A REVIEW ON THE USE OF DISTILLERS DRIED GRAINS WITH SOLUBLES (DDGS) IN AQUACULTURE FEEDS

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Abstract

Distillers dried grains with solubles (DDGS) to replace fishmeal (FM) or other plant protein sources and its effects on an aquatic animal's growth performance and health system is rarely discussed. In recent times FM use in aquafeed has been a problem for the aquaculture industry because of the depletion of fish resources in the ocean and the high cost of FM. For that reason, researchers are focusing on reducing the cost of aquafeed production by lowering the inclusion of FM in aquafeed. One of the low-cost alternative protein sources to replace FM is DDGS. The present review discusses the compilation of available literature review and other works on the use of DDGS to replace FM in aquafeed and how it improves the growth performance, immune health system, and disease resistance in an aquatic animal. It focuses on the production of DDGS, the nutritional composition of DDGS, the chemical and physical characteristics of DDGS, and the effects of the DDGS inclusion in aquafeed on the aquatic animal. Apart from improving the growth performance of the aquatic animal when included in aquafeed, it also helps improve the immune health system of the animal. It also helps boost the immune system of the aquatic animal to fight diseases and increase disease resistance when included in aquafeed. Furthermore, DDGS also contains some constituents like carotenoids that help improve the meat quality of the animal when included in aquafeed. The current review also discusses the management and use of technology to improve the aquaculture industry, educate farmers on reducing diseases during the culturing periods, and increase productivity and profit in a friendly environment.

Key words: aquafeed, aquatic animals, distillers dried grains with solubles (DDGS), growth performance, immune health system, mycotoxins

The new millennium has seen an increase in the global aquaculture production but not with the pace in the increase rate seen in the 1980s and 1990s (Gjedrem and Robinson, 2014). The annual aquaculture production has increased at an average rate

of 8% (Kord et al., 2021). The rate of increase is higher than all the food production industries in the world (Campbell and Pauly, 2013). The reduction in the wild catches of fishes and the global rise in aquaculture production made people consume more aquaculture products (Loder, 2003). Aquaculture is the fastest-growing segment in global food production. Researchers have predicted aquaculture to overtake capture fisheries globally as a source of fish food (FAO, 2018 a). Aquaculture is said to resolve the deficit in food fish consumption (Cunningham, 2005). The production of aquaculture in 2010 was 60 million tons (\$119 billion US dollars, approximately 90 billion euros), without the inclusion of aquatic plants, algae (FAO, 2012). In 2016, the production of aquaculture reached 80.0 million tonnes (USD 231.6 billion), excluding aquatic plants and algae (FAO, 2018 b; Khalila et al., 2018). The production of capture fisheries has been declining since the 1990s (Gyan et al., 2020 a). Human per capita protein production is 19 kg, and fish alone constitutes 17% of all the animal food protein sources eaten by human beings (FAO, 2014). The increase in food fish production and its consumption requires more scientific research, education, and government intervention to meet its objective and improve food security globally.

Various culturing systems, including extensive culture, semi-intensive culture, and strictly intensive culture, are some of the culturing systems used to culture almost 600 different aquatic species with the various sophisticated technology in marine (Mansour et al., 2017; Khalil et al., 2019), freshwater, or brackish water environment (FAO, 2012). The culturing system primarily used globally is the intensive system for culturing carnivorous and omnivorous aquatic species in the marine environment (Campbell and Pauly, 2013). The inputs needed to culture some of these species, including fishes and shrimp, are aquafeed, water, energy, and available land (Trujillo, 2007). Furthermore, adequate aquafeed supply is the primary input that fish farmers need to consider in aquaculture production (Khalil et al., 2018; Kord et al., 2021). The feed is the holy grail in aquaculture production, and supplying it to the fish in enough quantities will go a long way to ensure absolute success in production. Feed use in aquaculture is on the rise, representing 55% of the global aquaculture production (Tacon and Metian, 2008). This information shows feed is one of the most critical inputs in aquaculture production. The increased demand for aquafeed in recent years results from the increase and development of the aquaculture industry in the world.

Fishmeal (FM) is the expensive protein source used in aquafeed due to its high composition of amino acids (AA), high protein content, palatability, and adequate source of n-3 highly unsaturated fatty acids (Allam et al., 2020). Feed formulators mainly incorporate FM as a major protein source in aquafeed (Khalila et al., 2018). However, the production of world FM has been declining at an average of 5 and 7 million metric tons (Mmt) since the 1990s, as reported by the International Fish Meal and Fish Oil Organization (IFFO – The Marine Ingredients Organisation, 2013). The trend in FM production with increased aquafeed demands for aquaculture and other animal feeds, including poultry feed, has raised the FM price in recent years, which has affected aquaculture production and its profit (Allam et al., 2020). Researchers have predicted that the price of FM will increase at a high record in the year 2025, which might affect aquaculture production and its investment (Allam et

al., 2020). This problem makes feed scientists research into finding an alternative protein source with similar nutritional qualities as FM and is less expensive than FM and is available everywhere in the world (Khalila et al., 2018). IFFO – The Marine Ingredients Organisation (2013) reported that aquaculturists used 10% FM produced in the 1980s for aquafeed. Fast forward in 2012, aquaculturists used 70% FM for aquaculture production, which is an increase of more than 200% of FM used.

