

# Fish feed response to essential and nonessential amino acid supplementation for low crude protein diets

### Suliman A.Alkandri

Email:k3000w@hotmail.com

Principal Research Technician

### Barrak F.Alazmi

Principal Research Technician

Email: bazmi@kisr.edu.kw

### maha B.sinan

Email: msinan@kisr.edu.kw

Principal Research Technician



#### **Abstract**

The interest in the cultivation of many fish species has led to research to study their nutritional needs, as industrial feeding is an indispensable necessity in the case of intensive fish farming. As the rapid development in animal nutrition science and its research into minute details and its ramifications in the field, it has imposed development, accuracy and complexity in the technique of analyzing materials and nutrients, methods and various methods in forming relationships and selecting materials for them. This study aims to investigate the response of fish feed to to essential and nonessential amino acid supplementation in low-raw protein diets due to the fact that the protein requirements in the starting feed for fish are relatively high, especially with the high prices of protein sources. Reducing the protein content in fish feed is one of the most important strategies required to increase the sustainability of fish farming by reducing the environmental impact as well as reducing the feed costs using less nitrogen inputs. Methionine (Met) followed by lysine (Lys) are the first amino acids to be restricted in plant and animal by-products. It was found through the research that protein is the most important factor and the most valuable component on which the cost of the component feed is calculated. Therefore, the type of protein required must be carefully chosen and that it is easy to digest and absorb and contains the amino acids required for fish.

Keywords: Fish, feed, amino acid, supplementation crude protein, diets.

#### ملخص البحث

أدى الاهتمام باستزراع العديد من أنواع الأسماك إلى إجراء أبحاث لدراسة احتياجاتها الغذائية ، حيث أن التغذية الصناعية هي ضرورة لا غنى عنها في حالة الاستزراع المكثف للأسماك. نظرًا للتطور السريع في علوم التغذية الحيوانية وأبحاثها في التفاصيل الدقيقة وتداعياتها في المجال ، فقد فرضت التطور والدقة والتعقيد في تقنية تحليل المواد والمغنيات والأساليب والطرق المختلفة في تكوين العلاقات واختيار المواد لها . تعدف هذه الدراسة إلى التحقق من استجابة أعلاف الأسماك لمكملات الأحماض الأمينية الأساسية وغير الأساسية في الانظمة الغذائية منخفضة البروتين الخام نظرًا لحقيقة أن متطابات البروتين في العلف الأولي للأسماك مرتفعة نسبيًا ، خاصة مع ارتفاع أسعار مصادر البروتين. . يعد تقليل محتوى البروتين في علف الأسماك أحد أهم الاستراتيجيات المطلوبة لزيادة استدامة تربية الأسماك من خلال تقليل التأثير البيئي وكذلك الأسماك أحد أهم الاستخدام مدخلات أقل من النيتروجين. الميثيونين (Met) متبوعًا باللايسين (Lys) هي الأحماض الأمينية الأولى التي يتم تقييدها في المنتجات الثانوية النباتية والحيوانية. وجد من خلال البحث أن البروتين هو أهم عامل وأهم مكون يحسب على أساسه تكلفة مكونات العلف. لذلك يجب اختيار نوع البروتين المطلوب بعناية وأنه سهل الهضم والامتصاص ويحتوي على الأحماض الأمينية اللازمة للأسماك.

الكلمات المفتاحية: أسماك ، أعلاف ، أحماض أمينية ، مكملات بروتين خام ، أعلاف.



#### 1. Introduction

The rapid and amazing development in the science of animal and human nutrition and its research in minute details and its ramifications in the field has imposed development, accuracy and complexity in the technique of analyzing materials and nutrients, and the various methods and methods of forming relationships and selecting materials for them, despite that for an approximate analysis to determine some basic nutrients such as: Moisture, protein, fat, fiber, etc in foodstuffs were used extensively in the early to mid-nineteenth century. However, the expansion of knowledge of the details of the nutritional requirements of animals and the increase in knowledge of their interactions and cycles, and their biological filters inside the body of the organism, alerted the vital relationship between substances Nutritional and body systems are due to the limitations of the approximate methods used (which focus mostly on chemical reactions) to determine the nutrients more precisely and to detail their quantity and quality (FAO, 2012).

Today, nutrition science is heading towards global limitations and transformations in setting standards and classifications for foodstuffs in terms of: composition and uses and the development of methods of devising them and methods of preparing them to be included in the framework of their use as an acceptable food, and this is the result of the multiplicity of food processing products, the development of methods for extracting foodstuffs chemically and mechanically and the multiplicity of their scientific names (NRC, 2011). There is also a new product resulting from an in-depth knowledge of animal experiences in terms of behavioral and physiological terms in feeding on new food compounds whose components have intertwined from natural materials (grains - legumes - etc.), industrial materials (urea - Bayourt ... etc. "and manufacturing wastes (Alberto, 2014).

