CHAPTER – VII

FEED TECHNOLOGY

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INTRODUCTION

Aquaculture world-wide is an emerging agricultural sector with an average growth of more than 10% per annum. According to the FAO (2014) statistics, 154 million metric tons of fish were produced in 2012 in the world, of which 41% was produced from aquaculture. It is expected that aquaculture will produce 120 to 130 million metric tons by the year 2020 (FAO 2014). Fish consumption per capita is growing rapidly due to its importance in human nutrition

and the need for more food protein sources. Recent data show that 85% of the total fish farmed or wild caught is used for human consumption and remaining 15% is used for fish meal and fish oil production. The future fish demands will be met by intensification of fish culture practices in existing operational units, by introducing more flow-through and recirculation fish culture systems, cage culture and pen culture. In sustainable intensive and semi-intensive aquaculture operations, feed costs represent up to 60 - 75% of the total operating costs. Intensification of fish culture is the most viable option to adopt for higher production and to bridge the widening gap between demand and supply. This high production oriented approach cannot be accomplished without availability of quality feed in adequate amounts.

The manufacturing of aqua feeds is more complex than the common feed manufacturing for terrestrial species of livestock. Fish demand high protein content feeds in well bound forms with minimum leaching of the nutrients into the aquatic environment. Therefore both chemical and physical attributes of manufactured feed play an integral role in sustainable and commercially viable aquaculture. Feed quality promotes good growth with efficient feed conversion, help in maintaining good water quality and health of fish. Selection of quality ingredients is the foremost step for production of quality feed. Every possible measure should be adopted to ensure quality and safe delivery of the ingredients to the production unit. Prior to formulation make sure that ingredient will provide the concentration of nutrients we are expecting. It is advisable to chemically analyze all the ingredients in all the new supplies to avoid uncertainty.

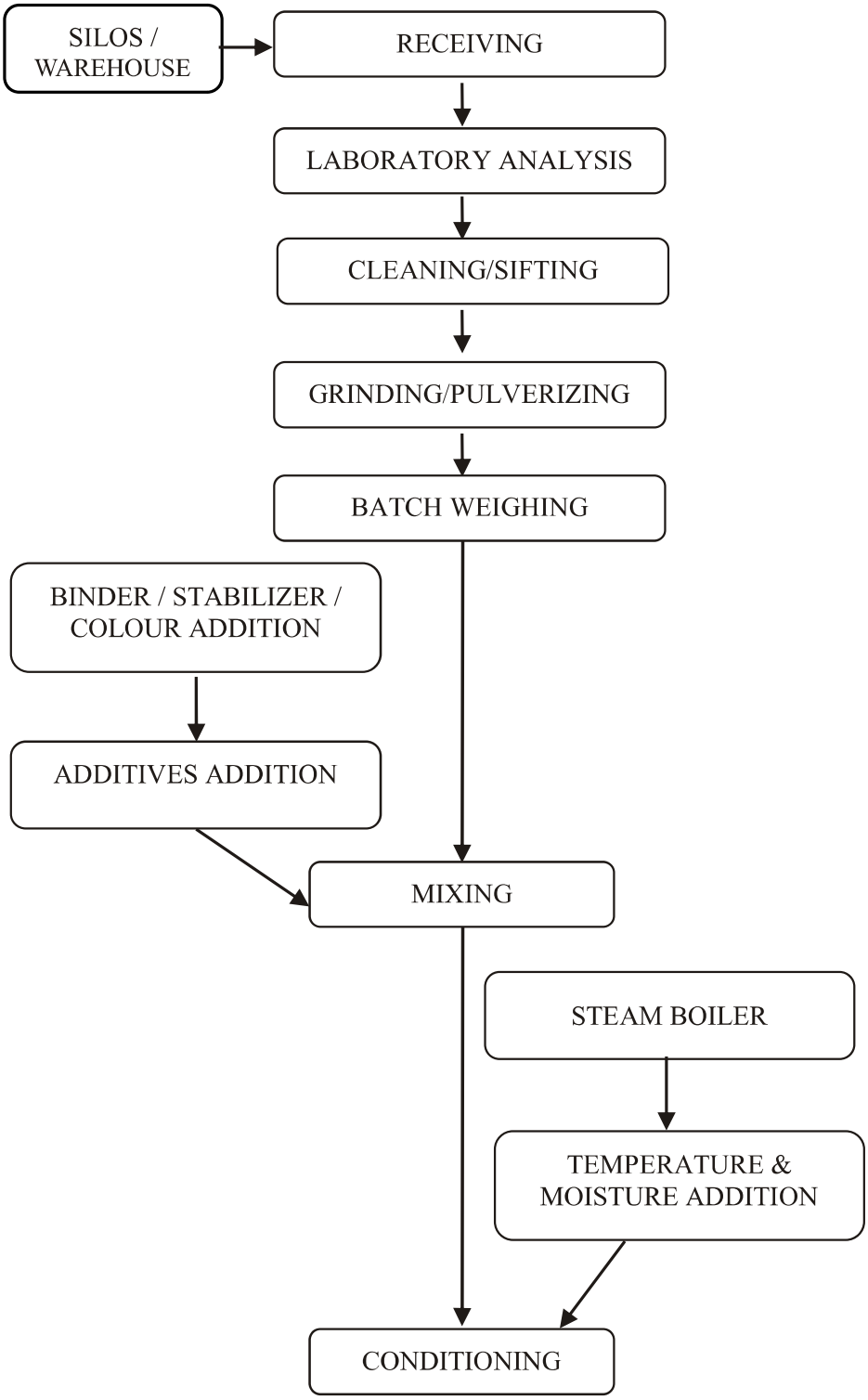
The diets produced for aquaculture enterprises can be in powder, flake, mash, pellet, crumble, or extruded floating forms. The buoyancy can be directed in the processing for sinking, slow sinking or floating feeds. The physical forms of feed in aquaculture are very important and certain modifications are required in conventional

production methods to manufacture feed which suits the type of species cultured and life stage. The following table shows a typical relationship of feed particle size with age and weight of a farmed fish.

Table 1. Relationship of feed particle size with fish age and weight

Fish Weight	**Crumbs Size mm**
2 to 30 days	0.5 mm
2 gram	1 mm
20 gram	1 – 2 mm
20 – 100 gram	2 – 3 mm
100 – 250 gram	3 – 4 mm
Above 300 gram	4 – 6 mm

The ingredient composition of the feed formula has an impact on the nature of feed and manufacturing process for aqua feeds. Feed types require various processing conditions and equipment to acquire different levels of temperature exposure, gelatinization and liquid addition in form of moisture and fats. Different feed ingredients like cereal grains, agro industrial by-products and animal protein sources pass through various milling and processing techniques to manufacture quality and efficient aqua feeds. The feed production process for aqua feeds generally is comprised of different processing steps (Fig. 1) to produce good quality feeds.



