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Growth, Forage Yield, and Quality of Berseem (*Trifolium alexandrinum* L.) Genotypes under Different Phosphorus Levels

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Background: The shortage of green and dry fodder remains a significant challenge for the livestock and dairy sectors. Currently, the country is facing a deficit in both green fodder and dry fodder. This is primarily due to the limited area under fodder cultivation, which directly impacts overall fodder availability. To address this issue, there is a pressing need either to expand the area under fodder crops or to adopt advanced production technologies that maximize yield per unit area. Among various fodder crops, berseem (*Trifolium alexandrinum* L.) stands out as a crucial legume cultivated during the winter season globally. Fertilizers, particularly phosphorus, play a key role in optimizing crop productivity. Phosphorus is essential for energy transfer in root nodules and also contributes significantly to root development, nutrient uptake, and overall plant growth.

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Aim: The present study aimed to evaluate the performance of different promising berseem genotypes under varied phosphorus application levels for growth, yield and quality parameters.

Study Design: The field research experiment was undertaken in the Split-Plot Design.

Place and Duration of the Study: The study was conducted during *rabi* season of 2023-24 under the AICRP on Forage Crops at BAIF's Central Research Station, Urulikanchan, Pune, MS.

Methodology: The experiment was undertaken with four promising genotypes of berseem along 2 checks as main treatment and three phosphorus levels as sub main treatments. A flat bed of size 4x3 m was prepared and seeds of each genotype were sown at 30 cm spacing. A full dose of phosphorus (as per treatments) supplemented with 20kgha⁻¹ urea were applied to each treatment before sowing. Irrigation was given as per the crop requirement and other agronomic package of practices were kept similar to all the treatments.

Result: Among the genotypes evaluated, JB-08-17 recorded the tallest plants with 66.10 cm, while PC-114 exhibited the highest leaf stem ratio of 0.772. In terms of green fodder yield, JHB-20-2 recorded the highest yield of 967.36 q/ha, followed by PC-114 with 966.72 q/ha. PC-114 also produced the significantly highest dry fodder yield (134.19 q/ha) and crude protein yield (28.79 q/ha). Application of 100 kg P_2O_5 per hectare resulted in the maximum average plant height of 63.08 cm, whereas the highest leaf stem ratio (0.69) was observed with 80 kg P_2O_5 per hectare. Phosphorus application at 100 kg/ha led to the maximum yield of green fodder (959.29 q/ha), dry matter (114.38 q/ha), and crude protein (23.68 q/ha). These results highlight the potential of specific berseem genotypes and optimal phosphorus application levels to enhance fodder productivity and contribute to addressing the ongoing deficit in green and dry fodder availability.

Keywords: Berseem; genotypes; fertilizer; phosphorus; green fodder yield.

1. INTRODUCTION

In rural India, livestock rearing has traditionally played a crucial role in supporting household livelihoods alongside agriculture. Over the decades, its role has grown increase significantly, due to the steady and sustainable cash income. The growing demand for milk and milk-based products, has made dairy farming an emerging and profitable enterprise in the country.

Selection of high-yielding varieties is one of the most important steps in any crop production. In the case of fodder berseem there are few varieties which are widely cultivated in India for green fodder production. More than sixteen berseem varieties have been developed and released in India for its cultivation in different parts of the country (Singh *et al.* 2019). Furthermore, a lot of effort is being taken to develop new varieties aiming for high green fodder yield, nutritionally rich, resistance to biotic and abiotic stresses etc. by scientists working on forage crops.

Despite these positive developments, India continues with a longstanding shortage of green fodder. One of the key contributors to this deficit is the limited land allocated for fodder cultivation (Kale and Takawale, 2023). According to Singh et al. (2022), At present, India faces a net shortfall of 35.6% in green fodder, 10.5% in dry fodder, and 44% in concentrate feed

components, posing a significant challenge to livestock productivity. Given the constraints on expanding the area under fodder crops, enhancing fodder availability remains a critical challenge (Takawale *et al.*, 2022).

