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Effect of Seed Pelleting with *Rhizobium* and Nutrition Management on Growth Parameters of Cowpea [*Vigna unguiculata* (L.) Walp.]

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

An experiment was laid out in a Randomized Block Design (Factorial) with different pelleting material i.e. no pelleting (P₁), *Rhizobium* liquid (P₂), *Rhizobium* powder (P₃) and *Rhizobium* liquid with jaggery (P₄) and 5 levels of nitrogen application i.e. 0% N of RDF(F₁), 40% N of RDF (F₂), 60% N of RDF (F₃), 80% N of RDF (F₄) and 100% N of RDF (F₅), altogether 20 treatment combinations of seed pelleting and nitrogen replicated thrice to find out effect on growth parameters i.e. emergence %, days to 50% flowering, plant height, number of branches per plant, number of pods per plant and number of seeds per pod of cowpea cv. Himachal Lobia-1. Among different pelleting treatments, seed pelleting with *Rhizobium* liquid exhibited better results over other treatments and all growth parameters affect significantly by pelleting treatments. Different level of nitrogen application also had significant effect on growth parameters. 80% N of recommended dose of fertilizer was best treatment over other treatments. Interaction effect of seed pelleting and nitrogen application was non-significant on growth parameters except number of branches per plant. Maximum number of branches was found in P₂F₄ (*Rhizobium* liquid + 80% N of RDF) treatment.

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1. INTRODUCTION

Cowpea (Vigna unguiculata (L.) Walp.) is one of the most important crops which is grown commercially as a grain crop, animal fodder and vegetable crop [1] in the semi-arid regions of Africa, Asia, Southern Europe, Southern United States, and Central and South America [2-4]. It belongs to family Leguminosae and is native of South-Africa [5]. The vast majority of the world's cowpea production (over 95%) takes place in sub-Saharan Africa with about 12.5 million hectares under cultivation worldwide in 2014. Asia is the second largest producing region. representing less than 3% of the global production in average over the 1993-2014 periods. In world, approximately over 5.59 million metric tons during the year 2014 was produced [6]. It is a crop of major importance to the nutrition of poor rural households whose diets tend to heavily rely on starchy foods such as millet, sorghum, maize and cassava. Therefore, it has a tremendous potential to contribute to the alleviation of malnutrition because it contains carbohydrate and proteins in huge quantity along with less quantity of fats, fiber, B-vitamins such as folic acid, thiamine, riboflavin and niacin and micronutrients such as iron and zinc [7].

Cowpea has ability to fix atmospheric nitrogen in soil at the rate of 56 kg/ha in association with symbiotic bacteria under favorable conditions [8,9] and biological nitrogen fixation leads to noteworthy reduction in production cost incurred on nitrogen fertilizers [10]. With a view to make sure the buildup of sufficient bacterial population and to have adequate symbiosis between bacteria and plant roots, *Rhizobium* is applied artificially in many ways like soil and seed applications. The application of *Rhizobium* through seeds in the form of seed pelleting is one of the effective ways to ensure the optimum symbiosis for enhanced crop productivity. Seed pelleting focuses on better establishment and increased productivity by precision sowing and addition of various materials along with filler material that improve the seed quality in many ways [11]. Keeping in view the above prospective, this study had been planned to determine the effect of seed pelleting with *Rhizobium* and nitrogen application on vegetative growth parameters of cowpea crop.

2. MATERIALS AND METHODS

The experiment was carried out at farm of Department of Seed Science and Technology, Dr Y S Parmar University of Horticulture and Forestry, Nauni, Solan (HP) is located at an altitude of 1250 meters above mean sea level with latitude of 35.5°N and longitude of 77.8°E in the mid- hill zone of Himachal Pradesh, India. Climate of the area is generally sub-temperate and semi-humid characterized by cold winters. Generally, December and January are the coldest while; May and June are the hottest with moderate rainfall (1000-1300 mm). The soil texture was loam to clay loam having pH ranging from 6.85-7.05.

