# Ingredients, Formulation, and Processing

The practical application of fish nutrition is to produce feeds that support growth, health, and welfare of farmed aquatic animals. This objective is achieved by selecting appropriate feed ingredients, deciding how they should be combined to meet the nutritional requirements of farmed aquatic animals, and processing the combination or mixture of ingredients into a physical form suitable for practical use. Each step in the process of making fish and shrimp feeds requires specific information, judgment, and compromise. Complete information must be available for feed ingredients being considered as components of feeds, including proximate composition, nutrient content, quality and potential variability among sources or producers, antinutrient content, contaminant level, and the digestibility of nutrients to farmed fish or shrimp. The nutritional requirements of the fish or shrimp at the life stage for which the feed is being prepared must be known. Potential interactions among feed ingredients that might influence the bioavailability of essential nutrients must be considered, as well as the physical characteristics of a feed ingredient mixture that might affect how it can be processed into pellets. Processing feed ingredient mixtures into pellets also requires careful consideration. Physical characteristics of pellets, such as hardness and durability, water stability, and porosity, are determined by the blend of ingredients used in a feed mixture and by the processing techniques used during conditioning of the ingredient mixture and pelleting. Further, processing techniques increase the availability of some nutrients in feeds but decrease the availability of others. The specific application in which a feed pellet will be used also affects the choice of processing method, the ingredients used in the feed mixture, and the way in which pelleting equipment is operated. For example, the buoyancy of feed pellets can be varied to produce pellets that float or sink. Overall, therefore, feed manufacture is a complicated undertaking that cannot be reduced to a series of steps that can then be applied to multiple situations. In nearly all cases of feed manufacturing, judgment and compromise are made. However, several straightforward considerations can serve

as a guide to feed manufacturing that illustrate both the complexity of manufacturing feed and the relatively simple principles upon which feed production is based. Readers seeking more information on fish feed manufacturing are encouraged to consult Hardy and Barrows (2002).

#### FEED INGREDIENTS

Feed ingredients are selected and combined to supply energy and essential nutrients such as amino acids, vitamins, minerals, and essential fatty acids to support fish growth, health, and reproduction, as well as product quality. Ingredients are also selected and combined based on how they affect the physical characteristics of pellets. Ingredient cost, palatability, physical characteristics, and availability also factor into ingredient selection for feeds. Many feed ingredients are byproducts of human food production, such as soybean meal and corn gluten meal, both byproducts of cooking oil production. Fish meal, in contrast, is now produced exclusively for use in animal feeds. Most ingredients used in fish feeds are also used in livestock or poultry feeds, but some are used exclusively in fish feeds, such as squid liver meal.

Ideally, each ingredient selected as a component of feeds has a specific role to play in the feed. Protein supplements, defined as protein-rich (> 35% crude protein) ingredients, are selected to supply protein and/or specific amino acids to a blended protein mixture. Oil sources are selected to supply dietary energy, essential fatty acids, and, in feeds for crustaceans, sterols. Starch sources, such as ground wheat or corn (maize) starch, are added to provide dietary energy and to act as nutritional binders, plus to allow extruded pellets to expand during starch gelatinization. However, all ingredients are complex mixtures of nutrients, nonnutrients, bioactive compounds, and, in some cases, compounds that interfere with digestion or metabolism, e.g., antinutrients. Antinutrients are mainly found in plant products. Given their complexity, feed ingredients possess both benefits and risks to fish and animals. Risks can be minimized by processing

or supplementation; however, to produce high-quality, nutritious feeds, producers must understand both the benefits and risks of ingredients. For more information on antinutrients in feed ingredients, see Chapter 11, as well as Francis et al. (2001), Gatlin et al. (2007), and Krogdahl (2010).

Feed ingredients are sourced from marine resources (fish meal, fish oil, hydrolysates), plant seeds (grains, oilseeds, pulses, and others), rendered animal proteins (poultry byproduct meals, meat and bone meal, blood meal, and others), seafood processing byproducts, and single-cell organisms. Each feed ingredient has a specific definition that includes a description of its source, how it is processed, stage of maturity in the case of forages, and nutritional composition. Information on specific feed ingredients and how they are regulated can be obtained from the Association of American Feed Control Officials (AAFCO, www.aafco.org) or in AAF-CO's official publication (AAFCO, 2010). Other sources of information on feed ingredient definitions and composition include Hertrampf and Piedad-Pascual (2000) and the Atlas of Nutritional Data on United States and Canadian Feeds (NRC, 1972). All sources have advantages and disadvantages as ingredients in feeds. Advantages are generally associated with nutrient content, while disadvantages include antinutrient contents, presence of contaminants, propensity for becoming moldy and developing aflatoxins, poor or variable quality, potential for oxidation, sustainability or cost.