The use of other plant protein (PP) such as wheat gluten meal (WGM), sunflower meal (SFM), soybean meal (SBM), or soy protein concentrate (SPC) has some special effects on the health of the animal species to some degree (Hassaan et al., 2018; Ray et al., 2020; Ray et al., 2019). Consequently, in replacing FM with other PP, not only the growth of the animal should be a worry; instead, health is also another vital issue to think through (Ray et al., 2020). The increasing use of PP also causes competition in demand and supply with human consumption and international trade, increasing prices and raising questions on its sustainability concerning the environment (Matos et al., 2017). For these reasons mentioned above, using by-products obtained from pharmaceuticals and fermentation industries is an excellent source to consider in replacing FM and other PP in aquafeed (Matos et al., 2017).

Distillers dried grains with solubles (DDGS) are among the best-underutilized by-products that can substitute FM in an aquafeed (Khalila et al., 2018). They are acquired from the derivatives of cereal fermentation and following distillation for alcohol production (Goda et al., 2019). The use of DDGS to replace FM or other plant protein sources and its effects on an aquatic animal's growth performance and health system is one of the hottest topics discussed in aquaculture nutrition (Khalila et al., 2018). Thus, this review aims to discuss other articles to explain how DDGS can affect and modulate growth performance and immune health function for aquatic animals in aquafeed. This review also discusses the physical and chemical characteristics of DDGS and its nutritional composition, and how it affects disease resistance in an aquatic animal.

Distillers dried grains with solubles

The distillers dried grains with solubles are produced in larger quantities by the dry-grind fuel ethanol industry. It is one of the cheapest, excellent, and alternative feed ingredients in aquafeed (Allam et al., 2020). The ethanol industry involved in the production of cereal grain of alcohol has become prominent in recent times (Gallagher, 2004). The ethanol industry can produce enough DDGS to support the aquaculture industry when the demands become high. The DDGS is made due to the depletion of starch when ethanol is processed, leading to increasing unfermented nutrients (Liu, 2011). Wet milling or dry-grind (DG) processing can produce ethanol by converting maize into ethanol (Belyea et al., 2010). After converting the maize into ethanol, the DG plant further has a coproduct known as DDGS and ethanol. The leading equipment used in ethanol production with large quantities of DDGS is the DG plant (Liu, 2011). DG plant is mainly used during this process because it is less expensive than the wet milling plant. Also, the DG plant encompasses less equipment than wet milling, involving less technical know-how. The nutritional composition of DDGS varies according to the grain used. Also, the causes of the variable DDGS

composition are enormous, including the technique used in the production, the quantity of the condensed soluble added to the grain, fermentation yeast impact on the soluble, and the methodology used as well (Liu, 2011). However, DDGS contains a crude protein of range between 268 and 337 g/kg, crude lipid of range between 35 and 128 g/kg, and an ash content in the range of 28–98 g/kg (Ayadi et al., 2012; Gatlin et al., 2007; Kluth and Rodehutsord, 2010; Oliva-Teles et al., 2015). The inclusion of DDGS in aquafeed makes it possible to replace the traditional protein source: fishmeal (FM) or soybean due to the content of high protein, energy, and phosphorous in it. The inclusion of DDGS in a well-formulated aquafeed could improve the growth performance, health system, and disease resistance in the aquatic animal. DDGS also helps improve the animal's product quality and meat due to some of the constituents in DDGS, such as carotenoids and yeast. These constituents are some of the qualities obtained from DDGS that make it a popular protein source to replace FM and other plant protein in an aquafeed.

The production of ethanol and distillers dried grains with solubles using dry-grind technology

Ethanol and distillers dried grains with solubles are produced mainly by dry-grind (DG) technology because of its ability to produce large quantities of ethanol and to reduce the particle size of the grain, such as maize using its hammer mill (Allam et al., 2020). A hammer mill crushes the maize grain into a flour form. The flour form is mixed with water to form a mash. Saccharification and fermentation (SSF), using the mash, is further processed to produce ethanol. The ethanol yield is affected mainly by the particle size. Due to this reason, producers mostly use grains with fine texture and size to maximize the production of ethanol and DDGS (Kelsall, 1999). *Saccharomyces cerevisiae* is the common microorganism used in the fermentation process (Lin et al., 2014; Lopes et al., 2017). The distillation process is further used in ethanol production afterward. Distillation separates the ethanol produced from water and any other solid suspension, including the mash. Mash is also known as stillage. The separation makes the stillage. The stillage is composed of fiber, oil, proteins, water and this must be further processed and stored for easy transportation.

In the production of DDGS, the first processing step, centrifugation of the stillage, separates the thin stillage (solubles) containing yeast cells and some soluble nutrients. Some of the solubles have high oil levels, increasing the energy density when added to the aquaculture feed. The solubles with other solid residues, known as wet distillers grains with solubles (WDGS), is mixed with the help of a rotary process. Lastly, the by-product of the solubles and WDGS is heated to remove the moisture content at a minimum of 10–12%. Finally, DDGS is produced.

On the other hand, one of the problems facing the production of DDGS is the influence of heat on the amino acids (AA) profile and other qualities in DDGS (Böttger and Südekum, 2018). Excessive heat can degrade the content of AA composition, chemical and physical characteristics of the DDGS. Therefore heat should be regulated and monitored to an optimum temperature during production to avoid the destruction of the chemical and physical qualities of the DDGS. Figure 1 below

shows the processing technique involved in ethanol production and its coproduct DDGS using dry-grind technology.

