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Effect of nitrogen and phosphorus levels and Nano-DAP application on growth and yield of soybean {*Glycine max* (L). Merr.}

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Abstract

A field experiment was conducted in Randomized Complete Block Design with ten treatments, replicated thrice during *kharif* 2022 to study the response of nitrogen and phosphorus levels in combination with seed treatment and/or foliar spray of novel nano-fertilizer 'Nano-DAP' in soybean. Application of recommended dose of nitrogen and phosphorus (100% NP; 20:60 kg/ha) recorded significantly taller plants at all stages, at par with application of 75% NP + seed treatment (ST) with Nano-DAP + foliar spray (FS) of Nano–DAP, 75% NP application + FS of Nano–DAP, and application of 75% NP + ST with Nano-DAP + its FS @ 0.4%. These treatments were also statistically similar for dry matter accumulation. Application of 100% NP, 75% NP + ST and FS of Nano–DAP, and 75% NP + FS of Nano–DAP, showed similar performance for seed yield but recorded significantly higher seed yield than control (no N and P).

Key words: Nano-DAP, Soybean, Growth, Yield attributes

Soybean {*Glycine max* (L.) Merr} is one of the major oilseed crops being the world's top-ranked edible oil and India's third-largest oilseed crop, after rapeseed mustard and peanut. Due to its superior quality, soybeans are a miracle crop since they are high in vitamins, minerals, salts, and other vital amino acids. They also contain 40–42% protein and 18–20% oil (Pravalika *et al*. 2024). Lysine, which is lacking in most cereals, is present in soybean protein (about 5%), and its use enriches the nutritional quality of cereal flour. Being a good source of flavones, it helps in the prevention of diseases like heart disease, cancer and HIV (Verma *et al*. 2017). It is considered a natural fertilizer factory because of its high nitrogen-fixing ability as it can fix about 35–40 kg N/ha. By producing large amount of biomass, it also increases organic matter in the soil. Globally, soybean is grown over an area of 136.03 m ha and annual production of 369.72 m t with a productivity of 2720 kg/ha (Anonymous, 2023a). In India, it occupies an area of 13.00 m ha with a production of 12.04 m ton and productivity of 930 kg/ha (Anonymous 2023b). In Himachal Pradesh, soybean was grown on an area of 520 hectare in 2022-23 with production of 410 metric tonnes and productivity of 799 kg/ha (Anonymous 2023c).

It has been established that continuous use of high analysis chemical fertilizers leads to deficiency of secondary and micronutrients, soil salinity and environmental pollution (Aziz *et al*. 2015). However, there is a need for the application of different sources of nutrients for sustaining the required crop productivity. In contrast, a relevant amount of fertilizers is released into the environment annually, resulting in eutrophication and groundwater contamination that threatens environmental resources, public health and economic investments. Thus, it will be essential to introduce agro technological innovations and revolutionary agri-inputs that can prevent the degradation of natural resources including environment.

Nanotechnology is the science of manipulating materials at the nano scale. Application of nanotechnology in agriculture has vast and sustainable benefits over many problems. In agriculture this

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technology of using nano – sized particles can be used in fertilizers, pesticides, herbicides, and growth regulators. This unique property enables it to enter easily inside the seed surface or through stomata and other plant openings. Nano – DAP is one such nano – fertilizer that has the ability to reduce the losses of nitrogen and phosphorus, besides increasing productivity of different crops (Poudel *et al*. 2023). This nano – fertilizer comes in sizes ranging from 10 to 30 nanometers and contains 8 per cent nitrogen and 16 per cent phosphorus. Also nano clusters of nitrogen and phosphorus in Nano-DAP are functionalized with bio-polymers and other excipients. Better spread ability and assimilation of Nano-DAP inside the plant system leads to higher seed vigour, more chlorophyll, photosynthetic efficiency, better quality and increase in crop yields. Apart from this, Nano-DAP through precision and targeted application fulfil the nutritional requirement of crops without harming the environment (Prakash *et al*. 2023). Keeping these points in mind the investigation was carried out to study the response of soybean to graded nitrogen and phosphorus levels and seed treatment and / or foliar spray of this nano – fertilizer in soybean.