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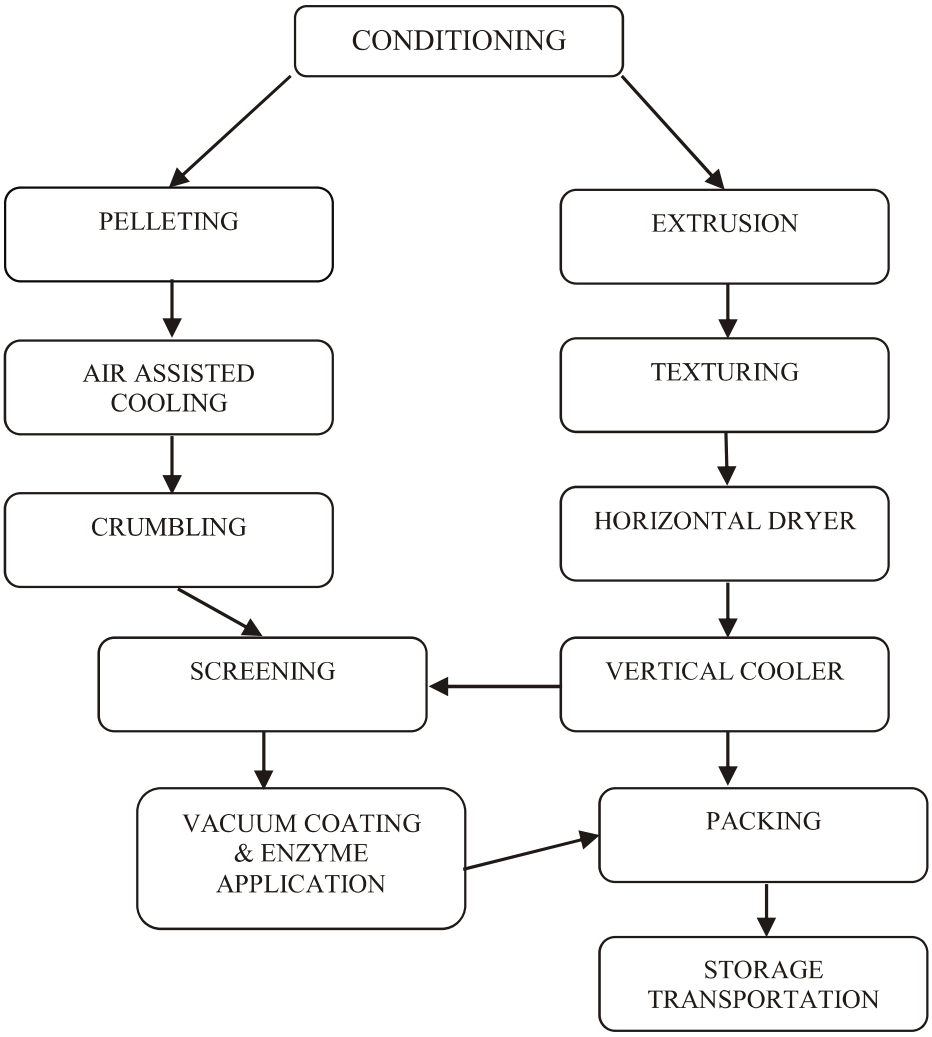

Figure 1. Flow Diagram of Aqua Feeds Manufacturing Processes.

FEED PLANT DESIGN AND SITE

The design and structure of feed plants vary considerably due to area, transportation / logistics, availability of the raw material, nearest fish markets and approaches, zoning restrictions, and type of labor available. Small modular turn-key plants can serve the new area of aquaculture development. The capacity of the plant involves selection of the processing equipment and plant design. The equipment selection is very important and sometimes the purchase cost of the equipment is insignificant in comparison to its

operational cost. For instance, on-farm feed manufacturing can be operated in a simple warehouse with the manual operation of batching, mixing and processing but it is much more complicated when we move to commercial production. Commercial plants entail complex factors like capital investment and pay back periods, feasibility, comprehensive surveys, grain storage silos, warehouses, up-stream and down-stream state of the art equipment, forward contracting for bulk ingredients, inventory controls, utility connections, flexibility in producing various types of feeds, future expansion capability, quality control, skilled manpower, commercial competitiveness, value addition, branding, environment friendly, extension work, marketing, and after sales technical services. When working commercially the processing equipment has the following key segments to accomplish the pelleting process.

- Grinding
- Screening
- Mixing
- Pelleting
- Extrusion
- Drying
- Cooling
- Crumbling / Sifting
- Coating

GRINDING (Particle Size Reduction)

The grinding of the raw materials is one of the major pre-requisites in aqua feeds manufacturing. Grinding of raw materials reduces particle size and increases surface area, reduces moisture, facilitates mixing, reduces density variations and increases digestibility, increases palatability, and acceptability. Grinding should be organized to achieve requisite sized material as large particles are undesirable and even dangerous for small fish and fry. Coarse or

ultra-fine grinding depends on feed type and supports pelleting or extrusion process by lowering the extra wear and tear of the equipment and consequently increasing the production output. As a general rule of thumb, material grinding level should be around 1/3 of the openings of the die. While producing feeds below 1mm size, 80 to 85% of the material should be less than 1/5th, the size of the die opening. For crumbles of less than 500 micron size the spectrum of the meal should have 80% less than 100 micron size. Particle size reduction depending on the equipment used, can involve immense energy utilization and will depend on several factors that will be mentioned below for each specific equipment. Grain type, size of sieve size, hole diameter, tip speed of the hammers, single or double pass pulverizing and degree of grinding will also have an impact.

The equipment used for particle size reduction include:

- Hammer mills
- Roller mills
- Pulverizers

HAMMER MILLS

Hammer mills accomplish size reduction by impact on a slow moving cereal grain with a fast moving hammer. Hammer mills work on the principle that most materials will crush, shatter or pulverize upon impact. The target has low kinetic energy and is struck with a heavy force of the high speed moving hammers which are attached to a shaft which rotates at high speed inside the chamber. The hammer tip travels at 15,000 to 18,000 ft. per minute. (76.2 to 91.4 m/s). The quantity, size, arrangement and speed of the hammers, screen size, distance among the screen and hammer produces coarse to fine grist spectrum as well as determines the mill capacity. The energy utilization depends upon degree of grinding. Hammer mills are less capital intensive compared to the roller mills

and pulverizers but at the same time are less efficient users of energy in comparison to the other mentioned machines. The effective screen / sieve size of hammer falls among 1 to 3 mm scattered holes for aqua feeds grinding requirements.

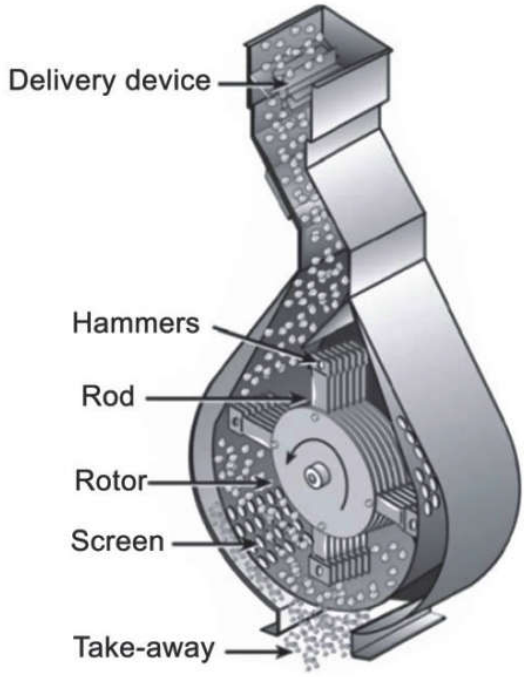

Figure 2. Internal view of a hammer mill

ROLLER MILLS

Roller mills accomplish particle size reduction through combination of cutting and crushing by corrugated rollers. The rollers moving at uniform speed create compression while rollers moving at different speeds create two way action of compression, shearing and tearing action. Roll grinding is economical and efficiently works on dry ingredients low in fat. Roller speed also matters for throughput of the grind. The typical speed ranges from 1,300 to 3,000 ft. per minute (6.6 to 15.2 m/s). Particle size can be achieved from 500 to 900 microns for cereal grains. Roller mills on an average grind size

are 50% more efficient than hammer mills in terms of kilo-watt per hour of energy.