Dry-grind Technology

Figure 1. Process for the production of maize distillers dried grains with solubles (DDGS) using dry-grind technology

Physical qualities and composition of distillers dried grains with solubles

One of the physical quality factors that is of high value and buyers look out for when buying distillers dried grains with solubles for the export market is its color. Buyers use this to check and analyze the high-quality DDGS from the low quality, and it also helps determine its value. Some of the available colors that are mostly linked or seen on DDGS include golden yellow, light, and dark brown. The instrument used in observing the color of the DDGS in the laboratory includes Hunter Lab or Minolta colorimeters. These are also used more in the food and animal feed industries. They are also sometimes used to determine heat damage during food processing and different feed ingredients (Cromwell et al., 1993; Ferrer et al., 2005). The ethanol industry in the United States usually uses these colorimeters to determine the color variations in the DDGS. Stars (*) measures the darkness or lightness reading (0 = dark, 100 = light). The red color of the DDGS is read by the (*) in the formula. The yellow color is red with the help of b*. In measuring the colors of the DDGS, a* values must range between 5.2 and 10.8, b* must also range between 12.5 and 23.4, and lastly, the values of the L* must range from 36.6 to 50.2 in various grains of the DDGS sources (Rosentrater, 2007). There is no linear correlation between the nutritional composition of the DDGS and its color.

Nevertheless, studies (Cromwell et al., 1993; Fastinger et al., 2006) reported a lower nutritional value of dark DDGS compared to the light DDGS. Less digestibility is mainly observed in DDGS with a dark color than light DDGS (Cromwell et

al., 1993). Low digestibility in DDGS may also lead to reduced growth performance in the aquatic animal when added to the aquafeed. The color of the DDGS is mostly affected by various factors, including dryer type, the temperature used in drying, the quantity of grain added to the solubles before and during the drying, and lastly natural color of the grain used (Welker et al., 2014 a). Also, the color of the DDGS can vary depending on the maize kernels' color since the color of the maize color varies among different varieties. The color of maize DDGS is mostly darker than the blended maize-sorghum DDGS because many sorghum varieties have bronze color on the outside. Furthermore, DDGS that is of high quality, is the one that has a sweet-smelling and a fermented smell (Lim et al., 2011). Other DDGS varieties with dark, cloudy, and a burned smell result from overheating (Lim et al., 2011).

Another physical quality, the particle size of the ingredients, including the grain and particle size uniformity, is essential when selecting the grain source for the DDGS processing and formulation for the aquatic animal (Allam et al., 2020). The particle size of the DDGS is influenced mainly by most factors, including efficient mixing of ingredients, ingredients segregation during handling, palatability, quality of the pellet, bulk density (Welker et al., 2014 a). According to researchers from the University of Minnesota in 2004, the average particle size of DDGS should range between 665 and 737 μm (Liu, 2009). This indicates that the particle size of DDGS above 737 μm is not optimum for use; therefore, the sizes have to be reduced by feed formulators to be highly utilized when included in aquafeed (Khalila et al., 2018). The particle size of DDGS is likewise an essential factor. Feed processors must address it because particle size can lead to segregation during transportation, which might affect the chemical composition of the DDGS during distribution (Clementson and Ileleji, 2012; Ileleji et al., 2007; Kingsly et al., 2010). Studies done by Fardisi et al. (2019) reported that larvae of fish fed DDGS with smaller particle size grow faster than fish fed with large DDGS size particles. Therefore, the larval period reduces when larvae are fed aquafeed with DDGS made of smaller particle size. Feeding larvae larger particle size of relative humidity (r.h.) of 30% increases the larval development stage, which interns upsurge in the culturing period, affecting the production cost when culturing fish (Fardisi et al., 2019). When feeding their fishes with a smaller particle size of DDGS, farmers may reduce the production cost and increase profit. DDGS, when incorporated in aquafeed and pelleted, is best in comparison to the granulated form when used to feed aquatic animals because it has an improved flowability, reduction in wastage, high bulk density, improving palatability, and high digestibility of energy (Khalila et al., 2018). A study by Rosentrater (2007) measured and observed the feasibility of pelleting DDGS on a laboratory scale and a commercial scale. The authors stated that the nutrients composition, including the amino acid composition in the DDGS, was not affected when they pelleted the DDGS before and after the study (Allam et al., 2020). Furthermore, there was negligible heat damage to the DDGS after pelleting DDGS. Most of the physical qualities of DDGS are almost similar to other feed ingredients such as wheat gluten meal, soybean meal (Khalila et al., 2018). Nutrients digestibility of DDGS is affected by the particle size, temperature, drying process, heating time, and the type of ethanol plant used (Belyea, 2004).

Chemical composition of the distillers dried grains with solubles

Bhadra et al. (2007) reported that the chemical composition and qualities of distillers dried grains with solubles could differ depending on the ethanol plant used. Böttger and Südekum (2018) also reported that the analysis of the qualities of DDGS could vary due to the nature of the production procedure. The use of a drill-mill plant during ethanol production produces ethanol of 1.02 L, a carbon dioxide of 0.28 kg, and a coproduct of 0.82 kg DDGS whenever 2.54 kg of maize has been used in the product. Moreover, maize contains two-thirds of starch, leading to maize converted into ethanol during fermentation (Moreira de Magalhães, 2013). The proximate nutritional composition of the DDGS, including crude protein, ash content, phosphorus, and fiber content, is approximately twice as much contained in maize DDGS than sorghum (Lim and Yildirim, 2008). The authors further observed that the fiber content in maize is not converted to ethanol, increasing the fiber content in DDGS that is approximately an insoluble fiber of 35% and a soluble fiber of 6% (Stein and Shurson, 2009). The apparent total tract digestibility (ATTD) of fiber is 43.7%.