To overcome this, forage scientists must focus on maximizing fodder yield from the existing land efficient utilization and advanced agronomic practices. This includes the adoption of integrated cropping systems, use of improved fodder varieties, crop rotation, cultivation on marginal or degraded lands, and strategic fertilizer management. Among the various fodder crops, berseem (Trifolium alexandrinum L.) plays a crucial role, particularly during the rabi season. Its widespread adoption by farmers is due to its multi-cut nature, prolonged fodder availability, high green biomass yield, superior digestibility, palatability, and nutritional value. Berseem contains approximately 20% crude protein and 62% total digestible nutrients, making it highly nutritious for livestock (Singh et al., 2018).

In addition to being a valuable fodder source, berseem significantly contributes to soil fertility. As a leguminous crop, it fixes atmospheric nitrogen in the soil, enhancing its fertility when used in rotation with other crops (Singh *et al.*, 2019). Given the importance of berseem in both livestock nutrition and soil health, effective nutrient management becomes vital for achieving optimum yields. Among the various agronomic

inputs, fertilizer management plays a pivotal role in enhancing plant growth by improving nutrient availability. Most chemical fertilizers are water-soluble, allowing for quick absorption by plant roots.

Phosphorus, in particular, is an essential macronutrient for root development and facilitates the uptake of other nutrients. In leguminous crops like berseem, phosphorus is important as it supports especially nodulation and enhances biological nitrogen fixation. It is also a key component of ATP (Adenosine triphosphate), the energy molecule that drives nearly all plant cellular processes. In addition, phosphorus is essential for the synthesis of DNA and RNA, thereby facilitating cell division and the development of tissues (Rawat et al., 2021).

Although various studies have examined berseem production and fertilizer strategies, the current research focuses specifically on the performance of AVT-2 (Second Advanced Varietal Trial) berseem genotypes under different phosphorus levels. This study aims to identify the optimal phosphorus requirement for maximizing both fodder yield and nutrient content while supporting sustainable agricultural practices.

2. MATERIALS AND METHODS

A field experiment was conducted during the *Rabi* season of 2023-24 under the All India Coordinated Research Project on Forage Crops at BAIF's Central Research Station, Urulikanchan, Pune. The objective was to evaluate the performance of berseem genotypes under different phosphorus levels.

The experiment was conducted in a Split Plot Design, with six AVT-2 berseem genotypes assigned to the main plots and three phosphorus levels (60, 80, and 100 kg ha⁻¹) allocated to the sub-plots. Each treatment was replicated trice. Seeds were manually sown with an inter-row spacing of 30 cm. Crop management practices were followed as per the recommended package of practices for berseem cultivation, except for the phosphorus treatments, which were applied as per the experimental design.

Observations were recorded on various growth parameters, including plant height and leaf stem ratio, as well as yield parameters such as green fodder yield, dry matter yield, and crude protein content. The first harvest was carried out at 60 days after sowing (DAS), with subsequent cuts

taken at 30-day intervals. Plant samples collected at 50% flowering stage were used for laboratory analysis for dry matter and crude protein content. A 500 g fresh sample was collected from each net plot for dry matter estimation. These samples were dried in a hot air oven at $70^{\circ}\text{C} \pm 2^{\circ}\text{C}$ until a constant weight was achieved. The dried samples were then ground using a Wiley mill to pass through a 1 mm sieve. Total nitrogen content was determined using the Kjeldahl digestion and distillation method. Crude protein was calculated by multiplying the nitrogen percentage 6.25 (AOAC, 1965).

3. RESULTS AND DISCUSSION

3.1 Performance of AVT-2 Berseem Genotypes under Varying Phosphorus Levels in Terms of Growth and Quality Parameters

3.1.1 Effect of berseem genotypes

Growth parameters such as plant height and leaf stem (L:S) ratio were recorded at each cutting, and the average data is presented in Table 1. The results revealed that berseem genotypes exhibited significant differences in plant height, whereas phosphorus levels did not show any significant difference in plant height. Among the genotypes, JB-08-17 recorded the maximum plant height of 66.10 cm, which was significantly higher than other genotypes. However, Wardan (NC) (63.52 cm) and JHB-20-2 (63.25 cm) were statistically at par. These findings align with previous studies by Nanda et al. (2022) and Godara et al. (2016), which reported significant genotypic variation in plant height, L:S ratio, and crude protein (CP) content in berseem. Such variability can be attributed to the inherent genetic potential of each genotype, as also noted by Naveen et al. (2021). Laboratory analysis of quality parameters revealed that both genotype and phosphorus levels had a positive impact on dry matter content and crude protein content. Among the genotypes, the highest dry matter content of 13.88% was recorded in both PC-114 and BB-2 (CZ), followed by JHB-20-1 with 12.27%. In terms of crude protein, PC-114 (21.45%) and Wardan (NC) (21.52%) exhibited significantly higher dry matter and crude protein content.