Materials and Methods

The experiment was carried out at the Research Farm of Department of Agronomy, College of Agriculture, CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur during *kharif* 2022, to study the effect of foliar spray of Nano-DAP on growth and yield of soybean {*Glycine max* (L.) Merr.}. Soil of the experimental site was silty clay loam in texture, acidic in reaction, low in available nitrogen and medium in available phosphorus and available potassium content. The experiment was conducted in a Randomized Block Design (RBD) with ten treatments and three replications. The treatments comprised of T_1: 0 % NP, T_2: 100 % NP T_3: 0 % NP+ seed treatment (ST) with Nano-DAP @ 5 ml/kg seed, T_4: 0 % NP+ ST @ 10 ml/kg seed, T_5: 0 % NP+ foliar spray (FS) @ 0.6 % at 30 days after germination, (DAG) T_6: 0 % NP + (ST) @ 5 ml/kg seed + FS @ 0.2 % at 30 DAG, T_7: 75 % NP + FS @ 0.2 % at 30 DAG, T_8: 75 % NP + ST @ 5 ml/kg seed + FS @ 0.2 % at 30 DAG, T_9: 50 % NP + FS @ 0.4 % at 30 DAG and T_{10}: 50 % NP + ST @ 5 ml/kg seed + FS @ 0.4% at 30 DAG. Potassium was uniformly used in all treatments. The RDF for soybean was 20:60:40 kg/ha and *Harit soya* variety was used. The data on plant height and dry matter accumulation by the crop was recorded at periodic intervals while yield attributes and yield was recorded at the time of harvest.

The data obtained was statistically analyzed as per the procedure outlined by Gomez and Gomez (1984). The critical difference (CD) was estimated for parameters with significant impacts at the 5% probability level.

Results and discussions

Growth parameters

The data pertaining to the effect of nitrogen and phosphorus levels and Nano-DAP application on plant height at periodic intervals and at harvest (Table 1) revealed that the plant height of soybean was significantly impacted by varying NP levels as well as application of Nano-DAP across all stages of observation (Table 1). Significantly taller soybean plants were recorded with application of 100 % NP (T_2) at all stages of observation though this treatment was statistically at par with application of 75 % NP in combination with Nano-DAP as ST @ 5 ml/kg seed and FS of Nano-DAP @ 0.2 % (T_8), application of 75 % NP along with FS of Nano-DAP @ 0.2 % (T_7) and with the treatment where 50 % NP was applied along with ST @ 5 ml/kg seed and FS of Nano-DAP @ 0.4 % (T_{10}). Nitrogen is an essential constituent of chlorophyll as well as protein synthesis which are necessary for cell division and cell elongation (Karimah *et al*. 2024). Higher amount of nitrogen applied in the treatment receiving 100 @ NP resulted in its enhanced availability which ultimately resulted in taller plants. Similarly, phosphorus plays an important role in root development besides being involved in different metabolic processes as well as energy storage and transfer and higher availability of phosphorus, as a result of its higher application, resulted in higher cell division (Sharma *et al*., 2003) and ultimately higher plant height. Similar result showing improved plant height and other growth parameters of mung bean as a result of higher nitrogen application has been reported by Kaur *et al.* (2024) while the beneficial effect of higher phosphorus application on plant height of soybean has been reported by Shilpa *et al.* (2021).

