Research Article

Phosphorus Influenced Nutritive Value, Yield and Economics of Berseem (*Trifolium alexandrinum* L.) Genotypes

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Abstract

A field experiment was conducted at CCS Haryana Agricultural University, Hisar (Haryana) to study the effect of different phosphorus levels on the yield, quality and economics of berseem (*Trifolium alexandrinum* L.) genotypes for fodder during winter season (*Rabi*) of 2016-17. The experiment was conducted in split plot design in three replicates; five genotypes (JB-04-23, JB-04-21, Wardan, Mescavi and BB-2) were kept in main plot and three P levels (60, 80 and 100 kg P_2O_5/ha) in split plot. Among genotypes, JB-04-21 recorded highest green fodder yield (71.19 t/ha) which was on a par with BB-2, Mescavi and JB-04-23. The genotype BB-2 was superior in terms of crude protein and digestible dry matter yields (1.72 and 6.88 t/ha) being on par with JB-04-21 and Mescavi. The average crude protein and IVDMD content of four cuts were not affected significantly among all genotypes. However, the crude protein and IVDMD ranged from 17.37 to 17.75 and 67.22 to 69.62 per cent, respectively. The mean crude protein and IVDMD content increased from first cut to second cut and then decreasing trend was observed in third and fourth cut. Maximum net returns (Rs. 32537/ha) and B:C (1.58) were fetched with JB-04-21 followed by BB-2. Among different P levels, significantly highest total green fodder, dry matter, crude protein and digestible dry matter yields (73.48, 10.50, 1.86 and 7.29 t/ha, respectively) were recorded with 100 kg P_2O_5/ha which were 9.0, 12.1, 16.3, 14.4 and 21.8, 28.4, 38.8, 35.3 per cent higher over 80 and 60 kg P_2O_5/ha, respectively in total of four cuts. Maximum net return (Rs. 34451/ha) and B:C (1.60) were also fetched with 100 kg P_2O_5/ha. In nutshell, optimization of P fertilization improved the productivity and nutritive value of the fodder in berseem.

Keywords: Berseem, fodder yield, quality, crude protein, IVDMD content and P levels

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Introduction

Berseem or Egyptian clover (*Trifolium alexandrinum* L.), is one of the best Rabi (winter season) fodder crop in entire North West Zone, Hill Zone and part of Central and Eastern Zone of the country, occupy more than two million hectare [1]. Berseem is popular due to its multicut (4-8 cuts) nature, providing fodder for a long duration (November to May), very high quantum of green fodder (85 t/ha) and better quality of fodder (20% crude protein), high digestibility (up to 65%) and palatability [2]. It is also a well known green forage crop that stimulates milk production in dairy animals due to its succulence and nutritiousness with high crude protein and total digestible nutrients [3]. Being a leguminous crop, it fixes atmospheric nitrogen and thus restores soil fertility. For higher green fodder yields, vegetative growth of the crop is very important. Although the vegetative growth of any crop largely depends upon the potential of the genotype, nutrient supply system, capacity of the soil to supply the nutrients and capacity of the plants to take and use the nutrients in a unit time. Among the primary nutrients, phosphorus is one of the key mineral nutrients for rumen microbes. The vast majority of phosphorus in concentrates is in the form of phytate. For stomach, phytate degradation is crucial for phosphorous utilization in ruminants [4].

P deficiency in Indian soils is wide spread (98% of districts), and crop responses to its application are highly profitable. All indications are that P removal will continue to exceed net P additions, and P deficiency will accentuate further with time [5].

Most of the soils of Haryana are low to medium in phosphorus and if the required amount of phosphorus of any forage crop is not supplied in sufficient amount then the deficiency is reflected in green and dry fodder. Since P is an essential constituent of nucleic acids, nucleoproteins, amino acids, proteins, phosphotides, phytin and several co-enzymes, and deficiency of P in leguminous fodder crops may cause severe disorders in animals. It is essential in transfer and utilization of energy. Initial symptoms of P deficiency are decreased appetite, lowered blood P and "pica", in which the animals have a craving for unusual foods such as wood or other materials. Hence, the present study was undertaken to find out the optimum dose of P along with suitable genotype to realize fodder yield potential of berseem in terms of quantity and quality.