The problem of providing suitable, inexpensive feed is the main problem that limits the expansion in the field of animal husbandry in its various departments, whether for the production of white and red meat or fish production. The problem of fish feeding is the determining factor for the expansion of this industry at the global level and in developing countries in particular (Taleb & Hassan., 2013).



Fish are living organisms that live in an aquatic environment and get their food through their aquatic environment, i.e. self-sufficient from their environment, but in fish farms, fish breeders are forced to use processed foods (fish diets) to feed large numbers of fish because the limited environment in fish ponds is unable to produce appropriate natural food with a high density of fish, and to increase the rates of fattening of the raised fish in order to obtain the highest productivity in the shortest period of time (Klein, 2007)...

Fish breeders are forced to use processed foods to feed large numbers of fish due to the inability of the limited environment in fish ponds to produce natural food commensurate with the high fish density. Special diets are used to feed the fish, which are characterized by their consistency and floating for a long time in the water until they are eaten by the fish, as their fall on the ground of the ponds leads to difficulty feeding on them and negatively affects the characteristics of the water. As is well known, it is not only important what the feed material contains of nutrients, as much as the amount of digested food content, and therefore it is not only important to analyze a feed material for its components, but these results must be supported by evaluating the feed material in terms of the parameters of its components to know the value of the feed material Productivity (Lemme, 2004).

Protein, energy metabolism, and amino acids together with feed additives are important in the cost that must be weighed in the feed in order to reduce as much as possible the costs of feed mix when following any diet (Klein, 2007).

Fish is one of the animal sources with high nutritional values due to its high protein content, reaching 18% in it, in addition to what it provides from long-chain essential unsaturated fatty acids, as well as many vitamins and mineral salts, and the global demand for fish meat is constantly increasing and per capita consumption Of fish meat in developed countries is about 9.25 kg / year, while the per capita share in the Arab world generally reaches 2.5 kg (FAO, 1995).

Since fish breeding needs to provide good food and relying on an adequate protein source that provides the fish needs of the nutritional components necessary for their growth and reproduction, so many international institutions interested in feeding and raising fish have started using modern scientific breeding methods, adopting balanced feeding programs and introducing biological enhancers products (Al-Janabi, 2011).



Extensively as one of the growth-stimulating food additives, as well as its role in increasing resistance to disease microorganisms and improving the cellular and humoral immune response (Cellular Immunity Humeral and), since it was noted that there is a significant role of these enhancers in improving fish productivity also as they contribute to increasing the absorption of protein consumed as protein The single cell has a high biological value as well as its role in protecting fish from pathological micro-organisms and improving their health status (Ringo, 1995). Therefore, this study aims to investigate the response of fish feed to to essential and nonessential amino acid supplementation in low-raw protein diets due to the fact that the protein requirements in the starting feed for fish are relatively high, especially with the high prices of protein sources.

### 2. Literature review and previous studies

#### 2.1 Industrial Nutrition

Industrial feeding is an indispensable necessity in the case of intensive fish farming, but in the case of an extensive or semi-intensive culture system, fish get a part of their food from the surrounding natural environment and thus their need for artificial feeding is reduced, taking into account that the use of fertilizer in semi-intensive fish farms leads to increased production Natural food (plankton) and thus fish need less industrial food (FAO, 2012).

Industrial feeding constitutes approximately 50% or more of the total running costs of the farm. Therefore, it is wise to pay special attention to the issue of industrial feeding with the aim of reaching the optimal food at the lowest costs. In general, with regard to fish feed, the following specifications must be met (FAO, 2012):

- 1. It contains the nutrients that the body needs protein, fats, carbohydrates, vitamins and mineral salts.
- 2. Accepted by fish.
- 3. It consists of elements that are (locally if possible) always available.
- 4. It is easily manufactured and stored.
- 5. The costs are cheap.
- 6. It does not contain harmful substances to fish such as pesticides, microbes and toxins.



Industrial feed for fish must contain protein, fats, carbohydrates, vitamins, and mineral salts, and it should be noted that fats and starches are the main source of energy, while protein represents the basic building blocks of the body.

