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Effect of supplementary feeding of fresh azolla (*Azolla pinnata*) on dry matter intake and digestibility of lactating cows

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Abstract
The present investigation entitled "Effect of Supplementary Feeding of Fresh Azolla (Azolla Pinnata) on The Dry Matter Intake and Digestibility of Lactating Cows" was undertaken in the Department of Animal Husbandry and Dairy Science, Dr. P.D.K.V. Akola, Maharashtra. The present investigation was carried out with the main objective to effect of *Azolla* feeding on dry matter intake and digestibility to standard methods of statistical analysis and presented in this chapter in the forms of tables with the implications of the results to the impacts of *Azolla* feeding on dry matter intake and digestibility in cows. For this purpose, 20 lactating cattle of were selected for study and randomly distributed into four groups. The control (T_1) group practice was fed, soyabean straw, Hybrid Napier (Grass) and concentrates whereas treatment (T_2) group was fed same quantity of roughage, concentrate along with 1kg fresh Azolla/cattle/day, (T_3) group was fed same quantity of roughage, concentrate along with 1.5kg fresh Azolla/cattle/day, (T_4) group was fed same quantity of roughage, concentrate along with 2kg fresh Azolla/cattle/day, (T_5) group was fed same quantity of roughage, concentrate along with 2.5 kg fresh Azolla/cattle/day. The feeding trial tasted for four months. During trial daily feed intake and water intake of animal was recorded and found the feed intake was significantly higher in treatment group Also digestibility was significantly higher in treatment group. The cows from azolla incorporated group received significantly more DMI and digestibility over that of control (T_1) group. The average DMI was 8.508, 8.648, 8.706, 8.963 and 8.917 kg/day/cow in T_1, T_2, T_3, T_4 and T_5 groups respectively.

Keywords: *Azolla*, feeding, supplementary, lactating cows, Cattle, feed intake, water intake, digestibility, dry matter

Introduction
In the livestock farming feeding constitutes the largest item of cost for milk production. Therefore, it is important that feed costs are to be possible at lowest level in order to make production profitable. In India ruminants play a vital role in providing nutritional and livelihood security for millions of rural households. Among many factors governing the livestock productivity, feeding accounts for more than 60-70 percent of the total recurring cost and hence qualitative and quantitative improvement in this aspect will usually improve productivity. Conventional sources of feeds are not enough to mitigate the shortage of feeds and fodder and to make animal production viable and profitable in tropical countries. Net deficient of green fodder is around 60 percent, apart from the feed deficit of around 64 percent. In order to fulfil this gap and to ensure optimum production of livestock throughout the year, we have to use of unconventional fed resources as supplement or replacement of conventional feed, but without compromising the quality of nutrients supply. Many researchers have identified many unconventional feed and fodder to maintain the milk production particularly in off season. The search of alternatives to green fodder and concentrates led to a wonderful plant Azolla, which holds the promise of providing a sustainable feed for livestock. Azolla has attracted the attention of scientists as feed resources for livestock and even called it as Green gold mine or super plant due to its high nutritive value and faster growth. Azolla is a small free floating aquatic fern (Varma *et al.*, 2021) [34].
Azolla is very in proteins, essential amino acids, vitamins (vitamin A, vitamin B12, Beta Carotene), growth promoter intermediaries and minerals including calcium, phosp

horous, potassium, ferrous, copper, magnesium. It grows naturally in stagnant water of drains, ponds, rivers and marshy lands. It is most economic and efficient feed substitute for dairy animal and increases milk production in animal. High protein and low lignin content contribute for its high digestibility by dairy cows. Azolla may serve as a valuable protein supplement for many species. (Singh *et al.*, 2017) [30]

Azolla is a promising water fern which is having high production rate and nutritive value. Its use as animal feed has been studied recently. In world, at least eight species are known of Azolla, 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. It can easily be grown with minimum cost of investment. It grows naturally in stagnant water of river, canal, pond etc. Azolla contain relatively high levels of nitrogen due to its symbiosis with blue green algae Anabaena azolla which fixes atmospheric nitrogen. Costly source of protein in ruminant feed i.e., oil seed cake or meal. Moreover, Azolla is an accumulator of mineral matter and contains all macro and micro elements responsible for animal growth and production. Azolla contains all essential amino acids and carotene. Feeding trials on pigs and poultry reported improved feed conversion efficiency, average daily gain, dry matter intake and other growth performances on Azolla supplementation (Roy *et al.*, 2016) [28].

Materials and Methods

The data for present investigation entitled "Effect of Supplementary Feeding of Fresh Azolla (Azolla Pinnata) on The Dry Matter Intake and Digestibility of Lactating Cows" was collected from different non-descript cows which was rearing in the department of animal husbandry and dairy science, Dr. PDKV Akola. Daily feed intake of each animal was recorded for the period of four month.

Cultivation of Azolla

Establish a small pit of 2.0 x 5.0 x 0.2-meter size under the tree shade. Cover this pit with a layer of plastic sac to protect from the intruding root system and then with silpauline sheet (120 G) thickness. Apply 80-90 kg of soil into this silpauline sheet lined pit. Fill the pit with water 10 cm and mix 10-15 kg of cow dung into this. Two kg of *azolla pinnatta* into this water bath and mix this bath once in 2-3 days and remove any excessive fallen tree leaves in order to maintain the pH only in neutral pH the azolla will grow well. Azolla will multiply at a faster rate. once the entire water bath is fully covered by floating azolla then it is ready for harvest. It gives nearly 9-10 kg *azolla* per pit after every 7-8 days, to fulfil daily requirement of *azolla* 20 pits was constructed. (Kumar *et al.* 2020) [21].