Premixes are used to supply vitamins and minerals to feeds; these concentrated mixes are added in small amounts to provide appropriate levels of essential vitamins and minerals. Generally, vitamin premixes are added to meet all essential vitamin requirements in feeds used in intensive aquaculture systems where natural food is not available. The potential contribution of vitamins present in feed ingredients is normally not considered. In semiintensive aquaculture systems, vitamin premixes are not expected to supply all essential vitamin requirements.

Additives are supplied to feeds in small amounts to increase digestion of specific feed components, impart color, alter physiology of fish, supply essential nutrients, increase feed intake, and prevent microbial spoilage during storage of feed. Example of additives used to increase feed component digestibility are enzymes, e.g., phytase and amylase. Carotenoid pigments are supplied to feeds to impart color to skin, muscle, or eggs. Examples include astaxanthin, Haematococcus pluvialis (an algal species), krill, and crustacean processing byproducts. Supplements added to alter physiological functions include immunostimulants, prebiotics, probiotics, and products designed to enhance smolting and enhance the success of seawater transfer in salmon. Feed-grade amino acids are added to feeds to balance levels of lysine, methionine, or threonine. Feed intake can be improved in some feeds by adding palatability-enhancing materials, e.g, fish hydrolysate fractions, betaine, or mixtures of phosphorylated nucleotides. Feed additives are covered in more detail in Chapter 10.

## **FEED FORMULATION**

Feed formulation is the process of combining feed ingredients to meet the nutritional needs of farmed animals, birds. or fish to produce a mixture that can be pelleted, shipped, and stored; is relatively water-stable; supports growth, health, and wellness; and is economical to use. Formulation begins with establishing target levels for protein, energy, and essential nutrients in the feed. These levels are based upon the established or estimated requirements of the target species. The second step is to select ingredients that are appropriate sources of nutrients and appropriate choices based on the criteria discussed above. The nutrient content in each ingredient being considered must be known, and, if available, the apparent digestibility of nutrients in each ingredient. Most feed formulation is done using least-cost computer programs that calculate the best and most economical combination of ingredients to meet the dietary levels of nutrients specified for a feed. The cost of each ingredient must be known to produce a least-cost feed. Least-cost formulations are somewhat a misnomer in that they are the lowest cost formulation within the constraints imposed on the formulation. For example, a relatively expensive feed ingredient may be required at a low percentage to ensure optimum fish growth. In this case, a minimum level of this ingredient is established, even though removing this minimum constraint would lower the cost of the feed. Feed formulation using least-cost formulation programs is increasingly referred to as "precision formulation" to address this point.

Feed formulation programs are useful, time-saving tools, but they are also notorious for computing extremely efficient blends of ingredients that would fail if made because they cannot be pelleted or sustain rapid fish growth and health. To avoid problems generated by least-cost formulation programs, upper and lower limits are placed on feed ingredients to ensure that the resulting mixture is practical. For example, fish and/or plant oils are excellent sources of dietary energy. Although there are limits to the amount of oils that can be added to feeds, least-cost formulations do not automatically take this limitation into consideration. Likewise, starch is needed to make durable, water-stable pellets, so there has to be a minimum amount of starch in a feed mixture. As mentioned above, for some farmed fish species, a minimum percentage of fish meal has to be included in the feed; without it, growth rates are reduced. In the case of antinutrients in some feed ingredients, allowances must be made to ensure that levels are below the threshold known to reduce fish growth or affect health. Experienced feed formulators are able to set upper and lower limits for ingredients and for dietary nutrient levels in the final feed, and also to make allowances for partial losses of essential nutrients, such as vitamins, that occur during feed pelleting and storage. Another potential pitfall associated with the use of feed formulation programs is failure to take into account changes in dietary energy, protein, or digestibility values when using requirement levels expressed as mg (or g) per kg diet. Experienced

operators make adjustments in nutrient requirement levels, especially essential amino acids and phosphorus, to account for differences in dietary energy levels in feed formulations. Establishing minimum levels in feed formulation programs of essential nutrients on a mg or g/kg diet basis can lead to deficiencies in the final formulation unless such adjustments are made.