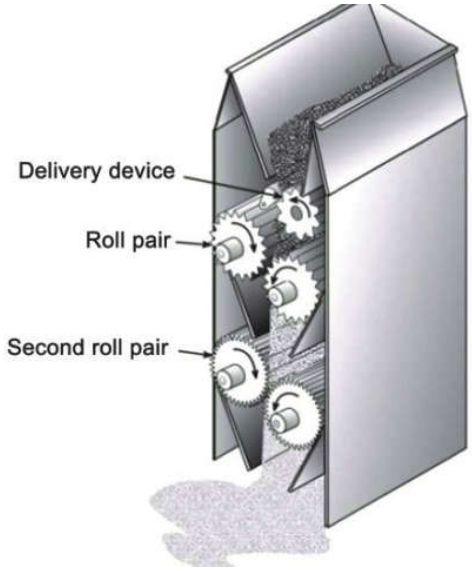

Figure 3. Grinding process is a roller mill

PULVERIZERS

Pulverizers are used for ultra-fine grinding of aquatic feed raw materials required for special processing. Pulverizers work by impacting particles with their beater plates at very high speed by moving them to corrugated liners and reduce size of the material in microns. The process of hitting continues again and again altering density reduction ultimately leaving the machine through the air stream to a product collector.

Pulverizers are used for tilapia, larval and shrimp feed production, which generally utilize 0.5 mm to 1 mm maximum in particle size. When grinding screen size of 1 mm or below is used in hammer mills, the capacity due to the screen's hole size significantly drops and may result in reduced quality, production efficiency and increases the maintenance cost of the unit.

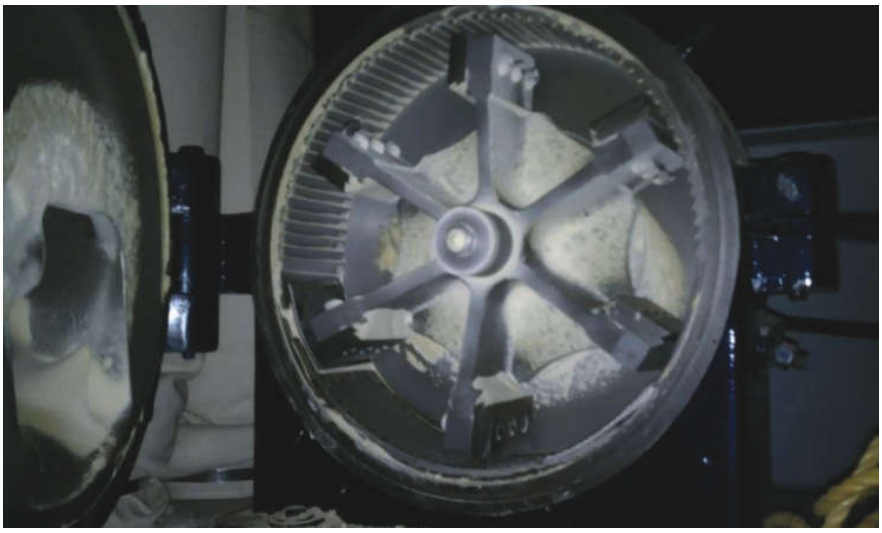

Figure 4. A typical Pulverizer for fish feed production

In this case it is more efficient to use an air classified mill (pulverizer) to get dramatically higher throughput of fine grind. Pulverizers are cost intensive at installation. Advantages of ultra-fine grinding are;

- Meets special feed and processing requirements
- Improves water stability of the feeds
- Identical density keeps nutritional balance intact
- Efficient through put grind of mixed feeds can be attained
- Reduce wear on extruder dies
- Energy efficient in comparison to other grinding systems
- Produces less heat during particle size reduction

SCREENING

The type and quantity of ingredients selected are based upon two factors; fish species and its size or age. Feeding habits are also to be considered. Some species learn quickly to eat floating feed whereas others, more used to searching for food at the bottom of the pond, may take longer to train or like shrimp, may need sinking diets. Size

and age of fish will determine the type of nutrition to be added as well as the size of the feed pellet.

In order to produce quality feed economically and to ensure desired feed conversion rate as well as disease free rearing of fish, the ground raw materials must be segregated as per their particle size by passing through a sieving system either composed of multiple sieves shaken through gyratory vibratory motion or of an inclined rotary sieve bed. Correct particle size is also necessary to ensure correct proportions of all ingredients as per the feed formula in each feed unit. The particles larger than the required size or "overs" are examined and then based upon their nature and requirement are either subjected to regrinding or expelled from the production line as rejects. One undesirable result of grinding is generation of excessive very fine particles, or dust, especially when dry raw material is being processed. Although quantity of these fines can be reduced by employing various methods such as spraying of oil or by using higher moisture content raw material, it is essential that these dust particles must be segregated from the bulk through properly designed dust collectors. The collected fines can then be processed appropriately either to prepare feed for fingerling or fry fish or utilized for other purposes. These raw material dust particles may separate from the feed in water and accumulate on the gills of the fish where they can create a breeding ground for bacteria and parasites resulting in gill disease.

MIXING

Mixing is carried out either to blend or to scatter the individual components in the mix so that each unit part of the mix contains all the ingredients either in the desired proportions or very close to it. Thus proper mixing is very important from the quality control point of view. Mixing is considered more of an art than any specific science and is conducted more on experience than anything else.

Within the context of this handbook, mixing involves mixing of solids as well as liquids both separately as well as combined. In addition to type of mixer, correct mixing depends upon differences in physical properties of the various ingredients of the mix. In the case of solids; size, density, shape, coefficient of friction and electrostatic charge determines distribution of ingredients, whereas in the case of liquids; viscosity, miscibility and base play vital roles. In fact, in solids the most important factor is the size distribution of the ingredients, which may lead to randomized mixing or segregation. It has been experimentally established that small uniform sized ingredients will result in a mixture that is close to the distribution as per the formula.

Mixing can be done manually in batches either with shovels on dust protected floors or in suitable containers. It can also be done as batch or continuous processes in mechanical mixers. Generally fish feed is prepared in batches after mixing in mechanical mixers. For this purpose the large volume ingredients are added first to the mixers followed by any medium volume ingredients like salt, calcium, or premixes, then the small quantity ingredients are added and in the end the liquids are added. These mixers are made in different configurations, shapes, and sizes depending upon the buyer's requirement, such as revolving drum, straight or tapered sided cylinder, or cone shaped, horizontal or vertical, top or bottom loading, ribbon, scrapper, paddle, plough or screw mixers. Rotation speed and mixing time are very important factors in ensuring proper blending free of lumps and with minimum possible size reduction of the contents. Methods employed to verify mixing uniformity (evaluation of the degree of mixing of each proportions of the formula's ingredients) are based on the measurement of coefficient of variance (CV). This quantification utilizes water soluble tracers in the mix that are quantified using laboratory. Another important mixing process applicable to fish feed preparation is called premixing. It involves mixing of micronutrients such as vitamins

and minerals. This process can also be utilized to include fish medications in the feed on required basis. Premixing is also carried out in batch type mixers for recommended preset time to ensure proper blending. A well-mixed batch of ingredients in powder form is often referred to as a "mash". In some extensive pond farming systems the mash may be broadcast by hand. Even though fish may aggressively consume the mash, many particles are lost and end up just fertilizing the pond ecosystem.

PELLETING

Pelleting is an agglomeration process that consists of changing mash feed into solids of given shape or texture by means of a mechanical process in combination with moisture, heat and pressure. The moisture and heat are inducted through saturated steam (steam at almost 100% quality) and/or mechanical pressure. The pressure obtained mechanically by pressing the feed by the rollers through the die, is around 300 to to 600 kg/cm2. In the steam addition process during the conditioning of the mash, the steam will increases the temperature and moisture of the mash. The rule of thumb is that for every 10 0C rise in temperature, it will typically increases the mash moisture by 0.7 to 1 %. Hence, the overall goal of the conditioning is to add between 4 to 6 % moisture into the mash in order to initiates gelatinization of starches in the outer layer of the particles. The pelleting processes started a century ago based on a ring die principle applying specific mechanical energy and still continues as a major processing technique in feed processing and product densification for various species of livestock and sinking aqua feeds.