Materials and Methods

The promising genotypes of berseem were screened for two years (winter season of 2014-15, 2015-16) in the breeding trials. The best two selected elite genotypes (JB-04-23 and JB-04-21) were then promoted and tested against three checks (Wardan, Mescavi and BB-2) for advance agronomy trial during the third year (winter season of 2016-17) at Forage Section Research Farm of CCS Haryana Agricultural University, Hisar, Haryana, India (29° 10' N of 75° 46' E, at an average elevation of 215.2 m above mean sea level). The site has semi-arid and sub-tropical climate with hot dry summers and severe cold winters. Average annual rainfall is about 450 mm, 75 per cent of which is received from July to September. July and August are the wettest months. The crop received 51.8 mm rainfall during crop period. **Figure 1** represents the weekly weather parameters *i.e*. temperature °C, relative humidity (%) and rainfall (mm) during the study. The physico-chemical properties of the soil of the experimental field are given in **Table 1**. The experiment was conducted in split plot design in three replicates, keeping genotypes in main plot and phosphorus levels in sub plots. The experiment consisted five berseem genotypes *i.e.* JB-04-23, JB-04-21, Wardan, Mescavi and Bundel Berseem-2 (BB-2) and three phosphorus levels, *viz*. 60, 80 and 100 kg P_2O_5/ha. The berseem genotypes as per treatment were sown manually on 13 November 2016 using the seed rate of 25 kg/ha. All the other standard agronomic practices for the cultivation of berseem were followed uniformly in all the treatments. Total four cuts were taken across different treatments. First cut was taken 65 days after sowing and subsequent three cuts were taken at 30, 30 and 25 days interval after the first cut, second cut and third cut, respectively. The harvested green fodder from each plot was weighed in situ in kg/plot and then converted into q/ha. A random sample of 500 g was taken from each plot at the time of green fodder harvest and oven dried till constant weight achieved. On the basis of these samples, the green fodder yield was converted into dry matter yield (q/ha). Crude protein content and *in-vitro* dry matter digestibility (IVDMD) were estimated in dry and grinded samples (2 mm sieve size) at each cut analyzed by micro-Kjeldhal method [6].