Least-cost formulation is based on two assumptions that those operating the computer programs often do not realize. First, there is no "ideal" formulation. Many possible formulations can meet the specifications established for nutrient content of the ingredient blend. The key to making a useful ingredient blend are the upper and lower limits placed on ingredients and on nutrient levels. Second, feed formulation programs "assume" that nutrients in various feed ingredients are equal in value, unless otherwise specified. In other words, total lysine or phosphorus in fish meal is equal to lysine or phosphorus in soybean meal unless the apparent digestibility or bioavailability of the nutrient is accounted for. It is wise to remember these assumptions when using least-cost feed formulation programs.

### FEED MANUFACTURING

Feed manufacturing is the physical process of forming feed ingredient mixtures into particles used to feed fish or shrimp. Another term for feed manufacturing is pelleting, but this term excludes other types of feeds produced for larval fish, such as flaked feeds, microbound feeds, microextruded marumerized (MEM) pellets, feeds produced by particleassisted rotational agglomerization (PARA), spray beadlets, microencapsulated feeds, and complex feeds, which are feed particles containing smaller particles in which various nutrients are enclosed (Hardy and Barrows, 2002). Regardless of the technology used or the kind of feed produced, the aim of the manufacturing process is to use physical and mechanical forces to make particles that are practical to ship, store, and use, plus are acceptable to fish. For the most part, commercial (other than larval) feeds are manufactured as pellets by cooking extrusion, compression pelleting, or cold extrusion.

Ingredient mixtures, regardless of the type of pelleting, undergo a series of steps in the process of feed manufacturing that include grinding, mixing, conditioning, pelleting, cooling and drying, top-dressing, packaging (sacking), storing, and shipping. Briefly, grinding is done to reduce particle sizes of ingredients or mixtures prior to pelleting to increase surface area of ingredient particles, and to reduce differences in the average size of particles from different feed ingredients. If the average particle size of feed ingredients is dissimilar, ingredients can segregate during mixing and affect the homogeneity of a mixture. Grinding is also important to reduce particle size to facilitate conditioning and pelleting, and to ensure that particles are small enough to ensure that individual feed pellets contain all nutrients. Mixtures for fish feeds are ground to smaller particle sizes than is neces-

sary for livestock or poultry feeds. For most fish feeds, the mixture is ground to pass through a 200 µm screen. Because some ingredients used in fish feeds containing relatively high lipid contents, grinding to this small screen size can cause the screens to become blocked. To minimize this, grinders that use high air flow to force particles through screens, called windswept pulverizers, are often used in commercial feed production. Ball mills are another approach to reduce particle size of ingredients containing > 5% lipid, such as fish meal. Feed producers can grind individual ingredients and/or feed mixtures. Concerning mixing, the goal is to produce a homogenous blend of dry ingredients. Mixing is an art in itself in that overmixing (mixing for too long a period) can be just as deleterious as undermixing because particles begin to segregate based on material density. Appropriate mixing times can be determined by adding iron filings to the mix, taking samples at intervals and determining the weight of iron filings in samples removed from the mixer using a magnet to remove the filings. Mixers can be batch mixers or continuous mixers; most fish feed producers prefer batch systems.

After mixing, feed mixtures are conditioned in a chamber to which steam and physical agitation are introduced. Conditioning prepares a feed mixture for pelleting, increasing the moisture content, heating the mixture, and adding energy to activate gluten proteins. This process occurs at ambient pressure. After 30 seconds to 5 minutes of conditioning, depending on the feed formulation, type of pelleting and rate of steam addition, the mixture enters either a compression pelleting chamber or an extrusion barrel to form the mixture into pellets. Compression pelleting involves the use of a static roller to force a feed mixture that has been exposed to dry steam for 10-30 seconds through tapered holes in a rotating metal die that resembles a doughnut or a tire. Heat, moisture, and pressure compress the mixture into dense threads that are cut off by a stationary knife as they emerge from the outside surface of the rotating die. Pellet diameter is determined by the diameter of holes in the die, and pellet length is determined by the adjustment of the knife. Compression pellets are often referred to as sinking pellets because of their high density compared to extruded pellets. A variation of the usual compression pelleting system is to install an expander between the conditioner and compression pelleter. The expander is simply a cone through which the feed mixture is forced, creating pressure that gelatinizes starch before pelleting to increase starch digestibility. After pellets exit the pelleting die, they move along a belt through a drying chamber where forced air removes moisture and cools the hot pellets. No added heat is required to reduce the moisture level to < 12%, the target moisture level to prevent mold growth.

