Nutritional evaluation of berseem
1- Effect of nitrogen fertilizer on yield and quality response of berseem

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Abstract: The experimental work was conducted during the two successive seasons to study the effect of the N fertilization rate of 0, 23.8 and 47.6 kg ha^{-1} during implantation on forage yield, composition, protein and energy yield, nitrogen recovery and output green forage per hectare. Results revealed that the yield of green and dry whole plant, leaves and stems of berseem forage during both the 1st and 2nd seasons increased significantly (P<0.05) with increasing the level of N fertilization. The percentage of leaves increased significantly (P<0.05), but the percentage of stems decreased significantly (P<0.05) with increasing the level of N fertilization. The contents of CP, EE and ash increased, but DM, OM, CF and NFE contents decreased in whole plant, leaves and stems with increasing the level of N fertilizer. Protein and energy yield, nitrogen recovery and output of forage yield increased significantly (P<0.05) with increasing the level of N fertilizer in both the 1st and 2nd seasons.
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1. Introduction

In Egypt, berseem (*Trifolium alexandrium L.*) is the main forage crop for animal feeding. It usually used fresh in winter and spring seasons from December till the end of May and as hay or silage in summer season (Saleh 1986). The berseem area in Egypt was about 1.09 million hectare produce about 80 million ton green, 6.6 million ton TDN and 1.30 million ton DCP, which contribute 55.9% of TDN and 82.9% of DCP produced throughout the year. Berseem covers 45.9% of TDN and 124.5% of DCP of animal's requirements throughout the year or 91.8% of TDN and 249% of DCP of animal's requirements during winter feeding. The balancing of using berseem in animal's feeding were - 2.42 million tons TDN and + 0.45 million tons DCP (Abou-Slim and Bendary 2005).

Dry matter production increased significantly at a rate of 75 kg N/ha but decreased significantly as the rate was doubled. Plant height, number of leaves per plant, leaf area per plant and leaf-stem ratio increased significantly in response to nitrogen fert1lization (Assaeed 1994). Vuckovic et al (2005) found different rates of nitrogen fertilizer (0, 40, 80, 120 and 160 kg ha^{-1}) had strong positive effect on increasing the yield, crude protein, ash and fat contents of meadows, while, the negative on crude fiber content. The maximum DM yield was obtained in the test with 160 kg ha-1 nitrogen, amounting 4.44 ton ha-1, which was an increase of 2.04 ton ha-1 or 85% over the control.

Vuckovic et al (2006) reported that the investigation included five nitrogen fertilizer rates (0, 40, 80, 120 and 160 kg ha^{-1}) and undersowing with red clover and birdsfoot trefoil. Increase in nitrogen fertilization level resulted in corresponding increases in the quantity and quality of grass, while legumes and other herbages decreased. Maximum two-year average dry matter yield of 4.44 t ha^{-1} was achieved with the highest N rate (160 kg ha^{-1}). The increase was 2.03 t ha^{-1} or 85.0% compared with the control. Increasing N rates increased the contents of protein, ash, fat, Cu, Co, and NO3, and reduced the contents of cellulose, K, P, Ca, Mg, S, Zn, and B.

Almodares et al (2009) use four nitrogen treatments (50, 100, 150 and 200 Kg urea / ha) found that treatment of 200 kg/ha urea had the highest biomass (64.8 t/ha) and protein content (8%) and it had the lowest soluble carbohydrates (12.8%) and fiber contents (31.9%). Ahmadi et al (2009) reported that nitrogen fertilizing systems had significant effects (P<0.05) on DM yield and CP (crude protein) concentration of snail medic. Aydin and Uzun (2005) found that overall mean dry matter yield increased with increase in N application. The highest dry matter yield production was 3407 kg ha-1 with 200 kg ha^{-1} of N as compared to control treatment which had a 1152 kg ha^{-1}. Tolera et al. (2006) stated that the application of 69 kg N/ha appears to be the optimum level of nitrogen fertilizer in terms of dry matter yield and nutritive value of the grass. Leto et al. (2005) found grass DM yield was the highest at N200 and N150 (7.25 and 7.12 t ha^{-1} respectively).

Nichols et al. (1990) indicated that yields increased quadratically as N levels increased.

The objective of this study was to investigate the effect of nitrogen fertilizer on the forage yield, composition, protein and energy yield, nitrogen recovery and output green forage per hectare.

2. Materials and methods

The current work was carried out at the Department of Animal Production, Faculty of Agriculture, Kafrelsheikh University to investigate the effect of nitrogen fertilization on green and dry yield, chemical composition, protein and energy yield, nitrogen recovery and output of green berseem forage. The experimental work was conducted during the two successive seasons. The soil texture of the experimental site was clay loam in the first and second seasons. Rate of seeding was 47.6 kg ha^{-1}. Seeds were of local berseem from Kafrelsheikh Governorate. Berseem was planted during the middle of October in the two seasons. Four hundred and seventy six kg of superphosphate (15.5% P2O5) per hectar was added during the land preparation. Ammonium sulphate (20% N) was added before the first irrigation at rate of 0, 119 and 238 kg ha^{-1} to give the N fertilization rate of 0, 23.8 and 47.6 kg ha^{-1}, respectively. Four cuts of berseem were taken every season during the experiment, 1st cut on 28-29th of December, 2nd cut on 13-14th of February, 3rd cut on 1st -2nd of April and 4th cut on 13-14th of May.

