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Effect of Partial Replacement of Protein Sun Flower Meal by Azolla Meal as Source of Protein on Productive Performance of Growing Lambs

Abou El-Fadel, M. H.; Hanan A. M. Hassanein* and Heba A. El-Sanafawy



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ABSTRACT



The evaluation of the effect of partial replacing protein of sun flower meal (SFM) by protein of azolla meal in concentrate feed mixture (CFM) on growth performance, digestibility, feed intake and conversion and economic feed efficiency of growing lambs it was the aim of the present study. 15male crossbred (Osimi X Flelandi) were divided into 3 equal groups T1, T2 and T3 fed on CFM containing 0, 10 and 20% azolla meal replacing 0,25 and 50% protein of SFM respectively. Results showed that, integrated azolla meal with experimental rations resulted in decreasing the digestibility of DM,OM,CP and NFE and feeding value as TDN and DCP with R2 and R3 rations compared to those of R1 which had highest means (65.61,72.08, 69.69, 77.45,69.81 and 8.37,respectively). While, digestion coefficients of CF and EE increased with integrated azolla in rations with highest value in R3.T1 recorded the highest average of final body weight (40 kg) and T3 recorded the lowest average (38.4kg) with insignificant differences among tested groups. Also total body weight gain and average daily gain decreased from T1 up to T3 with insignificant differences among experimental groups. Economic feed efficiency(%) recorded the best value(2.28%) in T2 which had 10% azolla meal followed by T3 which had 20% azolla meal and the lower value (2.15) in T1. The results concluded that dietary insert of azolla meal up to level 10% instead of SFM had positive impact on economic feed efficiency of growing crossbred lambs without any adverse effect on the performance of the animals.

Keywords: Azolla meal, Sun flower meal, Growth performance, Economics efficiency, lambs.

INTRODUCTION

In Egypt as in other developing countries, there is a serious problem of feed shortage especially protein sources. (Badawy, 2014) noticed that provision of the quality of protein in lamb's diet does not only improve animal performance but also ensures profitable animal production. Sheep is a multi functional animal and plays a significant role in the economy and nutrition of landless, small and marginal farmers in the country. Sheep rearing is an enterprise which has been practiced by a large section of population in rural areas. Sheep can efficiently survive on available shrubs and trees in adverse harsh environment in low fertility lands where no other crop can be grown.

There are many various varieties of water fern Azolla viz., Azolla pinnata, A. nilotica, A. maxicana. The most important variety of them is Azolla pinnata, it can be grown easily with less initial investment cost. In tropics and subtropics it is commonly found and grows naturally in stagnant water of canals, ponds, drains, rivers and marshy lands. Azolla is a free floating fresh water fern belonging to the family Azollaceae and order Pteridophyta. The water fern Azolla (Azolla pinnata) is an unconventional feed ingredient. It is a potential feed ingredient for growing lambs. It is very low in oil content, but rich in protein the content of total protein is 25-30%, 10-15% mineral content and 7-10%, a combination of amino acids, bio-active substances, biopolymers and carbohydrate (Kathirvelan, et al., 2015). So, Lumpkin (1984) reported that azolla was a potential source of nitrogen and is a potential feed ingredient for livestock. Ahirwar and Leela (2012) found that azolla is containing 28% crude protein and has a potential to be used as a protein supplement in ruminants.

Alalade and Iyayi (2006) found that, arginine, isoleucine, lysine, leucine, glycinephenylalanine, and valine were predominant. On the other hand, the sulphur-containing amino acids did not meet the recommended value of protein. Mandal *et al.* (2012) also reported azolla as a good source of protein (21.6%) with all essential amino acids, including a rich source of lysine, along with arginine and methionine.

Pillai *et al.* (2002) reported that azolla is a free floating water fern which content high level of protein, essential amino acids, vitamin A, B12 and β -carotene, growth promoter intermediate and minerals like calcium, phosphorus, potassium, iron, copper, manganese etc. More over due to high fiber and low lignin content it is easily digested by livestock (Sujatha and Jeyakumar, 2009). Aquatic plant species, because of their growth habit, appear not to accumulate secondary plant compounds and therefore offer a great potential than tree leaves as a source of protein for animals (Becerra *et al.*, 1995). Azolla as a good protein source can partially replace the concentrate for livestock feeding (Shital *et al.*, 2012).