#### 2.2 Protein

Protein is the basic building block of the organism, and proteins are composed of units called amino acids that aggregate in a chain, the type of protein depends on the numbers and types of amino acids, the amino acid is mainly composed of carbon, hydrogen and nitrogen, and the amino acids that make up protein are divided into basic and other non-essential amino acids. There are many sources of protein as following (Klein, 2007):

### a) Animal protein

After animal protein, the most valuable protein sources in terms of containing the essential amino acids, but the main problem facing the use of animal protein in fish diets is its high prices and lack of production in the face of the increasing demand for it. Therefore, it is necessary to calculate the amount of animal protein that the diet must contain. Strictly, this amount should be used as a source of constructive growth, not destructive energy. Fish meal, poultry manure, slaughterhouse residues, blood meal, fish and snail waste are among the most important sources of animal protein used in fish diets. The quality of protein varies according to the source and its content of amino acids, the method of preparation and preservation, and fish meal is the best type of animal protein, as it contains high quantities of all the essential amino acids compared to other sources.

### b) Vegetable protein

Oil crops such as soybeans, cottonseeds, sunflower seeds and sesame are among the main sources of plant protein, after the seeds are squeezed and the oils are extracted from them, and the seeds of these plants contain a high percentage of most of the essential amino acids. However, you find that the reference to some plant protein sources lack lysine and therefore this acid must be added when using it as a protein source in a diet of farmed fish. In general, the missing acids are often added by adding another source that contains a high amount of these missing acids.



### c) Single cell protein

This protein is produced (as it appears from the name) from single-celled plant or animal sources such as yeast, algae and protozoa, and it is known that these organisms multiply and multiply millions of times in short periods of time, and this can be cultured intensively and in a small space, in order to be used To feed fish, especially in the early stages of life (larval stages). These organisms are characterized by their high protein content, so they can be added in different proportions to the artificial diets for fish.

#### d) Industrial and agricultural waste

Many industrial and agricultural wastes and other wastes are important sources of protein in diets of farmed fish. Industrial food wastes such as industrial juices, preserved foods, yeast and starch contain varying proportions of vegetable or animal protein, and thus can be added in certain proportions according to the culture conditions and protein content for the additive (ZUO, 2017).

#### 2.3 Sources of feed materials used to feed fish:

There are many sources of plant protein feed used to feed fish, such as oilseeds, soybeans, cottonseeds, peanuts, rapeseed, sunflower, sesame, cobra, yeast, leftovers from processing corn, and legumes. These sources are characterized by their ability to produce large amounts of protein in a short time (40-80% protein) (FAO, 2006).

There are also many sources of animal protein feed used in feeding fish, as animal-source feed materials can cover the deficiency in the necessary amino acids and vitamins, and for this reason, animal proteins, even if used in small quantities, can improve the nutritional value of the feed as a whole, and among these sources is poultry manure, Dried fish meal, slaughterhouse remnants, invertebrate animal powders, and dairy waste. The process of producing artificial feed for fish is especially important when the natural food in the water becomes so few that it does not meet the needs of the existing fish numbers. It is necessary to rely on feed that contains all the elements of the food in sufficient quantities to reach the best and highest growth rates of fish, the lowest death rates and highest reproduction rates (Al-Jalibi & Muhammad, 2017).



Aquaculture is one of the fastest growing meat production sectors in the world, with an annual growth rate of about 8.8%, which exceeds the growth of the rest of the meat producing sectors (Tacon, 2004). This significant growth in aquaculture is offset by an increase in the demand for feedstuffs to cover the needs of fish, especially protein, whose percentage in fish feed is between 25-45% (Lim & Dominy, 1989). The cost of feed represents more than 50% of the cost of producing aquaculture in semi-intensive farming (Silva De, 1993) and 70% of intensive farming (Thompson, 2005). Therefore, researchers have tended to search for non-traditional sources of fodder to reduce the cost of feeding such as residues of oilseeds and legume crops (Mohammad, 2016).

#### 2.4 Amino acids as feed additives

Today, commercial food systems need to be fine-tuned to meet the nutrient requirements of species that target specific stages of development, the culture system, and farm production levels. Aquatic feed for farmed fish is made from raw materials similar to those used for livestock animal feed. A large number of protein components can be used to manufacture aquatic feed, but fishmeal has historically been the preferred protein source. The reason is due to its nutritional value, especially in relation to high levels of digestible CP and balanced essential amino acid (EAA) content that approaches the requirements of fish and crustaceans. However, with the rapid growth of aquaculture, global supplies of fishmeal have reached the stage of stability. As a result, it is becoming less available and more expensive to use as the main protein ingredient in aquatic feed (Al-Jalibi & Muhammad, 2017).

Hence, there was an urgent need to increase the nutritional use of alternative protein sources. Reasonable options for replacing fish meal included byproducts from agriculture or fisheries or the slaughter of land-based production animals (for example: soybean meal, canola meal, corn gluten meal, meat and bone meal, soybean or pea protein concentrate, and poultry secondary meal., Feather meal, blood meal, bycatch of fisheries and waste meal preparation). Some of these ingredients can have similar CP levels to a fish meal, but values usually range from 40 to 75% (as is the basis) (FAO, 2012).