Table 1. Effect of nitrogen and phosphorus levels and Nano-DAP application on plant height and dry matter accumulation of soybean at periodic interval

Sr. No.	Treatment	Plant height (cm)				Dry matter accumulation (g/m^2)			
		30 DAS	60 DAS	90 DAS	Harvest	30 DAS	60 DAS	90 DAS	Harvest
T_1	N_0P_0	24.0	45.5	64.6	65.3	41.3	132.1	305.9	396.7
T_2	N_{100} P_{100}	35.1	64.1	83.2	86.8	49.7	169.2	390.3	496.9
T_3	N_0P_0 + Seed treatment (ST) Nano DAP @ 5 ml / kg seed	25.6	48.6	67.7	68.8	43.4	139.9	320.4	413.3
T_4	N_0P_0 + ST Nano DAP @ 10 ml / kg seed	26.8	50.9	68.8	70.0	44.8	142.7	325.5	421.5
T_5	N_0P_0 + Foliar spray (FS) Nano DAP @ 0.6 % at 30 days after germination (DAG)	26.0	50.5	68.4	69.7	41.4	137.3	316.1	407.3
T_6	N_0P_0 + ST Nano DAP @ 5 ml / kg seed + FS Nano DAP @ 0.6 % at 30 DAG	27.5	52.7	69.8	72.1	43.5	143.1	329.2	423.4
T_7	$N_{75}P_{75}$ + FS Nano DAP @ 0.2 % at 30 DAG	34.6	61.8	80.4	84.3	47.3	163.6	383.3	494.9
T_8	N_{75} P_{75} + ST Nano DAP @ 5 ml / kg seed + FS Nano DAP @ 0.2 % at 30 DAG	34.8	63.2	82.6	86.5	49.7	169.4	394.4	505.5
T_9	N_{50} P_{50} + FS Nano DAP @ 0.4 % at 30 DAG	32.7	57.4	77.9	80.1	45.4	155.2	359.6	467.9
T_{10}	N_{50} P_{50} + ST Nano DAP @ 5 ml / kg seed + FS Nano DAP @ 0.4% at 30 DAG	33.4	60.2	79.9	83.0	46.2	158.6	371.8	477.3
	SEm±	0.7	1.4	1.7	2.1	1.0	3.7	8.0	10.2
	CD (P = 0.05)	2.1	4.2	5.0	6.1	3.0	11.1	23.8	30.3

Treating the soybean seed with Nano-DAP could have resulted in better germination as well as root growth of soybean allowing the roots to mine nutrients from a deeper and wider soil profile which ultimately resulted in taller plants (Shaikh *et al*. 2024). Also the foliar spray of Nano-DAP done at 30 days after germination could have resulted in enhanced availability and better absorption of both nitrogen as well as phosphorus during the active growth period of the crop which would have resulted in better growth and higher plant height. Baghla and Sharma (2023) reported the beneficial effect of bio nano P and K in wheat. Thakur *et al*. (2024a) have also reported better root and shoot development of different crops due to seed treatment with Nano-DAP. Significantly lower plant height of soybean was recorded with no NP application which could be due the inadequate availability of both nitrogen and phosphorus. Although soybean is a leguminous crop that has the ability to fix atmospheric nitrogen but inadequate availability of nitrogen and phosphorus, due to its non-application, could have resulted in poor nodulation, root and shoot growth, and this could have resulted in lower plant height all through the life cycle (Chandel *et al*. 2023).

Dry matter accumulation (g/m^2) in soybean refers to the total biomass produced over time, including leaves, stems, pods, and seeds. The data on the effect of nitrogen and phosphorus levels and Nano-DAP application on dry matter accumulation (g/m^2) at periodic intervals and at harvest (Table 1) revealed that dry matter accumulation (g/m^2) of soybean was significantly influenced by different nitrogen and phosphorus levels as well as by the application of Nano-DAP. Significantly higher dry matter accumulation at all the stages of observation was recorded with the application of 75 % NP along with seed treatment @ 5ml/kg seed supplemented with foliar spray of Nano-DAP @ 0.2 % at 30 DAG, though this treatment was at par with 100 % NP application as well as 75 % NP application along with foliar spray of this nano fertilizer @ 0.2 %. Seed treatment with Nano-DAP could have resulted in better germination and root growth which could have allowed the plants to absorb nutrients from a wider profile of soil resulting in higher availability of nutrients to the crop. Further application of Nano-DAP as foliar spray, owing to the small size of this nano fertilizer, allowed better absorption through the leaves as well as stomata (Shaikh *et al*. 2024) which resulted in increased availability of nitrogen and phosphorus for various biochemical processes, primarily photosynthesis. This higher availability of nutrients from the soil as well as through foliar spray could have resulted in higher leaf area as well as chlorophyll content (Kaur *et al*. 2024) leading to better photosynthetic activity and dry matter production in treatments where seed treatment was practiced along with foliar spray of Nano – DAP. Similar results have also been resulted by Fathi (2022). Also, the combined use of Nano-DAP for seed treatment as well as foliar spray along with no NP application resulted in significantly higher dry matter accumulation as compared to no NP indicating the beneficial effect of both these approaches, when used simultaneously.

