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Effect of Nano Zinc Oxide Seed Treatment on Physiological Growth and Biophysical Traits and Seed Quality of Soybean [*Glycine max* (L.) Merrill]

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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

The field experiment was conducted at Seed Technology Research Center, Department of Plant breeding and Genetics, JNKVV-Jabalpur *Kharif*-2021. Growth analytical attributes studies indicated superior performance of seed treatment with nano-ZnO @500mg L^{-1} for LAI @60 DAS (days after sowing), @70 DAS, @80 DAS, LAD, CGR, RGR and BMD w.r.t. 24.26%, 21.39%, 28.50%, 12.09%, 21.42%, 20.17% and 20.56% enhancement over control. Varietal studies revealed

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superior performance of variety JS 20-116 for LAI, LAD, CGR, RGR and BMD. Biophysical parameters studies revealed superior performance of seed treatment with nano –ZnO @500mg L^{-1} for chlorophyll content index and energy interception with 5.19% and 26.51% enhancement over control respectively. With respect to variety superior performance of variety JS-20-116 for chlorophyll content index and energy interception. Seed quality parameters studies indicated superior performance of seed treatment with nano–ZnO @500mg L^{-1} for final plant stand, germination, and seed vigour index I and seed vigour index II with respect to 29.16%, 14.11%, 26.40% and 21.43% enhancement over control. With respect to variety, superior performance of variety JS-20-116 for final plant stand, and seed vigour index I and seed vigour index II and JS 20-34 for seed germination. This results justifies that the seed treatment with nano formulation in the form of ZnO @ 500mg L^{-1} will improve growth analytical attributes, physiological efficiency and seed quality of the soybean. Hence the seed treatment with nano-ZnO @500mg L^{-1} is recommended to farmers for maximum productivity, physiological efficiency and sowing seed quality.

Keywords: Growth analytical attributes; micro-nutrient; nanoparticles; nanotechnology; seed treatments.

1. INTRODUCTION

Soybean is, designated as a wonder crop, has confirmed its potential as an industrially important and valuable oil seed crop in many areas of India. Soybean cultivation has placed India on the world map in recent past soybean has not only gained the vital importance in Indian agriculture, but also plays an important role in oil economy of India [1]. Though, soybean is a legume crop, yet it is widely used as oilseed crop. It is grown in varied agro-climatic conditions. It is the most important crop in terms of protein and fat content. It contains about 35-40% good quality protein, 20 % oil having about 85% saturated fatty acids, including 55% polyunsaturated fatty acids (PUFA) 25 – 30% carbohydrates. It is an abundant source of protein and oil, and it is also known as vegetarian meat. It is similar to cow's milk and animal proteins as it also contains all the essential amino acids, including glycine, tryptophan, and lysine. Soybean seeds are high in phosphorus, potassium, sulphur, iron, vitamin A, D, E, K, unsaturated fatty acids with anti-cholesterol properties, lecithin, 30% carbs, 4% saponins, 5% fibre, and 18-20% oil. Sprouts include a significant amount of vitamin C, which is commonly found in fresh fruits and vegetables [2].

Nano-technology has the potential to revolutionize the agriculture and plays a significant role in enhancing food and crop production. During the past decade, a number of patents and products incorporating engineered nano-particles (NPs) into agricultural practices, viz., nano-pesticides, nano-fertilizers and nano-sensors, have been developed with the collective goal to enhance the precision and minimize the input and enhancing farm income than conventional products and approaches [3]. In recent years, several metal based nano-particles viz., Ag NPs [4], Au NPs [5], Cu NPs and Fe NPs [6], FeS_2 NPs [7], TiO_2 NPs, Zn NPs [8] and ZnO NPs [9] have been applied as seed pre-treatment agents for promoting seed germination, seedling growth and stress tolerance in some crop plants.