3.1.2 Effect of phosphorus levels

Although phosphorus application did not significantly influence plant height, a numerically higher average was observed with 100 kg P_2O_5

ha⁻¹ (63.08 cm), followed by 80 kg ha⁻¹ (62.12 cm). The leaf stem ratio measured at each cut showed no significant variation among the genotypes or phosphorus treatments. In case of quality parameters phosphorus treatments, the application of 60 kg ha⁻¹ and 80 kg ha⁻¹ resulted in significantly higher dry matter contents (12.72% and 12.20%, respectively) and crude contents (20.92% and respectively). These findings are consistent with Roy et al. (2015), who reported a significant influence of phosphorus application on the L:S ratio and nutritional composition of berseem. Phosphorus plays a crucial role in plant physiology, particularly in enzyme activation, DNA and RNA synthesis, and energy transfer processes (e.g., ATP, ADP), all of which are essential for protein synthesis and plant growth (Satpal et al., 2020).

3.2 Performance of AVT-2 Genotypes to Varied Phosphorus Levels in Terms of Green Fodder, Dry Matter, and Crude Protein Yield of Berseem

3.2.1 Effect of berseem genotypes

The data on green fodder, dry matter, and crude protein yields of AVT-2 berseem genotypes under different phosphorus levels were recorded at each harvest and are presented in Table 2. The results indicate that both genotypes and

phosphorus application had significant effects on the yield attributes. Among the evaluated genotypes, JHB-20-2 recorded the highest green fodder yield during the first (290.32 q/ha) and second (281.81 q/ha) cuts. In contrast, PC-114 produced a superior green fodder yield in the third (265.69 q/ha) and fourth (182.75 q/ha) cuts. The total cumulative green fodder yield was marginally higher in JHB-20-2 (967.36 q/ha) compared to PC-114 (966.72 q/ha).

For dry matter yield, PC-114 recorded significantly higher dry matter yield in the second (38.16 q/ha), third (36.88 q/ha), and fourth (25.37 q/ha) cuts, culminating in a total dry matter yield of 134.19 q/ha. In the first cut, BB-2 (CZ) produced the highest dry matter yield (39.65 q/ha), while PC-114 remained statistically at par. Other high performers included BB-2 (CZ) (36.51 q/ha) and JHB-20-1 (34.36 q/ha) in the second cut, and JHB-20-2 (29.79 q/ha) in the third cut. Cumulative dry matter yield for BB-2 (CZ) was 119.33 q/ha, statistically comparable to PC-114.

Regarding crude protein yield, PC-114 recorded significantly higher yields in the second (8.18 q/ha), third (7.91 q/ha), and fourth (5.44 q/ha) cuts, with a total of 28.79 q/ha. BB-2 (CZ) had the highest crude protein yield in the first cut (7.83 q/ha) and also performed well in subsequent cuts.

Table 1. Effect of phosphorus levels and performance of AVT-2 genotypes in terms of growth and quality parameters

Treatments	Plant height (cm)	Leaf Stem ratio	Dry matter content (%)	Crude Protein Content (%)								
AVT -2 genot	AVT -2 genotypes of berseem											
JB-08-17	66.10	0.607	11.29	20.52								
JHB-20-1	58.93	0.688	12.27	19.63								
PC-114	60.91	0.772	13.88	21.45								
BB-2 (CZ)	61.27	0.630	13.88	19.86								
Wardan	63.52	0.649	11.10	21.26								
(NC)												
JHB-20-2	63.25	0.679	11.24	21.52								
SEm±	1.55	0.042	0.001	0.003								
CD@5%	NS	NS	0.003	0.008								
Phosphorus	levels											
60 kg/ha	61.56	0.641	12.72	20.92								
80 kg/ha	62.12	0.693	12.20	20.60								
100 kg/ha	63.08	0.679	11.96	20.61								
SEm±	0.398	0.030	0.001	0.002								
CD@5%	1.17	NS	0.002	0.006								
Interaction	NS	NS	0.005	0.014								