Azolla consists of various varieties viz. *Azolla pinnata*, *A. maxicana*, *A. nilotica*. Among them *Azolla pinnata* is the most common Azolla species in tropics and subtropics. It can easily be grown with minimum cost of investment.

Selection of Lactating animals

Twenty Lactating cows were selected from Livestock Instructional Farm, Dept. of AHDS, Dr. PDKV, Akola on the basis of nearness of lactation period and body weight. Selected cows were randomly divided into 5 groups of 4 cows in each Treatment.

Treatment details

The experimental animals feed as per feeding standard to meet the requirement nutrient and details as follow

T_1: Dry roughages ad lib. + Green roughages + Concentrate (Control)

T_2: Dry roughages ad lib. + Green roughages + Concentrate + 1 kg of fresh Azolla

T_3: Dry roughages ad. lib. + Green roughages + Concentrate + 1.5 kg of fresh Azolla

T_4: Dry roughages ad. lib. + Green roughages + Concentrate + 2 kg of fresh Azolla

T_5: Dry roughages ad. lib. + Green roughages + Concentrate + 2.5 kg of fresh Azolla

(Green roughages and concentrates required was calculated and provided on the basis of thumb rule).

Results and Discussion

The present study was conducted to know the effect of supplementary feeding of fresh *Azolla* (*Azolla Pinnata*) on the dry matter intake and digestibility of lactating cows under field condition.

1. Chemical composition of feed stuffs

It is essential to include quality feeds in the ration of animals for maintaining their productivity. The term quality implies to feeds physical texture and chemical composition which has direct impact on its palatability and in tum supply of nutrients to animal body. Therefore, chemical composition is one of the most important indices of nutritive value of feeds. Hence the chemical compositions of different feeds used in the present study are discussed. The chemical composition of different type of feed and fodders ingredients on the laboratory is given in table 1.

Table 1: Chemical composition (%) of experimental feed and fodder (on DM basis)

Sr. No.	Attributes	Soybean straw	Hybrid Napier (Grass)	Concentrate (Sugras)	Fresh Azolla	Dry Azolla
1.	Moisture	9.64	75.73	9.46	89.20	9.70
2.	DM	90.36	24.27	90.54	10.80	90.30
3.	CP	6.80	10.63	18.45	2.67	24.78
4.	CF	39.24	28.38	11.18	0.35	13.09
5.	EE	1.69	2.51	4.79	1.41	3.12
6.	TA	11.96	11.62	3.80	2.06	19.10
7.	NFE	40.31	46.86	61.78	4.31	39.91

The average crude protein content in soyabean straw, napier grass, concentrate, fresh azolla, on dry basis was 6.80, 10.63, 18.45, 24.78 percent respectively. Crude protein content of azolla was more than soyabean straw, napier grass & concentrate content. It was observed from the table that DM content of soyabean straw, napier grass, concentrate, fresh azolla was 90.36, 24.27, 90.54, 10.80 percent respectively. DM of concentrate was more than soyabean straw, napier grass, fresh azolla. The percentage of CF in soyabean straw, napier grass, concentrate, fresh azolla was 39.24, 28.38, 11.18 and 0.35 respectively. CF percent of azolla was less than the soyabean straw, napier grass but more than the concentrate mix. The percentage of EE and NFE in soyabean straw, napier grass, fresh azolla, concentrate was 1.69, 2.51, 1.41, 4.79 and 40.31, 46.86, 4.31, 61.78, respectively. The EE and NFE content of concentrate was more than of fresh azolla, soybean straw, napier grass. The TA of soyabean straw, napier grass,

concentrate, fresh azolla was 11.96, 11.62, 3.80, and 2.06 respectively. The TA percentage of napier grass is more than soybean straw, fresh azolla and concentrate.

The Dry Matter (DM) content (%) of sun-dried *Azolla pinnata* was 90.30 percent which was similar to the values reported by Cherryl *et al.*, (2014) [12] i.e. 89.73 percent, Navnath (2015) i.e. 89.91 percent and D. Srinivas *et al.*, (2015) i.e. 93.00 percent in sun dried azolla. Crude Protein (CP) content (% DM) of sun-dried *Azolla pinnata* was 24.78 percent which was almost similar with values observed by Anitha *et al.*, (2016) but higher values were reported by Indira and Ravi (2009) [17] and Roy *et al.*, (2016) [28] (28.24 and 32.05 percent). The lower values have been reported by Tamang and Samanta (1993) [32], Alalade and lyayi (2006) [2].

The Ether Extract (EE) content (% DM) of sun-dried A. pinnata was 3.12 percent. The EE value 3.35 percent, 3.33 percent and 3.24 percent was reported by Ghodake *et al.*, (2012) [16]. The lower values (2.36 and 2.71 percent) have been reported by Kumar *et al.*, (2016) [28]

2. Water intake

According to Chaudhary and Gupta (2002) water intake of animal was dependent on many factors like type of diet, quality of roughage, type of animal and physical environment. While Anonymous (2011) mentioned that water consumption was influenced by DM content of feed, level of milk production and climatic conditions. On this background, the results on water intake of cows under different treatments are tabulated in Table 2.