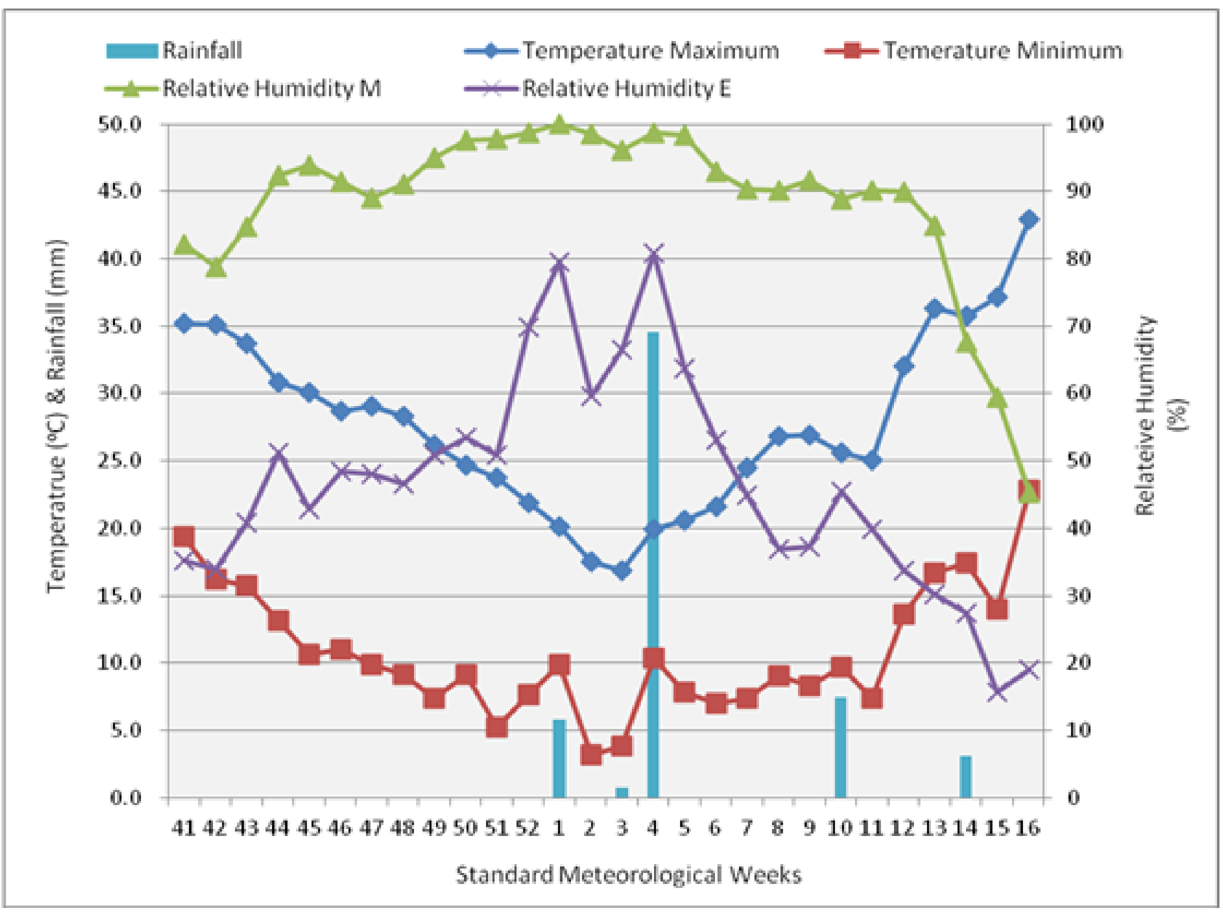

Figure 1 Weekly weather parameters during the crop season

Table 1 Physico-chemical properties of soil before sowing

S.No.	Particulars	Results	Category	Method used
1	Soil texture	Sandy loam	Sandy loam	Bouyoucos hydrometer method [17]
2	Soil pH value	8.20	Moderately alkaline	Determined in soil : distilled water suspension (1:2) [18]
3	EC (dS/m)	0.43	Non saline	
4	Soil organic carbon (%)	0.52	Medium	Partial oxidation method [19]
5	Available Nitrogen (kg P/ha)	154.8	Low	Alkaline permanganate method [20]
6	Available Phosphorus (kg P/ha)	12.0	Medium	Sodium bicarbonate method [21]
7	Available Potassium (kg K/ha)	252.0	High	Neutral normal ammonium acetate method [18]

Structural carbohydrates viz. neutral detergent fibre (NDF), acid detergent fibre (ADF), cellulose, and hemicellulose were estimated in leaves and shoots according to Van Soest *et al*. [7] without addition of sodium sulphite. NDF was assayed without a heat stable amylase and expressed inclusive of residual ash. ADF was also expressed inclusive of residual ash. Sulphuric acid lignin was determined by the method of Robertson and Van Soest [8]. The hemicellulose content was estimated in same sample in sequential manner. In vitro dry matter digestibility was determined by the method of Barnes *et al*. [9].

Crude protein yield (CPY) was calculated by the following formula

$$CPY = \frac{CP \times DMY}{100}$$

Where, CP = Crude protein content (%), DMY = Dry Matter Yield

Digestible dry matter (DDM) yield was calculated by the following formula

$$DDM = \frac{IVDMD \times DMY}{100}$$

Where, IVDMD = *In vitro* Dry Matter Digestibility (%), DMY = Dry Matter Yield

Economics was worked out on the basis of prevailing market prices of inputs and outputs in the local market. The experimental data were analyzed by using OPSTAT software [10]. Treatments were compared with CD values at 5% level of significance.

