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A Review on the Use of Azolla Species in Poultry Production

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ABSTRACT

Aquatic plants are receiving a lot of attention in nutrition research, which is due to their broad range of uses in animal and human food. *Azolla* is one of the commonly used these floating plants. Currently, different *Azolla* species are used as maintainable feed alternatives for cattle, pigs, poultry, and fish as dried flakes or fresh form. In addition to high protein and essential amino acid content of *Azolla*, the fern is rich in other nutrients such as minerals, vitamins, and pigments. There is a discrepancy in the use of *Azolla* to improve the productivity of poultry. However, most studies have indicated that the incorporation of broiler ration with certain levels of *Azolla* promotes feed intake, body weight gain, feed conversion rate, and general health conditions as *Azolla* comprises growth promoters. In layers, the use of *Azolla* improves productivity in terms of egg quantity and quality as *Azolla* contains pigments, minerals, and essential amino acids. Moreover, *Azollas* improves carcass traits at processing. Therefore, this review article provides information on *Azolla* plant, its composition, and the significance of its supplementation for different poultry species.

Key words: Azolla, Broilers, Immunity, Layers, Performance

INTRODUCTION

As a result of declining food resources around the world and increasing their costs, the search for unconventional resources for poultry production has become increasingly important. In addition, there is a serious effort to replace the conventional feed ingredients of poultry ration with cheap items to reduce the costs. Aquatic plant species do not appear to accumulate secondary plant compounds due to their growth habits and therefore offer greater potential than tree leaves as a source of protein for monogastric animals (Bacerra et al., 1995; Wagner, 1997). Several studies had been done on the nutritive value of aquatic plant Azolla. Azolla is a small aquatic and floating fern of the family Azollaceae and the order Pteridophyta. The name is referred to azo (to dry) and allyo (to kill) as the fern can be killed after exposure to drought conditions. In the world, at least eight species of Azolla are known, namely Azolla pinnata, Azolla nilotica, Azolla caroliniana, Azolla japonica, Azolla circinata, Azolla microphylla, Azolla rubra, and Azolla Mexicana, of which most common is Azolla pinnata (Mathur et al., 2013). Azolla is an abundantly available aquatic fern in the stagnant water of ponds, drains, rivers, canals, marshy fields and wetland paddy in tropical and subtropical countries of the world. This fern can be cultivated under natural and controlled environmental conditions (Senthilkumar and Manivannam, 2016). The blue-green symbiotic cyanobacterial partner alga (Anabaena azollae) grows in Azolla leaves cavities (Becking, 1979). Azolla, in turn, provides carbon source, nutrients, and a protective cavity to Anabaena colonies in exchange for fixed high amount of atmospheric dinitrogen as well as growth promoters (Pillai et al., 2005). This special symbiotic relationship makes Azolla a great and protein-rich plant (Mooventhan et al., 2019).

Azolla was originally used as green manure but is now used as mosquito inhibitor, herbicide, water saver and purifier, fertilizer saver (Van Hove and Lejeune, 1996), cough medicine (Raja et al., 2012), saline soils reclaimer (Raja et al., 2012), biogas producer (Van Hove, 1989; Das et al., 1994) and bioremediator (Sood et al., 2012; Yadav et al., 2014). *Azolla* fern is promising in terms of the ease of cultivation, the minimal water for propagation, the rapid biomass production, the growth in unexploited niches, productivity and nutritive value (Singh and Subudhi, 1978; Lumpkin and Plucknett, 1982; Van Hove and Lejeune, 1996; Pillai et al., 2002; Alalade and Iyayi, 2006; Prabina and Kumar, 2010). It is reported that the addition of *Azolla*

in rations significantly reduces the feed cost (Escobin, 1987; Bacerra et al., 1995; Lawas et al., 1998; Sujatha et al., 2013). The bio-composition of *Azolla* makes it one of the cheapest, economic, potential, efficient, and maintainable feed alternatives for livestock and poultry (Pannaerker, 1988; Kathirvelan et al., 2015).

