42	Shrimp, process residue, meal	5-04-226	39.9	2.52	.96	1.68	2.68	2.17	.82	.59	1.59	1.33	1.42	.36	1.83
43	Sorghum, milo, grain	4-04-444	10.2	.37	.22	.49	.1.39	.22	.15	.16	.48	.43	.34	.10	.52
44	Soybean, protein concentrate, more than 70% protein	5-08-038	84.3	7.34	2.41	4.60	6.33	5.61	.88	.92	4.33	3.10	3.34	.88	4.38
45	Soybean, seeds, heat processed 177°C	5-04-597	38.0	2.80	1.01	2.18	2.8	2.40	.54	.55	2.10	1.2	1.69	.52	2.02
46	Soybean, seeds, meal mech extd	5-04-600	42.9	3.07	1.14	2.63	3.62	2.79	.65	.56	2.20	1.55	1.72	.61	2.28
47	Soybean, seeds, meal solv extd	5-04-604	44.8	3.03	1.07	2.03	3.27	2.68	.52	.75	2.11	1.33	1.66	.64	2.02
48	Soybean, seeds without hulls, meal solv extd	5-04-612	49.7	3.67	1.22	2.46	3.73	3.17	.71	.75	2.44	1.68	1.94	.69	2.55
49	Sunflower, common, seeds without hulls, meal solv extd	5-04-739	46.3	4.42	1.23	2.25	3.83	1.92	1.16	.74	2.36	1.39	1.93	.61	2.60
50	Wheat, bran	4-05-190	15.2	.96	.39	.51	.92	.58	.19	.32	.55	.42	.46	.25	.69
51	Wheat, flour by-product, less than 7% fiber (Wheat shorts)	4-05-201	16.5	1.18	.45	.58	1.09	.79	.27	.36	.67	.47	.60	.21	.83
52	Wheat, flour by-product, less than 9.5% fiber (Middlings)	4-05-205	16.4	.92	.38	.67	1.08	.67	.18	.22	.64	.40	.54	.20	.75
53	Wheat, gluten	5-05-221	79.0	3.00	1.65	3.41	5.58	1.55	1.24	1.76	4.24	2.38	2.17	.72	3.93
54	Wheat, hard red winter, grain	4-05-268	12.7	.64	.30	.51	.89	.36	.21	.32	.63	.43	.37	.17	.59
55	Wheat, soft red winter, grain	4-05-294	11.5	.65	.32	.45	.90	.36	.22	.36	.64	.38	.39	.27	.58
56	Whey, dehy (Cattle)	4-01-182	13.3	.34	.17	.79	1.18	.94	.19	.30	.35	.25	.90	.18	.68
57	Whey, low lactose, dehy (Dried whey product) (Cattle)	4-01-186	16.7	.60	.27	.96	1.54	1.40	.41	.43	.55	.46	.95	.27	.87
58	Yeast, brewers, dehy	7-05-527	43.8	2.20	1.09	2.21	3.23	3.11	.74	.49	1.83	1.50	2.12	.52	2.36
59	Yeast, torula, dehy	7-05-534	49.1	2.64	1.32	2.85	3.52	3.74	.77	.61	2.85	2.00	2.64	.52	2.96

[a]The first digit is the feed class, coded as follows: (1) forages and roughages; (2) pasture, range plants, and forages fed green; (3) silages; (4) energy feeds; (5) protein supplements; (6) minerals; (7) vitamins; and (8) additives.

TABLE 14 Weight-Unit Conversion Factors

Units Given	Units Wanted	For Conversion Multiply by
lb	g	453.6
lb	kg	0.4536
oz	g	28.35
kg	lb	2.2046
kg	mg	1,000,000
kg	g	1,000
g	mg	1,000
g	μg	1,000,000
mg	μg	1,000
mg/g	mg/lb	453.6
mg/kg	mg/lb	0.4536
μg/kg	μg/lb	0.4536
Mcal	kcal	1,000
kcal/kg	kcal/lb	0.4536
kcal/lb	kcal/kg	2.2046
ppm	μg/g	1
ppm	mg/kg	1
ppm	mg/lb	0.4536
mg/kg	%	0.0001
ppm	%	0.0001
mg/g	%	0.1
g/kg	%	0.1

TABLE 15 Weight Equivalents

1 lb = 453.6 g = 0.4536 kg = 16 oz
1 oz = 28.35 g
1 kg = 1,000 g = 2.2046 lb
1 g = 1,000 mg
1 mg = 1,000 μg = 0.001 g
1 μg = 0.001 mg = 0.000001 g
1 μg per g or 1 mg per kg is the same as ppm

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