Results and Discussion

Effect of Genotypes

Being an introduced crop in India, genetic improvement in this crop is hampered by narrow genetic base and lack of variability in desirable traits which makes its breeding a tough task. The promising genotypes promoted through breeding trials were evaluated in advanced agronomy trial. Among the test genotypes, JB-04-21 performed better. The highest average number of tiller/m^2 (849.92) and plant height (65.31 cm) were recorded in JB-04-21 which was on par with all the other genotypes except Wardan (**Table 2**). Highest total green fodder yield (71.19 t/ha) was also recorded in JB-04-21 which was significantly superior over Wardan except other genotypes. Highest total dry matter yield (10.05 t/ha) was recorded in BB-2 which was significantly superior to Wardan and JB-04-23. Cut wise green fodder and dry matter yields of all the genotypes are shown in **Figures 2** and **3**, respectively. The average crude protein content ranged from 17.37 to 17.75 % and average IVDMD was 67.22 to 69.62 % among genotypes (**Table 3**). The average crude protein content of 1^{st}, 2^{nd}, 3^{rd} and 4^{th} cut was 18.76, 20.28, 16.05 and 15.18% and average IVDMD was 67.15, 72.77, 68.77 and 64.48%, respectively. The highest total crude protein yield (17.2 q/ha) and total digestible dry matter yield (68.8 q/ha) were recorded in BB-2 which were significantly superior over JB-04-23 and Wardan (**Table 4**). Variations among the genotypes of berseem for fodder yield and quality parameters were also reported by several researchers [11].

Effect of Phosphorus Levels

Berseem, being a leguminous crop, requires sufficient quantity of phosphorus in free form for better nodulation which might result in more nitrogen fixation in plant roots. Phosphorus also plays a fundamental role in number of enzymatic reactions. Besides this, it is an essential component of DNA, RNA needed for protein synthesis and plays a major role in energy transfer system (ADP, ATP).

Table 2 Effect of P levels on yield attributes, fodder yield and economics of berseem genotypes

Treatment	Average no. of tillers/m^2	Average plant height (cm)	Total green fodder yield (t/ha)	Total dry matter yield (t/ha)	Total Crude protein yield (t/ha)	Total Digestible dry matter yield (t/ha)	Gross return (Rs./ha)	Net returns (Rs./ha)	B:C ratio
Genotypes									
JB-04-23	782.14	62.34	64.81	8.91	1.51	5.95	81019	24571	1.43
JB-04-21	849.92	65.31	71.19	9.98	1.71	6.76	88984	32537	1.58
Wardan	705.49	58.50	59.53	8.51	1.44	5.73	74418	17971	1.32
Mescavi	827.70	62.70	68.97	9.30	1.61	6.42	86210	29762	1.53
BB-2	836.58	64.04	70.83	10.05	1.72	6.88	88535	32088	1.57
SEm±	21.33	1.18	2.46	0.31	0.06	0.22	3076	3076	0.05
CD (P=0.05)	70.77	3.90	8.15	1.01	0.20	0.74	10185	10185	0.18
P Levels (P_2O_5 kg/ha)									
60	723.26	58.73	60.31	8.18	1.34	5.39	75381	19813	1.36
80	807.70	63.04	67.41	9.37	1.60	6.37	84268	27892	1.49
100	869.91	65.96	73.48	10.5	1.86	7.29	91849	34451	1.60
SEm±	7.22	0.68	0.74	0.15	0.03	0.09	922	922	0.02
CD (P=0.05)	21.44	2.02	2.19	0.46	0.08	0.28	2740	2740	0.05
CV %	3.49	4.20	4.26	6.37	6.32	5.66	4.26	13.04	4.26

Interaction detail: Interaction of **Entry x P levels** was found non-significant for all the above characters.