Methionine followed lysine are the first amino acids to be restricted in plant and animal by-products. Commercial feed for aquatic animals is usually rich in protein with many formulations containing excessive amounts of dietary crude protein (CP).



The use of the concept of ideal protein in formulating EAA requirements is gaining popularity especially in reducing dietary CP content and providing EAA requirements through synthetic amino acids (Alberto, 2014).

Amino acids: it are the building blocks of proteins, which are acids that contain an amine group, and amino acids, some of which are monoamines, monocarboxylic (neutral), such as glycine and leucine, and some of which contain sulfur such as cysteine and cystine, and some of which are dicarboxylic monoamines (acidic) such as glumatic and aspartic acids, including basic ones such as lysine and hydroxy-lysine, and aromatic ones (cyclic) such as phenylalanine and daiodotyrosine, and some of them are mixed-rings such as tryptophan and hydroxyproline. Some amino acids are added to animal diets if they are not available in the natural sources of the diet in the required quantity for the body, and the body does not originally form other acids. Among these additions are methionine and lysine (the amino acid glycine may be added to detoxify the benzoic acid in poultry) (Nang Thu et al., 2009).

In the event that the diet is deficient in a specific amino acid, the percentage of protein formation in the animal (which includes this deficient essential amino acid) decreases with the same percentage of deficiency of this acid, and it is called the specific amino acid. So some amino acids are added to complete this deficiency. Amino acids are biosynthesized in Japan, Europe and elsewhere for the large-scale production of amino acids, such as glumatic acid and its derivatives, and lysine, which is lacking in most plant fodder materials. The specific amino acid in grains is mainly lysine, while in legumes it is methionine. Therefore, green fodder is added to multiple protein sources such as fish powder and yeast to supplement the amino acids in general, in addition to specific amino acids (lysine, methionine) for feed mixtures (Fayez, 2018).

The relative balance between amino acids (AA) as a key factor in protein quality is as firmly established in fish as it is in other animals. For example, the chemical grading method (Mitchell and Block, 1946) used to estimate protein quality, relies on the premise that the performance of an animal fed an amino acid deficient diet depends only on the specified amino acid level, if all other nutrients are supplied in sufficient quantities (Langer & Fuller, 1996).



This hypothesis led to the concept that unlimited amino acid removal has no effect on nitrogen gain (N). According to this hypothesis, it appears that protein concentration and amino acid increase in the diet play a secondary role as long as the minimum requirements for amino acid reduction are met (Langer and Fuller, 1996). However, several reports indicate that excess amino acid reduces the growth performance of different animals (Yamamoto, 2004) suggest that the chemical point method should take into account the dietary protein concentration and certain excesses amino acid (Langer & Fuller, 1996).

Many studies concluded that the effects of protein excess on growth performance are due almost entirely to changes in voluntary feed intake (Nang Thu, 2007). The effect of dietary protein level on the efficiency of use of the first limit of amino acid is controversial in fish. This view is supported in some way by recent results by Nang Thu et al. (2007) that demonstrated the negative effect of unbalanced vegetable protein sources (corn gluten meal, sesame oil cake) on the retention efficiency of Lys above maintenance in rainbow trout fry, indicating a negative effect of some excesses of amino acid (leucine, alanine, arginine) on fish performance and Lys utilization efficiency. However, Abboudi et al. (2006) suggested that the retention efficiency of Lys is independent of protein level (Abboudi, 2006). Unfortunately, these authors worked in a limited range and with a limited number of dietary Lys levels and their experiments were not specifically designed to test the hypothesis of an effect of protein level on Lys utilization efficiency (Nang Thu et al., 2009).

#### 2.5 Amino Acids and Fish Health

The great development of antibiotic resistance in animals has put pressure on nutritionists to find suitable and alternative methods for obtaining high production and healthy animals. Nutritional supplements are a way to primarily strengthen the immune system and improve intestinal defense mechanisms against bacteria. Glutamine, threonine, and arginine are involved in many maintenance functions, particularly in the processes of repairing the intestinal mucosa and the immune system (Bequette, 2003).



The intestine is the first barrier to food-borne antigens and bacteria. The high rate of protein turnover in the gut ensures that damaged tissues are eliminated quickly and large amounts of mucin are excreted to create a barrier to bacterial transmission. Intraepithelial lymphocytes and mucosal cells contain glutamine requirements as an energy source and synthesize other non-essential amino acids and nucleotide bases. The dietary supply of glutamine may become limited during gut stress, and glutamine supplementation improves weight gain and reduces intestinal villi atrophy (SILVA, 2008). The effectiveness of glutamine may depend on whether the insult to the digestive tract poses challenges (Bequette, 2003).

