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Utilization of *Azolla Microphylla* as Feed Supplement for Crossbred Cattle

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Abstract

Azolla is a floating aquatic macrophyte belonging to the family of Azollaceae. The fern Azolla, hosts a symbiotic blue green algae Anabaena azollae, which is responsible for the fixation and assimilation of atmospheric nitrogen. Azolla has been reported to be a very good source of protein, essential mineral elements and vitamins for livestock. Out of several species of Azolla, Azolla microphylla has been reported to be best suited for tropical climate and livestock feeding. The mean concentration (% of DM) of organic matter, crude protein, crude fibre, ether extract, total ash, NFE, NDF, ADF and ADL in Azolla microphylla meal were 80.53±0.59, 24.06±0.35, 13.44 ± 1.20, 3.27 ± 0.18, 19.47±0.59, 37.71 ± 1.83,45.52 ±1.93, 30.16 ±1.12 and 8.96±0.56, respectively. A growth trial (105 days) was conducted on ten crossbred male calves. In the treatment group Azolla microphylla was supplemented equivalent to 1 kg fresh azolla/animal. There was no significant difference in DM Intake and digestibility of nutrients. The average daily live-weight gain (ADLG) was significantly ($P<0.05$) higher in the Treatment group (423.6±14.0 g/d)) than in Control group.(389.9±14.9 g/d). The feed conversion efficiency also improved significantly. The effect of Azolla microphylla supplementation in crossbred cattle was also studied in a lactation trial of 90 days. In the treatment group 2kg of fresh Azolla was supplemented over conventional ration. There was no significant difference in total DMI and CP intake. The average daily milk yield (kg/d) was significantly higher ($P<0.01$) in Treatment group by around 11%. The average percentage of milk fat, protein, SNF, total solid and ash showed no significant difference. Total dry matter intake per kg FCM yield was significantly ($P<0.05$) lower in the treatment group

indicating better feed conversion efficiency. It can be concluded from the present study that Azolla can serve as a potential green feed supplement for the dairy cattle for the improvement in productive performance specially where/when green fodder availability is scanty.

Keywords: Azolla, Feed Supplement, Cattle, Milk Production, Growth Performance.

1. Introduction

The estimates by different group of workers have consistently pointed out the deficit of the feed resources for livestock in terms of dry roughages, greens and concentrates. Conventional sources of feeds are not enough to mitigate the shortage of feeds and fodder and to make animal production viable and profitable. The gap between the demand and supply is also increasing. In order to bridge this gap, and to ensure optimum production of livestock throughout the year use of non-conventional feed resources as supplement or replacement of conventional feed without compromising the quality is the area of focus in recent years. There is ample scope for improving the productivity of livestock by better balancing of nutrients and optimizing the utilization of feed resources.

Primary consideration on feed resources must be to identify the feed resources in ample supply to provide the bulk of a ration for the local herd and the supplements (usually high in minerals, vitamins and/or non protein nitrogen and/or protein) needed to balance the animal's nutrition. The former resources are comprised largely of fibrous carbohydrates that require microbial fermentative activity for digestion. The other group of feed resources is the supplements which provide essential nutrients in high concentrations and therefore complement and balance the basal feed resources.

The supplementary resources in India include aquatic macrophytes which have rich nutrient and mineral profile. concentrated sources of minerals (e.g. residue after fermentation of molasses); non protein nitrogen Throughout the world, and particularly in Asia, farmers have harvested naturally produced aquatic plants for a number of purposes including animal feed, green manure and for their family feed resources. The best known of these include the free floating plants; water lettuce (Pistia), water hyacinth (Eichhcornia), duckweed (Lemna) and azolla and some bottom growing plants. In recent years commonly occurring aquatic plant, *Azolla* has become prominent, because of its ability to concentrate minerals on heavily polluted water such as that arising from sewage treatment facilities. However, it has also attracted the attention of scientists because of its apparent high potential as a feed resource for livestock. So some workers also called it as *Green gold mine* due to its high nutritive value and *Super plant* due to its fast growth. (Wagner, 1997).

2. The Fern- *Azolla*

Azolla (mosquito fern, duckweed fern, fairy moss, and water fern) is a small free floating aquatic fern native to Asia, Africa, and the America. It grows in swamps, ditches, and even in lakes and rivers where the water is not turbulent (Lumpkin and Plucknett, 1982).