After the conditioning process, the hot and moistened mash is compressed outward through cylindrical die by the rollers to produce the pellets. This process of forming the pellets, changes the mash density of 0.4 g/cc to a pellet bulk density of 0.5 to 0.6 g/cc.

Figure 5. A compression pelleting machine

Pelleting process can be accomplished in three stages:

- Conditioning
- Compressing
- Cooling

After cooling, in some cases it is required to have a smaller pellet size, therefore, in a Post-pelletizating process called crumbling, the pellet may be broken into smaller pieces. This crumbling process assures that the proper mix of ingredients is maintained and that the crumble is "bite" sized for small mouths in younger fish. At the same time, after crumbling, a sifting step is almost always required as the breaking process inevitably produces small particulates that would otherwise become fines in the feed. The material sifted is often fed back into the original pelleting process. The many advantages that pelleting process offers over mash feeds include:

- Pellets have better flowing characteristics
- Improves bulk density of the rations
- Reduces segregation of the materials
- Reduces feed wastage
- Reduces feed degradation by controlling bacterial and fungal populations
- Pellets provide a “mouthful” of balanced feed
- Improves nutrient utilization through better digestibility
- Improves uniformity and robustness of the fish harvests
- Pellet mills are available in capacities of 1 to 35 T/h of feed production and allows flexible selection for installed capacity

Pellet mill operation is considered very sensitive due to of the characteristics of its input and output variables. The input variables can be divided into conditioning and pelleting.

INPUT VARIABLES

- Mash flow(conditioning)
- Steam condition and pressure (conditioning)
- Particle sizes of the mash (conditioning and pelleting)
- Distance between die and roller shell (pelleting)

OUTPUT VARIABLES

- Size of the pellet
- Pellet durability
- Pellet moisture content
- Pellet temperature.

Pellet durability and hardness are quality factors that are considered objective, while color, texture, dust level and palatability are subjective.

PELLET MILL DIE SPECIFICATIONS

- Entry taper of the hole
- Size of the hole
- Depth of the taper
- Length of the hole
- Angle of the taper
- Angle and depth of the relief taper

CONDITIONING, PELLETING PROCESS AND COOLING DISTURBANCE VARIABLES

- Properties of the mash
- Moisture of the mash
- Temperature of the mash
- Relative humidity
- Ambient air temperature.
- Steam type, saturated, dry, over heated
- Velocity of the steam
- Roller shell, die type and condition

EXTRUDERS

Extrusion can be defined as a technological process of forcing feed raw material in one or more of the processing conditions (such as mixing, heating, cutting, etc.) by making some chemical and physical changes through the die to make material forming or eruption gasification. Extrusion allows buoyancy control to make floating, sinking, or slow sinking feeds. The bulk density of the extruded aqua feeds can be reduced by 0.25 – 0.3 g/cc compared with mash prior to extrusion. The extrusion process may or may not involve a simultaneous cooking process and this process functions in many ways including texture alteration, grinding, hydration,

expansion, homogenization, mixing, partial dehydration, shearing, protein denaturing, gelatinization and shaping. A properly conditioned mash that has substantial levels of water and temperature in a range of 100 to 1600C, will gelatinizes the starches and make proteins glutinous to increase the dough viscosity. After the mash is conditioned and cooked throughout the extruder, the mash can then be mechanically converted into expanded floating or sinking aquatic feeds.

The extrusion cooking for aqua feeds may involve using high temperature steam in the extruder barrel for 5 to 10 seconds in order to raise the mash to 2000C. Some of the benefits that are obtained are:

- Improved digestibility
- Improved feed conversion ratio
- Inactivates certain anti–nutritional factors
- Kills gram negative and aerobic bacteria
- Controls mold and yeast growth and destroys lipases
- Enhances energy liberation through breaking starch and fat molecules
- Enhances amino acids availability
- Improves palatability and
- Improves water stability of feeds up to 12 hours

Extruders have the characteristic that they can handle mash feed with moisture content between 20 and 40%. The extrusion mechanism depends on certain independent variables and dependent variables. Independent variables are further divided into two parts of conditioning and extrusion.

Figure 6. E-750 extruder machine with accessories.

The conditioning variables include critical parameters like temperature, moisture content, and retention time. In this phase the product responds with certain chemical and physical changes. While in extrusion phase the most important variables are: moisture content, oil addition, thermal energy, mechanical energy application, and retention time. The dependent variables include feed rate, lighter or heavier densities, and extent of cooking which can determine between the sinking and floating feeds because less cooking will lead to less expansion, high density and sinking feeds. High expansion on the other hand will result in low density floating feeds. The starch contents of the rations will also direct the ultimate feed type production. Starch is the primary form of carbohydrates found in aquatic feeds and levels like 10% starch tend to form sinking feeds while, 20% starch based formulations tend to turn into floating feeds. Amylose based starches have better binding and gelatinizing ability and convert into more water stable aqua feeds. The water activity is also an important factor in the shelf-life of extruded feeds. The ratio between vapor pressure of the water above the test sample and pure water or partial pressure of the water

at the same temperature should not exceed the level of 0.7, since it depicts the measurement of energy status of water in a system and at the same time, practically all the microbial activity stops at this point not allowing the growth of contaminants.

CONDITIONING

Conditioning refers to the preparation of the mash with the aid of steam added heat and moisture before it is compressed by the extruder mechanical force and pressure. Conditioning has a great bearing on compression process, palatability, durability and thus on quality of the extruded feeds. Proper conditioning of the mash before extrusion largely depends on the grist spectrum of the particles and generally the spectrum described below (Table 2) is considered suitable for efficient operation.

Table 2: Relationship between particle size and the maximum amount in a fish feed formula

Size Of The Particles.	% Level In The Meal.
3.5 mm	Up to 1 %
2 mm	Up to 5 %
1 mm	20 %
500 micron	30 %
250 micron	25 %
< 250 micron	Not less than 20 %

The purpose of the conditioning can be highlighted as,

- Equalizes moisture level of different raw materials
- Reduces wear and resistance during compression
- Reduces bacterial / fungal count
- Enhances adhesive properties
- Builds up bridges of solids

The conditioning process creates certain modifications for enhancing nutrient availability to the fish. It improves gelatinization of the starches, makes room for high fat and liquid addition, improves pellet quality, and intestinal digestibility.

Figure 9. A pre-conditioner with feed hopper

Crude fiber modification liberates more energy and creates more flexibility in using fibrous ingredients. Protein modification enhances digestibility through denaturizing process, reduces amino acids wastage as in high temperature steam treatment, reduces nitrogen excretion, alleviates restrictions in the use of synthetic amino acids, and plasticizes/coagulates proteins. The conditioning process kills most gram negative bacteria, molds, aerobic bacteria and improves feed hygiene.

Steam quality also affects the conditioning process such that the steam entering the conditioner should not just add temperature but should also add moisture. The pressure of the steam through the feeding lines should be between 5 to 7 bar, but when it enters the conditioner, its pressure should be reduced through a PRV (Pressure reducing valve) to 1 – 1.5 bar. At the same time, the velocity of the steam is also altered by increasing the pipe diameter by 2.5 times

just after the PRV to a recommended velocity is 20 m/s. Generally, the ideal quality of the steam should be as close to 100% as possible, therefore, as saturated steam. The conditioning process may be short or long term by means of retention time and type of the conditioner. Twenty seconds to 1 minute time is considered as short term conditioning while dwell time for long term conditioning can be 3 to 10 minutes increasing ripening of the mash and enhancing gelatinization prior to the extrusion process.