Mucins act as a barrier to infection and antigenic substances introduced into the gut. In fish, the skin secretes mucin for external protection, which is rich in threonine, cysteine, cysteine, proline, tannins, and indigestible fiber, and intestinal diseases increase mucin secretion and stimulate atrophy of the gut (Zootec, 2010).

### 2.6 The composition of the feed (fodder)

The interest in the cultivation of many types of fish, such as tilapia, catfish, and others, has led to research studies to study their nutritional needs. As a result, fish farmers use artificial feeds to feed their fish, which may be manufactured for other fish such as trout and catfish. This strategy may be successful in terms of both growth and health of fish, but it may lead to the formation of unnecessary residues when more or more elements and nutrients in the diet exceed the true needs of the species. It turns out that commercial feeds are formulated to contain a relatively high level of nutrients higher than required by the type of fish being cultured in order to maximize and increase the growth rate. These additional nutrients are added during the composition of the feed to ensure that the fish consumed and absorbed the nutritional components and is used as a safety method from the farmer's point of view to obtain Maximum growth rate. Unfortunately, these amounts in excess of the feed partly contribute to the production of excess waste in the farm's waste products (Klein, 2007).

Most of the nitrogen in the diet is found in the form of amino acids, which are the basic unit for protein formation, and the intestinal enzymes work to break down the proteins in the diet into amino acids that are absorbed and used to build new proteins such as muscle formation. The nitrogen (extracted) extracted from many sources includes non-food proteins.

Digested, unabsorbed, or intestinal cell residues, amino acids absorbed in greater quantities than can be used by fish were extracted from the excretory products of fish (Al-Janabi, 2011).



The nitrogenous waste can be reduced by developing the components of the feed to cover the nutritional needs of the fish quality and also by developing the fish feeding systems. However, most of the nitrogen excreted by the fish is lost through the gills in a soluble form and cannot be recovered again. The nitrogen extracted by the gills originates from the absorbed but unused amino acids and from the decomposing metabolites. Therefore, the balance of the group of amino acids to suit the needs of the species and avoid overnutrition is the best ways to reduce the nitrogen excretion through the gills (Al-Jalibi & Muhammad, 2017).

The optimal amount of nitrogen loss represented (metapolic N) ranges from 100-200 mg nitrogen / kg fish / day in salmon fish and cannot be avoided, of which most of the nitrogen is excreted in soluble form therefore, management methods for controlling solid farm products, such as suction or sedimentation of solid solids. Not efficient in reducing the nitrogen in wastewater from the farm. Therefore, reducing and reducing the amount of excess nitrogen entering the water system in the form of feeds is the only highly efficient way to control nitrogen in fish farm wastewater. Therefore, the quality and quantity of the feed protein are also the important factors that must be taken into account to control the dissolved nitrogen in the farm wastewater. Fish is referred to as protein quality in the diet, balance and digestibility of amino acids in the diet (Thompson, 2005).

There are also essential amino acids (EAA) that most fish cannot form inside the body, and we must obtain them from high-quality feed or diets that will provide and supply essential amino acids from high-quality ingredients such as fish meal and soy powder, and these protein sources must be added in the necessary proportions and quantities. To meet the specific needs of fish in terms of metabolism and muscle growth. Feeding at appropriate levels of protein consisting of balanced amino acids specific to another fish species leads to an increase in the level of ammonia in the products of farm drainage. In addition, low-quality diets consist of large quantities of unbalanced protein sources to meet and meet the minimum nutritional needs of essential amino acids similarly to diets.

Manufactured with essential amino acids that are suitable for one fish species, they do not necessarily be suitable for another fish species (Alberto, 2014).



This leads to the absorption of some of the amino acids in greater amounts than the fish can use for muscle growth. When the amino acids are absorbed in greater amounts than those used by the fish, the removal and excretion of nitrogen is through the gills in the form of non-static dissolved ammonia (NH3). Fish size, protein consumption, and water temperature all affect the amount of ammonia excreted from the fish. Young fry are fast growing and need high protein diets. Nevertheless, a relatively small amount of nitrogen in the feed enters the water medium in relation to the first feeding and the largest proportion of it is from the output of fish because the nitrogen output is inversely proportional to the weight of the fish (Abboudi, 2006).