Azolla pinnata meal has long been used successfully for broiler and layer chickens (Castillo et al., 1981; Querubin et al., 1986; Bhuyan et al., 1988; Sreemannarayana et al., 1993; Basak et al., 2002; Alalade and Iyayi, 2006; Balaji et al., 2009; Dhumal et al., 2009; Naghshi et al., 2014), ducks (Escobin, 1987; Bacerra et al., 1995; Lawas et al., 1998; Sujatha et al., 2013; Acharya et al., 2015), quails (Rathod et al., 2013; Shamna et al., 2013; Varadharajan et al., 2019), fish (Nwanna and Falaye, 1997) and rabbits (Wittouck et al., 1992; Sreemannarayana et al., 1993; Sadek et al., 2010; Anitha et al., 2016a). Moreover, dietary Azolla pinnata has been applied for feeding of shrimps, goats, cattle and buffalo calves (Sudaryono, 2006; Hossiny et al., 2008; Indira et al., 2009; Mandal et al., 2012; Rawat et al., 2015), while Azolla filiculoides has been added to diets of sows as protein source replacer (Leterme et al., 2010) and for growing fattening pigs (Bacerra et al., 1995).

Therefore, this review article provides an overview of *Azolla* plant composition and the significance of its supplementation for different poultry species.

Composition of Azolla

Azolla is a rich and potential source of protein (25-35%), nitrogen (Lumpkin, 1984), almost all essential amino acids (7-10%) especially lysine (Van Hove, 1989), essential minerals such as iron, calcium, phosphorous, magnesium, manganese, potassium, iron and copper (10-15%), vitamins like vitamin A and vitamin B12 (Bacerra et al., 1995; Lejeunea et al., 1999), carotenoids, chlorophyll a and b, bio-polymers, probiotics, and growthpromoting intermediates (Ivan and Thuget, 1989; Tamany et al., 1992; Lejeune et al., 2000; Pillai et al., 2005; Alalade and Lyayi, 2006; Lakshmi and Sailaja, 2012; Mathur et al., 2013; Parashuramulu et al., 2013; Cherryl et al., 2014; Henry et al., 2017). Thus, Azolla is considered a significant source of nutrients. Furthermore, cyanobiont of Anabaena azollae contains chlorophyll phycobiliproteins, and carotenoids (Tyagi et al., 1980). However, its contents of carbohydrate and oil are low. Azolla is highly digestible because of lignin and high protein contents (Anitha et al., 2016b). A study by Namra et al. (2010) revealed the presence of high levels of energy in Azolla which is important for both digestion and availability of nutrients. *Azolla* appears to be a possible biofertilizer due to nitrogen contribution to rice crops (Kannaiyan, 1992).

Effects of *Azolla* supplementation on production performance

Broiler chickens

The addition of *Azolla* to the poultry diet economizes production. *Azolla* is a cheap and plentiful alternative plant protein source that improves FCR, energy efficiency, and performance with no adverse effects on livestock, poultry, and humans (Lejeunea et al., 1999; Alalade and Lyayi, 2006; Namra et al., 2010). Although there are inconsistent results, the majority of the data show enhancement of production and reproduction of livestock and poultry fed with diet containing *Azolla*.

Many studies have demonstrated the efficiency of using Azolla at different replacement concentrations in the ration of broiler chickens. However, Ali and Leeson (1995) found that feeding of broilers on Azolla resulted in similar body weight and growth like those kept on a maize-soybean meal. Also, Sarria and Preston (1995) found an increase in the growth of broilers when soybean protein was replaced by Azolla up to 15% level. Sundararaju et al. (1995) investigated the performance modulating effects of dietary Azolla, Sesbania, and Leucaena proteins on broilers and found that the addition of them to broiler chicken diet at a level of 6% significantly improved the body weight at 4, 6 and 8 weeks of age. Improvement in the FCR as a result of Azolla feeding was also reported by Ardakani et al. (1996). Seth et al. (2013) found an increase in efficiency at 5-15% Azolla level in the ration of broilers, while Balaji et al. (2009, 2010); Namra et al. (2010); Ara and Adil (2012); Sujatha et al. (2013); and Naghshi et al. (2014) found this improvement at 5% level. Incorporation of Azolla up to 5% as a feed ingredient to replace sesame meal in the ration of 2 to 6-week-old broilers improved growth rate, FCR, and energy efficiency without deleterious effect on palatability and mortalities (Basak et al., 2002), along with promising economic returns (Parthasarathy et al., 2002). The use of 20% fresh Azolla was suggested as a substitute for commercial feed in chicken diets as it could increase the body weight (Subudhi and Singh, 1978). A significant difference in the feed intake was observed when Azolla increased up to 15% in the diet (Querubin et al., 1986). The study by Dhumal et al. (2009) demonstrated a significant increase in feed intake with an increase in Azolla levels up to 30%. However, Bacerra et al. (1995) indicated a benefit from Azolla supplementation at a low