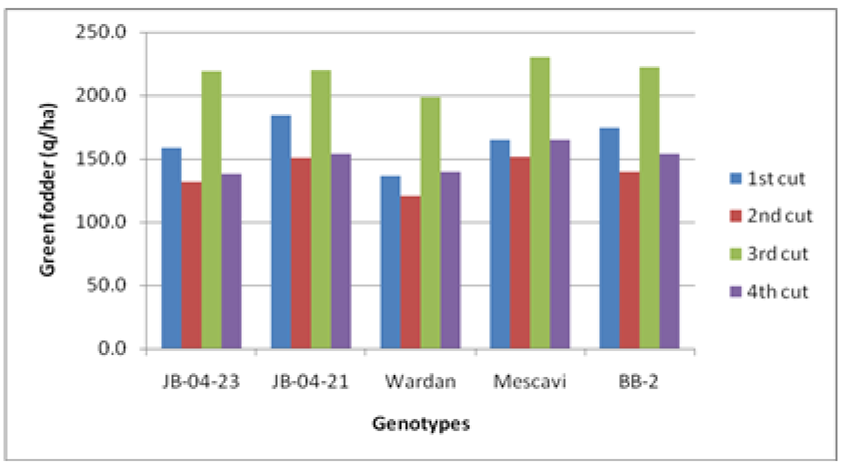

Figure 2 Cut wise GFY of genotypes

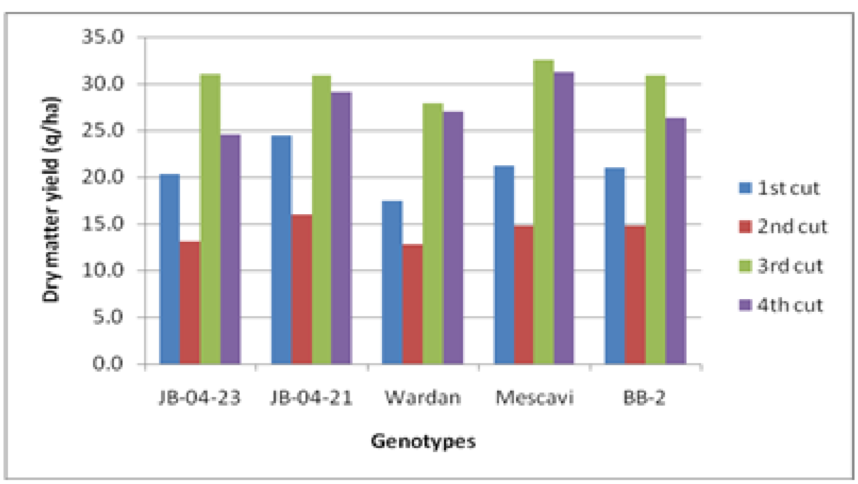

Figure 3 Cut wise DMY of genotypes

Table 3 Effect of phosphorus levels on crude protein content and IVDMD of berseem genotypes

Treatments	Crude protein content (%)					*In vitro* dry matter digestibility (%)				
	1st cut	2nd cut	3rd cut	4th cut	Average	1st cut	2nd cut	3rd cut	4th cut	Average
Genotype										
JB-04-23	18.56	19.93	15.71	15.57	17.44	63.40	72.70	67.77	65.00	67.22
JB-04-21	19.27	20.81	16.32	14.61	17.75	67.13	71.68	70.47	63.43	68.18
Wardan	19.14	19.64	16.34	14.35	17.37	67.08	70.57	69.80	62.80	67.56
Mescavi	18.63	20.47	16.86	14.89	17.71	70.07	75.24	67.13	66.03	69.62
BB-2	18.22	20.54	15.01	16.46	17.56	68.07	73.67	68.67	65.13	68.88
SEm±	0.31	0.08	0.16	0.09	0.10	1.81	1.42	1.54	0.87	0.60
CD (P=0.05)	NS	0.29	0.53	0.03	NS	NS	NS	NS	NS	NS
P Levels (P_2O_5 kg/ha)										
60	19.00	19.94	15.22	14.36	17.02	64.56	70.62	66.20	64.04	66.36
80	19.00	20.20	16.04	15.34	17.57	67.88	72.72	69.02	64.18	68.45
100	19.00	20.69	16.90	15.83	18.12	69.01	74.98	71.08	65.22	70.07
SEm±	0.18	0.10	0.11	0.08	0.06	1.11	0.83	0.54	0.59	0.45
CD (P=0.05)	NS	0.29	0.31	0.24	0.17	3.29	2.46	1.60	NS	1.32
CV %	3.74	1.85	2.55	6.58	1.29	6.38	4.41	3.03	3.57	2.52

Interaction detail: Interaction of **Genotype × P levels** was found non-significant for all the above characters except CP (%) for 2nd, 4th cut and IVDMD (%) for 3rd cut.