Zinc is an essential micronutrient for plant growth and reproduction. It has several functions in the plant, such as enzyme activation and regulation, protein formation, photosynthesis, carbohydrate assimilation, fertility, and production of seeds [10]. Zinc sulfate and chelate were used as zinc fertilizers added to plants, whether as soil or foliar, but their efficiency is low. Moreover, zinc sulfate fertilizer has highly cost compared with zinc oxide as a source of Zn. We hypothesized that nano-scale ZnO, due to its smaller size and higher specific surface area penetrates better and enhances the seed germination, plant establishment, vigor, growth rate, physiological efficiency, and productivity. Only a few researchers have seen the effect of nano-ZnO on seed germination and yield. However, our effort is to decipher the effect of nano-ZnO on the growth analytical attributes, physiological efficiency and seed quality of the soybean.

2. MATERIALS AND METHODS

The research work was conducted at Seed technology research center, Department of Plant breeding and Genetics, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur (Madhya Pradesh) during *kharif* season 2021.The topography of the experimental field area was fairly uniform. All facilities viz., labourers, agrochemicals, equipment and irrigation etc., were adequately available on the research farm to carry out the field experiment. The treatments were laid out in Factorial RBD (Randomized Blocks Design) with eight replications. The different treatments were randomized within each replication using a random Table 1. Seed of soybean varieties JS 20-116 and JS 20-34 were sown in the field by hand dibbling maintaining a distance of 0.40m between rows and 0.05m between plants. The experimental plots were kept weed free and hand weeding was done as and when required.

The physiological observations (growth analytical parameters) include leaf area index (LAI), leaf area duration (LAD), crop growth rate (CGR), relative growth rate (RGR) and biomass duration (BMD). Leaf area was estimated at 60, 70 and at 80 days' stages using Laser Area Meter (model CI 203). LAI expresses the ratio of leaf surface (one side only) to the ground area occupied by the plant or a crop stand worked out as per specifications of Gardner et al. [11]. Leaf area duration expresses the magnitude and persistence of leaf area or leafiness during the period of crop growth [12]. It reflects the extent of seasonal integral of light interaction and correlated with yield. The daily increment in plant biomass is termed as crop growth rate [12]. The Relative growth rate expresses the dry weight increase in time interval in relation to initial weight. CGR and RGR were determined as per the following formula suggested by Watson, [12]. It was calculated as per formula given by Williams, [13]. Biomass Duration is the parameter that represents dry weight losses or gains during a unit time period.

Biophysical parameters includes chlorophyll content index (CCI), energy interception and light transmission ratio (LTR). Chlorophyll content index which is expressed as grams of chlorophyll per unit ground area and it was determined in the 4th leaf of five weeks old plant using a non-destructive method that uses an optical instrument called chlorophyll meter (Model: CCM 200 Made in USA). The total incident light at the canopy crown and transmitted light within the crop were converted into average incident and transmitted energy on the basis of value reported by Gaastra [14], 71 KLux = 1 Cal cm^{-2} min^{-1}. The efficiency of the crop canopy for solar energy interception (Ei) was calculated as per the formula given by Hayashi [15].

Ei = Total incident - Transmitted energy

The light intensity, incident on crop canopy surface and infiltration profile within the canopy at the ground level was recorded by Lux-meter (Model – LX - 105). The LTR was calculated as per formulae given by Golingai and Mabbayad's [16].

Seed quality parameters included final plant stand m^{-2}, seed germination (%), seedling shoot length (cm), seedling root length (cm), seedling dry weight (g), seed vigour index I and seed vigour index II (Fig. 1 and Fig. 2). Three replication of 100 seeds from respective treatments were used for germination by using paper towel methods (BP) at 25 ± 20°C in seed germinator for 8 days at 90% relative humidity. The seeds were categorized as normal seedlings, abnormal seedlings, hard seed, and dead seed. The germination percentage was recorded based on normal seedlings only. Seedling length and root length of 10 normal seedlings in cm is measured during the final count. The weight of seedling excluding the cotyledons is taken on 10th day after drying them at 60-80˚C in an oven for 24 hrs in g. The lot exhibiting maximum dry weight is considered as vigorous. A combination of standard germination test with seedling length provides evaluation for seed vigor [17].