Genetically pure seed of cowpea cv. Himachal Lobia-1 was purchased from Directorate of Extension Education, Dr Y S P University of Horticulture and Forestry, Nauni, Solan. Liquid and powder form of Rhizobium culture was obtained from Soil microbiology laboratory of Department of Soil Science and water management, UHF Nauni, Solan. Seed pelleting was done at CSIR-IHBT (Institute of Himalayan Bioresource Technology) Palampur, Himachal Pradesh. For seed pelleting clay and adhesive (45% Gum Arabica) was used. There were total 20 treatment combinations (P \times F) and each treatment was replicated three times so there were total 60 plots of $1.8 \text{m} \times 1.5 \text{m}$ (i.e. 2.7 m²) size.

Table 1. Meteorological data on rainfall, temperature and relative humidity during the course of
investigation

Month	Mean temperature (°C)	Mean rainfall (mm)	Relative humidity (%)	
June	38.05	91.1	60	
July	38.20	294.40	79	
August	38.05	102.20	80	
September	36.75	41.60	68	
October	19.65	34.60	58	

Source: Meteorological Observatory, Department of Environmental Sciences, Dr. Y S Parmar University of Horticulture and Forestry, Nauni-173 230, Solan (HP) There were total 40 plants in every plot and spacing was 45cm × 15cm. Urea is applied to these plots and recommended dose for cowpea is 45 kg/hectare. Cultural practices were done according to package of practices for vegetable crops, Directorate of Extension Education, Dr Y S Parmar University of Horticulture and Forestry, Nauni, Solan (HP). Recorded observations were emergence %, days to 50% flowering, plant height, number of branches per plant, number of pods per plant and number of seeds per pod. The statistical analysis for RBD was done as per design of experiment as suggested by [12].

3. RESULTS AND DISCUSSION

Emergence percentage of cowpea seeds was significantly affected by different seed pelleting with Rhizobium treatments and different dose of nitrogen application. When seeds were pelleted with Rhizobium (liquid form), emergence percentage was maximum (93%) and nonpelleted seeds shown only 89.67% emergence in field condition. Seed pelleted with dry form of Rhizobium gave 91.17% emergence. Among different dose of nitrogen application, 80% N of RDF (F_{4}) gave maximum (92.50%) emergence and minimum (89.79%) emergence was found in control (0% N of RDF). Interaction effect of seed pelleting and nitrogen application was nonsignificant on emergence of cowpea seeds. It might be due to continuous supply of nutrients to seeds present in pelleting material which helped in survival of seedlings. Similar increased emergence due to seed pelleting was also found by Raj [13] in cowpea. Such beneficial influence

(higher germination percentage) of seed pelleting with *Rhizobium* was also found in Bengal gram, Moth beans, and Green gram and peas by Pawar et al. [14]. The hike in germination also could be attributed to higher cytokinin production due to biofertilizer which led to higher cell division. Ahmed et al. [15] also reported increased germination in French bean due to seed pelleting with *Rhizobium*.

Different types of pelleting and different doses of nitrogen had significant effect on days to 50% flowering of cowpea. Among different pelleting methods, minimum (40.33) days for 50% flowering was taken when seed were pelleted with liquid form of Rhizobium and maximum (41.08) days was taken when seeds were not pelleted. Among different dose of nitrogen application, 50% of flowering was first occurred when 80% N of RDF was applied and maximum days for 50% flowering when 40% N of RDF was applied. Interaction effect of seed pelleting and nitrogen application was non-significant on days to 50% flowering. Reduction in days to 50% flowering might be attributed to the fact that pelleted seeds germinated early leading to faster vegetative growth of plant and flowering growth started early in comparison to plants which were germinated from non-pelleted seeds. Nitrogen is known to stimulate vegetative growth therefore, days to 50% flowering increased with increase in nitrogen dose. Reduced days to 50% flowering due to seed pelleting with Rhizobium was also reported by Patel and Jadav [16] in cowpea cv. Pusa Phalguni.