Table 2: Average weekly daily water intake (lit) over experimental period on different treatments

Weeks	Week Wise Mean Values of Treatment (Water Intake)					Pooled Mean
	T1	T2	T3	T4	T5	
Week 1	29.30	28.44	27.75	27.94	27.55	28.20
Week 2	29.45	27.94	27.66	26.88	27.44	27.87
Week 3	30.21	27.64	27.94	26.84	27.32	27.99
Week 4	28.54	26.64	26.76	26.63	27.28	27.17
Week 5	27.69	25.66	26.92	27.74	26.97	27.00
Week 6	26.31	26.55	26.07	25.99	26.86	26.36
Week 7	25.31	27.64	26.95	27.83	26.65	26.88
Week 8	29.64	26.98	26.74	27.77	26.89	27.60
Week 9	28.71	27.97	26.59	27.94	26.86	27.61
Week 10	27.31	27.98	26.65	26.99	25.87	26.96
Week 11	24.10	25.85	27.75	25.65	25.86	25.84
Week 12	28.46	26.85	26.84	26.94	26.97	27.21
Week 13	29.34	28.94	27.94	26.79	26.94	27.99
Week 14	27.12	27.91	27.88	26.87	25.97	27.15
Week 15	26.15	27.71	26.77	26.95	26.64	26.84
Week 16	23.94	25.97	27.87	26.65	26.97	26.28
Week 17	24.15	26.54	26.55	25.8	26.96	26.00
Pooled Mean	27.40	27.25	27.15	26.95	26.82	GM = 460.95

F Test	NS
SE(M)±	0.827

The feeding trial was conducting during early months of winters; therefore, it was though necessary to record the observation on water intake of the cows. Water was offered 2 times daily during the period of trial. The data obtained in this regard was statically analyzed.

It is noticed that in the Table 3. daily water intake of the cow was differed significantly between the feeding group, the cows from T_1 group intake more 27.40 than that T_2 (27.25), T_3 (27.15), T_4 (26.95), T_5 (26.82). This trend indicate that the level of azolla had little effect on the water consumption of cow. The feeding of azolla reduced the water intake as fresh azolla contains nearly 89.20 percent moisture is the first reason and second reason is the feeding trail was conducted during early months of winters therefore the water intake seems to be a little bit lower.

Table 3: Effect of supplementary feeding of fresh azolla on water intake of lactating cows

Treatment	Average body weight (kg)	Water intake/day/cow (lit)	Water intake/cow/100 kg body weight (lit)
T_1	301.25	27.40	9.09
T_2	302.5	27.25	9.00
T_3	300	27.15	9.05
T_4	303.75	26.95	8.87
T_5	301	26.82	8.91
F Test		NS	NS
S.E (±M)		0.95	0.67

The present intake values similarly trend was observed nearer to the values reported by past research work Jadhav (1997) reported that the effect of Anjan tree leaves on quality and quantity of milk in crossbreed cows and observed that the average daily water intake was 24.475, 28.256 and 29.561 liter in T_0, T_1 and T_2 treatments respectively. The difference among different treatment groups were statistically significant (p<0.05) the water intake was noticed more in T_2 followed by T_1 and T_0 when

crossbred heifers were fed lucerne green, jowar straw and concentrate mixture with azolla.

Mehla *et al.*, (2005) observed that water consumption in buffaloes was not significantly influenced by source of protein (Cotton seed or mustard cake), which supports the present results. On the contrary Chaudhary and Gupta (2002) noticed higher water intake in buffaloes with 5 kg green Jowar over that of increasing level of green roughages in diet. This observation does not agree with present trend.

3. Feed intake

3.1 Intake of azolla and other feeds

The intakes of different feeds are directly related to supply of different nutrients to animal body and subsequently its influence on the performance of the animal. In view of this the intake of different feeds consisting soybean straws, green hybrid napier and concentrate under different treatments are recorded in order to know the response of lactating cows to azolla which is shown in table 4

Table 4: Actual feed intake by experimental cows (kg/day)

Treatment	Soybean Straw			Hybrid Napier		Concentrates	Azolla		Total feed Intake
	Offered	Leftover	Intake	Offered	Intake	Intake	Offered	Intake	
T_1	6	1.51	4.49	5	5	3.58	0	0	13.07
T_2	6	1.48	4.52	5	5	3.59	1	1	14.11
T_3	6	1.43	4.57	5	5	3.54	1.5	1.5	14.61
T_4	6	1.00	5.00	5	5	3.60	2	2	15.60
T_5	6	1.11	4.89	5	5	3.55	2.5	2.5	15.94

Soybean straws, green hybrid napier and concentrate of which greens and concentrates were given in restricted quantity while the cows had liberal opportunity of straws. All the cows were fed with 6 kg of green hybrid napier, out of which negligible amount of refusal was noticed hence the consumption of greens was considered cent percent. Whereas, the concentrates were fed to fulfill the nutritional requirements, therefore the amount offered was consumed cent percent by the cows. On the average the cows consumed 3.58, 3.59, 3.54, 3.60 and 3.55 kg of concentrates, 4.49, 4.52, 4.57, 5.00 and 4.89 kg soyabean straw, 5 kg hybrid napier under T_1, T_2, T_3, T_4 and T_5 treatments respectively, indicating there was decrease in the concentrates as a result of inclusion of azolla in the ration of lactating cows. Thus, considering the intake of different feeds, on an average the cows consumed a total feed on fresh basis as 13.07, 14.11, 14.61, 15.60 and 15.94 kg per day under T_1, T_2, T_3, T_4 and T_5 treatments respectively. This trend indicates that the total feed consumption was more in T_5, followed by T_4, T_3, T_2 and T_1. It was observed that the inclusion of azolla in the diet increased the total feed intake.

The data in table 5 indicates that straw intake in cows differed significantly over the experimental period. The straw intake reached to 4.79 kg/day/cow in 1st week of trial from the intake of 4.77 kg/day/cow in 20th week. The straw intake over the weeks was found significantly increasing as well as declining. However, within the experimental period it was noticed that the straw intake was more or less similar in the range of 4.79 to 4.59 kg/day/cow up to 4th week of trial as the differences did not reach the level of significance. The cows increased their intake from 3rd week onward till 6th week of trial and again showed a decreased from 14th to 16th week of the trial.