Vigor index- I = Germination percentage x seedling length at final count

Vigor index- II = Germination percentage x seedling dry weight at final count

3. RESULTS AND DISCUSSION

3.1 Physiological Growth Parameters

The results revealed that significant variations was observed for LAI except with respect to treatment at 80 DAS and with respect to treatment and variety interaction showed non-significant differences for LAI during entire period of crop growth; non-significant variations was observed for LAD during crop growth stages except with respect to variety LAD at 70-80 DAS of crop growth. The CGR, RGR and BMD in varieties (V), treatments (T), varieties and treatments interaction (V x T) varied non-significantly at different crop growth stages (Table 2).

LAI (Leaf Area Index): For determining light interception and transpiration, the leaf area index is one of the most essential plant growth indicators. As a result, LAI is a crucial element in many crop development models that employ net photosynthesis, assimilate partitioning, canopy mass, and energy exchange to estimate yield.

The leaf area index describes the ratio of leaf surface to cropped ground area, and it is a practical way of capturing solar energy and transforming it into food and other useful components. The range of LAI at 60 days, 70 days and 80 days were found to be 3.29 to 6.59. The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 24.26%, 21.39% and 28.50% in LAI at 60 days, 70 days and 80 days respectively over control. In terms of seed treatment, seed treatment with ZnO nano-particle @ 500mg L^{-1} was found to be superior over untreated control. The present study indicated that the maximum LAI was obtained at 70 DAS later on it decreases. This is because of the decline was attributed to the reduction in the quantum of assimilatory surface area due to drying and senescence of leaf and movement of photo assimilates to other sinks of the plant, particularly economic sinks that generally have higher sink demands. The leaf area index was significantly increased in soybean from six weeks after sowing. Our finding is in consistent with Hassan et al. [18] reported that Leaf Area Index was substantially improved and was observed maximum during middle of growth period but that was constant when crop reached towards maturity. Raj and Chandrasekhar, [19] also reported that among seed treatments, LAI, LAD, and SPAD chlorophyll meter values were recorded with nano ZnO seed treatment than seed priming with nano zinc solution and chelated ZnSO4 seed treatment. Same result were observed with Razzaq et al. [20] who reported that nano seed treatment increases LAI and LAD in maize.

Leaf Area Duration (LAD): The leaf area duration is a crucial factor in contributing to photo-assimilate production because it signifies an active phase of leaf growth and leaves survival period. The longer the leaves are active, the more photosynthates they produce. If this food is adequately translocated to developing sinks, it will make a significant contribution to increasing economic productivity. The range of LAD 60-70 days and 70-80 days was found to be 17,809.28 to 22,241.56 and 16,890.08 to 21,164.57 respectively. The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 13.00% and 14.56% in LAD 60-70 days and LAD 70-80 days respectively over control. In terms of seed treatment, seed treatment with ZnO nano-particle @ 500mg L^{-1} was found to be superior over untreated control. The present study showed that the maximum LAD was observed at 60-70 DAS thereafter, it showed reduction during the subsequent growth phase. This reduction in leaf area duration because crop is maturing. Our result is in conformity with Raj and Chandrasekhar [19] reported that among seed treatments, higher seed cotton yield (2842 kg ha^{-1}), LAI, LAD, and SPAD chlorophyll meter values were recorded with nano-ZnO seed treatment. Same result were observed with Razzaq et al. [20] reported that nano seed treatment increases the LAI and LAD in maize.

Crop Growth Rate (CGR) (g cm^{-2} day^{-1}): Crop growth rate is the rate of dry matter accumulation per unit area per unit time. It is a measurement of how much crops grow in size and bulk over time. Grain yield was found to have a strong relationship with LAI, LAD, and CGR. The range of CGR 60-70 days and 70-80 days were found to be 0.00183.28 to 0.00268 and 0.002441 to 0.00341 respectively. The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 28.57% and 26.35% in CGR @60-70 days and CGR @70-80 days over control. In terms of seed treatment, seed treatment with ZnO nano-particle @ 500mg L^{-1} was found to be superior over untreated control. This might be due to efficient dry matter accumulation, photosynthesis, vegetative and reproductive growth. Tariq et al*.* [21] observed that foliar applied Zn and B increased the photosynthesis and chlorophyll production that ultimately increases the CGR. Nano-fertilizers boost crop growth, yield, and quality by increasing nutrient use efficiency, reducing fertilizer waste, and lowering cultivation costs [22].