The name *Azoll*a is derived from the two Greek words, *Azo* (to dry) and *Ollyo* (to kill) thus reflecting that the fern is killed by drought. *Azolla* is a genus of six species of aquatic ferns, the only genus in the family Azollaceae. It grows naturally in stagnant water in drains, canals, ponds, rivers and water bodies including marshy lands with temperature range of 15-35°C.

***Azolla* Taxonomy and Distribution** (Lumpkin and plucknett, 1982)		
Kingdom: Plantae	Division: Pteridophyta	Class: Pteridopsida
Order: Salviniales	Family: Azollaceae	Genus: *Azolla*
The genus *Azolla* consist of two subgenera and six living species		
Subgenus Euazolla include four species:	*1) Azolla filiculoides 2) Azolla caroliniana, 3) Azolla microphylla 4) Azolla mexicana.*	
Subgenus Rizosperma include two species:	*1) Azolla pinnata 2) Azolla nilotica.*	

The fern Azolla has a symbiotic blue green algae *Anabaena azollae*, which is responsible for the fixation and assimilation of atmospheric nitrogen. This fact makes the *Azolla* tend to contain relatively high levels of nitrogen and be an attractive protein source for animal feed, not only the livestock and poultry (Buckingham *et al.*, 1978) but also in aquaculture species (Pantastico *et al.*, 1986). Azolla, in turn, provides the carbon source and favourable environment for the growth and development of the BGA symbiont.

3. Nutritional Assessment of Azolla

3.1 Chemical Composition

As cited by various authors the crude protein content of *Azolla* varies between 15.4 to 27.93, crude fibre content of between 9.07-22.25%, on an average the ether extract value for various species varies between 1.60-5.05 % while total ash was in the range of 10.15-36.10% and NFE values were found to vary between 30.08-52.46%.

Van Hove and Lopez, (1983) noted that the crude protein content of *Azolla* might vary from 13.0 to 34.5%. These variations in the nutrient composition of *Azolla* meal is due to differences in the response of *Azolla* strains to environmental conditions such as temperature, light intensity and soil nutrients which consequently affect their growth morphology and chemical composition.

The cell wall composition of *Azolla* is highly variable depending upon the species and the season of cultivation of Azolla. NDF content of *Azolla* was found to be in the

range of 36.88-70% while ADF was reported to be in range of 25.24-47.08%. Cellulose and hemicelluloses content was found to range between 6.8-36.7% and 10.09 to 17.8 respectively. Lignin was reported to vary between 9.27-28.24% and silica content varies between 4.8-16%.

An experimental azolla production unit have been established at NDRI ERS campus. The fresh Yield of Azolla was around 200-250 g/ sq m/ day. Chemical analysis indicated that it was a fair source of plant protein (210.7-296.7 g kg^{-1} DM). The mean concentration (% of DM) of organic matter, crude protein, crude fibre, ether extract, total ash, NFE, NDF, ADF and ADL in *Azolla microphylla* meal were 80.53±0.59, 24.06±0.35, 13.44 ± 1.20, 3.27 ± 0.18, 19.47±0.59, 37.71 ± 1.83,45.52 ±1.93, 30.16 ±1.12 and 8.96±0.56, respectively.

3.2 Amino acid composition

Sanginga and Van Hove (1989) compared the total nitrogen and amino acid composition of seven Azolla strains at four different growth phases. An *Azolla microphylla* strain was the best source of amino acids so best use as animal feed and an *A. filiculoides* strain the poorest under the cultural conditions used for green manure. Data on the amino acid analysis repoted by Alalade and Lyayi (2006) indicated that lysine, arginine, isoleucine, leucine, phenylalanine, glycine and valine were predominant. However the sulphur-containing amino acids did not meet the recommended value of 3.5g/100g protein. Mandal *et al.* (2012) also reported *Azolla* as rich source of protein (21.6%) with all essential amino acids, including a rich source of lysine, along with arginine and methionine.

3.3 Mineral and vitamin composition

In general *Azolla* was reported to be rich in mineral profile, the fern was found to be a rich source of calcium, phosphorous, potassium, ferrous, copper, magnesium and zinc. Calcium content of Azolla varies between 0.8- 4.99 %, while phosphorus between 0.3-1.3%. Querubin *et al*., (1986) reported the following mineral composition, Ca-2.07%, P-0.77%, Mn-0.27%, Fe-0.25%, Mg-0.17%, Na-0.49%, K-4.93%, cu-17.6 ppm, Zinc-71.8 ppm in *A.microphylla,* Srinivas *et al* (2012) found calcium and phosphorus content of 1.32 and 0.86 % respectively.