High temperature affects nitrogen excretion through its effect on increasing food consumption and food movement through nitrogen through its effect on increasing food consumption and food movement through the intestine while working to reduce nutrient use, and this is the problem if feeding is done until satiety or using food as needed Since fish will consume more quantities than required for optimal growth rate and nutrient utilization, some nitrogen loss cannot be avoided even if high quality diets are used due to protein turnover processes in fish and causes the enzymes that break down proteins are usually constantly active in Fish, however, the use of protein for lack of muscle formation was reduced by substituting calories used in the form of protein with calories from carbohydrates or fats. This is called protein sparing, because fish will use more fats and carbohydrates than protein for their energy needs, therefore, it provides protein for muscle growth (Alberto, 2014).

### 2.7 Reducing dietary protein by amino acid supplementation

Reducing the protein content in fish feed is one of the most important strategies required to increase the sustainability of trout farming by reducing the environmental impact as well as basically reducing the feed costs using less nitrogen input. An animal's growth is determined by the first environmental, dietary, or genetic determinant. The physiological condition of the animal also keeps nutrient requirements in constant flux as requirements may change with disturbances in health status, environmental conditions, and age or size of the animal (Lovell, 2002).

Protein requirements are the sum of the amino acid requirements for protein deposition, substrates for energy acquisition and biosynthesis of metabolic media. Hardy (2002) has observed multiple research groups that amino acid supplementation will improve trout performance (Yamamoto T. S., 2005).



However, the exact amino acid balance that allows for maximum growth and production efficiencies has not been established (Hardy, 2002).

One effective method for determining the amino acid balance and estimating their requirements was to compare the proportions of dietary amino acids with those found in the tissues of the cultured species. This so-called "ideal protein" has been widely studied and used in the productive animal industries (Dari, 2005).

Gaylord and Rawls (2005) conducted a study supporting the use of the ideal protein concept in aquaculture feeds. A pet food-grade poultry meal based diet was formulated on an optimal protein basis, and were fed hybrid striped bass for 10 weeks. Fish fed on amino acids, a diet devoid of fish meals, had a growth equivalent to seabass fed a diet based on fish meal (Gaylord, 2005).

The ability to improve fish growth and feeding efficiency has been demonstrated by adding one or more essential amino acids to reduce total dietary protein. Viola et al. (1992) showed that total dietary protein can be reduced when lysine is added to fish, and Cheng et al. (2003b) that dietary crude protein could be reduced from 42 to 37% in fishmeal based feed for rainbow trout when methionine, tryptophan, lysine, and threonine are added (Viola, 1991). Other research demonstrating the ability of lysine supplementation or lysine and methionine supplementation (Li and Robinson, 1998) to reduce total nutritional raw protein requirements in channel catfish have been ineffective. One reason might be that in some cases another amino acid was limiting first instead of methionine or lysine (Li, 1998).

#### https://naldc.nal.usda.gov/download/28815/PDF

Reducing the protein content of a diet is essential to formulating appropriate diets in terms of cost and pollution, especially when the fish are raised in an intensive system, where the fish's requirements are met from the nutrients provided by the food. Nitrogen is one of the most polluting nutrients in fish diets, and digestible high protein diets are required to reduce environmental pollution without negative effects on fish performance (Zootec, 2010).

https://www.scielo.br/scielo.php?script=sci\_arttext&pid=S1516-35982010001300010



The increasing cost of strategic use of fishmeal for high-value fish species means that it is essential to take into account the economical diets of aquaculture and this is particularly important for non-carnivorous fish, other than sea bass and salmon that require high-specification diets and where fish meal is used on the wide range. Therefore, scientists' attention has recently focused on evaluating the use of plant protein sources (Zootec, 2010).

Body weight and age are important factors for determining protein content in diets to support efficient fish production. However, some studies suggested that the total substitution of fishmeal with vegetable protein affected the growth performance of rainbow trout and Atlantic salmon. Nonetheless, it was possible to substitute 100% fishmeal in the salmon diet without any negative effect on growth (Zootec, 2010). Feed consumption on diet supplemented with amino acids in salmon was better compared to highly concentrated fishmeal feeds (AYISI, 2017).

Knowledge of protein requirements for fish during a period of rapid growth is essential to the practice of aquaculture to maximize feed conversion and utilization, save some costs, and reduce protein load in the aquatic environment (ABDEL-TAWWAB, 2010). Several researchers have attempted to determine the substitution of fishmeal in tilapia feed with inexpensive sources of plant and animal protein (ZUO, 2017). Nevertheless, further investigations are needed to evaluate the performance of different combinations of alternative ingredients for preparing a balanced fish diet. Webster et al. (1999) show that the combination of animal and plant protein sources with additional amino acid traits may enhance the potential for insufficient nutrients that can adversely affect fish growth (WEBSTER, 1999). Fish meal is being successfully substituted with other plant protein sources in fish feed, and there is some concern that current assessments of essential amino acid requirements are an unsatisfactory target, and some AAs may turn into a limiting factor and thus need to be supplemented in species feed important fishes (LIU, 2017.).