Relative Growth Rate (RGR) (g g^{-1} day^{-1}): RGR is the basic parameter that gives one of the most ecologically important and valuable plant growth indices. The relative growth rate shows the increase in existing biomass, which could be a key component in increasing production during a given growth period. The range of RGR @60-70 days and RGR @70-80 days were found to be 0.047 to 0.067 and 0.040 to 0.055 respectively. The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 22.44% and 28.57% in RGR @60-70 days and RGR @70-80 days respectively over control. In terms of seed treatment, seed treatment with ZnO nano-particle @ 500mg L^{-1} was found to be superior over untreated control. Alabdallah and Alzahrani [23] reported the addition of (bulk ZnO) increased the (RGR) non-significantly in all seawater concentrations. However, (ZnO-NPs) increased these measures significantly as compared to their corresponding controls.

Table 1. Factor and treatment combination

	Factor 1 – Varieties
V1	JS 20-116
V2	JS 20-34
	Factor 2 – Seed treatments
T1	: Control
T2	: State recommended package of practice (Rhizobium @ 5g kg^{-1} seed and Vitavax @ 5g kg^{-1} seed)
T3	: Seed treatment with nano-ZnO @ 500mg L^{-1}
	Treatment combinations
V1T1	JS 20-116+ Control
V1T2	JS 20-116+ state recommended package of practice (Rhizobium @5g kg^{-1} seed and Vitavax @5g kg^{-1} seed)
V1T3	JS 20-116 + seed treatment with nano-ZnO @ 500mg L^{-1}
V2T1	JS 20-34 + Control
V2T2	JS 20-34 + state recommended package of practice (Rhizobium @5g kg^{-1} seed and Vitavax @ 5g kg^{-1} seed)
V2T3	JS 20-34+ seed treatment with nano-ZnO @ 500mg L^{-1}

Where: varieties (V), treatments (T), varieties and treatments interaction (V x T)

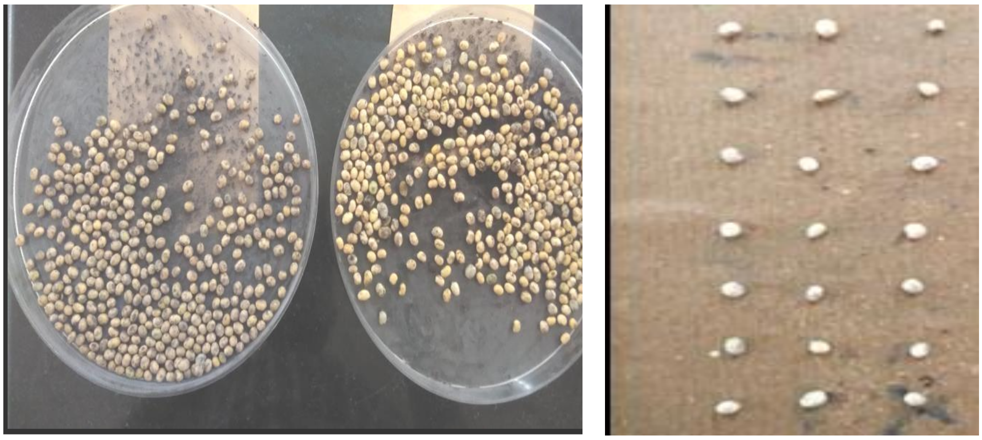

Fig. 1. Assessment of Post-Harvest Seed Quality

Fig. 2. Seeds quality - seedling shoot and root length

Table 2. LAI, LAD, CGR, RGR and BMD in soybean during successive growth intervals under the effect of varieties, seed coating treatments and its interaction