Different types of conditioners are used for varying temperature and moisture content levels of the mash prior to extrusion.

- Small barrel conditioner mixer with paddles
- Two stage barrel conditioner with paddles.
- Long conditioner LLX type
- Differential diameter cylinder with varying speed in each chamber

The differential diameter cylinder conditioner has more popularity for conditioning aqua feeds. It improves average retention time/distribution in the conditioner. The shaft speed also affects mixing as the small chamber shaft operates on high RPM in comparison to the large side which works on a lower RPM. It offers more flexibility for adding more water and oil during conditioning. Steam in the conditioner is added from the bottom while other liquids are added from the top. In terms of filling of the conditioner, 40% filled gives more homogenized ripening and gelatinization of the meal prepared for extrusion.

Gelatinization is described as the swelling of starches with heat and moisture. Starch granules are generally insoluble in water below 500C. When a suspension of starches in water is heated beyond critical temperature, the granules absorb water and swell to many times of their original size. The suitable temperature for this purpose

will be 60 to 800C. The swelling of starches due to colloidal dispersion makes starch paste defining gelatinization. True gelatinization of starch molecules occurs when a paste is cooked at 100 to 1600 C (HTST) which is usually required for mash having 20% and above starch content for floating aqua feeds.

FLOATING FEEDS

To understand the floating characteristics of the pellets, its density should be below 1g/cc in comparison to water density in order to float. If the pellet has a density of 1.05g/cc in freshwater, and 1.12 g/cc at 3 % sea water, it will sink. Therefore, in order to make the pellets float, certain processing parameters should be reached. Following is an example of operating parameters that can be used to produce floating feed in an extruder. The conditioning time may range from 120 to 140 seconds and the mash should have a temperature between 95 to 1100C with a moisture content between 22 to 26%. The temperature of the mash at the discharge end of the extruder should be 1300 C, moisture content between 21 to 24%, and a pressure at the discharge between 34 to 37 atm. The bulk density should be less than 480 and 440 g/l in fresh water and sea water, respectively.

Figure 7. Floating feeds of various shapes and sizes

SINKING FEEDS

For sinking feeds, the operating parameters of the extruder are different when compare to producing floating feed. For example, the conditioning time may range from 90 to 120 seconds and the mash should have a temperature between 85 to 950C, and a moisture content between 25 to 32%. The temperature of the mash at the discharge end of the extruder should be at 1200C, the moisture content of the mash between 24 to 27%, and the pressure at discharge between 20 to 29 atm. The bulk density of mash to be extruded into pellets should be less than 600 and 640 g/l in freshwater and sea water, respectively.

Generally, there are two types of extruders that are used for manufacturing aqua feeds:

- Single screw extruder
- Double screw extruder

Also, the extrusion process may be dry or wet cooking depending on the addition of water in the conditioning process.

Single screw extruders have three types:

- Dry cooking extruders
- Wet cooking extruders
- Cold forming extruders

Generally dry cooking extruders are operated without steam and are used in processing of low moisture highly expanded starch products. These are also used for processing whole soybean to full fat soybean meal or flour and are not commonly used in the production of aqua feeds. Due to dry cooking, the post extrusion step of drying can be eliminated. These extruders are not cost intensive and are sometimes used for small operations at the farms.

Wet cooking extruders are equipped with pre-conditioners and are largely used in the aqua feed and pet food industry. These extruders can handle 20 to 30% moisture mashes with the high capacity of production from 1 to 20 T/h with variety of raw materials. Wet extrusion cookers can handle direct water and fat injection and require a post extrusion drying and cooling process. Wet cooking extrusion improves digestibility and eliminates most pathogens. Cold forming extruders have a very limited use in producing moist aquatic feeds. These function like a meat grinder with the ability of blending certain raw materials. This extrusion does not benefit like pasteurization.

Twin screw cooker extruders are used in wet extrusion and are operated for specialized aquatic feeds manufacturing. Initial capital investment, operational and maintenance costs are higher in

comparison to single screw extruders. Twin screw extruder of identical capacity as a single screw extruder can have a purchasing price 1.5 to 2 times higher and at the same time, the same factor applies in maintenance costs. Twin screw extruders can handle water and fats injection during the course of cooking extrusion and can use very small dies of 0.7 to 1.5 mm in diameter. They also offer the advantage of a resulting more clean equipment at end of the operation. They can handle formulations having 30 to 40% meat based ingredients. Overall, these extruders offer great flexibility and produce extruded products with even size and uniformity.

Extrusion cooking is not only cost intensive regarding the initial capital investment but is also expensive in operation due to the high electrical energy input requirement. For instance, small cooking extruders can use up to 0.5 kWh in producing just 1 kg of the aquatic feeds. Special emphasis should be given in selection for proper capacity installation of the equipment.

Figure 8a. An assembly of twin screws.

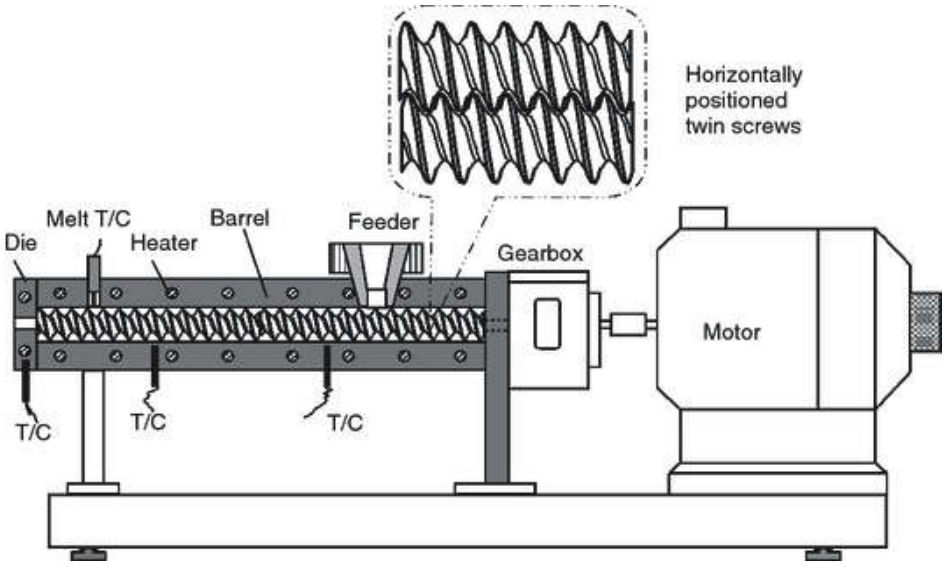

Figure 8b. An assembly of twin screws and their arrangement in the extruder.

DRYERS

The drying process in aqua feed production is more relevant in case of feeds manufactured through extrusion since their moisture contents are relatively higher. Drying with heated dry air is used for removal of excess moisture thereby increasing the shelf life of the feed. Generally, horizontal dryers equipped with single or double stage perforated conveyor belt or trays allowing flow of hot air through the thin layer of properly spread feed are employed for this purpose. Single stage horizontal dryers are a better option because it results in generation of fewer fines but they require more floor space and better control on processing parameters. Whereas double stage dryers are compact and efficient in controlling the moisture content, mechanical handling results in more fines which then have to be reprocessed to control production cost. Air temperature, speed of movement of feed through the dryer are determined by the moisture content of the feed entering the dryer and the desired moisture level of the feed leaving the dryer.