Table 5: Average weekly dry feed (soybean straw) intake (kg/day/ lactating cow) over experimental period on different treatment

Weeks	Week Wise Mean Values of Treatment (Feed Intake)					Pooled mean
	T_1	T_2	T_3	T_4	T_5	
Week-1	4.59	4.59	4.6	5.24	4.92	4.79
Week-2	4.45	4.58	4.59	5.26	4.96	4.77
Week-3	4.32	4.45	4.47	4.51	4.89	4.53
Week-4	4.49	4.46	4.48	4.53	4.98	4.59
Week-5	4.52	4.57	4.57	4.58	4.87	4.62
Week-6	4.57	4.59	4.57	5.16	4.86	4.75
Week-7	4.38	4.26	4.27	5.05	4.73	4.54
Week-8	4.57	4.36	4.6	4.75	4.87	4.63
Week-9	4.71	4.57	4.58	4.89	4.89	4.73
Week-10	4.58	4.49	4.52	4.85	4.89	4.67
Week-11	4.37	4.59	4.67	5.24	4.89	4.75
Week-12	4.52	4.62	4.65	5.10	4.97	4.77
Week-13	4.47	4.35	4.45	5.12	4.96	4.67
Week-14	4.49	4.51	4.69	5.19	4.89	4.75
Week-15	4.49	4.59	4.68	5.12	4.78	4.73
Week-16	4.51	4.52	4.57	5.16	4.86	4.72
Week-17	4.42	4.63	4.69	5.25	4.87	4.77
Pooled Mean	4.50	4.51	4.57	5.00	4.89	G.M=4.69

F Test	Sig.
SE(M)±	.0528
CD(p<0.05)	0.15850

The data in 6 table indicates that straw intake in cows differed significantly over the experimental period. The straw intake reached to 1.59 kg/day/cow in 1st week of trial from the intake of 1.54 kg/day/cow in 20th week. The straw

intake over the weeks was found significantly increasing as well as declining. However, within the experimental period it was noticed that the straw intake was more or less similar in the range of 1.49 to 1.59 kg/day/cow up to 4th week of trial as the differences did not reach the level of significance. The cows increased their intake from 3rd week onward till 6th week of trial and again showed a decreased from 11th to 13th week of the trial.

Table 6: Average weekly dry feed (soybean straw) intake (kg/day/100 kg body weight) over experimental period on different treatment

Weeks	Weeks Wise Mean Values of Treatment (Feed Intake)					Pooled mean
	T_1	T_2	T_3	T_4	T_5	
Week-1	1.52	1.52	1.53	1.73	1.63	1.59
Week-2	1.47	1.51	1.52	1.73	1.63	1.57
Week-3	1.43	1.46	1.48	1.48	1.62	1.49
Week-4	1.48	1.47	1.48	1.49	1.64	1.51
Week-5	1.49	1.50	1.51	1.50	1.61	1.52
Week-6	1.50	1.51	1.51	1.69	1.60	1.56
Week-7	1.44	1.40	1.41	1.65	1.56	1.49
Week-8	1.50	1.43	1.52	1.55	1.60	1.52
Week-9	1.54	1.50	1.51	1.60	1.61	1.55
Week-10	1.50	1.47	1.49	1.58	1.61	1.53
Week-11	1.43	1.50	1.54	1.70	1.61	1.56
Week-12	1.45	1.51	1.53	1.66	1.63	1.55
Week-13	1.46	1.42	1.46	1.66	1.62	1.52
Week-14	1.46	1.46	1.54	1.67	1.60	1.55
Week-15	1.46	1.49	1.53	1.65	1.56	1.54
Week-16	1.46	1.46	1.49	1.65	1.58	1.53
Week-17	1.43	1.49	1.52	1.68	1.57	1.54
Pooled Mean	1.47	1.48	1.50	1.63	1.60	1.54

F Test	Sig.
SE(M)±	0.0144
CD(p<0.05)	0.0432

3.4 Total feed intake

The azolla was supported with green hybrid napier and concentrates in order to fulfill nutritional requirements of lactating cows. Hence the observations on total feed intake of cows on fresh basis under soyabean straw, hybrid napier, concentrates are tabulated in table 7.

It is seen from table 7 that total feed intake was differing significantly between the feeding treatments. The overall average of total feed intake in cows was 13.07, 14.11, 14.61, 15.60 and 15.93 kg/day/cow under the treatments T_1, T_2, T_3, T_4 and T_5, respectively.

Moreover, irrespective of the treatments the total feed intake showed a significant increase from 14.51 Kg/day/cow in 1st week to 14.69 Kg/day/cow in 17th week of the trial, indicating increase in total feed intake with the advancement of trial. The overall average of total feed intake in cows was 13.07, 14.11, 14.61, 15.60 and 15.93 kg/day/cow under the treatments T_1, T_2, T_3, T_4 and T_5, respectively. This trend does indicate that the total feed intake exhibited a steady increase up to 4th week of the trial and thereafter there was decrease in last 10th week of the trial. The overall averages Total Feed Intake/day/cow under different feeding treatments are tabulated in table 7.

Table 7: Average weekly total feed intake (kg/day/lactating cow) over experimental on different treatments.