Furthermore, Stein et al. (2005) reported that high fiber content in DDGS might lead to increased energy in DDGS. It eventually leads to increased fish growth when incorporated in aquafeed. Moreover, DDGS contains a phosphorus value of approximately 0.60 to 0.70% (Pedersen et al., 2007). A study by Fardisi et al. (2019) reported that the chemical composition of DDGS, such as the protein, fiber, and ash content, does not affect the larval development of *T. castaneum*.

Furthermore, the chemical characteristics of DDGS are affected by many factors, including the characteristics of the grain used, settings of the hammer mill, different additives used, chemicals used during the production of the DDGS, conditioning (Belyea, 2004). There is a linear correlation between the concentration of protein and non-starch carbohydrates and between the maize used and the DDGS (Liu, 2009). However, as said earlier, there could be vast changes in the nutritional composition and quality of the DDGS produced due to the ethanol plant used and the processing technique (Belyea, 2004). Rosentrater studied the chemical composition and qualities of different DDGS, including the water activities, thermal conductivity, thermal diffusivity, and angle of repose in the United States in 2004 (Rosentrater, 2006). The author stated that the moisture content ranges from 13.4% to 21.2%, the activity of the water in DDGS also was between 0.53 and 0.63. The variations in the chemical qualities of DDGS resulted from several factors such as the processing methods, the proportions used in the production of the DDGS during blending, and drying (Belyea et al., 2004; Cromwell et al., 1993). Different types of DDGS are available in the market because of the grain used during production (Iram et al., 2020). Varieties of grains used to produce DDGS include maize, sorghum, and wheat. In ethanol production, maize is used chiefly during production. Moreover, the nutritional composition of sorghum and barley DDGS, including crude protein and ash content, is slightly lower than the maize DDGS (Lim and Yildirim, 2008). Table 1 below shows the nutritional composition of FM, maize, and sorghum DDGS. Table 1 showed that FM's dry matter content was higher than in the maize and sorghum DDGS. However, maize DDGS also had higher crude fiber than that of the FM and sorghum DDGS. Crude protein content was also high in sorghum DDGS as compared to maize DDGS.

Table 1. Proximate composition for menhaden fish meal (FM) and distillers dried grains with solubles (DDGS) from different grain sources (maize and sorghum) (g/kg dry matter)

Dietary	FM (g/kg)	Maize DDGS (g/kg)	Sorghum DDGS (g/kg)
Dry matter	920	910	890
Crude protein	640	280	340
Fat	92	100	98
Crude fiber	10	110	97
Ash	190	52	42
Phosphorus	30	9	7

Source: Feedstuffs (2011).

Table 2 presents the EAA content of DDGS made from maize, sorghum, and menhaden fish meal. Grains used in the production of DDGS influence its essential amino acids (EAA) composition. Lysine, methionine, and arginine are deficient in DDGS as compared to fishmeal. Yet, supplementation of synthetic AA could help improve the AA profile in DDGS (Welker et al., 2014 a). The EAA, such as lysine, limiting EAA is lower in maize DDGS and sorghum DDGS than in the FM. However, lysine content in maize DDGS was higher than the lysine content in sorghum DDGS. There was more tryptophan in sorghum DDGS as compared to the maize DDGS.

Table 2. The essential amino acid composition of menhaden fish meal (FM), sorghum DDGS and maize DDGS (g/kg)

Dietary	FM (g/kg)	Maize DDGS (g/kg)	Sorghum DDGS (g/kg)
Arginine	38	12	11
Histidine	15	7	7
Isoleucine	27	11	14
Leucine	45	30	42
Lysine	47	9	7
Methionine	18	5	5
Phenylalanine	24	13	17
Threonine	25	10	11
Tryptophan	7	2	4
Valine	32	13	17

Values are expressed as g/kg of dry matter (Feedstuffs, 2011).

Table 3 presents the vitamin and mineral contents of FM and maize and sorghum DDGS. Vitamin and mineral concentrations vary significantly among the maize DDGS and the FM. Maize DDGS contains higher levels of thiamin, riboflavin, and niacin as compared to the FM. Also, maize DDGS contains higher vitamin A than in FM.

Table 3. Vitamin and mineral content of menhaden fish meal (FM) and maize distillers dried grains with solubles (DDGS)

Nutrients	FM	Maize DDGS	Sorghum DDGS
Vitamins			
A (IU)	0	2700	2610
D (IU)	0	0	0
E (mg/kg)	5.27	40	41
B ₁₂ (mg/kg)	0.15	0	0
Thiamin (mg/kg)	0.2	3.5	4
Riboflavin (mg/kg)	4.8	9	5.6
Folic acid (mg/kg)	1	0.88	0.75
Niacin (mg/kg)	55	79.9	80
Pantothenic acid (mg/kg)	8.8	11.4	12.5
Pyridoxine (mg/kg)	0	0	0
Biotin (mg/kg)	0.15	0.3	0.5
Choline (mg/kg)	3080	3400	3200
Minerals			
Sodium (g/kg)	6.8	2	3
Phosphorus (g/kg)	30	7.8	6.2
Potassium (g/kg)	9.6	10	9
Chloride (g/kg)	8	1.7	1.8
Magnesium (g/kg)	2.1	4.2	4.6
Sulfur (g/kg)	4.5	3	2
Manganese (mg/kg)	40.8	30	28
Iron (mg/kg)	880	300	350
Copper (mg/kg)	8	50	50
Zinc (mg/kg)	92	85	85
Selenium (mg/kg)	2	0.35	0.36

Sourced from Welker et al. (2014 b).