Cooking-extrusion pelleting differs from compression pelleting in that more conditioning (added steam and agitation), higher moisture in the feed mixture (~ 20–23%), and much more force and pressure along the course of the extruder barrel are involved. The extruder barrel is pressurized

and additional steam is added to the mixture as it moves through the barrel. Inside the barrel is a screw on which the flights become closer, resulting in increased pressure and force. This keeps the added moisture in a liquid state. As a result, starch gelatinization is more complete. When the feed mixture passes through a die shaped like a plate with holes at the end of the barrel, pressure drops to ambient external air pressure and entrapped moisture instantly converts to a gas (steam), creating micropockets within pellet strands that are then cut to length with a rotating knife. Pellet expansion from micropockets reduces bulk density of the pellets. Pellet density can be varied by adjusting conditions (pressure, moisture, heat) in the barrel of the extruder to produce floating, neutrally buoyant, or slowly sinking pellets. Pellets are then conveyed through a forced air, heated dryer to reduce moisture to < 12%.

Both compressed pellets and extrusion pellets can be coated with liquid additives, a process called top-dressing. Typically, fish or plant oil blends are added to pellets after pelleting; adding too much to the mixture before pelleting interferes with pellet compression. Other liquid additives, such as probiotics or enzymes such as phytase, can be added by top-dressing. Extruded pellets can soak up higher amounts of oil than compressed pellets due to differences in density. Vacuum chambers are used to produce high-fat extruded salmon feeds. These chambers are operated on batches by lowering the pressure, then adding the fat, which replaces air in the pellets, soaking fat throughout the pellet rather than adding to the surface, as is the case with compressed pellets.

Cold extrusion simply refers to extrusion without addition of heat or steam. For cold extrusion to produce stable pellets, binders must be included in the feed mixture, and the mixture must contain 28–32% moisture. Cold-extruded pellets should be used within a short period unless they are dried or frozen. The first commercial salmon feeds, such as the Oregon moist pellet, were produced by cold extrusion. Marumerized pellets are also produced by cold extrusion (see below).

Larval feed manufacturing presents several challenges. First, feed mixtures must be very finely ground because the size of feed particles is very small. Second, particles must be water-stable and neutrally buoyant. Third, the techniques used to make water-stable particles must not reduce nutrient digestibility. Fourth, particles must not foul rearing water. Finally, particles must be recognized, palatable, and accepted by fish as food.

Larval feeds can be categorized as microbound, on-sized, microencapsulated, and complex feeds. Microbound feeds are those that use special combinations of feed ingredients to produce extruded or compressed pellets that are then crumbled and sized by screening to yield particles within specific size ranges. Flaked feeds can also be broken up and sized to various small particle sizes. Such particles are commonly used as starter feeds for salmonids and other farmed species not having a larval stage. For larval fish, microbound, crumbled feeds are sometimes effective, but

there are limitations to their effectiveness. On-sized feeds, in contrast, are micropellets produced by techniques used to make small pharmaceutical products. They are produced using MEM or PARA processes. Microencapsulated feeds are produced by coating a feed mixture containing nutrients with an impermeable coating or a coating designed to dissolve (controlled release). Examples include cross-linked proteins and lipid-walled microcapsules, the latter often referred to as a "complex feed." These are micropellets produced by MEM or PARA processes that contain even smaller microcapsules produced by other methods distributed within the micropelleted material. The advantage of complex-feed particles is that the interior particle components can be made to have different properties than the main body of the particle. For example, small vehicles can be made to deliver watersoluble nutrients that would otherwise leach from particles, or designed to release nutrients in the intestine after passing through the stomach.

# FEED QUALITY ASSESSMENT

Feed production is an accurate but not always exact process. Ingredients vary in composition and quality from batch to batch. Levels of some essential nutrients are reduced during pelleting, drying, and storage. To account for this, feed manufacturers always formulate feeds to supply a slight excess of protein, lipid and other essential nutrients to ensure that sufficient levels of essential nutrients are present when the feed is used. Feed manufacturers also retain samples of feeds for quality testing and in case any dispute about feed quality arises after the feed is used by fish farmers. Feeds are routinely tested for proximate composition and the status of lipid oxidation. Shrimp feeds are also routinely tested for water stability. Abusive storage conditions, such as exposure to moisture or excessive heat, or prolonged storage beyond the manufacturers' recommended shelf life of feeds are the primary causes of feed quality problems at the farm. Feeds showing any signs of mold upon visual inspection or oxidation detected by heat production of feed in bags or by smell should be discarded to prevent fish health problems.