A little work has been done to utilize azolla in poultry ration. But no systematic study has been done to utilize azolla as ruminant animal feed. So in the present study attempt was taken to utilize azolla in growing lambs ration replacing high cost protein source to reduce cost of feeding. The present study aimed to assess the effect of replacing protein of sun flower meal with different levels of azolla meal in CFM on

^{*} Corresponding author. E-mail address: drhanan165@hotmail.com DOI: 10.21608/jappmu.2020.95833

growth performance, feed intake, feed conversion and economic feed efficiency of growing crossbred lambs.

MATERIALS AND METHODS

At Sakha Animal Production Research Station, belonging to Institute Animal Production Research (APRI), Agricultural Research Center (ARC), Egypt this study was conducted, during the period of August till December 2019.

Azolla was collected from canals which sun dried for 15 days. Then it was grinded manually to reduce the particle size and for homogenous mixing with other feed ingredients used for preparation of concentrate feed mixtures. Three concentrate mixtures containing ground azolla at different levels replacing protein of sun flower meal. Fifteen crossbred (Osimi X Flelandi) male lambs weighing 21.3±0.5 kg (6 months of age) were divided into 3 equal groups (five in each group) and designated as T1, T2 and T3 in feeding trail which lasted about 120 days. T1 group was kept as a control and fed on concentrate feed mixture (CFM) containing 20% sun flower meal without azolla meal whereas T2,T3, groups were fed on concentrate feed mixtures containing 10 and 20% azolla meal respectively, replacing 25 and 50% from protein of sun flower meal of control group. All the rations were isonitrogenous and isocaloric. Ingredients composition of different concentrate feed mixtures is presented in Table(1). Measured quantities of concentrate feed mixtures were fed in the morning and in the evening, good quality yellow corn silage was offered in all the groups to meet the nutrient requirement according to NRC (2007). Clean drinking water was provided twice daily. Body weight was recorded (every 14 days) before feeding and amount of feeds were adjusted as per the body weight.

At the end of the feeding trial, three digestibility trials were carried out using three animals/group to estimate the nutrient digestibility coefficients and nutritive values of the tested diets using internal marker (acid insoluble ash AIA) described by Van Keulen and Young (1977). From the rectum of each lamb in each group, faces samples were taken twice daily at (8 a.m. and 6 p.m.) for 5 days collection period. According to AOAC (2005), the representative samples of feed and faces were taken for proximate analysis of CP,CF, EE, DM, and ash

At the end of feeding trials from three lambs in each group (3 animals/group) rumen liquor samples were collected using a rubber stomach tube connected with drawing automatic machine. Collected samples before the morning feeding and 3, 6 hours after the morning feeding, then strained through double layers of cheesecloth. Digital pH meter was using to determined directly ruminal pH value, while ammonia–N (NH3-N) concentration was determined as described by AOAC (1990). Total volatile fatly acids (TVFA's) concentration were determined by steam distillation method as described by Warner (1964).

From juggler vein at the end of the experimental period (3 animals/group) blood samples were taken. For 20 min samples were centrifuged at 3000 rpm to obtain blood plasma which frozen and stored at -20°C to subsequent analysis. Assay of plasma total protein was estimated according to Gornal *et al.* (1949). According to Doumas *et al.* (1971) plasma albumin was determined. By the differences plasma globulin was calculated. Total cholesterol was measured according to Richmond (1973).

Urea estimated according to Fawcett and Scott (1960). According to Young (1990) AST and ALT activities were measured according to Reitman and Frankel (1957).

Table	1.	Ingredient	composition	(%)	of	different
		concentrate	feed mixtures	(CFN	Ð.	