		T1	T2	T3	Mean(V)	S.Em±	CD (5%)
				LAI			
60 DAS	JS 20-116	5.17	5.66	6.00	5.62	T= 0.27	T= 0.80
	JS 20-34	3.73	4.34	5.03	4.37	V=0.22	V=0.65
	Mean (T)	4.45	5.00	5.53		V×T=0.39	V×T=NS
70 DAS	JS 20-116	5.85	6.16	6.59	6.20	T= 0.26	T= 0.77
	JS 20-34	3.87	4.26	5.20	4.44	V=0.21	V=0.63
	Mean (T)	4.86	5.21	5.90		V×T=0.37	V×T=NS
80 DAS	JS 20-116	5.13	5.69	6.02	5.62	T=0.45	T= NS
	JS 20-34	3.29	4.12	4.81	4.07	V=0.37	V=1.06
	Mean (T)	4.21	4.91	5.41		V×T=0.64	V×T=NS
				LAD (cm^2 $days^{-1}$)			
60-70 DAS	JS 20-116	19,922.67	20,886.88	22,241.56	21,017.04	T= 1,209.09	T= NS
	JS 20-34	17,809.28	18,564.58	20,395.09	18,922.98	V=987.22	V= NS
	Mean (T)	18,865.97	19,725.73	21,318.32		V×T=1,709.91	V×T=NS
70-80 DAS	JS 20-116	17,892.38	19,774.62	21,164.57	19,610.53	T= 753.96	T= NS
	JS 20-34	16,890.08	17,596.20	18,684.02	17,723.43	V=615.61	V=1774.9
	Mean (T)	17,391.23	18,685.41	19,924.29		V×T=1,066.27	V×T=NS
				CGR (g cm^{-2} day^{-1})			
60-70 DAS	JS 20-116	0.00209	0.0022	0.00268	0.00232	T= 0.00028	T= NS
	JS 20-34	0.00183	0.00201	0.00235	0.00206	V=0.00022	V= NS
	Mean (T)	0.00196	0.0021	0.00252		V×T= 0.00039	V×T=NS
70-80 DAS	JS 20-116	0.00273	0.00306	0.00341	0.00307	T= 0.000412	T= NS
	JS 20-34	0.00244	0.00277	0.00312	0.00277	V= 0.000337	V=NS
	Mean (T)	0.00258	0.00292	0.00326		V×T= 0.000583	V×T=NS
				RGR (g g^{-1} day^{-1})			
60-70 DAS	JS 20-116	0.051	0.053	0.067	0.057	T= 0.010	T= NS
	JS 20-34	0.047	0.049	0.053	0.05	V=0.008	V= NS
	Mean (T)	0.049	0.051	0.06		V×T=0.014	V×T=NS

		T1	T2	T3	Mean(V)	S.Em±	CD (5%)
70-80 DAS	JS 20-116	0.044	0.054	0.055	0.051	T= 0.008	T= NS
	JS 20-34	0.04	0.045	0.053	0.046	V=0.007	V= NS
	Mean (T)	0.042	0.049	0.054		V×T=0.011	V×T=NS
				BMD (g days)			
60-70 DAS	JS 20-116	67.26	82.86	85.52	78.54	T= 4.60	T= 13.27
	JS 20-34	65.61	77.61	82.64	75.29	V=3.75	V= NS
	Mean (T)	66.43	80.24	84.08		V×T=6.51	V×T=NS
70-80 DAS	JS 20-116	111.68	128.39	144	128.02	T= 4.96	T= 14.30
	JS 20-34	106.69	118.6	129.99	118.42	V=4.05	V= NS
	Mean (T)	109.18	123.49	136.99		V×T=7.01	V×T=NS

Where: LAI (leaf area index), LAD (leaf area duration), CGR (crop growth rate), RGR (relative growth rate), BMD (biomass duration)