The cumulative protein yield of BB-2 (CZ) was 23.58 q/ha, ranking second after PC-114. On the lower end, Wardan (NC) recorded the lowest cumulative yields for green fodder (821.06 q/ha), dry matter (91.20 q/ha), and crude protein (19.42 q/ha). These variations in productivity are largely attributed to the genetic potential of each genotype. Previous studies by Naveen *et al.* (2021), Devi & Satpal (2019), and Singh *et al.* (2020) have similarly reported significant differences in fodder yield performance across berseem varieties, reinforcing the present findings.

3.2.2 Effect of phosphorus levels

In case of Phosphorus levels, application significantly influenced all yield components. Application of phosphorus @ 100 kg ha⁻¹ consistently produced the highest green fodder yield across all four cuts: 294.71, 277.43, 252.48, and 134.68 q/ha in the first to fourth cuts, respectively. The total green fodder yield was 959.29 g/ha, representing 12.81% and 8.07% higher yields than the 60 and 80 kg P₂O₅ ha⁻¹ treatments. respectively. Yield remained consistent up to the third cut before declining. likely due to increasing temperatures affecting plant growth.

For Dry matter yield highest values were observed at 100 kg P_2O_5 ha⁻¹ in the first (34.95 q/ha), third (30.16 q/ha), and fourth (16.24 q/ha) cuts. Interestingly, the second cut recorded a higher yield (33.50 q/ha) under 60 kg P_2O_5 ha⁻¹. The total dry matter yield with 100 kg P_2O_5 ha⁻¹ was 114.38 q/ha, showing an increase of 5.51% and 5.47% over 60 and 80 kg ha⁻¹, respectively.

For crude protein yield, the 100 kg P_2O_5 ha⁻¹ treatment resulted in the highest yields in the first (7.23 q/ha), third (6.25 q/ha), and fourth (3.38 q/ha) cuts. The second cut saw a slightly higher yield (7.02 q/ha) at 60 kg P_2O_5 ha⁻¹. Cumulatively, phosphorus application at 100 kg/ha increased crude protein yield by 6.52% over 80 kg/ha and 4.27% over 60 kg/ha. These findings highlight the essential role of phosphorus in plant metabolic processes, including energy transfer (ATP), photosynthesis, and protein synthesis. Particularly in leguminous crops like berseem, adequate phosphorus is critical for maximizing biomass and protein output.

The results align with those of Arif *et al.* (2022), who reported improved forage yields with 120 kg P_2O_5 ha⁻¹, Kumar *et al.* (2021) and Kumar *et al.*

(2016), who confirmed that $80-100 \text{ kg P}_2\text{O}_5 \text{ ha}^{-1}$ significantly enhances green and dry fodder yields. Similarly, Samanta *et al.* (2023) emphasized the role of phosphorus and potassium in promoting overall forage productivity and quality.

3.3 Performance of AVT-2 Genotypes to Varied Phosphorus Levels in Terms of Per Day Production of Green Fodder, Dry Matter, and Crude Protein Yield of Berseem

3.3.1 Effect of berseem genotypes

Per day productivity for green fodder, dry matter, and crude protein was calculated for each cut by considering the respective yields and the number of days between harvests. The results are summarized in Table 3. The data revealed a positive influence of phosphorus levels on per day productivity across cuts, except for the second cut. In contrast, the effect of different berseem genotypes on green fodder yield per day was non-significant in most cuts, except the fourth.

Among genotypes, PC-114 recorded the highest cumulative green fodder productivity, averaging 6.45 q/ha/day, followed by JHB-20-1 with 5.97 q/ha/day. Individual cuts showed peak values as BB-2 (CZ) in the first cut: 4.77 q/ha/day, JHB-20-1 in the second cut: 9.33 q/ha/day, PC-114 in the third and fourth cuts: 8.86 and 6.09 q/ha/day, respectively.

3.3.2 Effect of phosphorus levels

Phosphorus levels, a significant effect was observed on per day green fodder productivity across all cuts except the second. The application of 100 kg P_2O_5 ha⁻¹ consistently resulted in higher productivity, recording yield increases of 17.46%, 5.59%, 10.35%, and 25.42% over the 60 kg P_2O_5 ha⁻¹ treatment in the first, second, third, and fourth cuts, respectively.