INGREDIENTS	T_1	T_2	T 3				
Undecorticated sun flower meal (SFM)	20	15	10				
Azolla meal	-	10	20				
Soybean meal	-	-	2				
Yellow corn grains	40	31	29				
Wheat bran	31	35	30				
Molasses	5	5	5				
Limestone	3	3	3				
common salt	1	1	1				
Total	100	100	100				

Statistical analyses:

According to the general linear models procedure of SAS (1999) all data were analyzed. By using Duncan's multiple range tests (Duncan, 1955) means were separated for the comparison among them of the tested diets when the main effects were significant. Data of percentages were subjected to arc-sin transformation to approximate normal distribution before being analyzed The model used was:

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where: Y_{ij} = the observation of ij, e_{ij} = the experimental random error, μ =overall mean of Y_{ij} . T_i = Effect of i (treatments),

RESULTS AND DISCUSSION

Chemical composition

The chemical composition for concentrate feed mixtures, azolla meal, sun flower meal, corn silage and different tested rations are presented in Table (2).It showed that the content of EE, CF and ash increased by increasing level of replacing sun flower meal with azolla meal. While, contents of OM and NFE decreased with increasing azolla meal in T_1 up to T_3 . On the other hand, there were slightly differences in contents of DM and CP in T_1 up to T_3 . The chemical composition of three tested rations are showed in Table (2), it was important to show that R₃ had lowest value of DM, OM, CP and NFE, although it had the highest value of CF and ash percentages. The same results obtained by Ahmed et al.(2016) who found that the values of azolla increased in the rations, OM content decreased but CF level increased. This may be due to higher total ash and crude fiber content of azolla.

 Table 2. Chemical composition of tested rations and ingredients (% on DM basis).

Item	DM	OM	СР	CF	EE	NFE	Ash		
Feedstuffs									
Azolla meal	85.08	72.86	18.58	32.17	3.35	18.76	27.14		
SFM	86.94	95.54	37.68	19.89	1.52	36.45	4.46		
Corn Silage	24.64	94.46	4.85	31.30	2.48	55.83	5.54		
Concentrate feed mixtures									
T ₁	88.38	93.00	15.56	7.97	2.86	66.61	7.00		
T ₂	85.01	92.26	15.77	11.72	2.86	61.91	7.74		
T ₃	88.05	83.58	15.64	14.21	3.11	50.62	16.42		
		Expe	rimental	Rations					
\mathbf{R}_1	49.56	93.38	12.01	15.71	2.74	62.92	6.62		
R ₂	49.27	92.93	11.98	18.54	2.73	59.65	7.10		
R ₃	49.22	87.55	11.60	20.62	2.87	52.46	12.45		
T ₁ =Concentra	T_1 =Concentrated feed mixture contained 0% azolla meal,								
Γ_2 =Concentrated feed mixture contained 10% azolla meal,									
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T₂=Concentrated feed mixture contained 10% azolla meal, T₃=Concentrated feed mixture contained 20% azolla meal, SFM= Sun Flower meal, R₁= Ration contain T₁+Corn silage, R₂= Ration contain T₂+Corn silage R₃= Ration contain T₃+Corn silage.

Feeding values and digestion coefficients of tested rations:

Feeding values and nutrients digestibility of tested diets are showed in Table (3). Inserted azolla meal with tested rations resulted in decreasing the digestibility coefficients of DM, OM, CP and NFE and feeding value of TDN and DCP with R_2 and R_3 rations as compared to those of R_1 (control group) which had the highest values (65.61, 72.08, 69.69, 77.45, 69.81 and 8.37, respectively). Control group (R_1) defer significantly (P<0.05) with other tested groups in digestion coefficients of DM, OM, NFE and TDN.

 Table 3. Feeding values and digestion coefficients of tested rations.

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Itom	Tes	+SE					
Item	\mathbf{R}_1	\mathbf{R}_2	R 3	TOL			
	Digestibility	%					
DM	65.61 ^a	59.74 ^b	59.74 ^b	1.29			
OM	72.08 ^a	67.78 ^{ab}	65.24 ^b	1.38			
CP	69.69	66.82	63.39	3.39			
CF	52.25	54.97	60.31	2.69			
EE	72.98	68.71	73.08	3.06			
NFE	77.45 ^a	71.84 ^b	67.17 ^c	0.86			
Nutritive values %							
TDN	69.81 ^a	65.33 ^b	59.75°	1.27			
DCP	8.37	8.02	7.35	0.45			

^{ab} and ^c means in the same row for each parameters with different superscripts are significantly different (P<0.05). SE=Standard error.