Treatment	Total	Days to	Plant	Number of	Number of	Number of	Number of
	emergence	50%	height	branches	seeds per	pods per	seeds per
	(%)	flowering		per plant	pod	plant	plant
Seed pelleting							
P 1	89.67 (9.52)	41.08	83.80	3.86	12.25	12.90	158.05
P ₂	93.00 (9.70)	40.33	90.18	4.44	12.90	14.05	181.24
P3	91.17 (9.60)	40.61	89.28	4.25	12.64	13.39	169.31
P ₄	90.67 (9.57)	40.85	85.54	4.24	12.39	13.36	165.51
CD _{0.05}	0.06	0.38	0.43	0.08	0.20	0.16	3.08
Nitrogen application							
F ₁	89.79 (9.53)	40.98	86.32	4.00	12.27	13.06	160.40
F ₂	90.63 (9.57)	41.01	86.74	4.07	12.43	13.31	165.45
F ₃	91.25 (9.60)	40.93	87.10	4.18	12.50	13.43	167.98
F ₄	92.50 (9.67)	40.33	88.11	4.41	12.87	13.72	176.58
F ₅	91.46 (9.62)	40.34	87.72	4.31	12.65	13.61	172.21
CD _{0.05}	0.07	0.43	0.48	0.09	0.23	0.18	3.44
Interaction ($P \times F$)							
CD _{0.05}	NS	NS	NS	0.17	NS	NS	NS

Table 2. Effect of seed pelleting and nitrogen application on growth parameters of cowpea

P₁-Control, P₂-Rhizobium liquid + clay + adhesive, P₃- Rhizobium powder + clay + adhesive, P₄- Rhizobium liquid formulation + jaggary, F₁-0 % N of RDF, F₂ -40 % N of RDF, F₃ -60 % N of RDF, F₄ -80 % N of RDF, F₅ -100 % N of RDF Figures in parenthesis are square root transformed

Plant height of cowpea was increased when seeds were pelleted with Rhizobium and maximum (90.18 cm) plant height was obtained when seed were pelleted with liquid form of Rhizobium and minimum (83.80 cm) was found in control (P1). Different level of nitrogen application had also significant effect on plant height of cowpea. 80% N of RDF gave maximum (88.11 cm) plant height and minimum (86.32 cm) was found in control (0% N of RDF) treatment. Interaction effect of seed pelleting and nitrogen application on plant height of cowpea was nonsignificant. This may be attributed to the fact that pelleting material maintained nutritive reserve and dehydrogenase activity in seeds and nitrogen is an essential part of chlorophyll and nucleic acids, which might have played major role in promoting plant growth in terms of plant height, number of branches per plant, number of seeds per pod etc. hike in plant height due to seed pelleting with Rhizobium was also reported by Khaitov et al. [17] in chickpea. Increased plant height due to nitrogen application was also reported by Upadhyay and Singh [18] in cowpea and by Starling et al. [19] in soybean.

Number of branches per plant was affected significantly by seed pelleting and different dose

of nitrogen application. Minimum number of branches was obtained when seeds were not pelleted and 0% N of RDF was applied. When seeds were pelleted with liquid form of Rhizobium, maximum (4.44) branches per plant was obtained. In non-pelleted seeds there were only 3.86 (minimum) branches per plant. Increment in nitrogen dose up to 80% N of RDF also increase number of branches per plant. Minimum number of branches was found when nitrogen was not applied. Interaction of seed pelleting and nitrogen application also affected number of branches significantly. Maximum number of branches per plant (4.67) was found in P_2F_4 (*Rhizobium* culture pelleting + 80% N of RDF) which was at par with P_2F_3 (4.53) and P_2F_5 (4.58). However, minimum number of branches per plant (3.80) was found in P_1F_1 (control + 0%) N of RDF). This might be due to constant and optimal supply of nitrogen and other nutrients under Rhizobium treatments which led to more plant height and plants with more height bear a greater number of branches. Similar results of increased number of branches per plant due to seed inoculation with Rhizobium and nitrogen application was reported by Yadav and Malik [20] in cowpea.

 Table 3. Interaction effect of seed pelleting and nitrogen application on growth parameters of cowpea