Mycotoxins in distillers dried grains with solubles

Mycotoxins are secondary metabolites produced by filamentous fungi, highly toxic to animals, and structurally diverse (Hussein and Brasel, 2001). Mycotoxins such as deoxynivalenol, aflatoxins, zearalenone, T-2 toxins are mostly found in grains including maize, wheat, sorghum (Ensley and Radke, 2019). Mycotoxins found in grain products, including distillers dried grains with solubles, are a highly toxic constituent in crops (Zhang et al., 2009). When DDGS used to feed fish or

shrimp contains these mycotoxins, it may cause high disease outbreak in fish or shrimp, leading to increased economic loss in the aquaculture industry (Jantrarotai et al., 1990; Jantrarotai and Lovell, 1990). Krogh (1978) stated that approximately 25% of the grain supply in the world contains mycotoxins. A previous study on mycotoxins in soybean (SB), maize, and wheat across the different continents reported 81% of the grain samples contained one or two major mycotoxins, including fumonisins, aflatoxin, deoxynivalenol, and zearalenone (Rodrigues and Naehrer, 2012). The major mycotoxin contaminant known as deoxynivalenol is mainly found in DDGS. Wang et al. (2008) reported that DDGS made from maize is likely to contain more mycotoxins than DDGS made from sorghum. Several mycotoxins in DDGS can negatively affect the growth performance of an aquatic animal. Aquatic animals, including rainbow trout, are susceptible to mycotoxins (aflatoxins) as compared to a terrestrial animal; moreover, low concentration of aflatoxins (LD50 = 0.81 and 1.90) can be harmful to the growth and health of rainbow trout (Bauer et al., 1969).

Storing DDGS in any unhygienic condition can increase mold growth and produce more mycotoxins. The production of ethanol does not destroy the content of mycotoxins, neither does it also destroy it during the production of DDGS during the drying process. Nonetheless, the content of mycotoxins is dependent on the area where the grain is grown. Tropical regions with high temperatures produce maize grain containing low content of mycotoxins (Chukwudi et al., 2021). This is because the climatic condition of tropical areas is not favorable for mycotoxin growth, leading to reduced mycotoxin content in DDGS (Chukwudi et al., 2021). However, few processes can help reduce the use of grains containing mycotoxin in the production of DDGS. During the production of DDGS, the ethanol plant used in production can monitor the grain quality and detect and reject any grain containing mycotoxins. This method helps to reduce the presence of mycotoxins in DDGS during production (Tangendjaja, 2008). That said, we recommend researchers to do more studies on the understanding of mycotoxins and their toxicity and threat they pose to the aquaculture industry. That information will help minimize the impact of mycotoxins and their effects on the aquaculture industry when those reports are implemented.

The effects of distillers dried grains with solubles when incorporated in aquafeed on growth performance in aquatic animal

There is an increasing interest for feed formulators about using distillers dried grains with solubles in aquafeed worldwide because of its protein contents, low cost, and high availability worldwide compared to fishmeal. DDGS has attracted more interest in the use of aquafeed because it contains less or no antinutritional factors (ANF) as compared to other plant protein sources, including soybean meal (SBM) and cottonseed meal (CSM). The increase in the ethanol industry indicates there will be enough DDGS produced to meet the demands of DDGS in the aquaculture industry in the years to come. Several studies have reported the use of DDGS in aquafeed and its impact on growth performance in fish and shrimp (Cheng et al., 2003; Coyle et al., 2000; Diógenes et al., 2019; Oliveira et al., 2020 b; Tidwell et al., 1993 a, b).

Distillers dried grains with solubles are highly palatable to aquatic animals, including tilapia and catfish. DDGS, when included in the fish diets, increased feed intake in channel catfish (*Ictalurus punctatus*), striped catfish (*Pangasianodon hypophthalmus*), and tilapia (*Oreochromis niloticus*) (Lim et al., 2009; Pham et al., 2007; Robinson and Li, 2008). The authors reported that the increase in feed intake in fish could be a result of the use of DDGS in the fish diet (Li et al., 2010; Robinson and Li, 2008). A study done by Wu et al. (1994) observed an increase in weight gain for tilapia when 29% of DDGS was used to replace FM in fish diets. Also, when pelleted DDGS was used to feed tilapia in a polyculture system including fresh prawn, authors observed that growth performance, including weight gain, survival rate, and feed conversion ratio, was increased as compared to the fish fed unpelleted DDGS (Tidwell et al., 2007). In another study, Li et al. (2012) fed catfish (*Ictalurus punctatus*) with diets including wheat DDGS at various levels: 0 (diet 1), 10, 20, 30, and 40% without supplementing the feed with an amino acid such as lysine, however, diets 6 to 9 were supplemented with lysine for 12 weeks. The study results observed that the inclusion of wheat DDGS at 20% is suitable for channel catfish and could replace other plant protein sources such as soybean and cornmeal mixture without any adverse effect on the growth performance such as weight gain (Li et al., 2012). The authors suggested that wheat DDGS can be included in the catfish feed up to 40% when AA, such as lysine, is supplemented (Li et al., 2012). The authors proposed that the improvement in the growth performance of the fish could be because of the presence of β -glucan and nucleotides found in DDGS.