Yield attributes and yield

A close perusal of data revealed that application of 100 % NP resulted in significantly higher numbers of pods per plant (Table 2). Sufficient supply of both nitrogen and phosphorus enhance pod setting and lessen blossom drop. Further enhances nitrogen supply promotes vigorous leaf and stem growth, supporting more branches and flowering sites, which could develop into pods. Mere *et al*. (2013) reported that application of 100% NPK produced more pods per plant of soybean. Besides phosphorus also plays a key role in root formation and nutrient uptake, ensuring that the plant has a strong foundation to support reproductive growth (Sharma *et al*. 2014). Phosphorus and nitrogen are also required for protein synthesis and photosynthesis which is important for improving synthesis of plant hormones like cytokinins and auxins, which ultimately resulted in enhanced pod initiation and development. The data further revealed that application of 75 % and 50 % NP along with Nano-DAP seed treatment and foliar spray was also found to be statistically at par with 100 % NP application. Seed treated with Nano-DAP could have resulted in enhanced early availability of these nutrient which promoted better germination and early root development that enhanced water and nutrient uptake. Besides enhanced availability it also increased photosynthetic activity and resulted in enhancement in chlorophyll synthesis and energy metabolism, leading to better reproductive development (Karimah *et al.* 2024). Foliar spray with Nano-DAP also improved the uptake of these nutrients because of its small size would result in efficient use of nutrients which promoted plant growth. Further data revealed that significantly lower

Table 2. Effect of nitrogen and phosphorus levels and Nano-DAP application on yield attributes and seed yield of soybean

Sr. No.	Treatment	No. of pods/plant	No. of seeds/pod	1000-seed weight (g)	Seed yield (q/ha)
T_1	N_0P_0	50.3	1.96	136.0	12.38
T_2	N_{100} P_{100}	59.7	2.37	146.8	17.46
T_3	N_0P_0 + Seed treatment (ST) Nano DAP @ 5 ml / kg seed	51.2	2.05	137.9	13.12
T_4	N_0P_0 + ST Nano DAP @ 10 ml / kg seed	51.7	2.08	138.6	13.42
T_5	N_0P_0 + Foliar spray (FS) Nano DAP @ 0.6 % at 30 days after germination (DAG)	51.5	2.09	138.0	12.96
T_6	N_0P_0 + ST Nano DAP @ 5 ml / kg seed + FS Nano DAP @ 0.6 % at 30 DAG	53.0	2.16	140.5	13.58
T_7	$N_{75}P_{75}$ + FS Nano DAP @ 0.2 % at 30 DAG	59.0	2.30	145.7	17.02
T_8	N_{75} P_{75} + ST Nano DAP @ 5 ml / kg seed + FS Nano DAP @ 0.2 % at 30 DAG	59.5	2.33	146.7	17.39
T_9	N_{50} P_{50} + FS Nano DAP @ 0.4 % at 30 DAG	57.1	2.20	144.0	15.82
T_{10}	N_{50} P_{50} + ST Nano DAP @ 5 ml / kg seed + FS Nano DAP @ 0.4% at 30 DAG	57.8	2.26	144.5	16.32
	SEm±	1.3	0.06	1.7	0.37
	CD (P = 0.05)	3.9	0.16	4.9	1.09

numbers of pods per plant were recorded with 0 % NP application, could be due to the inadequate supply of these major nutrients.