Weeks	Week Wise Mean Values of Treatment (Total Feed Intake)					Pooled mean
	T_1	T_2	T_3	T_4	T_5	
Week-1	13.10	13.85	14.51	15.20	15.89	14.51
Week-2	13.14	13.95	14.59	15.40	15.96	14.61
Week-3	13.00	13.97	14.60	15.59	15.96	14.62
Week-4	13.12	14.16	14.61	15.45	15.98	14.66
Week-5	12.80	14.20	14.59	15.06	15.84	14.50
Week-6	13.12	14.28	14.63	15.35	15.82	14.64
Week-7	13.19	14.20	14.45	15.96	15.96	14.75
Week-8	13.05	14.16	14.59	15.87	15.87	14.71
Week-9	13.10	14.14	14.62	15.89	15.89	14.73
Week-10	13.08	14.26	14.68	15.96	15.96	14.79
Week-11	13.07	14.10	14.68	15.75	15.99	14.72
Week-12	13.02	14.06	14.62	15.65	15.96	14.66
Week-13	13.05	14.12	14.65	15.50	15.94	14.65
Week-14	13.06	14.10	14.67	15.60	15.89	14.66
Week-15	13.08	14.05	14.59	15.60	15.93	14.65
Week-16	13.18	14.14	14.60	15.70	15.96	14.72
Week-17	13.08	14.05	14.67	15.70	15.97	14.69
Pooled Mean	13.07	14.11	14.61	15.60	15.93	14.66

F Test	0.0062
SE(M)±	0.0186
CD (p<0.05)	Sig.

3.5 Dry matter intake

The actual intakes of different nutrients through feeds are important to cow for expressing their productivity. Generally wide variation in OM content of feed is observed which has a direct influence on OMI in cows. Therefore, nutrient intake in animals is dependent on DMI rather than intake of fresh basis total feed. On this background the dry matter intake of cows under different treatments over the experimental period were worked out from the total intake of different feeds like dried azolla, soybean straw, hybrid napier and concentrates offered to cows. The overall averages DMI/day/cow under different feeding treatments are tabulated in table 8.

Table 8: Average DMI kg/day of lactating cows in comparison to feeding standard (4.4% BW)

Treatment	Average body weight (kg)	Feed intake dry feed + Green feed + conc.(kg) + Azolla		Percent intake
		Actual Intake (kg)	Requirement as per thumb rule (4.4% BW)	
T_1	301.25	13.07	13.25	102.37
T_2	302.5	14.11	13.31	94.33
T_3	300	14.61	13.20	90.34
T_4	303.75	15.60	13.36	85.64
T_5	301	15.94	13.24	83.06

The value of average dry matter intake kg/body weight of the treatment group T_1, T_2, T_3, T_4 and T_5 were 13.07, 14.11, 14.61, 15.50 and 15.94 kg respectively. The dry matter intake (DMI) through roughages and concentrate with azolla was affected significantly. The cows received 102.37 percent of the requirements under T_1 groups, While T_2, T_3,

T_4 and T_5 cows received 94.33, 90.34, 85.64 and 83.06 percent feed in relation to their requirements indicating a marginal shortage of 5.67, 9.66, 14.36 and 16.94 percent. In contrast T_1 group cows had advantage of excess consumption of 2.37 percent over that of their requirements as per thumb rule norms based on fresh basis. The dry matter intake was noticed more in T_5 followed by T_4, T_3, T_2 and T_1. It indicates the influence of incorporation of azolla in concentrate mixture improves the dry matter intake of the experimental lactating cow. The lowest dry matter intake kg/day was observed under T_1 treatment due to standard feeding.

Nik-Khan and Motaghi (1992) [24] reported inclusions of 35 percent azolla in diet of lactating cows without any adverse effect. Sreemannayana *et al.*, (1993) [31] reported 20 percent of azolla meal in the ration of rabbits without having an adverse effect. Ventura *et al.*, (1994) [33] and Zahid *et al.*, (2001) [36] reported inclusion of seaweed in concentrate mixture without any adverse effect in broilers. Becerra *et al.*, (1995) [9] reported replacement of concentrate mixture by green azolla in diet of ducks and found increased feed intake and weight gain with increase in level of green azolla. Becerra *et al.*, (1990) [8] reported inclusion of azolla in concentrate mixture without any adverse effect in pigs. Tamang and Samanth (1993) [33] reported 20 percent of azolla meal in ration of goats without having any adverse effect. Wadhawani *et al.*, (2007) reported 20 percent of azolla meal in ration of lambs without having any adverse effect. Shital *et al.*, (2011) [29] reported that the average daily dry matter intake (0.35kg) per Osmanabad kid was higher in 15 percent concentrate was replaced with azolla meal without any adverse effect. Debashis *et al.*, (2016) [15] reported that the average dry matter intake (4.51 kg) in control and (4.20 kg) in treatment in 3 months.

It is seen from table 9, dry matter intake (DMI) in lactating cows were influenced significantly by the treatments. The cows from azolla incorporated group received significantly more DMI over that of control group. The average DMI was 8.508, 8.648, 8.706, 8.963 and 8.917 kg/day/cow in T_1, T_2, T_3, T_4 and T_5 groups respectively. This trend goes increases with azolla incorporation, that DMI in cows fed with fresh azolla.