The fresh berseem forage weight estimated for every plot (1 m^2) in each cut in triplicate and the green forage yield per hectare was calculated. Samples of about 0.5 kg of fresh green forage were taken from each plot spread in a tray and dried in forced air oven at 70 ^oC for 48 hours to determine dry matter content (DM) and calculate dry forage yield per hectare. Other samples were taken for the separation of leaves and stems to estimate the percentage of each part from the whole plant, then leaves and stems samples were dried in a forced air oven at 70 ^oC for 48 hours to determine DM content. The dried samples were ground and analyzed for determination of crude protein (CP), ether extract (EE), crude fiber (CF), ash and NFE (by difference) according to AOAC (1995). The output of green berseem forage yield = green forage yield x price of 1 ton green berseem forage (120 LE, according to prices 2010). The obtained data were statistically analyzed using general liner models procedure adapted by SPSS (2008) for one-way ANOVA. The Duncan multiple range test was used to compare difference between means.

3. Results and discussion

The yield of green and dry whole plant, leaves and stems of berseem forage are presented in Table (1). The yield of green and dry whole plant, leaves and stems of berseem forage during both the 1st and 2nd seasons increased significantly ($P<0.05$) with increasing the level of N fertilization. The high N fertilization level (47.6 kg ha^{-1}) recorded the highest green and dry whole plant, leaves and stems yield during the both seasons averaging 121.6, 28.1 and 93.5 and 15.2, 6.2 and 9.0 ton ha^{-1}., respectively. The mean green whole plant, leaves and stems yield increased quadraticlly by 13.8, 48.2 and 7.7 and 41.7, 95.2 and 30.9% for the N fertilization levels of 23.8 and 47.6 kg ha^{-1}, respectively (R^2= 0.79, 0.90 and 0.69, respectively). The corresponding values for dry yield were 32.7, 40.7 and 27.6 and 60.3, 78.6 and 49.2%, respectively (R^2= 0.83, 0.88 and 0.79, respectively). There were strong positive correlation between N fertilization and the yield of green and dry whole plant, leaves and stems of berseem forage being 0.87, 0.95 and 0.80 and 0.92, 0.94 and 0.88, respectively. These results showed that the nitrogen fertilize is more effective on the yield of leaves than stems and also the yield of dry stems is more affected than green stems. These results agree with those obtained by Polat et al (2007) who found that overall mean forage dry matter yield increased with increase in N application. Ahmadi et al (2009) reported that nitrogen fertilizing systems had significant effects ($P<0.05$) on forage DM yield.

The percentage of leaves increased significantly ($P<0.05$), but the percentage of stems decreased significantly ($P<0.05$) with increasing the level of N fertilization (Table 1). The quadratic response of the percentage of leaves and stems for nitrogen fertilization was R^2 = 0.82. The percentage of leaves was positively correlated (r = 0.91), but the percentage of stems was negatively correlated (r = - 0.91) with the level of N fertilization. Plant height, number of leaves per plant, leaf area per plant and leaf-stem ratio increased significantly in response to nitrogen fert1lization (Assaeed 1994).

Results in Table (2) showed that the contents of CP, EE and ash increased, but DM, OM, CF and NFE contents decreased in whole plant, leaves and stems with increasing the level of N fertilizer. The quadratic response of DM, OM, CP, CF, EE, NFE and ash for the N fertilization were R^2 = 0.81, 0.11, 0.91, 0.90, 0.90, 0.24 and 0.79, respectively. Moreover, N fertilizer positively correlated with CP, EE and ash contents being 0.95, 0.94 and 0.89 and negatively correlated with DM, OM, CF and NFE contents being - 0.90, - 0.33, - 0.94 and - 0.49, respectively. These results reflect the differences in leaves to stems ratio as affected by N fertilizer. These results are in accordance with those obtained by Vuckovic et al (2005) who found strong positive effect of N fertilizer on crude protein, ash and fat contents of meadows, while, the negative on crude fiber content. Vuckovic et al (2006) reported that increasing N rates increased the contents of protein, ash

and fat and reduced the contents of cellulose. Almodares et al (2009) found that treatment of 200 kg/ha urea had the highest protein content and the lowest soluble carbohydrates and fiber contents.