level of inclusion. Similarly, Ara et al. (2015) found a linear reduction in feed intake with increasing Azolla levels in the diets of broiler chickens. It was found that Azolla pinnata meal can be safely included up to 15% in growing pullet ration with no health issues (Alalade et al., 2007) but 10% inclusion level in pullet chick diet has given the best performance. Incorporation of the concentrate feed with Azolla at 7.5% level resulted in an increase in the body weight up to 2.6% (1.99 kg) compared to control diet (1.93 kg). In addition, the consumed feed was lower in broilers fed on 7.5% Azolla (Prabina and Kumar, 2010). Seth et al. (2013) also detected increased weight gain in Vanaraja chicken fed on 5% or 10% Azolla over control. Saikia et al. (2014) demonstrated that the highest body weight gain was in the group supplemented with 5% Azolla while the lower one was in the group fed with 15% level and they explained that increasing the fiber content in high concentrations of Azolla negatively affects the appetite of the birds and consequently reduces the growth rate. They also concluded that Azolla can be added to the broiler diet by 10% level without adverse effect in the performance. Improvement in body weight gain as well as FCR was detected after the inclusion of Azolla at 5% or 10% level in the feed (Acharya et al., 2015) and up to 7.5% level (Kumar et al., 2018). Moreover, the feed cost of production was considerably reduced by the inclusion of Azolla at either level. The inclusion of Azolla at 10% level showed the maximum economic benefit. A study carried out by Henry et al. (2017) indicated that fresh Azolla supplementation (30)g/bird/day) reduced consumption with no effects on growth performance parameters in 7-week-old turkeys, which might be due to high protein level and mineral content of Azolla. The addition of dried Azolla to Vencobb broilers ration can be used safely up to 5% without any adverse effects, however, 2.5% addition level of Azolla is effective in improving both growth and biochemical parameters (Rana et al., 2017). It was revealed that inoculation of Azolla at levels of 5% or 7% is appropriate for safe and profitable production of broilers due to reduced FCR, mortalities, and production costs, as well as improved net profit (Islam and Nishibor, 2017). The addition of Azolla to the basal ration could improve FCR with no adverse effects on blood biochemistry and immune parameters (Shukla et al., 2018). Recently, Samad et al. (2020) suggested that the addition of Azolla up to 15% level enhances the growth performance traits without negative effects on the nutrient digestibility of broiler chickens.

On the other side, some researchers found no or low effects of Azolla supplementation on poultry production performance. Higher levels compared to low levels of aquatic plants result in lower body weight, which may be related to the high level of neutral detergent fiber (Buckingham et al., 1978) and tannin contents (Tamany et al., 1992) as limiting factors for FCR as well as efficient nutrient utilization (Muzlar et al., 1978). The decrease in the consumed feed may be due to reduced palatability (Bested and Morento, 1985) and increased bulkiness of Azolla (Bacerra et al., 1995) which reduces its utilization. Castillo et al. (1981), Bhuyan et al. (1988), and Sreemannarayana et al. (1993) found that inclusion of Azolla by 15% in broilers ration did not affect feed consumption. The discrepancy in the trend of feed intake could be attributed to the differences in the nutrient composition of the diets and Azolla species used. Moreover, another study on Azolla revealed a proportional decrease in the body weights of broilers (Parthasarathy et al., 2002). The results of Biplob et al. (2002) and Balaji et al. (2009) indicated no changes in the production performance of broiler chickens after feeding on Azolla compared to control. In addition, Alalade and Iyayi (2006) attributed the lower growth rate of broilers fed on high levels of Azolla to the lower feed intake and subsequently the reduction of metabolizable energy intake. Alalade et al. (2007) demonstrated that addition Azolla to the ration of pullets up to 10% level induced non-significant differences in growth parameters.

Limited researches have been conducted on evaluating the effects of *Azolla* supplementation on the carcass traits. Higher dressing percentage of broiler chicken with giblet percentage at 15% was reported in the treatment group fed on 5% *Azolla* (Basak et al., 2002) and this improvement was attributed to the higher body weight gains. Feeding on 5% *Azolla* powder significantly increased the carcass yield percentages of broiler chicks, while lowest percentage was observed in 15% *Azolla* supplemented group (Naghshi et al., 2014) However, Ara et al. (2015) found no adverse effect on carcass traits on feeding *Azolla* up to 20% level.