On the other hand, there were no significant differences between R₁ and other groups in digestion coefficients of CF. CP. EE and DCP. Digestion coefficients of CF and EE for ration R₃ had higher than the others which had 20% azolla meal (60.31 and 73.08, respectively). These results may be due to the azolla meal had higher content of curd fiber and ash. Shital et al. (2012) reported that integrated azolla meal by level 0, 15 and 25% with experimental rations of Osmanabadi kids resulted in decreasing the digestibility of DM, CP, CF, EE and NFE in diets, with increasing azolla meal level. Ahmed et al. (2016) found that, highest DM digestibility was observed in ration had 6% azolla meal but lowest DM digestibility was observed in ration had 24% azolla meal. However, Reddy et al. (2009) reported significantly higher DM digestibility in azolla based diet than control group. Samanta and Tamang (1995) also observed profuse diarrhea in Black Bengal goats when they were fed on diets containing 50% azolla. They also reported significantly lower NFE digestibility in Black Bengal goats when 20% of the concentrate mixture was replaced by azolla on equi weight basis.

Feed Intake

Average daily feed intake by growing lambs fed the experimental diets is showed in Table (4). As the level of azolla increased in the CFM, DM intake from CFM decreased. Highest average of dry matter (DM) intake from concentrate feed mixture was observed in T_1 (control) group and lowest DM intake from concentrate feed mixture was observed in T_2 group with slightly differences with T_3 group. These results agreement with those reported by Ahmed et al.(2016) Who observed that increasing level of azolla meal in CFM reduced CFMI, DMI. Lowest average of DM intake from corn silage was also observed in T_2 group. This may be due to lower growth rate and body weight in T_2 and T_3 groups in comparison to control group. Becerra et al.(1995) also

reported lower feed intake in growing ducks as the percentage of azolla increased in the diet. Results showed in Table (4) indicated that TDN, CP and DCP intake were higher in T_1 , while T_3 containing 20% Azolla had the lowest averages significant differences (P<0.05) between T_1 and T_3 in TDN intake. These results may be due to the azolla meal had higher percentages of curd fiber and ash. From these results, it could be that introducing Azolla meal in ration of growing lambs tended to lower feed intake as DM, TDN and DCP amounts.

Table 4. Daily fe	ed intake of lambs	from tested rations.
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Item	T_1	T_2	T 3	±SE
CFMI (kg/h/d))	0.758	0.630	0.636	
Corn silage (DM) intake (kg/h/d))	0.380	0.310	0.320	
DMI (kg/h/d))	1.140	0.940	0.950	
TDNI (kg/h/d))	0.688^{a}	0.536 ^{ab}	0.494 ^b	0.05
CPI (kg/h/d))	0.118	0.098	0.095	0.009
DCPI (kg/h/d))	0.080	0.066	0.058	0.008
^a and ^b means in the same	row for a	each naram	eters with	different

superscripts are significantly different (P<0.05). SE=Standard error.

Growth performance

Table (5) were show data of performance of growth and feed conversion ratio (FCR). As the level of azolla increased in the experimental diet, final body weight decreased slightly which T₁ (control group) recorded highest average (40 kg) and T₃ recorded the lowest average (38.4kg) with insignificant differences among tested groups. Also total gain of body weight and average of daily gain of T1 recorded the highest value compared to others. However, significant differences among different groups were not significant as show in Table (5). These results were agreement with those obtained by Ahmad et al. (2016) they noticed that the average of body weight gain decreased with increased level of azolla in growing lambs diets. FCR increased, because though the total DM intake reduced with increased level of azolla in the tested diets. With no significant differences among groups in different FCR parameters. The results showed that adding Azolla meal to ration of growing lambs appeared to lower body weight with higher feed efficiency, especially T2 which containing 10% Azolla.

 Table 5. Growth performance parameters of growing lambs fed the tested rations.

Item	T 1	T_2	T 3	±SE			
Growth performance							
Initial of body weight (kg)	20.20	20.80	22.00	1.26			
Final of body weight (kg)	40.00	39.40	38.40	2.36			
Total body weight gain (kg)	19.80	18.60	16.40	1.71			
Daily of body weight gain (kg)	0.165	0.155	0.137	0.014			
Relative of daily body weight gain%	100	93.94	83.03	-			
Feed	l conversi	on					
DMI kg/kg gain	7.14	6.31	7.19	0.61			
TDNI kg/kg gain	4.38	3.59	3.76	0.58			
CPI kg/kg gain	0.76	0.65	0.72	0.10			
DCP kg/ kg gain	0.518	0.432	0.446	0.075			