COOLERS

Coolers extract heat and surplus moisture created during extrusion/pelleting making feed harder and suitable for handling and storage. The pellets leaving the cooler should be 5 to 80C above the ambient temperature and moisture uptake by finished feed should be around 0.5 to 1% as compared to the mash compounded for conditioning. Generally, depending on their design, coolers can take 10 minutes for cooling after extrusion process. Cooling mechanism depends upon the ambient air temperature coming in intimate contact with outer surface of the pellets. The temperature imparted to the pellets by steam is a major factor in subsequent drying. The ambient air blown through the newly formed hot pellets will absorb heat and thus increases its water holding ability leading to drying initiation. The following types of coolers are used in aqua feeds manufacturing:

1. Vertical cooler
2. Horizontal belt cooler
3. Counter flow cooler

VERTICAL COOLER

In a vertical cooler the free flowing pellets by gravity makes a bed in the cooler at the full level to attain its maximum efficiency and discharge is governed by the feeding rate. Cooling is achieved by drawing ambient air cross wise through the column of pellets and exhausts through a cyclone leaving the fines for recycling.

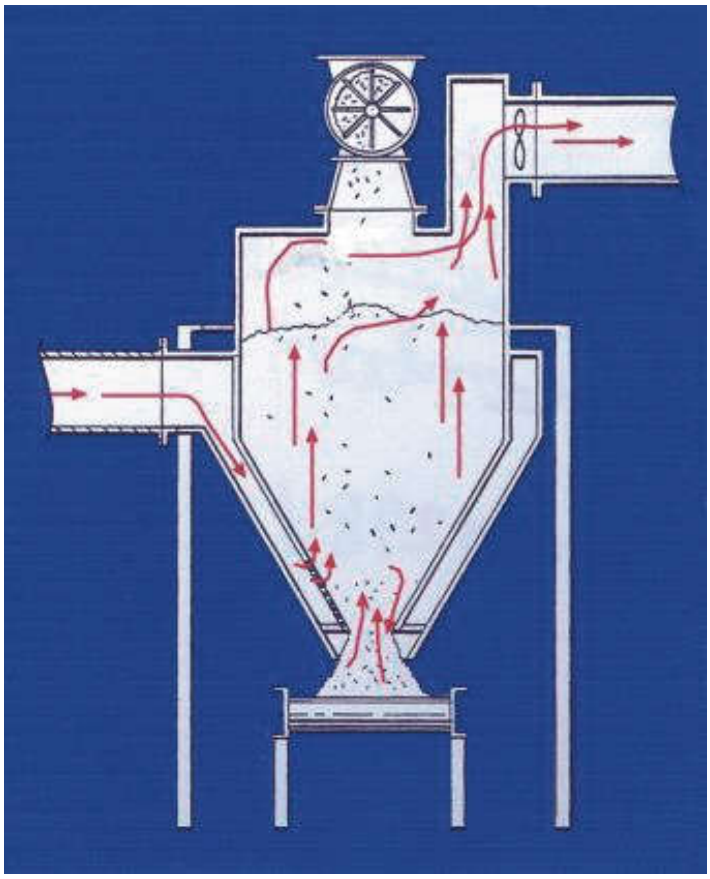

Figure 10. Air flow in a vertical cooler

HORIZONTAL BELT COOLER

Horizontal coolers are more flexible for cooling/drying extruded feeds which usually contain more moisture by controlling retention

Figure 11. A horizontal belt cooler

time of feeds. Bed depth and air flow can be controlled and better efficiency of the cooling can be achieved by completely covering the cooler bed with an even depth of the pellets to allow the air to pass through for uniform dispersal and uptake of moisture from the feeds.

COUNTER FLOW COOLERS

In a counter flow cooler the pellets and cooling air flow in opposite directions. The coolest pellets contact the cool air and the hottest one contact with the warmest air. These coolers are becoming more popular in the feed industry. The counter flow cooler provides constant retention and an even flow over the entire discharge area. These coolers are energy efficient and cover less space for installation due to vertical orientation.

Figure 12. A view of a counter flow cooler

CRUMBLING AND SIFTING

In order to reduce their size, pellets can be passed through a crumbler after cooling. This is done for obtaining a smaller feed unit more suitable according to age and type of small fish. The crumbling process provides flexibility of producing special feeds with selected granulations, eases product flow in difficult feeding systems, higher production efficiency in comparison to small pellet manufacturing using more electrical energy, and allows more economic formulations. Crumbling mechanism occurs through

cutting by rollers having different flutings and running on speed differentials. The cutting of the pellets is flexible by adjusting the roller gap.

The crumbs are transferred to reciprocal multi-deck screen operation or rotary type screen operation for separation of desired crumbs for packing and removing the fines for recycling of pellets. Removal of fines is critical both to maintain economic efficiency of reusing the material, but also as fines can end up suspended in the water column thereby not only wasting the nutrients but also contributing to poor quality of water. Fines can also result s from rough handling of feed after it leaves the feed plant. In transport to, or on the farm, throwing bags of feed, piling additional heavy materials on top or walking on full bags, will grind the pellets together and produce fines.

COATING

The pellets manufactured through pellet machine or extruder are exposed to high temperatures of conditioning and compression leading to loss of potency of vitamins and enzymes. Feeds normally contain 5 to 10% fat and to achieve higher levels, in some cases a coating is necessary with oils using low heat. In the same way, certain heat sensitive vitamins and enzymes (thermo-labile products) that would not normally withstand the conditioning and extrusion temperatures can also be added later during the coating process.

QUALITY CONTROL AND PRODUCT ANALYSIS

Aqua feeds production and farming in Pakistan is in its infancy compared to higher terrestrial animals. The sector being in a development stage will further define its parameters as industry expansion begins and commercial capitalization continues. Seafood preferences in the urban population and high income groups,

provides hope for rapid development of industrial and quality norms. Pakistani feed act compliance and follow up of instructions pertaining to restricted medicines, heavy metals and prohibited feed additives. Further labeling will improve the quality and robustness of the aqua culture feed industry. New feed requirements in general, notably those related to traceability (of ingredients used within aqua feeds), higher degree of compliance with legislation, package recycling, carbon foot-prints, sanitation programs, environmental effects of feeds and factory effluents/ smoke, quality assurance, after sales close cooperation with fish farmers, R&D cooperation development in local area farming, and analytical state of the art laboratory testing facilities will serve as milestones in quality operation of aqua feeds manufacturing.

QUALITY ASSURANCE AND LABORATORY ANALYSES

For future export orientation the following certification programs will be applicable.

- Hazard Analyses at Critical Control Points (HACCP)
- International Standards Organization (ISO) Certifications
- Quality and Safety System for Specialty Feed Ingredients and their Mixtures (FAMI QS)
- Good Manufacturing Practices (GMP's)

The quality control programs in feed mills pertaining to ingredients, in-process, and finished products quality control that will rely on a sophisticated laboratory. The laboratory testing will further elaborate the measuring of specific components of feed ingredient sample to assure its quality. It may be chemical, physical or electronic measurement to determine quality of a product to compare with the predetermined or established standard. The testing data may be used to:

i. Evaluate ingredients and sources
ii. Nutrient evaluation of formulation
iii. Establishing product standard deviation variation and trend lines
iv. Assist in troubleshooting
v. Assures that feed meets the label guarantee
vi. Incorporates psychological satisfaction