Table 9: Average weekly dry matter intake (kg/day/ lactating cow) over experimental period on different treatment

Weeks	Week Wise Mean Values of Treatment (D.M Intake)					Pooled mean
	T_1	T_2	T_3	T_4	T_5	
Week-1	8.14	8.12	7.58	8.98	9.01	8.37
Week-2	8.24	8.35	7.80	8.94	8.95	8.46
Week-3	8.48	8.79	8.07	8.68	8.83	8.46
Week-4	8.63	8.47	8.22	8.95	8.74	8.56
Week-5	8.56	8.56	8.47	8.56	8.92	8.61
Week-6	8.82	8.84	8.75	8.72	9.61	8.95
Week-7	8.49	8.82	8.95	8.59	9.38	8.85
Week-8	9.06	8.75	9.03	8.95	8.56	8.87
Week-9	9.03	8.49	9.23	9.27	9.70	9.14
Week-10	8.01	8.65	9.44	9.16	8.92	8.75
Week-11	8.15	8.78	9.37	8.82	8.28	8.68
Week-12	8.37	8.60	9.78	8.54	9.89	9.03
Week-13	8.75	8.79	8.53	9.28	8.63	8.80
Week-14	8.42	8.71	8.82	9.56	8.77	8.85
Week-15	8.20	8.63	8.38	8.75	8.33	8.45
Week-16	8.63	8.87	8.73	9.35	8.65	8.85
Week-17	8.66	8.79	8.85	9.27	8.42	8.80
Pooled Mean	8.508	8.648	8.706	8.963	8.917	8.73

F Test	Sig.
SE(M)±	0.0152
CD(p<0.05)	0.0456

Hₒwever, enrichment of soyabean straw with 2 kg of azolla resulted in improving significantly DMI in cows by 5.34 percent as compared to feeding without azolla. This means incorporation of azolla was beneficial to improve the acceptability and thereby the DMI intake in cows. The daily dry matter intake was significantly (p<0.05) higher in T_5 cows as compared to T_1, T_2, T_3 and T_4 group of cows. The increase in dry matter intake is due to improve by balance ration with additional protein of fresh azolla. The similar result was reported by Basak *et al.* (2002) [7] and Alalade and Lyayi (2006) [2], who recorded significant (p<0.05) increased in dry matter intake in azolla supplemented group. The higher dry matter intake was observed by Wadhwani *et al.* (2010) [35] in weaner lambs. The lower and nonsignificant (p>0.05) effect was reported by Roy *et al.* (2016) [28] who recorded the non-significant effect on Hariana heifers when added 5 percent level of Azolla in treatment groups.

Similarly, the results have been found, Kahate (2012) [20] indicated higher DMI by 3.23 and 4.14 percent in kids and crossbred calves, respectively, on feeding untreated SBS over that of feeding with urea treated ration. These observations disagree with the present trend. On the other hand, Bansod (2008) [6] observed higher DMI by 1.15 and 10.54 percent in crossbred heifers and cows respectively, on SBS against Jowar straw based ration and support of greens and concentrates to both the rations. Whereas Mule *et al.*, (2008) [23] and Adangale *et al.*, (2009) [1] reported that feeding of untreated SBS with concentrates had achieved OMI in kids and CB cows as that of offering Jowar straw with concentrates. Similarly, Mudgal *et al.*, (2010) [22] reported that feeding of wheat straw or untreated SBS and wheat straw in 75:25 combinations had no negative effect on DMI in lactating cows. While Chainda *et al.*, (1997) [11]

observed higher DMI in lactating cows on SBS diet against feeding of rice straw ration.

Bandeshwaran *et al.*, (2004) [5] reported that in spite of lower DMI (11.4 kg) on SBS based feed in lactating buffaloes in reference to offering green sorghum and concentrates diet (12.8 kg) was sufficient to meet out ICAR standards. To confirm this contention the DMI in cows was compared with standard norms of 2.5 kg DM/100 kg.

The results of the foregoing pages clearly demonstrated that the cows from all the treatments received the dry matter in sufficient quantity to fulfill the ICAR feeding norms.

However, it seems essential to ascertain whether this intake was proportionate to their body weight or otherwise. In view of this the data on DMI was converted to unit body size and are shown in table 10.

Table 10: Average weekly dry matter intake (kg/day/lactating cow/per 100kg body weight) over experimental period on different treatment

Weeks	Weeks Wise Mean Values of Treatment (Dry matter Intake)					Pooled mean
	T_1	T_2	T_3	T_4	T_5	
Week-1	2.70	2.68	2.52	2.95	2.99	2.77
Week-2	2.72	2.75	2.59	2.93	2.96	2.79
Week-3	2.79	2.89	2.67	2.84	2.91	2.78
Week-4	2.84	2.89	2.72	2.93	2.88	2.82
Week-5	2.82	2.81	2.80	2.80	2.94	2.83
Week-6	2.89	2.95	2.89	2.85	3.16	2.94
Week-7	2.78	2.89	2.95	2.81	3.09	2.90
Week-8	2.97	2.86	2.98	2.93	2.81	2.91
Week-9	2.95	2.89	3.04	3.03	3.19	3.00
Week-10	2.62	2.83	3.11	2.99	2.93	2.87
Week-11	2.66	2.87	3.08	2.88	2.72	2.84
Week-12	2.74	2.81	3.21	2.77	3.24	2.95
Week-13	2.85	2.86	2.79	3.00	2.82	2.86
Week-14	2.74	2.87	2.88	3.08	2.86	2.88
Week-15	2.66	2.87	2.73	2.80	2.71	2.74
Week-16	2.79	2.86	2.83	2.99	2.80	2.85
Week-17	2.80	2.82	2.87	2.95	2.72	2.83
Pooled Mean	2.80	2.85	2.90	2.90	2.90	2.86

F Test	Sig.
SE(M)±	0.0386
CD(p<0.05)	0.1148

It was observed that the average DMI/100kg/BW was 2.80, 2.85, 2.90, 2.90 and 2.86 kg in T_1, T_2, T_3, T_4 and T_5 groups, respectively. However, the cows from T_2, T_3, T_4 and T_5 groups consumed high DM as compare to T_1 group, indicating that significantly highest percent BW, DMI was in T_4, T_3, T_5 followed by T_2 and T_1 groups. Moreover, DMI on feeding azolla was significantly more than that of control group. This means enrichment of azolla was advantageous to increase the DMI per unit body size**.**

It appears from Table 10 that the DMI in lactating cows of all the treatments was sufficient to meet out nutritional norms. On the contrary the lactating cows reared on soyabean straw and fresh azolla ration received more DM by 14.39, 16.08, 18.08 and 18.57 percent in comparison to control group (T_1) 12.98 percent. While, the DMI on control diet in lactating cows was just sufficient to fulfill the recommended norms i.e. excess by 12.98 percent and may not influence adversely the supply of nutrition to cows in order to express optimum performance. As a result, one could expect optimum performance of cows under all feeding treatments on the basis of DMI and in term supply of different nutrients in required amount to cows.