Feed ingredient adulteration, substitution, or mislabeling, either by accident or deliberately, is a concern to feed manufacturers. Adulteration refers to the addition of material to an ingredient to increase its economic value without increasing its nutritional value. An example is addition of melamine to increase the apparent protein content of an ingredient. Melamine contains 66.64% nitrogen on a molecular weight basis. Hence, adding melamine to a feed ingredient increases its nitrogen content, leading to an inflated protein content when Kjeldahl nitrogen is used to analyze protein content (N × 6.25). Suspected adulteration with melamine is easily detected if an amino acid analysis is conducted because the sum of amino acids will not match total protein in the ingredient when analyzed by Kjeldahl nitrogen. Other kinds of adulteration, such as adding soybean hulls to soybean

meal, can easily be detected by ingredient protein content. If analysis results of a common ingredient are widely different from tabled values, adulteration, substitution, or mislabeling is a possibility. Another useful technique to check ingredient quality is feed microscopy. Although training and experience is necessary to use feed microscopy effectively, it remains a powerful tool. Further details are available in the Manual of Feed Microscopy and Quality Control, Third edition (Khajarern and Khajarern, 1999).

#### **ENVIRONMENTAL AND SUSTAINABILITY CONCERNS**

Environmental and sustainability concerns have had a large impact on fish feeds over the past decade. Excess phosphorus in feeds, for example, leads to excessive excretion of phosphorus, thereby contributing to eutrophication of rivers and lakes receiving fish farm effluent water. Regulations limiting phosphorus levels in fish farm effluent water have led to more sophisticated feed formulation to match available phosphorus levels in feeds with dietary requirements of fish and to limit levels of unavailable phosphorus in feeds. In contrast, regulating phosphorus levels in feeds to achieve reductions in phosphorus levels in farm effluent water has limited innovative solutions associated with changes in feed formulation and phase-feeding strategies. Protein is the most expensive component of feeds, and protein metabolized for energy results in low retention of dietary protein as tissue protein and excessive nitrogen excretion into the aquatic environment. Feeds are increasingly being formulated to supply dietary energy needs of fish with nonprotein sources, resulting in higher protein retention and lower nitrogen losses. Fish farmers have an economic interest in minimizing nutrient losses to the environment and increasing their use to support fish growth. Sophisticated models have been developed to allow fish farms to predict phosphorus and nitrogen losses when different feeds are used (Hua and Bureau, 2006; Dumas et al., 2007; Bureau and Hua, 2010).

Marine resources used to produce fish meal and fish oil are finite resources that have been fully utilized for decades. Aside from higher recovery and utilization of seafood processing byproducts, there is no prospect of increasing fish meal and fish oil production from wild stocks of marine fish (Naylor et al., 2009). Therefore, continued growth of feed production depends on the development and use of alternative sources of protein and oil from sustainable sources. Fortunately, a growing body of knowledge exists to support rationale replacement of fish meal and fish oil with sustainable alternatives in major farmed species, such as salmonids, shrimp, and many marine species (see Chapter 16). However, lack of information on dietary nutrient requirements of many important farmed species and on the effects of ingredient substitution in production feeds limits the extent to which alternative sources of protein and oil can be used. As mentioned earlier, fish meal is a complex material containing a wide array of essential nutrients and biologically active compounds, many of which are absent in plant proteins. Moreover, plant proteins possess negative properties, such as antinutrients and nonsoluble carbohydrates, which have to be overcome to avoid adverse effects on fish growth, health, or reproduction. Replacing fish oil with alternative lipid sources in fish feeds generally does not affect fish performance as long as essential fatty acid requirements are met (see Table 19-3). However, the fatty acid profile of fish tissues reflects the fatty acid profile of the diet, so care must be taken to avoid lowering the content of long-chain polyunsaturated fatty acids by excessive use of alternative lipids in fish feeds. At present, fish oil is the only practical source of long-chain polyunsaturated fatty acids (PUFA) such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) for farmed fish and crustacean feeds (see Chapters 6 and 16). Further investment in research and development is needed to allow higher levels of replacement of marine resources with plant-derived feed ingredients and possible single-cell proteins to improve the sustainability of aquaculture production.

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