Table 4 Effect of phosphorus levels on crude protein yield and DDM of berseem genotypes

Treatments	Crude protein yield (q/ha)					Digestible dry matter (q/ha)				
	1st cut	2nd cut	3rd cut	4th cut	Total	1st cut	2nd cut	3rd cut	4th cut	Total
Entries										
JB-04-23	3.8	2.6	4.9	3.8	15.1	13.0	9.6	21.0	16.0	59.5
JB-04-21	4.1	3.1	5.3	4.6	17.1	14.3	10.6	23.0	19.8	67.6
Wardan	3.4	2.5	4.6	3.9	14.4	11.8	9.2	19.5	16.9	57.3
Mescavi	3.9	3.0	5.2	3.9	16.1	14.7	11.1	21.0	17.4	64.2
BB-2	4.5	3.3	4.7	4.8	17.2	16.7	11.8	21.3	19.0	68.8
SEm±	0.24	0.24	0.24	0.22	0.60	0.81	0.91	0.96	0.88	2.24
CD (P=0.05)	NS	NS	NS	0.74	1.98	2.69	NS	NS	NS	7.40
P Levels										
60	3.2	2.4	4.1	3.7	13.4	11.1	8.6	17.9	16.3	53.9
80	4.0	3.0	4.9	4.2	16.0	14.5	10.7	21.0	17.6	63.7
100	4.6	3.4	5.8	4.8	18.6	16.8	12.1	24.5	19.5	72.9
SEm±	0.13	0.10	0.13	0.14	0.26	0.39	0.39	0.53	0.58	0.93
CD (P=0.05)	0.38	0.30	0.38	0.42	0.78	1.16	1.17	1.57	1.72	2.75
CV %	12.79	13.62	10.04	13.13	6.32	10.68	14.51	9.70	12.63	5.66

Interaction detail: Interaction of **Entry × P levels** was found non-significant for all the above characters.

The soil of the experimental field being medium in P availability (Table 1) responded very well to berseem crop. Among different P levels, the highest number of tiller/m^2 (869.91) and plant height (66.0 cm) were recorded with the application of 100 kg P_2O_5/ha which were significantly superior to the lower phosphorus levels. Data presented in Table 2 reveal that significantly higher green fodder and dry matter yields were recorded with the application of 100 kg P_2O_5/ha (73.48 and 10.50 t/ha, respectively). Perusal of the data reveal that yield attributes and fodder yield increased with increasing levels of phosphorus from 60 to 100 kg P_2O_5/ha. Cut wise green fodder and dry matter yields at different phosphorus levels are shown in **Figures 4** and **5**, respectively. Researchers also reported highest green and dry fodder yield of berseem with 100 kg P_2O_5/ha but it was on par with 80 kg P_2O_5/ha across seven berseem genotypes [11]. Significantly highest crude protein content (18.12%), IVDMD (70.07%), total crude protein yield (18.56 q/ha) and total digestible dry matter yield (72.91 q/ha) were also recorded with the application of 100 kg P_2O_5/ha (Tables 3 and 4). The average crude protein content of 1st, 2nd, 3rd and 4th cut was 17.02, 17.57 and 18.12% and average IVDMD was 66.36, 68.45 and 70.07%, at 60, 80 and 100 kg P_2O_5/ha respectively (Table 3).

Table 5 shows the effect of phosphorus levels and genotypes on structural carbohydrates. In the present investigation, the dry matter and cell wall constituents (NDF, ADF, cellulose, lignin and hemicellulose) exhibited upward trend with plant maturity due to secondary depositions. NDF, ADF, hemicellulose and cellulose content ranged from 39.46 to 41.97 %, 30.58 to 32.63 %, 8.87 to 10.88 %, and 25.52 to 28.04 % respectively. The lignin content ranged from 3.93 to 6.21 %. Maximum lignin per cent was observed in Mescavi (6.21 %). These observations are in accordance with the results of [12].