Biomass Duration (g days): Biomass duration refers to the persistence of biomass over time, which reflects plant retention and higher dry matter accumulation capacity. The range of BMD @60-70 days and BMD @70-80 days were found to be 65.61 to 85.52 and 106.69 to 144.00 respectively. The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 26.56% and 25.47% in BMD@60-70 days and BMD @70-80 days over control. In terms of seed treatment, seed treatment with ZnO nano-particle @ 500mg L^{-1} was found to be superior over untreated control. ZnO positively enhanced the growth of the crop results in more biomass obtained with seed treatment with ZnO nano-particle @ 500mg L^{-1}. Our result is in consistent with Torabian et al. [24] who reported that Zinc oxide NPs (ZnONPs) had positive effect on biomass production of sunflower plants compared to the normal form. Awasthi et al. [25] observed that ZnO NPs, at 50 mg/L have positive effect on seed germination, number of roots, plant biomass and overall growth of roots, shoots and leaves.

3.2 Biophysical Traits

Chlorophyll Content Index: With increasing shade, the total chlorophyll content of the soybean crop increased, while the light compensation point (LCP) and light saturation point (LSP) declined [26]. The seed yield was favourably connected with the chlorophyll content index and photosynthetic parameters, indicating that different applied treatments increased, estimated or measured growth characteristics as well as the parallel increase of photosynthetic pigments and total chlorophyll [27]. The range of Chlorophyll Content Index was found to be 40.83 to 47.39 (Table 3 and Fig. 3). In terms of seed treatment, seed treatment with ZnO nano-particle @ 500mg L^{-1} was found to be superior over untreated control. The seed treatment with ZnO nano-particle @500 ppm leads to increment of 5.19% in Chlorophyll Content Index over control. Similarly Prasad et al. [28] found seed treatment with different concentrations of ZnO NPs in peanut seeds leads to enhancement of chlorophyll contents.

Energy Interception (cal cm^{-2} min^{-1}) and Light Transmission Ratio (%): The intercepted photosynthetically active radiation (PAR) and above-ground dry matter production have a linear connection, according to model simulations. Grace et al. [29] estimate intercepted radiation (Si) as the difference between incoming radiation (S) and that transmitted through the canopy to the soil (St). Crop productivity will grow as the amount of collected radiation energy increases. The temperature of the leaf surface rises when plant leaves absorb solar energy for photosynthesis. To cool the leaf surface, plants respond by releasing water via the stomata. The range of Energy Interception was found to be from 0.396 to 0.563 cal cm^{-2} min^{-1}. In terms of seed treatment, seed treatment with ZnO nano-particle @500 mg L^{-1} was found to be superior over untreated control. The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 26.76% in energy interception over control.

The range of Light Transmission Ratio was found to be 48.87 to 60.49%. In terms of seed treatment, seed treatment with ZnO nano-particle @ 500mg L^{-1} was found to be inferior over untreated control. The seed treatment with ZnO nano-particle @500 ppm leads to decrement of 16.48% in Light Transmission Ratio over control. As seed treatment enhances the both energy interception and light transmission ratio, which is in conformity with Pal et al. [30] conducted an experiment where they found out the combined effect of seed treatment with 25% cow urine and soil inoculation with 6 mL kg^{-1} pseudomonas were more superior over the other combinations with light transmission ratio (34.52%) and energy interception (0.44 cal cm^{-2} min^{-1}). Zhang et al. [31] reported that primed seeds recorded increase in yield of 15.3 %, leaf area index, photosynthetic active radiation interception (%) than non-primed seeds.

3.3 Seed Quality Parameter

Final plant stand m^{-2}: Seed nano-priming is an efficient process that may change seed metabolism and signaling pathways, affecting not only germination and seedling establishment but also the whole plant life cycle. Engineered nano-particles reach the seed coat through parenchymatous intercellular gaps, which aid in solution transport to the cotyledon [32]. The range of Final plant stand was found to be 25.18 to 65.12 (Table 4). In terms of seed treatment, seed treatment with ZnO nano-particle @ 500 mg L^{-1} was found to be superior and leads to increment of 61.33% in final plant stand over untreated control. Nano-treatment has the added advantage of triggering certain metabolic processes that are normally activated during the early phase of germination. The result was in consistent with Pandao et al. [33] the maximum viz., Germination (88%), plant stand per plot (635), seed yield kg ha^{-1} (1169), straw yield kg ha^{-1} (2660) and test weight (8.23 g) were found with seed treatment with nano ZnO 1000 ppm. Consequently, nano-priming enhances the rate of emergence and subsequent growth, yield, and crop quality [33].