#### 3. Conclusion

The protein represents the basic building blocks of the living organism, and proteins are composed of units called amino acids that aggregate in a chain. The type of protein depends on the numbers and types of amino acids, and the amino acid is composed mainly of carbon, hydrogen and nitrogen.

Raw protein is an essential nutrient in fish feed. Reducing the use of raw protein not only reduces the cost of feed, but also reduces environmental pollution during fish production. Thus, finding the minimum protein requirement in fish feed without compromising fish growth was the aim of this research. Supplements of the essential amino acids including methionine and lysine to the low protein diet have shown similar growth performance to a regular protein diet. Thus, reducing the crude protein level is possible if the essential amino acid balance is appropriate for fish growth.

We conclude from this research that the use of low-protein feeds improves growth better than the presence of diets with high protein content and reduces the costs of producing feed, even if these diets are made from simple feed materials with low protein content, whether they are animal or vegetarian proteins. The quality of protein varies according to the source and its content of amino acids, the method of preparation and preservation, and fish meal is the best type of animal protein, as it contains high quantities of all the essential amino acids compared to other sources.

In order to improve the competitiveness and effective application of advances in aquaculture nutrition, installers are now adopting modern and environmentally sound formulation techniques based on nutrient value, crystalline EAAs supplementation and animal nutrient requirements. It is important to conduct more research to determine the dietary amino acid balance that increases amino acid retention and protein gain, and to address the economic value of alternative protein sources. It is also necessary to develop models to simulate and predict responses of amino acid supplementation to fish performance and production cost under variable conditions.



#### Reference

Abboudi, T. M. (2006). Protein and lysine requirements for maintenance and for tissue accretion in Atlantic salmon (Salmo salar) fry. Aquaculture 261, 369–383.

ABDEL-TAWWAB, M. A. (2010). Effect of dietary protein level, initial body weight, and their interaction on the growth, feed utilization, and physiological alterations of Nile tilapia, Oreochromis niloticus (L.

Alberto, J. N.-A. (2014). Practical supplementation of shrimp and fish feeds with crystalline amino acids. Aquaculture Volume 431, Pages 20-27.

Al-Jalibi, A., & Muhammad, M. A. (2017). The effect of soaking in water and heat treatment of sativa Vicia raw grains as a partial or complete substitute for soybean meal on some productive traits of juvenile common carp fish L carpio C. . Volume 13, Issue 3, The Jordanian Journal of Agricultural Sciences.

Al-Janabi, M. (2011). EFFECT OF ADDITION DIETARY PROBIOTIC WITH TWO LEVELES OF PROTEIN CONTENT RATIONS ON SOME GROWTH PARAMETERS OF COMMON CARP CYPRINUS CARPIO L.

AYISI, C. Z. (2017). Growth performance, feed utilization, body and fatty acid composition of Nile tilapia (Oreochromis niloticus) fed diets containing elevated levels of palm oil[J]. Aquaculture and Fisheries, vol. 2, no. 2, pp.

Bequette, B. (2003). Amino acid metabolism in animals: an overview. In: D´MELLO, J.P.F. (Ed.) Amino acids in animal nutrition. Edinburgh: CABI, . p.87-101.

Dari, R. P. (2005). Use of digestible amino acids and the concept of ideal protein in feed formulation for broilers. J. Appl. Poult. Res.14,195–203.

FAO. (1995). Food and Agriculture Organization of the United Nations, "The State of the World's Fish Resources and Aquaculture", pp. 61, 1995.

FAO. (2006). Food and Agriculture organization. state of world aquaculture. fisheries technical paper No. 500. Rome. Italy. 134.

FAO. (2012). The State of the World Fisheries and Aquaculture 2012. FAO Fisheries and Aquaculture Department. Food and Agriculture Organization of the United Nations (FAO), Rome, Italy (2012) (209 pp.).



Fayez, M. (2018). A physiological and nutritional view on mineral elements and their feed additives> Faculty of Veterinary Medicine, Suez Canal University.

Gaylord, T. R. (2005). The modification of poultry by-product meal for use in hybrid striped bass Morone chrysops X M. saxatilis diets. J. World Aquacult. Soc. 36, 363–374.

Hardy, R. .. (2002). Rainbow trout, Oncorhynchus mykiss. In: Webster, C.D., Lim, C. (Eds.), Nutrient Requirements and Feeding of Finfish for Aquaculture. CABI Publishing, New York, NY, pp. 184–202.