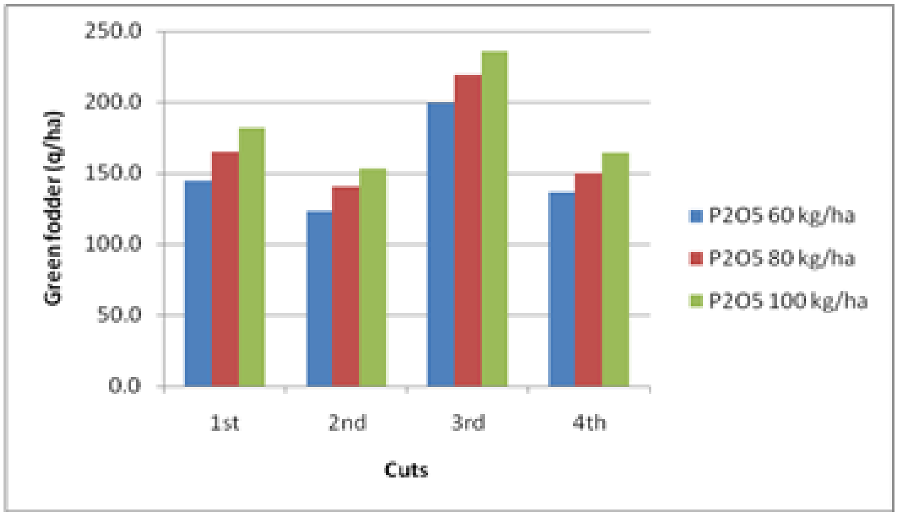

Figure 4 Cut wise GFY at different P levels

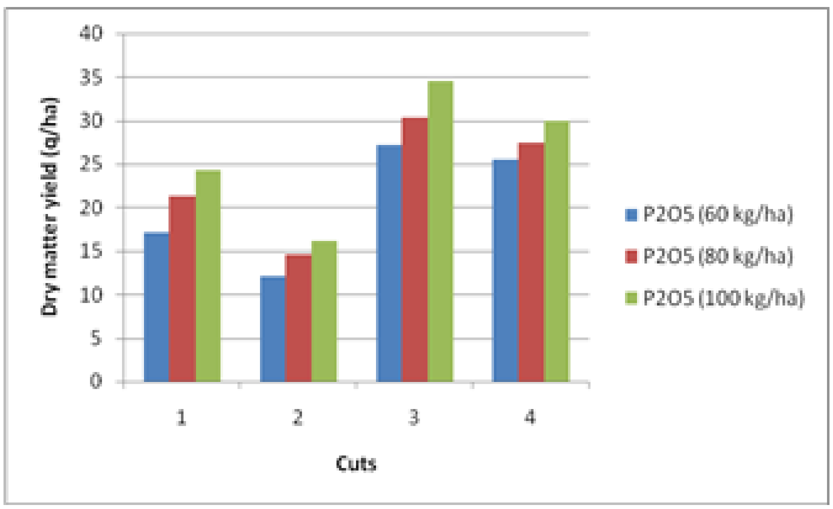

Figure 5 Cut wise DMY at different P levels

Table 5 Effect of phosphorus levels on quality of berseem genotypes (Mean data of 4 cuts)

Treatments	NDF (%)	ADF (%)	Hemicellulose (%)	Cellulose (%)	Lignin (%)
Entries					
JB-04-23	39.46	30.58	8.87	25.52	3.93
JB-04-21	41.65	31.35	10.31	25.76	4.58
Wardan	41.97	32.63	9.34	28.04	5.91
Mescavi	41.49	30.60	10.88	27.33	6.21
BB-2	41.28	30.66	10.62	25.58	3.99
SEm±	0.12	0.22	0.28	0.06	0.06
CD (P=0.05)	0.40	0.73	0.91	0.18	0.02
P Levels					
60	42.40	32.12	10.28	27.52	5.22
80	41.21	31.19	10.01	26.55	4.99
100	39.89	30.18	9.72	25.26	4.56
SEm±	0.07	0.11	0.13	0.05	0.04
CD (P=0.05)	0.21	0.33	0.38	0.14	0.11

CV %	0.65	1.38	4.96	0.72	2.95
Interaction detail: Interaction of **Genotype × P levels** was found non-significant for all the above characters except NDF (%) and cellulose (%).					