Protein and energy yield and nitrogen recovery increased significantly (P<0.05) with increasing the level of N fertilizer during the 1st and 2nd seasons as shown in Table (3). The high level of N fertilizer (47.6 kg ha^{-1}) recorded the highest means of protein yield (2.7 ton ha^{-1}), energy yield (60464 Mcal ha^{-1}) and nitrogen recovery (384.9 kg ha^{-1}). Protein and energy yield and nitrogen recovery response quadratically to N fertilization (R^2 = 0.89, 0.84 and 0.82, respectively). The high positive correlations excite between N fertilizer and Protein and energy yield and nitrogen recovery were 0.94, 0.92 and 0.90, respectively. These increases might be due to both increases the dry forage yield and the ratio of leaves with increasing N fertilizer. These results agree with those obtained by Ahmadi et al (2009) who found that protein yield and nitrogen recovery of snail medic increased with increasing N fertilizer (P<0.05). Kering et al (2010) reported that the yield of both CP and TDN increase with increasing N fertilization rate (0, 112, 224, 336, and 448 kg/ha).

The output of green forage increased significantly (P<0.05) with increasing the level of N fertilizer during the 1st and 2nd seasons (Table 3). The output of green forage improved by 16 and 42% during the 1st season and by 13 and 41% during the 2nd season for the N levels of 23.8 and 47.6 kg ha^{-1}, respectively. The quadratic effect of N fertilizer on output of green forage was 0.79. The positive correlation between N fertilizer and output of green forage was 0.87. The increase output was associated with increasing the yield of green forage (Table 1), protein and energy yield (Table 3).

Table 1. Green and dry forage yield (ton/ha), leaves and stems percentages.

Item	1st season				2nd season			
	N fertilizer (kg ha^{-1})				N fertilizer (kg ha^{-1})			
	0	10	20	Mean	0	10	20	Mean
Green yield								
Whole plant	84.5^{b}	97.8^{b}	120.0^{a}	100.9	86.9^{b}	98.5^{b}	123.0^{a}	102.8
Leaves	14.1^{c}	20.6^{b}	29.3^{a}	21.3	14.6^{c}	22.0^{b}	26.9^{a}	21.2
Stems	70.4^{b}	77.2^{b}	90.7^{a}	79.6	72.3^{b}	76.5^{b}	96.1^{a}	81.6
Dry yield								
Whole plant	9.3^{c}	12.1^{b}	15.9^{a}	12.4	9.6^{b}	13.0^{a}	14.5^{a}	12.4
Leaves	3.4^{c}	4.7^{b}	6.4^{a}	4.8	3.5^{b}	5.0^{a}	5.9^{a}	4.8
Stems	5.9^{c}	7.4^{b}	9.5^{a}	7.6	6.1^{b}	8.0^{a}	8.6^{a}	7.6
leaves and stems percentages (DM basis)								
Leaves	36.2^{b}	38.6^{ab}	40.7^{a}	38.5	36.3^{b}	38.6^{ab}	40.7^{a}	38.5
Stems	63.8^{a}	61.4^{ab}	59.3^{b}	61.5	63.7^{a}	61.4^{ab}	59.3^{b}	61.5

a, b, c: Values in the same row for each season with different superscripts differ significantly at 5% level.

Table 2. Chemical composition (%) of whole plant berseem and their leaves and stems (3rd cut, 1st season).

Item	DM %	Composition of DM %					
		OM	CP	CF	EE	NFE	Ash
Whole plant							
0 kg N/ha	17.8	85.4	15.7	23.3	2.32	44.1	14.6
10 kg N/ha	17.1	84.8	16.6	22.2	2.45	43.6	15.2
20 kg N/ha	16.4	84.2	17.8	20.7	2.61	43.1	15.8
Leaves							
0 kg N/ha	23.9	87.9	25.2	12.1	2.71	47.8	12.2
10 kg N/ha	22.8	87.5	28.2	10.8	2.86	45.6	12.5
20 kg N/ha	22.0	87.0	30.5	8.34	2.98	45.2	13.0
Stems							
0 kg N/ha	15.70	87.6	7.59	33.8	2.36	43.8	12.4
10 kg N/ha	14.8	87.2	8.34	32.2	2.50	44.2	12.8
20 kg N/ha	13.7	87.1	9.10	30.7	2.68	44.6	13.0

Table 3. Protein and energy yield and nitrogen recovery.

Item	1st season				2nd season			
	N fertilizer (kg ha^{-1})				N fertilizer (kg ha^{-1})			
	0	10	20	Mean	0	10	20	Mean
Protein (ton/ha)	1.48^{c}	2.02^{b}	2.83^{a}	2.12	1.52^{c}	2.17^{b}	2.59^{a}	2.09
Gross energy (Mcal/ha)	36785^{c}	47074^{b}	63118^{a}	48992	38006^{b}	50397^{a}	57810^{a}	48738
Nitrogen recovery (kg/ha)	234.4^{c}	298.7^{b}	403.9^{a}	312.3	242.3^{b}	321.3^{a}	365.8^{a}	309.7
Output of forage (LE/ha)	10144^{b}	11738^{b}	14406^{a}	12095	10436^{b}	11833^{b}	14754^{a}	12340
Output improvement %	100^{c}	116^{b}	142^{a}	119	100^{c}	113^{b}	141^{a}	118

a, b, c: Means in the same row for each season with different superscripts differ significantly at 5% level.

4. Conclusions

From this study it could be concluded that nitrogen fertilizer at rate of 47.6 kg ha^{-1} gave the highest yield of forage, protein, energy and output and the best forage composition.

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