Klein, H. (2007). Effect of lowered dietary crude protein content, adjusted balanced amino acid levels and altered feeding phases on performance and profitability of commercial turkeys-hessling. Turkey Production: Current challenges Proceedings of the 4th International Symposium on Turkey production Meeting of the Working Group 10 (Turkey). held in Berlin, Germany. 21st–23rd June 2007. Organized and Published by Prof. Dr. H. M. Hafez.

Langer, S., & Fuller, M. .. (1996). The effects of excessive amounts of protein on lysine utilization in growing pigs. Br. J. Nutr. 76, 743–754.

Lemme, A. U. (2004). Effects of reduced dietary protein concentrations with amino acid supplementation on performance and carcass quality in turkey toms 14 to 140 days of age. Int. J. Poult. Sci. 3:391-399.

Li, M. R. (1998). Effects of supplemental lysine and methionine in low protein diets on weight gain and body composition of young channel catfish Ictalurus punctatus. Aquaculture 163, 297–307.

Lim, C., & Dominy, W. (1989). Amirican soybean Association utilization of plant proteins by warmwater fish. USDA.ARS. Tropical aquaculture research unit.The oceanic institute. Hawai.

LIU, X. J. (2017.). Hic74, a novel alanine and glycine rich matrix protein related to nacreous layer formation in the mollusc Hyriopsis cumingii[J]. Aquaculture and Fisheries, vol. 2, no. 3, pp. 119-123.

Lovell, R. (2002). Diet and fish husbandry. In: Halver, J.E., Hardy, R.W. (Eds.), Fish Nutrition. Academic Press, New York, NY, pp. 703–754.



Mohammad, M. A. (2016). Effect of using soaking. germination and cooking for common vetch Vicia sativa seeds on growth performance of common carp Cyprinus carpio L. Ibn-Al-Haitham Journal for Pure and Applied Science. Vol. 29(1):7-15|Biology.

Nang Thu et al. (2009). Protein level does not affect lysine utilization efficiency at marginal lysine intake in growing rainbow trout (Oncorhynchus mykiss) fry. Aquaculture 288 (2009) 312–320.

Nang Thu, T. P. (2007). Comparison of the lysine utilization efficiency in different plant protein sources supplemented with L-lysine.HCl in rainbow trout (Oncorhynchus mykiss) fry. Aquaculture 272, 4.

NRC . (2011). Nutrient requirements of fish and shrimp Animal Nutrition Series, National Research Council of the National Academies. The National Academies Press, Washington, D.C., USA (2011). (376 pp.).

Ringo, E. S. (1995). Intestinal micro flora of Salmonids", a review . Aquacult. Res, 26:773-789, .

SILVA, L. (2008). glutamina e l-glutamato em dietas para tilápias do nilo (Oreochromis niloticus). . 50f. Tese (Doutorado em Zootecnia) - Programa de Pós-Graduação em Zootecnia, Universidade Estadual de Maringá, Maringá, 2008.

Tacon, A. (2004). Use of fish meal and fish oil in aquaculture: a global perspective. Aquatic. Resour. Cult. Devel. 1:3-14.

Taleb, & Hassan. (2013). The effect of using different levels of methionine on some of the Japanese yur's productivity indicators. Damascus University Journal of Agricultural Sciences - (2013) Volume (29) Issue 3 - Pages: 227-240.

Thompson, K. M. (2005). Evaluation of practical diets containing different protein levels. with or without fish meal. for juvenile Australian red claw crayfish (Cherax quadricarinatus). Aquaculture. 244 (1-4):241-.

Viola, S. L. (1991). Effects of lysine supplementation in practical carp feeds on total protein sparing and reduction of pollution. Isr. J. Aquacult.-Bamidgeh 43, 112–118.



WEBSTER, C. T. (1999). Effect of partial and total replacement of fish meal on growth and body composition of sunshine bass Moronechrysops × M. saxatilis fed practical diets. Journal of the World Aquaculture Society,.

Yamamoto, T. S. (2004.). Antagonistic effects of branched-chain amino acids induced by excess protein-bound leucine in diets for rainbow trout (Oncorhynchus mykiss). Aquaculture 232, 539–550.

Yamamoto, T. S. (2005). Essential amino acid supplementation to fish meal-based diets with low protein to energy ratios improves the protein utilization in juvenile rainbow trout Oncorhynchus mykiss. Aquaculture 246, 379–391.

Zootec, R. B. (2010). Nutritional innovations on amino acids supplementation in Nile tilapia diets vol.39 supl.spe Viçosa https://doi.org/10.1590/S1516-35982010001300010.

ZUO, R. H. (2017). Higher dietary protein increases growth performance, antioxidative enzymes activity and transcription of heat shock protein 70 in the juvenile sea urchin (Strongylocentrotus interm.