Table 10: Average DMI kg/day of lactating cows in comparison to feeding standard (2.5 kg% BW)

Treatments	BW (Kg)	DMI (Kg)	DM Requirement (kg) (2.5 kg% BW)	Per cent (excess/ deficit) intake
T_1	301.25	8.508	7.53	+ 12.98
T_2	302.5	8.648	7.56	+ 14.39
T_3	300	8.706	7.5	+ 16.08
T_4	303.75	8.963	7.59	+ 18.08
T_5	301	8.917	7.52	+ 18.57

3.9 Daily nutrient intake

The daily nutrient intake of different nutrients in lactating cows under different treatments were determined from the DMI made available from soyabean straw, green hybrid Napier, concentrates and azolla the composition of different feeds. The data in this regard is tabulated in Table 11.

Table 11: Daily intake of different nutrients under different treatments (Kg/day/lactating Cow)

Treatments	DM	CP	CF	EE	NFE
T_1	8.508	1.085	2.387	0.650	4.385
T_2	8.648	1.109	2.421	0.645	4.465
T_3	8.706	1.073	2.387	0.626	4.620
T_4	8.963	1.073	2.514	0.665	4.710
T_5	8.917	1.101	2.513	0.662	4.641

It indicates that, the lactating cows fed with 1kg azolla (T_2) and 2.5 kg azolla (T_5) received significantly more CP against the intake of CP in cows on feeding soyabean straw + green hybrid Napier + concentrates ration (T_1). The average CP intake was 1.085, 1.109, 1.073, 1.073 and 1.101 kg/day/lactating cow in T_1, T_2, T_3, T_4 and T_5 groups. respectively. This means the incorporation of azolla in the ration of cows did not impact on the CP intake in lactating cows, in spite of the lower intakes of DM and concentrates than that of control T_1 diets. Moreover, the CP intake on feeding of azolla to cows was more than offering control (T_1) diet. Borah *et al.*, (1987) [10] and Dahiya *et al.*, (1998) [14] observed more intake of N due to feeding of urea treated paddy straw in CB calves and urea ammoniated berseem straw in buffalo bullocks, respectively. Thus, the CP intake of cows from all the treatments was fulfilling the feeding standard requirement of 680 gm for a cow having 300 kg BW and yielding 4 kg of milk/day.

Crude Fiber intake on different rations it was observed that treatment with 2 kg azolla ration (T_4) supplied significantly more quantity of CF in relation to feeding of T_1 (control), T_2, T_3 and T_5. The average CF intake was 2.387, 2.421, 2.387, 2.514 and 2.513 kg/day in T_1, T_2, T_3, T_4 and T_5 groups respectively. Moreover, CF intake under T_4 group was significantly more as compared to T_1, T_2, T_3 and T_5 groups.

In reference to EE nutrient availability to cows on different treatments. It was noticed that the cows from treatment with 2 kg azolla ration (T_4) received more EE i.e. 0.665/kg/day/lactating cows from the rations. However, 0.650, 0.645, 0.626 and 0.662 kg/d/cow on T_1, T_2, T_3 and T_5 groups, respectively.

There were significant differences between the treatments in reference to providing NFE nutrient to cows. The average NFE intake was 4.385, 4.465, 4.620, 4.710 and 4.641

kg/d/cow under, T_1, T_2, T_3, T_4 and T_5 groups, respectively. This means T_4 cows received significantly more NFE as compared to T_1, T_2, T_3 and T_5 groups respectively.

4. Digestibility coefficient of different nutrients

It is already pointed out in the discussion of foregoing pages that the cows from all the treatments not only received the dry matter in adequate quantity to fulfill their requirement. The digestibility of different nutrients was worked out from the intake of nutrients and nutrients voided in faces. The results obtained in this respect are shown in table 12.

Table 12: Effect of different feeding treatments on digestibility coefficient different nutrients (%)

Treatment	DM	CP	CF	EE	NFE
T_1	57.43	63.32	50.40	71.54	54.96
T_2	61.08	70.78	52.71	78.60	60.60
T_3	61.41	67.31	51.32	79.40	63.68
T_4	65.84	74.09	55.57	81.05	67.30
T_5	63.74	68.66	53.95	76.44	65.76
F Test	Sig.	Sig.	Sig.	Sig.	Sig.
SE(M)±	0.362	0.380	0.385	0.356	0.378
CD(p<0.05)	1.085	1.132	1.150	1.069	1.128