Rumen fermentation

Rumen fermentation parameters of lambs fed tested rations are showed in Table (6). The pH means of rumen liquor at zero time were nearly similar and not significant in among different groups with highest value (6.93) in T₃. While, at 3 hour after feeding pH values increased with increasing azolla meal levels by 20% in tested rations with significant differences (p<0.05) between T₃ which had the highest average (5.84) as compared to T1 (control group) which had the lowest average (5.38). However, the differences between azolla meal rations (T₂ and T₃ were insignificant. The increased pH mean with increasing the level of azolla meal in tested rations was probably due to the decreasing fermentation of azolla diets in the rumen than the SFM ones. Also, at 6 hour after feeding pH values recorded the highest level (6.79) with T₃ which differ significantly (p<0.05) with T₁ and T₂.

The concentration of ammonia-nitrogen in rumen liquor of lambs fed T_3 recorded higher level at zero time, 3 and 6 hour after feeding compared with other groups with significant differences (p<0.05) as compared with control group (T_1) at zero time and 3 hour. While, at 6 hour after feeding there insignificant differences among tested groups.

The concentration of total volatile fatty acids (TVFA's) in rumen liquor, did not affected significantly by tested rations. T_2 had the highest values of (TVFA's) at zero time and 6 hour after feeding. While, control group (T_1) had the highest average of TVFA's at 3 hour after feeding. Also, data showed that the finger of pH, NH₃.N and TVF's, increased with passing time at 3hr. in different groups.

Table 6. Rumen parameters of lambs fed tested rations.

Item	Time	T_1	T_2	T 3	±SE
	0	6.89	6.53	6.93	0.30
pН	3	5.38 ^b	5.51 ^{ab}	5.84 ^a	0.12
_	6	6.30 ^b	6.29 ^b	6.79 ^a	0.13
NILI2 NI	0	7.19 ^b	9.18 ^{ab}	11.70 ^a	0.77
INFIO-IN mg/100ml	3	27.64 ^b	39.38 a	40.35 ^a	2.05
mg/100m	6	15.84	21.50	23.76	3.13
TVEAL	0	14.17	14.42	13.55	1.07
IVFAS	3	27.82	22.74	24.57	1.98
nii equiv/100111	6	17.08	18.10	16.53	0.96

 a and b means in the same row for each parameters with different superscripts are significantly different P<0.05). SE=Standard error.

Blood parameters

Some means of blood plasma parameters are showed in Table (7). The tested rations were effected significantly (P<0.05) on most parameters, total proteins, globulin, ALT and AST. Plasma total proteins were decreased with increasing azolla level in CFM with highest value (6.49 g/dI) in control group (T₁), also globulin recorded the same trend of total proteins. Plasma protein fractions are considered as biological index reflecting productive performance and health of animal (Singh and Jha, 2009). On the other side, the highest averages of ALT and AST were observed with T₃ which had 20% azolla meal (26.13 and 48.36 U/L, respectively).

 Table 7. Parameters of blood plasma as affected by feeding tested rations.

Item	T1	T2	T3	±SE
Total protein (g/dl)	6.49 ^a	5.70 ^b	5.68 ^b	0.12
Albumin (g/dl)	3.37	3.36	3.46	0.40
Globulin (g/dl)	3.12 ^a	2.35 ^b	2.22 ^b	0.12
ALT (U/L)	24.67 ^b	25.22 ^{ab}	26.13 ^a	0.28
AST (U/L)	44.81 ^b	46.86 ^a	48.36 ^a	0.58
Urea (mg/dl)	36.45	35.70	32.74	1.76
Total cholesterol (mg/dl)	165.27	171.98	165.12	5.45

 a and b means in the same row for each parameters with different superscripts are significantly different P<0.05). SE=Standard error.

The present data of AST and ALT showed normal activity of the animal hepatic tissue and consequently the integration of azolla meal in the present study could be used without any adverse effect on the liver functions. Singh *et*

al. (2013) reported that energy levels and dietary protein are the most effective factors related to the blood plasma picture. Blood plasma albumin, urea and total cholesterol did not affect significantly by azolla levels in tested rations. In general, the showed concentrations of blood contents are within the normal ranges for healthy lambs which noticed by Kim *et al.* (2012).