In another study, three different experiments were conducted on replacing SBM with various levels of corn DDGS in *P. mesopotamicus* diets and to assess its effects on the fish growth performance and feed utilization, the viability of the DDGS price in the production of the fish diet, and the release of the phosphorus content into the environment. The first experiment was conducted to have six replicates with the initial fish weight of 13 ± 0.3 g mean weight (Oliveira et al., 2020 c). The second experiment was also conducted to have four replicates with the initial fish weight of 21 ± 0.2 g mean weight and fed with various DDGS levels in diets (0, 100, 200, 300, or 400 g/kg) for 100 days (Oliveira et al., 2020 c). Lastly, the third experiment had five replicates with an initial fish weight of 27 ± 1.4 g mean weight and fed with various experimental diets to assess its effects and the amount of phosphorous released into its environment (Oliveira et al., 2020 c). The results from the study confirmed that DDGS is a suitable protein source, and we can use it to replace SBM in the fish diet at 400 g/kg (Oliveira et al., 2020 b). The authors further proposed that the inclusion of DDGS in the fish diet could improve fish's growth performance and reduce the negative impact aquaculture has on the environment (Oliveira et al., 2020 c). The authors explained that the inclusion of DDGS in the fish diet reduced the release of phosphorous in the water, thereby reducing the negative impacts aquaculture has on its environment (Oliveira et al., 2020 c).

Chatvijitkul et al. (2016) assessed the effect of replacing a high protein source cost SBM with a less costly protein source that is lipid extracted (LE)-DDGS in fish feed. LE-DDGS is produced by the extraction of oil from DDGS in ethanol operation (Rausch, 2006). Removing of oil affects the nutritional profile of DDGS primarily

by reducing fat and energy contents and increasing protein content (Rausch, 2006). Replacement of SBM with lipid extracted (LE)-DDGS is possible in hybrid tilapia (*Oreochromis niloticus* × *O. aureus*), and it could replace SBM in the fish diet at 300 g kg⁻¹ without having any negative impact on fish growth (Chatvijitkul et al., 2016). However, the authors observed that LE-DDGS could reduce growth performance in tilapia when a higher concentration is included in the fish diet (Chatvijitkul et al., 2016). The reduced growth in tilapia, when fed with higher LE-DDGS, is because LE-DDGS contained fewer lipids than the normal DDGS. In replacing FM or any other plant protein with DDGS, utilizing amino acids varies depending on the alternative protein source. Therefore the amino acid composition of the feeds has to be considered and analyzed well to avoid poor utilization of the diet by the fish. Allam et al. (2020) suggested that supplementation of amino acids such as lysine in higher DDGS in the fish diet is possible, and it would not have any adverse effects on the fish.

Two major experiments were conducted to evaluate two major co-products of ethanol using maize: maize DDGS and high protein distillers dried grains (HPDDG) as a protein source in fish feed (Overland et al., 2013 a). High protein distillers dried grains are a co-product of ethanol production that uses prefractionation technology and it contains high crude protein content, low fiber, and fat (Prachom et al., 2013). The study assessed the effects of maize DDGS and HPDDG on the growth performance for rainbow trout (*Oncorhynchus mykiss*) after 77 days of feeding trial (Overland et al., 2013 a). In experiment 1, it had a control diet consisting of FM, sunflower meal (SFM), rapeseed meal (RSM), and field peas, and two diets with 250 or 500 g kg⁻¹ DDGS, replacing 50 (DDGS50 diet) or 100% (DDGS100 diet) of the plant protein sources (PP), respectively (Overland et al., 2013 a). In experiment 2, the treatment groups contained SPC, SFM, and RSM, and two diets with 225 and 450 g kg⁻¹ HPDDG, replacing 50 (HPDDG50 diet) and 100% (HPDDG100 diet) of the PP sources, respectively (Overland et al., 2013). The fishes had an initial weight of 143 g. It was observed in experiment 1 that after feeding the fishes with their respective diets, growth performance, including weight gain, feed intake, and feed conversion ratio, was improved significantly in fish fed with DDGS50 diet compared to fishes fed the control diet (Overland et al., 2013). In experiment 2, it was observed that there was no significant difference in the fishes fed the treatment diets and the control diet on the growth performance (Overland et al., 2013). This shows that DDGS and HPDDG is a good protein source and it can be used to replace FM in the fish diet without having any negative impact on fish's growth performance.

Shrimp culture has been one of the fastest increasing species cultured in aquaculture. A study was done to assess the effects of replacing soybean meal with DDGS at various levels at 0%, 10%, 20%, 30%, and 40% replacement levels in shrimp diets. The result confirmed no significant difference in the final weight and survival rate when the authors replaced soybean meal with DDGS at a 40% replacement level (Adedeji et al., 2017). This indicates that optimum DDGS can replace soybean meal in a shrimp diet without negatively impacting growth performance in shrimp. Tidwell et al. did another research to evaluate the effects of DDGS in shrimp diets on the growth performance in freshwater prawn *Macrobrachium rosenbergii* (Tidwell et

al., 1993 a). The juvenile shrimp with an initial average weight of 0.66 ± 0.47 g were fed with various diets containing varying levels of DDGS at 0, 20, or 40%. The result from the study confirmed that survival rate, average yield, feed conversion ratio, and weight gain were not significantly different among the treatment groups (Tidwell et al., 1993 a). This indicates that DDGS is a suitable protein source and can replace FM in the shrimp diet up to 40% (Tidwell et al., 1993 a).