A perusal of data further revealed that application of 100 % NP resulted in significantly higher numbers of seeds per pod though this treatment was statistically at par with the treatments receiving 75 % NP and 50 % NP either with foliar spray or seed treatment supplemented with foliar spray of Nano-DAP (Table 2). Proper fertilization is essential for amino acids and protein synthesis, which directly supports flower and seed development of plant. Adequate nitrogen availability, particularly at flowering enhances pollen production and fertilization success, increasing the likelihood of full seed set in pods (Kaur *et al*. 2024). Further application of phosphorus is necessary for ATP formation, which fuels seed formation and development processes besides improve flower retention, leading to more successful fertilization and seed setting (Shinde *et al*. 2022). As discussed earlier the foliar spray as well as seed treatment along with foliar spray of Nano-DAP resulted in improved fertilization which resulted in better flowering, pod formation and seed setting. Significantly lower number of seeds per pod was recorded with 0 % NP application which could be due to the inadequate supply of nitrogen and phosphorus.

A glance of data presented in Table 2 revealed that significantly higher test weight (1000-seed weight) of soybean was observed with application of 100 % NP though no significant differences were observed between this treatment and other treatments which received either 75 % or 50 % of the recommended nitrogen and phosphorus as basal application along wither with seed treatment and foliar spray or foliar spray alone. Nitrogen is a major component of amino acids and proteins, which are important for seed development and gaining of seed weight. As has been discussed earlier higher availability of nitrogen as a result of higher basal application or due to foliar application of Nano-DAP increased the chlorophyll content besides keeping the leaves photosynthetically active for a longer period of time which could have resulted in better seed filling and ultimately higher 1000-seed weight (Kaur *et al*. 2024). Further energy transfers in crucial seed filling stage was enhanced by phosphorus supply, either due to its higher application dose (50-100 % of recommended P) or as a result of seed treatment and foliar spray, enhanced the energy transfer in better seed filling resulting higher 1000-seed weight. Thakur *et al*. (2024a) have also reported that the foliar spray provides a direct and quick source of nutrients to the plant leaves and other organs, especially during critical stages like pod development and seed filling, further enhancing seed weight. Also the above results are supported by the findings of McKnight *et al*. (2020). Significantly lower 1000-seed weight was observed with application of 0 % NP and this is might be due to the inadequate application of these major nutrients that are important for growth and yield of plants. The use of Nano-DAP as a foliar spray or seed treatment or its use both as seed treatment and foliar spray along with 0 % NP increased the 1000 seed-weight compared to 0 % NP application alone though the increase was not found to be significant.

A close perusal of data revealed that application of 100 % NP significantly recorded higher seed yield of soybean (Table 2) which was statistically at par with the application of 75 % NP along with seed treatment@ 5 ml/kg seed and foliar spray of Nano-DAP @ 0.2 % and 75 % NP along with foliar spray of Nano-DAP @ 0.2 %. The yield of any crop or treatment is a function of the yield contributing characters'/yield attributes. Significantly higher values of all the yield attributes of soybean *viz*., number of pods per plant, number of seeds per pod and 1000-seed weight recorded with application of 100 % NP resulted in significantly higher seed yield of soybean in this treatment. Thakur *et al*. (2024b) have also reported similar findings.

Conclusions

The present study clearly articulated the beneficial effect of use of nano – DAP in enhancing the growth and productivity of soybean crop. Application of 75 % NP along with seed treatment @ 5 ml/kg seed supplemented with foliar spray of Nano-DAP @ 0.2 % at 30 DAG, remaining at par with 100 % NP application, recorded higher values of yield attributes and seed yield indicating the possibility of substituting 25 % of recommended nitrogen and phosphorus through the integrated use of Nano – DAP as seed treatment and foliar spray.

Conflict of interest: The authors declare that there is no conflict of interest among the authors in this research paper.

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