Economic feed efficiency

Economics of feeding azolla to growing lambs is presented in Table (8). As the level of azolla increased in the diet, average daily feed cost (L.E.) decreased with highest value in T_1 (control) and the lowest value in T_3 . While, T_2 had lowest average (20.30 L.E.) of cost of feeding per kg of gain with insignificant differences among groups. Price of average daily gain (L.E.) decreased with increasing azolla meal in diets.

Table 8. Economical feed efficiency of growing lambs fed different tested rations.

Item	T_1	T_2	T 3			
Average of daily feed intake, as fed(kg)						
Concentrate feed mixture (CFM)	0.858	0.741	0.722			
Corn silage	1.538	1.278	1.290			
Av. Daily LBW gain(kg)	0.165	0.155	0.137			
Av. Daily feed cost (L.E.)	3.83	3.04	3.00			
Price of LBW gain (LE)	8.25	7.75	6.83			
Feed cost / kg weight gain (L.E.)	24.03	20.30	22.60			
Revenue (LE/ head/day)	4.42	4.71	3.83			
Gross margin above feed cost (LE)	1.15	1.55	1.28			
Economic efficiency (%)	2.15	2.55	2.28			
Improvement of economical efficiency (%)	0	18.60	6.05			
Price of one ton of CFM ₁ , CFM ₂ and CFM ₃ was = 4035, 3815 and 3700 L.E.						
respectively. One ton of SFM = 6000 L.E., Azolla meal = 3000 L.E. and corn						
silage fresh was 500 L.E Hover, price of live body v	veight ga	in =50 l	L.E.			

This may be due to decrease in average of body weight gain with increase level of azolla in the ration. Economic feed efficiency (%) recorded the best value (2.54%) in T_2 which had 10% azolla meal followed by T_3 which had 20% azolla meal and the lower value (2.15) in T_1 (control) group these results may be due to decreases of average daily feed cost with increasing of azolla meal level.

CONCLUSION

It could be observed that experimental animals of T_2 group fed on diet containing 10% of azolla meal replacing 25% from protein of sun flower meal performed tended to higher revenue and economical efficiency. So from the above findings it could be concluded that azolla meal can be added in the diet of growing lambs at 10% level replacing 25% from protein of sun flower meal without any adverse effect on the performance of the animals.

REFERENCES

- Ahirwar, M.K. and Leela, V.(2012). Nutritive value and in vitro degradability of *Azolla pinnata* for ruminants. Indian Veterinary Journal. 89(4):101-102.
- Ahmed, H. A.; Ganai, A.M. and Beigh, Y.A. (2016). Performance of growing sheep on azolla based diet. Indian J. Anim. Res, 50(5): 721-724.
- Alalade, O.A. and Iyayi, E. A. (2006). Chemical composition and the feeding value of Azolla (*Azolla pinnata*) meal for egg-type chicks. Int. J. Poult. Sci. 5: 137-141.
- A.O.A.C. (1990). Association of Official Methods of Analytical Chemists. Official Methods of Analysis. 15th ED Washington D.C.