It was observed that with the increasing level of phosphorus, the content of structural carbohydrates decreased. At 60 kg/ha, highest level of NDF, ADF, hemicellulose, cellulose and lignin were observed *viz*. 42.40, 32.12, 10.28, 27.52, 5.22, respectively. An inverse trend was observed with highest level of phosphorus *i.e*., 100 kg/ha. These results were at par with Dasci and Comakli [13] who observed that ADF and NDF content decreased with fertilization and varied significantly. Similar results were obtained by [14] and [15]. Balabanli *et al*. [16] studied the effects of nitrogen, phosphorus and potassium fertilization on the quality and yield of native rangeland and observed that fertilization significantly decreased the NDF, ADF and ADL content from 74.32 to 68.46%, 46.45 to 39.02% and 17.15 to 14.84%, respectively.

Economic analysis

Among genotypes, the maximum gross returns (Rs. 88984/ha), net return (Rs. 32537/ha) and B:C ratio (1.58) was fetched with JB-04-21 closely followed by BB-2. Among different phosphorus levels, maximum gross returns (Rs. 91849/ha), net return (Rs. 34,451/ha) and B:C ratio (1.60) was fetched with the application of 100 kg P_2O_5/ha and it was superior to lower doses (Table 2).

Genotype × P level Interaction

The interaction of Genotype × P levels was found non-significant for all the characters except crude protein content (%) for 2nd, 4th cut and IVDMD (%) for 3rd cut. The interaction **Tables 6-8** details the crude protein content (%) for 2nd, 4th cut and IVDMD (%) for 3rd cut.

Table 6 Interaction of genotypes and P levels for crude protein content at 2nd cut

Genotype	**P levels (P_2O_5 kg/ha)**			**Mean of genotypes**
	60	**80**	**100**	
JB-04-23	19.79	19.99	20.00	19.93
JB-04-21	20.60	20.40	20.61	20.54
Wardan	19.34	19.38	20.20	19.64
Mescavi	20.40	20.81	21.22	20.81
BB-2	19.59	20.40	21.43	20.47
Mean of P levels	19.94	20.20	20.69	
Interaction CD: P level at same level of Genotype (CD at 5% 0.66), Genotype at same level of P (CD at 5% 0.58)				

Table 7: Interaction of genotypes and P levels for crude protein content at 4th cut

Genotype	**P levels (P_2O_5 kg/ha)**			**Mean of genotypes**
	60	**80**	**100**	
JB-04-23	15.30	16.32	15.10	15.57
JB-04-21	15.50	16.12	17.75	16.46
Wardan	13.67	14.08	15.30	14.35
Mescavi	12.84	14.89	16.12	14.61
BB-2	14.49	15.30	14.89	14.89
Mean of P levels	14.36	15.34	15.83	
Interaction CD: P level at same level of Genotype (CD at 5% 0.56), Genotype at same level of P (CD at 5% 0.54)				

Table 8 Interaction of genotypes and P levels for IVDMD (%) at 3rd cut

Genotype	**P levels (P_2O_5 kg/ha)**			**Mean of genotypes**
	60	**80**	**100**	
JB-04-23	67.80	67.90	67.60	67.77
JB-04-21	64.40	70.40	71.20	68.67
Wardan	69.00	70.20	70.20	69.80
Mescavi	69.00	70.40	72.00	70.47

BB-2	60.80	66.20	74.40	67.13
Mean of P levels	66.20	69.02	71.08	
Interaction CD: P level at same level of Genotype (CD at 5% 3.87), Genotype at same level of P (CD at 5% 5.88)				

Conclusion

Berseem responded to phosphorus fertilization up to 100 kg P_2O_5/ha. Application of 100 kg P_2O_5/ha brought out an increase of 21.8 and 9.0 per cent in green fodder, and 28.4 and 12.1 per cent in dry matter over 60 and 80 kg P_2O_5/ha, respectively. Significant increase in fodder quality was estimated at 100 kg P_2O_5/ha in terms of crude protein content, IVDMD, crude protein yield, digestible dry matter and structural carbohydrates over the lower doses of phosphorus. The test genotypes JB-04-21 performed superior over all the genotypes except the check Bundel berseem-2 (BB-2) in terms of green fodder, dry matter, crude protein, digestible dry matter yields and structural carbohydrates.

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