The maximum cumulative green fodder productivity of 6.40 q/ha/day was also observed under the 100 kg P_2O_5 ha⁻¹ treatment, outperforming both 60 and 80 kg P_2O_5 ha⁻¹ applications. For dry matter and crude protein productivity per day, significant differences were noted among genotypes and phosphorus levels: The highest cumulative daily dry matter yield (0.763 q/ha/day) and crude protein yield (0.158 q/ha/day) were obtained with the application of 100 kg P_2O_5 ha⁻¹.

Table 2. Effect of phosphorus levels and AVT2 genotypes on yield parameters of berseem

Treatment		Green Fo	dder Yie	ld (q ha ⁻¹))		Dry Mat	ter Yield	(q ha ⁻¹)		Crude Protein Yield (q ha ⁻¹)				
	Ist cut	IInd	III rd	IV th	Total	st	IInd cut	IIIrd	IV th	Total	Ist cut	II nd cut	III rd	IV th cut	Total
		cut	cut	cut		cut		cut	cut				cut		
AVT-2 Berseem genotypes															
JB-08-17	278.77	241.04	238.25	124.84	882.90	31.42	27.18	26.87	14.01	99.48	6.44	5.58	5.51	2.87	20.41
JHB-20-1	259.34	280.04	234.95	121.29	895.60	31.56	34.36	28.75	14.90	109.57	6.21	6.1	5.68	2.95	21.65
PC-114	243.33	274.96	265.69	182.75	966.72	33.79	38.16	36.88	25.37	134.19	7.25	8.18	7.91	5.44	28.79
BB-2 (CZ)	285.88	263.27	232.67	79.12	860.93	39.65	36.51	32.27	10.91	119.33	7.83	7.21	6.37	2.17	23.58
Wardan	263.65	270.38	206.12	80.90	821.06	29.29	30.04	22.86	8.99	91.20	6.24	6.39	4.86	1.92	19.42
(NC)															
JHB-20-2	290.32	281.81	265.18	130.05	967.36	32.62	31.68	29.79	14.61	108.70	7.02	6.82	6.41	3.14	23.39
SEm±	10.45	12.66	18.33	11.35	35.49	1.33	1.62	2.41	1.28	4.64	0.27	0.32	0.48	0.27	0.93
CD @5%	NS	NS	NS	36.24	NS	4.26	5.17	7.70	4.09	14.82	0.86	1.03	1.54	0.85	2.96
							Phosphor	us level	S						
60 kg/ha	251.02	262.83	228.28	107.51	850.33	31.86	33.50	29.19	13.86	108.41	6.67	7.02	6.11	2.91	22.71
80 kg/ha	264.92	265.49	239.97	117.29	887.67	32.35	32.44	29.36	14.29	108.45	6.60	6.65	6.02	2.96	22.23
100 kg/ha	294.71	277.43	252.48	134.68	959.29	34.95	33.03	30.16	16.24	114.38	7.23	6.83	6.25	3.38	23.68
SEm±	3.19	4.68	5.41	3.91	12.31	0.41	0.56	0.6	0.50	1.54	0.09	0.12	0.14	0.11	0.32
CD @5%	9.35	NS	15.90	11.50	36.16	1.22	NS	NS	1.47	4.51	0.26	NS	NS	0.31	0.94
Interaction	NS	NS	NS	NS	NS	3.17	4.25	5.17	3.79	11.72	0.66	0.89	1.07	0.80	2.44

Table 3. Effect of phosphorus levels and AVT2 genotypes on yield parameters of berseem in terms of per day production