Interaction	Total	Days to	Plant	Number of	Number	Number	Number
treatment	emergence	50%	height	branches	of seeds	of pods	of seeds
	(%)	flowering	(cm)	per plant	per pod	per plant	per plant
P_1F_1	88.33 (9.45)	41.32	82.93	3.80	11.99	12.60	151.10
P ₂ F ₁	90.83 (9.58)	40.34	89.36	4.19	12.71	13.40	170.23
P ₃ F ₁	90.83 (9.58)	41.42	88.25	4.05	12.29	13.07	160.67
P₄F₁	89.17 (9.50)	40.82	84.74	3.96	12.10	13.19	159.61
P_1F_2	89.17 (9.50)	41.12	83.00	3.86	12.17	12.77	155.40
P_2F_2	91.67 (9.63)	40.75	89.46	4.21	12.83	13.82	177.28
P ₃ F ₂	90.83 (9.58)	40.94	89.13	4.11	12.33	13.39	165.10
P ₄ F ₂	90.83 (9.58)	41.22	85.38	4.10	12.39	13.24	164.03
P₁F₃	89.17 (9.50)	41.87	83.33	3.84	12.26	12.90	158.20
P_2F_3	93.33 (9.71)	40.75	90.14	4.53	12.88	14.07	181.20
P ₃ F ₃	90.83 (9.58)	40.37	89.41	4.15	12.44	13.44	167.23
P ₄ F ₃	91.67 (9.63)	40.72	85.52	4.21	12.43	13.30	165.31
P1F₄	91.67 (9.63)	40.31	85.22	3.95	12.49	13.14	164.05
P_2F_4	95.00 (9.80)	40.03	91.20	4.67	13.09	14.53	190.12
P ₃ F ₄	91.67 (9.63)	39.90	89.87	4.55	13.33	13.57	180.81
P_4F_4	91.67 (9.63)	41.08	86.16	4.47	12.56	13.65	171.35
P₁F₅	90.00 (9.54)	40.76	84.50	3.85	12.35	13.07	161.50
P ₂ F ₅	94.17 (9.76)	39.80	90.74	4.58	12.99	14.43	187.37
P₃F₅	91.67 (9.63)	40.39	89.73	4.37	12.80	13.49	172.74
P ₄ F ₅	90.00 (9.54)	40.42	85.92	4.46	12.45	13.43	167.23
CD _{0.05}	NS	NS	NS	0.17	NS	NS	NS

P₁-Control, P₂-Rhizobium liquid + clay + adhesive, P₃- Rhizobium powder + clay + adhesive, P₄- Rhizobium liquid formulation + jaggary, F₁-0 % N of RDF, F₂ -40 % N of RDF, F₃ -60 % N of RDF, F₄ -80 % N of RDF, F₅ -100 % N of RDF Figures in parenthesis are square root transformed Number of seeds per pod and pods per plant were increased when seeds were pelleted with Rhizobium. Maximum number of seed per pod and pods per plant was obtained when liquid form of Rhizobium was used for seed pelleting over other treatments and minimum numbers were found in control (P1). Different dose of nitrogen application also affected these growth parameters significantly. 80% N of RDF (F_4) produced maximum seeds per pods and pods per plant and in control (F₁) treatment. Interaction effect of seed pelleting and nitrogen application was non-significant on number of pods per plant and seeds per pod. The production of more pods per plant and seeds per pod with application of higher doses of nitrogen in comparison to lower doses and/or with no nitrogen also could be attributed to the fact that more biomass in terms of vegetative growth might have been put up with higher levels of nitrogen and nitrogen is known to stimulate vegetative growth and it might be also due to more number of flowers. Similar results of increased number of pods per plant due to seed pelleting with Rhizobium and nitrogen application were reported by Swaroop et al. [21] in cowpea cv. Arka Garima.

Different type of seed pelleting and different dose of nitrogen application had significant effect on number of seeds per plant. Number of seeds per plant was increased by 14.67% when seeds were pelleted with liquid form of Rhizobium over control. Among different dose of nitrogen, 80% N of RDF gave maximum (176.58) number of seeds per plant which were 10% more of control (F₁). Interaction effect of seed pelleting and nitrogen application was non-significant in number of seeds per plants. The increased number of seeds per plant might be due to seed pelleting with Rhizobium and nitrogen doses, more nodulation because of nitrogen and Rhizobium pelleting has occurred which led to a greater number of seeds per pod and pods per plant thereby, increase number of seeds per plant. Similar results of increased number of seeds per plant due to nitrogen application were reported by Farahvash and Mirshekari [22] in cowpea.

4. CONCLUSION

It is concluded that seed of cowpea shows better emergence% and vegetative growth in term of plant height, number of branches, number of pods per plant and number of seeds per pod when they are treated with Rhizobium.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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