These procedures can be accomplished either through the in-house laboratory or from any other laboratory of commercial repute. The tests for aqua feeds include; moisture content, particle size, bulk density, floatability, starch gelatinization, fat absorption, pellet durability, pellet hardness, water stability, and flow index. For these tests to be successful, a proper sampling procedure should be developed and followed for ingredient and finished feed. At the same time, it is important to establish a traceability procedure that starts at the ingredient receiving step and goes to the finished feed storage lot before it is sold or consumed at the farm. There are several testing equipment procedures that in some case utilizing high capacity sensors to quantify certain quality parameters. Among these equipment and procedures are:

i. Labeling, batching for traceability.
ii. Feed microscopy
iii. Wet chemistry application
iv. Chromatography
v. Spectrophotometry
vi. NIRS details reflectance spectroscopy
vii. Radio frequency
viii. Micro wave technology
ix. Capacitance and conductivity
x. DSC - Differential scanning calorimeter

Wet chemistry and NIRS are bench top technologies utilized in the scrutiny of the aqua feeds. The short wet chemistry procedures are included for the proximate analysis in the in-house laboratory. The following table (Reference) shows the equipment and procedure needed to quantify each of the main chemical compositions and its quality parameters present in the ingredients or finished feed:

Table 3: Equipment and procedure utilized for ingredient/feed chemical composition (AOAC Methods, 2012)

S. #	Test	Equipment(s)	Solutions required
1	Moisture/Dry matter	Silica dish, Oven, Electronic Weighing scale, Desiccators	NIL
2	Crude Protein	Weighing scale, Heater, Digestion flask, Volumetric flask, kjeldhal flask and apparatus. 100 ml beaker, Pipette, Hot plate	Concentrated Sulphuric acid, Sodium Hydroxide, 2% Boric acid, 0.1N HCl,$CuSO_4$, $FeSO_4$, K_2SO_4
3	Ether Extract	Weighing scale, Soxhlet apparatus, petri dish, Oven, dessicator, Water bath, Thimble	Diethyl Ether
4	Ash	Weighing scale, Crucible, Heater, Furnace, Desiccators	NIL
5	Moisture	Petri dish, Weighing scale, Desiccator, Oven	NIL
6	Acid Insoluble Ash (AIA)	Crucible, Hot plate, Whatman filter paper (#44), Heater, Furnace, Weighing scale	25 ml HCL (10%)

7	Crude Fiber	Weighing scale, 250ml Beaker, Hot plate, Oven, Muffle furnace, Water bottle , stirrer, Filter paper or cloth, Desiccators	1.25% H_2SO_4, 1.25% NaOH
8	True Protein	Weighing scale, Oven, Filter paper(#44), Digestion tube, kjeldhal apparatus	25 ml TCA (5%), Digestion mixture + 10 ml Sulphuric acid (Commercial), Sodium Hydroxide, 2% Boric acid, 0.1N HCl, $CuSO_4$, $FeSO_4$, K_2SO_4
9	Salt	Weighing scale, 500ml Conical flask, filter paper(#1), beaker, Shaker, Pipette 10ml, 1ml	Potassium Chromate (0.5%), $AgNO_3$ solution (0.1N) $ZnCH_3COOH$, Potassium Ferrocynide
10	Free Fatty Acid	Beaker, Hot plate, 10ml pipette, Magnetic Stirrer	Phenolphthalein, 0.1N NaOH, Ethanol
11	Peroxide Value	500 ml Beakers, funnel, Conical flask, Oven, Weighing Scale	Acetic Acid, Chloroform, Potassium iodide, Distilled water, Starch
12	KOH Solubility	Conical flask, Weighing scale, Digestion flask, Centrifuge machine & tubes, kjeldhal apparatus, Pipette, Mechanical shaker	KOH (0.2%), Digestion mixture, Commercial Sulphuric acid, (2%) Boric acid, 0.05 N Sulphuric acid

13	Aflatoxin Test	Weight scale, Blender jug, Graduated cylinder, Fluted filter paper, Microfiber filter paper, column, Aflatoxin apparatus, corvette, Auto pipette, vicam	NaCl, Methanol (HPLC grade), Bromine,
14	Mixing Test	Mechanical shaker, whatman filter paper#595, spectrophotometer	Ethanol, Methyl violet
15	Stability in water test	Screen Trays, Drying oven, Desiccators, balance	Nil

In the following section, all the quality parameters of each of the chemical composition are explained and detailed.

PROXIMATE ANALYSIS

Proximate analysis is performed for the determination of the major characteristics of feed to confirm that it is within its normal compositional parameters. This method partitioned nutrient characteristics of feed into several components: moisture content, ash (or acid insoluble ash), nitrogen free extract (or crude protein), crude fiber and crude fat.

MOISTURE CONTENT DETERMINATION

Moisture content is determined by the loss in weight that occurs when a sample is derived to a constant weight in an oven. About 20 grams of the feed sample is weighed into a silica dish previously dried and weighed. The sample is then dried in an oven for 1100C for 2 hours, cooled in a desiccator and weighed. The drying and weighing continues until a constant weight is achieved.

Calculation:

% Moisture Content = (wt. of sample + Petri dish before drying – wt. of sample + Petri dish after drying)/sample wt. × 100

DRY MATTER

Since the water content of feed varies widely, ingredients and feed are usually compared for their nutrient content on moisture free or dry matter (DM) basis.

Calculation:

% of DM= 100 - % Moisture Content

CRUDE PROTEIN

Crude protein is determined by measuring the nitrogen content of the feed and multiplying it by a factor of 6.25. This factor is based on the fact that most protein contains 16% nitrogen. Crude protein is determined by the Kjeldhal or Nitrogen Combustion method. The method involves: digestion, distillation and titration.

Digestion (AOAC 2012)

Weigh about 2 gram of the sample; add 25 ml of concentrated sulfuric acid and 5 gram of mineral mixture into digestion tube. Apply heat to digest it for 45 minutes until the digest becomes clear pale green. Leave until completely cool and rapidly add 100 ml of distilled water. Rinse the digestion flask 2-3 times.

Distillation (AOAC 2012)

Provide steam for the distillation apparatus and add about 10 ml of the digest into the apparatus via funnel and allow it to boil. Add 10 ml of NaOH from a measuring cylinder so that ammonia is not lost. Distill into 50 ml of 2% boric acid containing screened methyl red indicator.

Titration (AOAC 2012)

The alkaline ammonium borate (NH_4BO_3) formed is titrated directly with 0.1 N HCl. The titer value which is the volume of acid used is recorded. The volume of acid used, is fitted into the formula which becomes

Calculation:

% of Nitrogen = 0.0014 × 100 × 250 × 6.25 / 10 × 2

% of crude protein = % of Nitrogen × Titration reading value

ETHER EXTRACT

The ether extract of a feed represents the fat and oil in the feed. Soxhlet apparatus is the equipment used for the determination of ether extract. It consists of these major components.

Extractor: Comprises a thimble for holding a sample.

Condenser: For cooling and condensing the ether vapors.

Procedure (AOAC 2012)

About 150 ml anhydrous diethyl ether (petroleum ether) having boiling point of 40-60°C is placed in the flask. 2-5 gram of sample is weighed into thimble. As the heated ether vapors reaches the condenser through the side arm of extractor, the extraction continues for at least 4 hours and most of the solvent is distilled from the flask into the extractor. The flask is then disconnected and placed in an oven at 65°C for 2-4 hours, cooled in a desiccator and weighed.

Calculation:

% of ether extract = (wt. of Petri dish + extract – tare wt. of Petri dish)/ sample wt. × 100

ASH

Ash is the inorganic residue obtained by burning of the organic matter of feed stuff at 400-650°C in a muffle furnace for 4 hours. Five grams of the sample is weighed into a preheated crucible. The crucible is placed into muffle furnace at 400-650°C for 2-4 hours or until whitish grey ash is obtained. The crucible is then placed in the desiccator and weighed. (AOAC 2012)

Calculation:

% of ash = (wt. of crucible + ash – tare wt. of crucible) / sample wt. × 100

ACID INSOLUBLE ASH (AIA)

After estimation of ash, add 25 ml HCL (10%) in crucible. Heat it on a hot plate for 2-3 min. Filter through the Whatman filter paper #44. Burn it on heater and put it in furnace for 2 hours. Weigh the acid insoluble ash (AIA) with crucible (W2) (AOAC 2012).