J. of Animal and Poultry Prod., Mansoura Univ., Vol. 11 (4), April, 2020

- A.O.A.C. (2005). Chapter 4, Animal feed, Official Methods of Analysis of the Association of Official An alytical Chemist International, 18 th edition. Suit 500, 481 North Frederick Avenue, Gaithursburg, Meryland-20877-2417, USA.
- Badawy, H. S. (2014). Utilization of Canola meal as a source of protein for fattening lambs. Egyptian J. Nutrition and Feeds, 17(2):257-273.
- Becerra, M.; Preston, T. R. and Olge, B. (1995). Effect of replacing whole boiled soya beans with azolla in the diets of growing ducks. Livestock Research for Rural Development.7(3):Downloaded from http://www. Irrd. org/Irrd7/3 /7.htm
- Doumas, B.; Watson, W. and Biggs, H. (1971). Albumin standards and measurements of serum with bromocresol green. Clin. Chem. Acta, 3187-94.
- Duncan, D. B. (1955). Multiple ranges and multiple F-Test. Biometrics, 11: 42.
- Fawcett, J. K. and Scott, J. E. (1960). A rapid and precise method for the determination of urea. J. Clin. Path., 13:156-159.
- Gornall, A.G.; Bardawill, C. J. and David, M.M. (1949). Determination of serum proteins by Means of biuret reaction. J. Biol. Chem., 177(2):751-759.
- Kathirvelan C.; Banupriya, S. and Purushothaman, M.R. (2015). Azolla- An Alternate And Sustainable Feed For Livestock. International Journal of Science, environment and Technology, 4,(4):1153 – 1157.
- Kim, S.H.; Alam, M.J.; Gu, M.J.; Park, K.W.; Jeon, C.O.; Jong, K. Ha, ; Cho, K.K. and Lee, S.S. (2012). Effect of Total Mixed Ration with Fermented Feed on Ruminal *In vitro* Fermentation, Growth Performance and Blood Characteristics of Hanwoo Steers. Asian-Australas Journal of Animal Science (AJAS) 27(2):115-122.
- Lumpkin, T.A. (1984). Assessing the potential for Azolla use in the humid tropics. International Rice Commission news, 33: 30-33.
- Mandal R. N.; Pandey, B. K.; Chattopadhyay, D. N. and Ukhopadhyay, K. (2012). Azolla–an aquatic fern of significance to small-scale aquaculture. Aquaculture asia Volume XVII No.1 January – March.
- NRC,(2007). Nutrient Requirements of Small Ruminants: sheep, goats, Cervids and New World Camelids.National Academy of Sciences. Washington, DC.

- Pillai, P.K.; Premalatha, S. and Rajamony, S.(2002). Azolla: A suitable feed for livestock. LEISA India. 4(1): Downloaded from www.leisa.info.
- Reddy, Y.R.; Rao, K.S.; Sudhakar, K.; Guptal, D.R. and Prakash, M.G. (2009) Proceedings of Anima Nutrient Association Conference. Held on 14-17, February, New Delhi; 190
- Reitman, A. and Frankel, S. (1957). Calorimetric method for the determination of serum Alanine transferees and Aspartate transferees. Am. J. Clin. Path., 28:56.
- Richmond, W.(1973). Clin. Chem., 19:1350.*Samanta, G and Y. Tamang (1995). Feeding value of azolla in goats. Ann Zootech 44: 62.
- Samanta, G. and Tamang, Y. (1995).Feeding value of azolla in goats. Ann Zootech, 44:62.
- SAS (1999). SAS User's Guide: Statistics, SAS Institute Inc, Cary, N.C.
- Shital, S. Ghodake; Fernandes, A.P.; Darade, Rohini, V. and Zagade, B.G. (2012). Effect of different levels of Azolla meal on growth performance of Osmanabadi kids. Res. J. Animal Hus. & Dairy Sci., 3 (1): 13-16.
- Singh, V.K.; Pattanaik, A.K.; Goswami, T.K. and Sharma, K. (2013). Effect of varying the energy density of proteinadequate diets on nutrients metabolism, clinical chemistry, immune response and growth of *Muzaffarnagar* lambs. Asian-Australasian Journal of Animal Science, 26(8):1089-1101.
- Singh, A. and Jha, S.K. (2009). Developments in Technology for Fodder Densification. In Walli K., ed . Proceedings of the national symposium on fodder block technology, pp 93-98. Slovak Journal of Animal Science 47.2 (2014:90.99.
- Sujatha, T. and Jeyakumar, S. (2009). Azolla as feed supplement for livestock and backyard poultry. Indian Farming . 59: 22-24 and 36.
- Van Keulen, J. and Young, B.A. (1977). Evaluation of acidinsoluble ash as a natural marker in ruminant digestibility studies. J. Anim. Sci.,44:282-287.
- Warner, A. C. I. (1964). Production of volatile fatty acids in the rumen. Methods of measurements. Nutr. Abst., 34:339.
- Young, D.S. (1990). Effects of drugs on clinical laboratory tests. Third edition 3:6-12.

تأثير الاستبدال الجزئي لبروتين كسب عباد الشمس بنبات الأزولا كمصدر للبروتين علي الأداء الإنتاجي للحملان النامية مجدي حسن أحمد أبو الفضل ، حنان أحمد محمود حسنين و هبه عبد الرحيم الصنفاوي معهد بحوث الإنتاج الحيواني ـ مركز البحوث الزراعية ـ الدقي ــ مصر