Lejeune *et al.* (2000) reported that on fresh material, the carotene content ranged from 206 to 619 mg/kg on a dry matter (DM) basis and differed significantly between strains. Carotene content was maximal during the linear phase of growth and minimal during the stationary phase for the all strains. A 4 months storage at room temperature after 17 h of drying at 60 C lowered carotene content by 69% at a constant rate of 1% per day (from 259 to 79 mg kg/ DM).

4. Azolla as Feed Supplement for Crossbred Cattle

Although farmers, particularly in South East Asia and probably elsewhere had developed the use of *Azoll*a as a source of nutrients for livestock, the actual controlled

experimentation that has been typically used to develop such commercial crops as that of soyabeans or maize for livestock feed has not been undertaken. There are, however, some reports on the use of *Azolla* as feed supplements for fish and livestock. These report dealt with research on fish and domestic animals, in which normal feed protein sources have been replaced by Azolla meal on an iso-nitrogenous basis. The reports on supplementation of *Azolla microphylla* to cross bred cattle are scanty.

4.1 Supplementation of Fresh Azolla Microphylla to Lactating crossbred cattle

Pillai *et al (*2002) in a field trial showed an overall increase of milk yield of about 15 percent when 1.5 - 2 kg of fresh Azolla per day was combined with regular feed. He reported that the increase in the quantity of the milk produced was higher than could be expected based on the nutrient content of azolla alone. He concluded that it is not only the nutrients, but also other components, like carotenoids, bio-polymers, probiotics etc., that contributed to the overall increase in the production of milk.

The effect of fresh *Azolla microphylla* supplementation in crossbred cattle was studied in a lactation trial of 90 days at ERS of NDRI, Kalyani campus . Ten crossbred jersey cattle were distributed into two groups. The animals in control group (T0) were fed on conventional ration of paddy straw, green fodder and concentrate mixture as per recommendation of NRC 2001.In the treatment group (T1) 2kg of fresh Azolla was supplemented over conventional ration.

There was no significant difference in total DMI and CP intake between the two groups. The average milk yield (kg/d) over 6 fortnights was significantly higher ($P<0.01$) in T1(7.14±0.08) than T0 (6.42±0.13). Fat corrected milk(FCM) yield also showed similar trend with average values being 7.04±0.15 and 7.92±0.09 for T0 and T1, respectively. These results showed that supplementing of Azolla caused an increase in milk yield by 11.2% and FCM yield by 12.5%. Total dry matter intake per kg FCM yield was significantly ($P<0.05$) lower in the treatment group than in control group indicating better feed conversion efficiency. The average percentage of milk fat, milk protein , SNF, total solid and ash showed no significant difference between two groups.

4.2 Evaluation of Dried Azolla meal as a potential Feed Supplement for cattle

The fresh biomass of Azolla was harvested at regular interval. The harvested material was washed to remove the extraneous materials and dried under shade to remove moisture and then oven dried at 45^{O} C for 48 hours. The oven-dried sample was ground to prepare Azolla meal.

A growth trial (105 days) was conducted on ten crossbred male calves distributed into two groups. The animals in control group (T_0) were fed with paddy straw (around 45 %),Green fodder (around 15 %) and concentrate mixture (around 40 %). In the treatment group (T_1) dried *Azolla microphylla* meal was supplemented @ 60 g / animal (equivalent to 1kg fresh azolla) replacing 10 % of concentrate mixture. A digestibility trial of six days was conducted towards the end of the growth trial. There was no significant difference in DM Intake and digestibility of nutrients in crossbred calves

when Azolla was supplemented @ 60 g/animal/ d replacing 10 % of the concentrate mixture. The average daily live-weight gain (ADLG) was significantly ($P<0.05$) higher in T_1 (423.6 ± 14.0 g/d)) than in T_0 (389.9 ± 14.9 g/d). The growth rate (ADLG g/d) increased by around 9 % in the treatment group. The feed conversion efficiency has also improved significantly in the Treatment group.

5. Conclusion

Overall, it can be concluded that Azolla can serve as a potential alternative nutrient supplement for the crossbred cattle for the improvement of productivity in terms of growth, milk, meat etc. Further studies should be done on *in sacco* and *in vivo* digestibility of Azolla, in vitro methane production, more on farm/ field trials on Azolla supplementation in lactating animals to see the effect on milk yield and composition especially in terms of fat, protein, fatty acids and CLA content. The hidden factors in Azolla like micro minerals, fatty acid profile, proanthocyanidines, antioxidants *etc.* should be explored further.

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