Calculation:

% of AIA = W2-W1/ 5× 100

W1 (tare wt. of crucible)

W2 (Crucible wt. + AIA)

CRUDE FIBRE

The fat free sample is transferred into a flask/beaker and 200 ml of 1.25% H2SO4 is added. The solution is gently boiled for 30 minutes maintaining a constant volume of liquid by the addition of hot water. The residue is then washed several times with boiling water and transferred into a beaker. Then add 200 ml of 1.25 % NaOH and boil for another 30 minutes and put the sample in a crucible. The

residue is dried at 110°C in an oven for 2-3 hours and weighed. The residue is placed in a muffle furnace at 400-650°C for 2 hours. Then cooled in a desiccator and weighed.

Calculation:

% of crude fibre = (dry wt. of residue before ash – wt. of residue after ash)/ sample wt. × 100

NITROGEN FREE EXTRACT

NFE is determined by mathematical calculation. It is obtained by subtracting the sum of percentages of all nutrients already determined from 100 (AOAC 2012).

Calculation:

% of NFE = 100 – (% of moisture + % of crude fibre + % of crude protein + % of ether extract + % of ash)

NFE represents soluble carbohydrates and other digestible and easily utilizable non-nitrogenous substances in feed.

TRUE PROTEIN (AOAC 2012)

Take 1 gm sample and add to it 25 ml TCA (5%). Place it on magnetic stirrer for 15 minutes. Then shift it into the oven for 15 minutes with occasional shaking. Filter it with simple filter paper and wash it with distilled water. After filtration, transfer the filter paper into digestion tube. Now add 5 gram digestion mixture + 30 ml sulfuric acid (commercial). Next procedure is same as crude protein.

SALT

Weigh 1 gm well ground sample and put it in a ceramic dish. Heat the ceramic dish until the sample is free from smoke. Cool it at

room temperature and crush it with glass rod. Add ¾ of crucible volume with distilled water. Heat it on hot plate. Filter it in conical flask with the help of Whatman filter paper no. 1. Wash 2 to 3 times with distilled water. Take 10 ml of filtrate in 100 ml beaker. Add 2-3 drops Potassium Chromate (K2CrO4) as indicator (0.5%). Titrate with silver nitrate (AgNO3) solution (0.1) N. End point – yellow to brick red.

Calculation:

% of Salt = (Burette reading × factor of AgNO3) × 100

Factor of AgNO3 = 2.33

FREE FATTY ACID (AOAC 2012)

Take 10 gram sample of oil in 250 ml beaker. Take 50ml of ethanol. Boil it then add to it few drops of 1% phenolphthalein and change color with 0.1N NaOH. Color will be permanent light purple. Add it to sample and mix vigorously. Titrate it against 0.1N NaOH to bring light pink color, should be persisting for 30 Sec.

Calculation:

R × N × 282 × 100 / 1000 × 10 (SAMPLE WEIGHT)

(Should be <1)

R (reading) of 0.1N NaOH

While 282 is oil factor

PEROXIDE VALUE

Take 5 gm oil sample and add to it 30ml Acetic Acid: Chloroform (3:2). Add 0.5 ml saturated potassium iodide (KI) and shake vigorously for exactly 1 minute. Add 30 ml distilled water and titrate it against 0.1 N sodium thiosulfate ($Na_2S_2O_3$) (AOAC 2012).

Calculation:

R × N × 1000/ 5(sample weight)

KOH SOLUBILITY

Weigh 1.5 gm fine ground sieved sample in a beaker. Add 75 ml KOH (0.2%) and stir it for 15 minutes. After 15 minutes, take this sample in plastic or glass tube and centrifuge it for 15 minutes. When centrifuge is completed, take 5ml supernatant + 5 gm digestion mixture in digestion tube and add 30 ml commercial sulfuric acid in digestion tube. Digest for 60-90 minutes. Allow it to cool for 30 minutes and add 10-15 ml distilled water. Now take 10 ml (2%) boric acid solution in a 100 ml beaker and place it on distillation unit. Insert both pipes (NaOH and Distilled Water pipe) into its solutions. Turn on alkali switch, add suitable amount of NaOH (Until distillate becomes black) and turn it off. Ensure the water is always running. Turn on steam switch. Allow distillation of boric acid solution 30-40 ml. (takes 4-5 minutes). Titrate the distillate against 0.5 N sulphuric acid, color changes from green to light purple, read the end point (AOAC 2012).

Calculation:

% of KOH = (burette Reading) × 4375 / CP of this sample

AFLATOXIN TEST

Take 50 gm sample + 5 gm salt (NaCl) + 100 ml (80 ml methanol: 20 ml H_2O) HPLC grade in blender jug and shake it for 1 minute and filter. Take 10 ml filtrate sample in 50 ml cylinder and add 40 ml distilled water to make volume 50 ml. Filter the filtrated sample again with micro fiber filter paper. Now take the column, cut its tip and adjust it on stand. (Also adjust column and syringe on column). Now take 10ml filtrate sample and pass through column and 10 ml distilled water two times and then 1ml pure methanol (HPLC grade)

also passes through the column. Collect this methanol in cuvette. Now add 1ml developer in this cuvette, shake it and put it in the Aflatoxin Test Kit from VICAM, wait for 60 sec and take reading (AOAC 2012). For labs that do not have an HPLC, there are less accurate strip tests that will demonstrate a positive of negative presence to a threshold level.

MIXING TEST (on Methyl Violet basis) (AOAC 2012)

Use set of free samples. Take 10 gm of feed sample each + 30 ml ethanol; shake them for 30 minutes on mechanical shaker. Then filter them with Whatman filter paper no. 595. Now take the filtrates reading on spectrophotometer: (495 wavelength). Calculate the results through computer (mean, standard Deviation and percentage CV)

PHOSPHORUS (AOAC 2012)

Take 0.5gm sample in 100ml conical flask. Add 10ml Nitric acid to it. Heat the flask on hot plate until only a few drops are remaining. Cool the flask and add 5ml per-choleric acid. Then again heat this flask on hot plate until only a few drops are remaining. Cool the flask and make volume to 100 ml with distilled water in volumetric flask. Filter it with Whatman filter paper No.1. Take 1 gm sample in the test tube and add 9ml D.W. Take 1ml diluted sample in test tube and add 1 ml ammonium molybdate + 0.4ml ANSA+ 7.6 ml D.W. Take 1ml diluted standard in the test tube + 1 ml ammonium molybdate + 0.4ml ANSA+ 7.6 ml D.W.

Blank sample:

1ml ammonium molybdate +0.4ml ANSA+8.6 ml D.W.

After 8 minutes stir the samples and take reading on spectrophotometer (720 wavelength).

Calculation:

Standard reading- Blank = A

Sample reading – Blank = B

% of phosphorus = B/A × 16

Stability in Water Test

Ten grams of uniform sized pellets are spread on a screen tray of 1002 cm surface area. Floating feed pellets may require a screened top cover to keep them in place. The screen tray holes should be smaller than the pellet size. Two or more set of samples are immersed into still water tank for 10 minutes. After the set time the tray is raised and allowed to drain. The sample is then dried in a drying furnace at 130oC for 2 hours followed by cooling in a dessicator. The sample is again weighed and its ratio to initial weight (10 grams) is used as comparative measure of stability of pellet in water.

References

AOAC Methods (2012). Official Methods of Analysis of AOAC INTERNATIONAL, 19th Edition.