Effects of distillers dried grains with solubles on digestive enzyme activities in aquatic animals

Studies on digestive enzymes are used to predict growth changes and feed utilization in fish and shrimp fed with alternative protein sources (Rungruangsak-Torrissen, 2001). Digestive enzymes are linearly correlated to growth performance in aquatic animals (Hidalgo et al., 1999). The nutritional status of shrimp or fish may improve when digestive enzyme activities are improved, leading to efficient utilization in protein and lipid, resulting in improved growth performance in an aquatic animal (Iehata et al., 2009; Ziaei-Nejad et al., 2006). Nonetheless, few research studies have been done to assess the effects of DDGS on the digestive enzyme activities in fish and shrimp. We propose that researchers do more studies on distillers dried grains with solubles on digestive enzymes in fish and shrimp. This will help increase the utilization of DDGS in aquafeed in aquatic animals. Changes in the digestive enzymes affect the absorption and digestion of the feed in an aquatic animal (Lemieux et al., 1999). A study was done by Moreira de Magalhães (2013) and the authors observed no significant difference in digestive enzyme activities in seabass (*Dicentrarchus labrax*) when DDGS was included in its feed. However, protease activity in juvenile meagre (*Argyrosomus regius*) was significantly increased in the fish fed diet containing various levels of DDGS (Mansour et al., 2017; Khalil et al., 2019). *Piaractus mesopotamicus* juveniles fed various levels of DDGS diets for 100 days revealed that digestive enzyme activities were higher on the anterior intestine than the control group (Oliveira et al., 2020 a). Experimental diets including DDGS decreased the lipase, amylase, and trypsin activities, whereas no differences were detected for total protease activity (Oliveira et al., 2020 a). This shows that DDGS in the fish diet has no adverse effects on the digestive enzyme activities in the fish intestine.

Furthermore, higher amylase activity content was observed in the fish fed diet containing DDGS (Moreira de Magalhães, 2013). This indicates DDGS could improve the digestive enzyme activities in fish fed DDGS diets, leading to an increase in growth performance in fish. The authors reported that the increase in enzyme activities in fish fed DDGS diets could be due to the yeast in DDGS which also serves as a probiotic component, thereby improving the amylase activities (Moreira de Magalhães, 2013).

Beneficial effects of distillers dried grains with solubles on the health of aquatic animals

The welfare of aquatic animals can be determined using blood indices (Lemaire et al., 1991). Replacement of fishmeal with an alternative protein source such as distillers dried grains with solubles profoundly affects metabolism and immune health

in the aquatic animal. So it is essential to check the health condition of the shrimp and fish after it has been fed with an alternative protein source. Immune stress, nutritional and morphological indicators are mostly found in the shrimp hemolymph metabolite and fish blood (de Jesús Becerra-Dorame et al., 2014). The content of biochemical indices and antioxidant activities including acid phosphatase (ACP), alkaline phosphatase (AKP), glucose (GL), total protein (TP), cholesterol (TC), alanine transaminase (ALT), total antioxidant capacity (T-AOC), and superoxide dismutase (SOD), play an essential role and help improve the immune system of fish and shrimp (Sun et al., 2015). An increase in serum metabolites, including ACP and AKP, helps improve the immune system of aquatic animals (Gyan et al., 2006 b).

Some of the constituents contained in DDGS include yeast cells (Böttger and Südekum, 2018). There are approximately 3.9% yeast cells in the biomass of DDGS; however, the protein content of yeast makes up to 5.3% of the total protein of the DDGS (Ingledew, 1999). Some of the nutritional components found in yeast that help enhance the immune system of aquatic animals include proteins, β -glucans, and vitamin B-complex (Oliva-Teles, 2012). Biochemical indices and antioxidant activities, including ACP, AKP, and SOD, increase in aquatic animals when fed with diets containing β -glucans, yeast among others, and these constituents are primarily contained in DDGS as well (Oliva-Teles, 2012). However, the nutritional composition found in the component in DDGS is dependent on the grain used in the production of the DDGS. For example, the content of β -glucan found in wheat DDGS (1.76%) is approximately three times that of the maize DDGS (0.57%). The functions of β -glucan found in yeast in the purified form, or as a yeast by-product, or live yeast include stimulating the immune responses and increasing disease resistance in aquatic animals. Studies on the effects of DDGS in fish and shrimp health systems are few, and we suggest more studies be done in this area. This will help nutritionists understand the impacts of DDGS on fish health and benefit farmers to know the optimum quantity of DDGS that can be included in fish feed to improve fish health.

Corn DDGS in fish diets had significant effects on fish health, including the total serum immunoglobulin and antibody titres in channel catfish (*Ictalurus punctatus*) (Lim et al., 2009). The previous study further observed increased disease resistance when fish fed corn DDGS were infected with *E. ictaluri* compared to the fish fed control diet (Lim et al., 2009). This result shows DDGS has a positive effect on fish health and can also increase disease resistance when included in fish diets. The authors proposed that the increase in the disease resistance in the channel catfish could be because of brewer's yeast and β -glucan and nucleotides found in DDGS (Lim et al., 2009). Firmino et al. (2018) reported that juvenile turbot (*Scophthalmus maximus*) fed different levels of DDGS in fish diets, had no significant differences in total protein plasma, triglyceride, albumin, gluconeogenesis, glycolysis. Dietary treatment diets containing various levels of wheat DDGS did not affect the hematological and immunological parameters in channel catfish (*Ictalurus punctatus*) (Li et al., 2011). Also, the survival rate was not affected after the fish were challenged with *S. iniae* after four weeks (Li et al., 2011). Authors confirmed that 30% of wheat DDGS included in the diet of tilapia without lysine supplementation could help improve the immune health system of the fish and improve disease resistance (Li et al.,

2011). The differences in fish health observed after fish were fed with DDGS could be because of the diets formulation and composition, fish size and species, grain used in the production of DDGS, and the feeding habit of the animals (Pavasovic et al., 2007; Thodesen et al., 2001).