Treatments	G	reen Fo	dder Yie	ld (q/ha/d	lay)	Dry Matter Yield (q/ha/day)					Crude Protein Yield (q/ha/day)					
	Ist cut	IInd	III rd	IV th	Total	Ist cut	IInd	III rd	IV th	Total	I st cut	IInd	IIIrd	IV th	Total	
		cut	cut	cut			cut	cut	cut			cut	cut	cut		
						AVT-2	Berseem	genotyp	es							
JB-08-17	4.64	8.03	7.94	4.16	5.89	0.523	0.906	0.896	0.468	0.663	0.107	0.187	0.183	0.097	0.134	
JHB-20-1	4.32	9.33	7.83	4.04	5.97	0.527	1.15	0.960	0.498	0.731	0.103	0.227	0.189	0.099	0.144	
PC-114	4.06	9.16	8.86	6.09	6.45	0.563	1.27	1.23	0.847	0.893	0.122	0.274	0.261	0.181	0.192	
BB-2 (CZ)	4.77	8.78	7.76	2.64	5.74	0.660	1.22	1.07	0.36	0.794	0.129	0.242	0.213	0.071	0.159	
Wardan (NC)	4.39	9.01	6.87	2.70	5.47	0.489	1.00	0.762	0.300	0.609	0.104	0.213	0.163	0.063	0.130	
JHB-20-2	4.84	9.39	8.84	4.33	6.45	0.543	1.06	0.993	0.487	0.726	0.117	0.228	0.212	0.104	0.156	

Kale and Takawale; Arch. Curr. Res. Int., vol. 25, no. 9, pp. 118-126, 2025; Article no.ACRI.142187

Treatments	G	reen Fo	dder Yield	d (q/ha/da	ay)		Dry Mat	ter Yield	(q/ha/day	·)	Crude Protein Yield (q/ha/day)					
	Ist cut	IInd	III rd	IV th	Total	I st cut	IInd	III rd	IV th	Total	Ist cut	IInd	III rd	IV th	Total	
		cut	cut	cut			cut	cut	cut			cut	cut	cut		
AVT-2 Berseem genotypes																
SEm±	0.174	0.422	0.611	0.378	0.236	0.022	0.054	0.080	0.042	0.031	0.005	0.011	0.017	0.009	0.006	
CD@5%	NS	NS	NS	1.21	NS	0.072	0.172	0.255	0.135	0.100	0.015	0.034	0.055	0.028	0.020	
						Ph	osphorus	s levels								
60 kg/ha	4.18	8.76	7.63	3.58	5.67	0.532	1.12	0.972	0.462	0.722	0.111	0.234	0.203	0.097	0.152	
80 kg/ha	4.42	8.85	8.00	3.91	5.92	0.538	1.08	0.979	0.477	0.723	0.111	0.223	0.199	0.099	0.148	
100 kg/ha	4.91	9.25	8.42	4.49	6.40	0.583	1.10	1.00	0.543	0.763	0.119	0.228	0.208	0.111	0.158	
SEm±	0.053	0.156	0.180	0.130	0.082	0.007	0.019	0.022	0.017	0.010	0.002	0.004	0.005	0.003	0.002	
CD@5%	0.156	NS	0.530	0.383	0.241	0.020	NS	NS	0.049	0.030	0.005	NS	NS	0.010	0.006	
Interaction	NS	NS	NS	NS	NS	0.053	0.142	0.170	0.127	0.079	0.013	0.030	0.035	0.026	0.016	

This represented an increase of 5.68% and 5.53% over 60 and 80 kg P_2O_5 ha⁻¹ for dry matter productivity, respectively. For crude protein, the improvement was 3.95% and 6.76% over the same phosphorus levels.

These findings are consistent with the results of Arif *et al.* (2022), who reported maximum per day productivity at 120 kg P_2O_5 ha⁻¹, with green fodder and dry matter yields of 578 kg/ha/day and 64 kg/ha/day, respectively. Their study also indicated that 100 kg P_2O_5 ha⁻¹ was statistically at par with the higher dose. Overall, the observed variations in per-day productivity for green fodder, dry matter, and crude protein were largely attributed to differences in total yield and the duration between cuts, which were significantly influenced by genotype performance and phosphorus application levels.

4. CONCLUSION

To get maximum green fodder yield, dry matter yield, and crude protein yield, the genotypes JHB-20-2 and PC-114 were found significantly superior compared to the rest of the genotypes and check varieties. These genotypes also have superior per day productivity in terms of green fodder, dry matter and crude protein yield. In case of phosphorus application @100kg ha-1, the maximum yield parameters.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

The author(s) hereby declare that generative Al technologies were used during the editing of this manuscript. ChatGPT was used for the grammar check and clarity editing of the original draft.

COMPETING INTERESTS

Authors have declared that they have no known competing financial interests or non-financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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