The result revealed that dry matter digestibility differed significantly between the treatments. However non-significant difference in the digestibility co-efficient of DM between the different treatments and establish that inclusion of T_4 2 kg azolla in ration of cows was equally palatable and digestible as that of other dosage of azolla. T_4 is showing 65.84 percent digestibility of DM over the than treatments. This means incorporation of 2 kg azolla in diet of cows improve digestibility. Similarly, results have been shown by Arvindraj (2012) [4] conducted a digestibility trial on growing crossbred male calves supplemented or not supplemented with dried azolla meal. The DM digestibility was 59.25±0.77 & 57.46±0.66 percent and OM digestibility was 62.78±1.25 & 61.34±1.24 percent for the control (T_0) and treatment (T_1) groups, respectively. Statistical analysis of the data showed that the variations in the DM and OM digestibility between the two groups were non-significant. Ghodake *et al.*, (2012) [16] reported higher DM digestibility in Osmanabad kids fed azolla replacing 15 percent of concentrate mixture by weight. Indira *et al.*, (2009) [17] conducted a growth and digestibility trial on male buffalo calves. Control group was fed on hybrid Napier, paddy straw and concentrate mixture and in the treatment group 50 percent of GNC nitrogen of control diet was replaced by azolla nitrogen. The digestibility trial was conducted by TiO_2 indicator method. They obtained a statistically (p<0.01) higher dry matter digestibility coefficient in experimental group when compared to control group. Similarly, Tamang and Samanta (1993) [32] also reported a higher dry matter digestibility in case of experimental diet than control diet in case of black Bengal goats.

Crude protein digestibility differed significantly between the treatments. Inclusion of 2 kg azolla T_4 treatment in ration of cows was equally palatable and digestible as that of other dosage of azolla. T_4 is showing 74.09 percent digestibility of CP over the than treatments. This means incorporation of 2 kg azolla in diet of cows improve digestibility. Arvindraj (2012) [4] reported the average digestibility coefficients for the CP in crossbred male calves in the control group and treatment group were 60.2±5.07 and 59.4±3.02 percent, respectively. Statistical analysis revealed that the difference between the two groups were non-significant. Indira *et al.*, (2009) [17] obtained a statistically (p<0.01) higher crude protein digestibility coefficient in experimental group (58.0 percent) fed azolla than control group (52.0 percent) in case of buffalo calves. Similarly, Ghodake *et al.*, (2012) [16] also reported higher CP digestibility in Osmanabadi kids fed azolla replacing 15 percent of concentrate mixture by weight.

It appears from table 13, incorporation of 2 kg azolla in diet of cows improve digestibility. Indira *et al.*, (2009) [17] also obtained higher EE digestibility in treatment group then control group (57.1 vs. 55.1) on feeding *Azolla pinnata* meal to the buffalo calves. Arvindraj (2012) [4] reported no significant difference in ether extract digestibility in growing crossbred male calves in the control and treatment (azolla supplemented) groups. However, Ghodake *et al.*, (2012) [16] reported lower EE digestibility in Osmanabadi kids fed azolla replacing concentrate mixture @ 15 percent and 30 percent. Higher EE digestibility in the treatment group the present experiment might be due to alteration in rumen microbial activity due to supplementation of dried azolla meal.

Table 13: Details of Digestibility

Sr. No.	Particular	Nutrient	T_1	T_2	T_3	T_4	T_5
1	Average body wt. (Kg)		301.25	302.50	300.00	303.75	301.00
2	DMI/day (kg)		8.508	8.648	8.706	8.963	8.917
3	Daily nutrient intake (kg)	CP	1.085	1.109	1.073	1.073	1.101
		CF	2.387	2.421	2.387	2.514	2.513
		EE	0.650	0.645	0.665	0.665	0.662
		NFE	4.385	4.465	4.620	4.710	4.641
4	Average daily dung (kg)		18.67	18.70	18.75	19.17	19.07
5	DM in dung (%)		18.32	17.80	17.85	18.70	18.63
6	Daily nutrient voided in dung (kg)	DM	3.742	3.366	3.360	3.061	3.254
		CP	0.398	0.324	0.383	0.278	0.345
		CF	1.184	1.145	1.162	1.117	1.178
		EE	0.185	0.138	0.137	0.126	0.142
		NFE	1.975	1.759	1.678	1.540	1.589
7	Daily nutrient retention (kg)	DM	4.766	5.282	5.346	5.902	5.663
		CP	0.687	0.785	0.690	0.795	0.756
		CF	1.203	1.276	1.225	1.397	1.335
		EE	0.465	0.507	0.528	0.539	0.520
		NFE	2.410	2.706	2.942	3.170	3.052
8	Digestibility coefficient (%)	DM	57.43	61.08	61.41	62.46	63.35
		CP	63.32	70.78	67.31	74.09	68.66
		CF	50.40	52.71	51.32	55.57	53.12
		EE	71.54	78.60	79.40	81.05	76.44
		NFE	54.96	60.60	63.68	67.30	65.76

Similarly, Indira *et al.*, (2009) [17] in case of buffalo calves obtained significantly higher CF digestibility in azolla group (62.0 percent) in comparison to control group (58.0 percent). The values were also similar as obtained in the present study. However, Arvindraj (2012) [4] could not find any statistically significant change in CF digestibility on supplementation of azolla in crossbred male calves. Ghodake *et al.*, (2012) [16] also reported no significant difference in CF digestibility in Osmanabadi kids fed azolla replacing concentrate mixture @ 15 percent.

Indira *et al.*, (2009) [17] reported higher NFE digestibility value was (66.06 and 62.0 percent) in control and experimental group in case of buffalo calves. The NFE digestibility (%) in control and treatment groups value was

57.5±1.54 and 57.0±0.93 reported by Arvindraj *et al.*, (2012) [4] without any significant difference in case of growing crossbred male calves.

Conclusion

It is concluded that Daily intake of different nutrients like dry matter, CP, CF, EE, NFE is observed high in group feeding with 2 kg azolla among all other groups. Digestibility of all these nutrients is also high in group feeding with 2 kg azolla among all other groups.

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