Ducks

Some studies also showed the effects of *Azolla* as a supplement in the ration of ducks. *Azolla* as an unconventional nutrient source could be added by 10% to the basal diet of white Pekin broiler ducks (Acharya et al., 2015). In the study of Escobin (1987), the author found no differences in the production efficacy among different groups after partial replacement of growing Muscovy ducks ration with *Azolla* at levels of 20, 30, and 40%. In

addition, 15% replacement of soybean meal with *Azolla* increased the daily weight gains of ducks (Bacerra et al., 1995), however, at levels 20, 45, or 60% of *Azolla*, the growth rate was reduced. Similar findings on the production performance of ducks were reported (Lawas et al., 1998; Sujatha et al., 2013). In the same line, Lawas et al. (1998) observed that feeding of Mallard ducks with *Azolla* at levels of 75 or 250 g/bird/day lowered the FCR. In a trial with ducks, Bacerra et al. (1995) demonstrated that by addition of 15 to 60% *Azolla* in diet supplying 15.2-30.3% of the total protein, the FCR decreased with increasing *Azolla* consumption. However, studies with long periods are needed to determine the effect of *Azolla* supplementation on performance at older ages.

Quails

Shamna et al. (2013) reported that feeding of quails on *Azolla* at 5% replacement level of the basal ration enhanced the growth and FCR, also it was more economic than feeding on basal diet alone. Varadharajan et al. (2019) concluded that *Azolla* meal up to 6% can include in quails diet without affecting feed consumption and carcass traits. Rathod et al. (2013) reported non-significant effects of *Azolla* supplementation on feed consumption of Japanese quails.

Layer chickens

The pigmenting ability of group fed with Azolla was demonstrated by its higher Roche fan color score and this could be attributed to the ability of Azolla to pigment the yolk as it is rich in β carotene pigment. A similar effect on yolk color due to the inclusion of Azolla in layer ration was reported by Bastian (1987). Ali and Leeson (1995) found that the addition of Azolla powder to chicken feed significantly improved the carotene status of the chicken and increased egg production. Moreover, Khatun et al. (1999) investigated the replacement of sesame oil in the ration of layers by Azolla 200 g/kg and reported good egg mass output and FCR. A similar better egg production performance was detected by Kannaiyan and Kumar (2005) in terms of higher egg yield after Azolla inclusion at the level of 100 g/bird/day. Lakshmanan et al. (2017) stated supplementation of Azolla in layers diets resulted in increasing egg production, improving the nutrient value as well as saving the concentrated feed. The positive effect of Azolla on eggshell strength was referred to the high calcium content of Azolla. It has been found that the eggshell consists of minerals especially calcium carbonate that deposits in the organic matrix (Austic and Nesheim, 1990). It was reported that inoculation of Azolla in the ration of layer chickens at levels up to 20% had no effect on alanine aminotransferase and aspartate aminotransferase levels, which indicates no toxic effect of *Azolla cristata* supplementation (Ara et al., 2018).

Effects of Azolla supplementation on immunity

Prabina and Kumar (2010) demonstrated higher values of antibody titer against the Ranikhet virus in broilers fed on 10% dried Azolla in comparison with those received 7.5% dried Azolla. The authors explained increased immunity in Azolla treated birds by the ability of the fern to assimilate atmospheric dinitrogen through algal symbionts (Anabaena) in its leaves. The immunemodulatory effect of Azolla meal without any toxicity was detected by Mishra et al. (2016) as birds treated with 5%, 7.5%, and 10% Azolla showed higher cell-mediated immune responses. Contrary results were reported by Sujatha et al. (2013) who noted a non-significant difference between the mean hemagglutination-inhibition antibody titer and anti-mercaptoethanol antibodies titer between the control group and the group fed with 200 g fresh Azolla/chick/day from 45 to 60 weeks in Nicobari fowls. Moreover, the haemagglutinating and immunoglobulin M responses to 1% chicken red blood cells were comparatively better in the choice-feeding group compared to the 5% Azolla-fed group. In turkeys, Bhattacharyya et al. (2016) also reported that antibody response to sheep red blood cells was positively affected by 5.5% Azolla pinnata replacement in the conventional ration.

CONCLUSION

It can be concluded that *Azolla* species can be used as a source of protein and other essential nutrient elements for poultry species. Further research is required to determine the effects of using *Azolla* as a feed supplement in poultry ration.

DECLARATIONS

Competing interests

The author has no conflict of interest.

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