Research in two trials was done to assess the effects of replacing soybean meal with corn DDGS in juvenile *Piaractus mesopotamicus*. The test diets were formulated to include 0, 100, 200, 300, and 400 g of DDGS/kg in the replacement levels in the fish diet. In trial 1, five treatment groups were fed with the experimental diet to assess the effects on the digestibility coefficients (ADC). In trial 2, four treatment groups were provided with the experimental diet to evaluate the effect on digestibility, intestine morphology, and oxidative stress for 100 days. The study revealed that the morphology of the fish intestine was improved when fed with the DDGS diets (Oliveira et al., 2020 a). This actively demonstrates that SBM's harmful effects on fish health would be decreased when fish are fed with DDGS diets instead.

The undesirable effects of distillers dried grains with solubles in aquafeed

Distillers dried grains with solubles have a high content of lipid and crude fiber. This may be undesirable in the aquafeed formulation and affect the dietary protein-energy ratio in fish and shrimp when a high concentration of DDGS is included in aquafeed. Feed utilization may be limited to fish and shrimp because of the high crude fiber content in DDGS. Furthermore, a yellow pigment also known as xanthophyll is predominant in maize DDGS, and its concentration may vary from plant to plant and grain to grain (Lee, 1987). The high concentration of xanthophylls in DDGS may harm the flesh of the fish. A study done by Lee (1987) reported that channel catfish fed a diet containing 11 p.p.m. yellow pigments showed a yellow color in the fish fillet. This yellow color on the fish fillet is undesirable (Lee, 1987). It may affect the fish price, so this component must be considered when DDGS is being included in the fish diet. Another study on the yellow pigment was done, and it was observed that the xanthophylls content is acceptable by fish when it is 7 p.p.m. or lower (Li and Robinson, 2009). However, above 10 p.p.m. may have a negative impact on the fish fillet (Li and Robinson, 2009). Antibiotic residues in DDGS are another factor that has to be considered by feed formulators when DDGS is included in aquafeed. In the ethanol industry, antibiotics including virginiamycin, tylosin (tetracycline), penicillin, and erythromycin have been used to control bacteria growth during fermentation and increase the production of ethanol efficiency (Olmstead, 2009). These antibiotics can produce bacterial strains that are responsible for creating antibiotic resistance in distillers grains by-products. These products are used to feed any aquatic animal, and they might end up in the animal tissues where the final consumer buying the fish might eat it (Paulus, 2011). The national association of feed production has to test some of these components in DDGS before selling in the market. Doing so will help reduce and prevent most of the DDGS that contained these harmful substances in our market. Some ethanol-producing facilities are also using antibiotic-free alternatives in the production of ethanol. This will go a long way to also reduce the presence of antibiotics in DDGS.

Conclusion

In summary, distillers dried grains with solubles are suitable protein sources that can be included in aquafeed. The nutritional composition of DDGS varies depending on the grain used and the ethanol plant used during the fermentation and the drying process. This review revealed that the optimum concentration of DDGS that can be included in aquafeed is dependent on various factors, including the nutritional composition of the DDGS, animal species type, and the other ingredients used. Also, higher concentrations of DDGS could be used to cater to various aquatic animals' dietary needs, such as fish and shrimp, when amino acids such as lysine and methionine are supplemented in the aquafeed. Apart from improving the growth performance of the aquatic animal when included in aquafeed, it also helps improve the immune health system of the aquatic animal. Authors reported that the inclusion of DDGS in aquafeed helps boost the aquatic animal's immune system and increases disease resistance when included in aquafeed. Then again, the high content of lipid and crude fiber found in DDGS may affect the quality of the fish fillet. Several mycotoxins are contained in DDGS, and this constituent can negatively affect the growth performance of an aquatic animal. Therefore, mycotoxins and yellow pigment found in DDGS should be monitored and evaluated in aquafeed to prevent accumulating these toxic pigments in the fillet of the fish or shrimp. Antibiotic residues could also be found in DDGS due to the ethanol facility trying to use it to reduce bacteria growth. But the good news is in recent times, and ethanol producers are trying to use antibiotic-free alternatives, which might help reduce antibiotic residues in DDGS. The authors recommend that future research be done to include high DDGS content in aquaculture feeds and assess its effects on fish health for a more extended period.

Acknowledgments

We would like to thank all the laboratory mates for their immense support and experimental materials given. The Special Fund for Agro-scientific Research financially assisted this work in the Public Interest of China (201003020), the Guangdong Natural Science Foundation of China (2015A030313621; 2016A030313749), the Industry Technology and Development Special Fund Project of Guangdong Province (2013B021100017). We are grateful to the Key Laboratory of Aquatic, Livestock and Poultry Feed Science and Technology in South China, Ministry of Agriculture, for providing technical assistance.

Data availability statement

Data will be available from the corresponding author upon reasonable request.

Conflict of interest

The authors confirmed that there was no conflict of interest in this experiment.

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Received: 18 XI 2020

Accepted: 28 V 2021