Post-harvest seed quality attributes: Seed quality refers to the factors that make up a seed lot's attributes and influence seed or seed lot performance. Seed quality determine by the characteristics of seed such as shape, size and colour which give better seed germination and final plant stand [34]. Germination is one of the most crucial phases in the establishment of plants in agriculture, and it is critical for crop quality. Seedling growth is rapid, resulting in rapid expansion of the leaves and elongation of the roots, which favours nutrient intake, translocation through transpiration, and biomass production. The range of Seed germination was found to be 74.50 to 88.75% (Table 4). The seed treatment with ZnO nano-particle @ 500 mg L^{-1} seed leads to increment of 14.11% in Seed germination over control. The range of Seedling shoot length was found to be 11.46 to 16.17 cm (Table 4). The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 8.64% in Seedling shoot length over control. The range of Seedling root length was found to be 10.80 to 13.11 cm (Table 4). The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 14.13% in Seedling root length over control. The range of Seedling dry weight was found to be 0.65 to 0.77 g (Table 4). The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 5.88% in Seedling dry weight over control. In terms of seed treatment, seed treatment with ZnO nano-particle @ 500 mg L^{-1} was found to be superior over untreated control.

Table 3. Chlorophyll Content Index (CCI), energy interception and Light Transmission Ratio (LTR) in soybean under the effect of varieties, seed coating treatments and its interaction

		T1	T2	T3	Mean (V)	S.Em±	CD (5%)
Chlorophyll Content Index	JS 20-116	44.97	46.77	47.39	46.37	T= 0.70	T= NS
	JS 20-34	40.83	41.8	42.87	41.83	V=0.57	V=1.66
	Mean (T)	42.9	44.28	45.13		V×T=1.00	V×T=NS
Energy interception (Cal cm^{-2} min^{-1})	JS 20-116	0.455	0.52	0.563	0.513	T= 0.012	T= 0.035
	JS 20-34	0.396	0.508	0.518	0.474	V=0.034	V=0.029
	Mean (T)	0.426	0.514	0.54		V×T= 0.071	V×T=NS
Light Transmission Ratio (%)	JS 20-116	57.92	53.19	48.87	53.33	T= 0.84	T= 2.44
	JS 20-34	60.49	53.26	52.79	55.51	V=0.69	V=1.99
	Mean (T)	59.21	53.22	50.83		V×T=1.19	V×T=NS

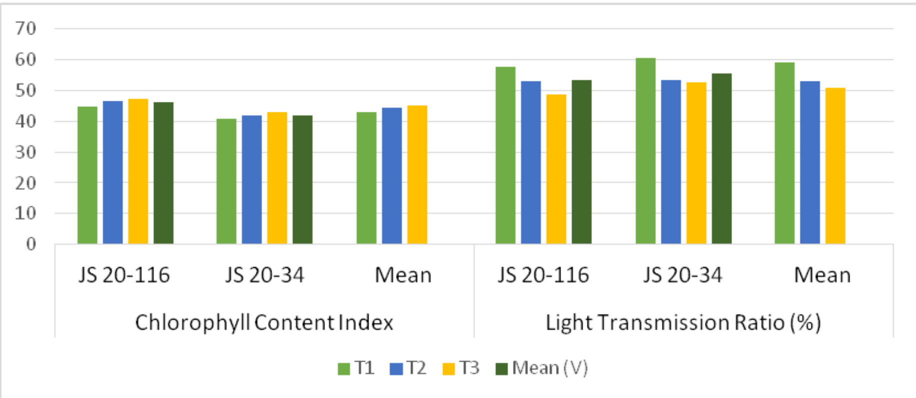

Fig. 3. Chlorophyll Content Index (CCI) and light transmission ratio (%) in soybean under the effect of varieties, seed coating treatments and its interaction.

Table 4. Effect of varieties, seed coating treatments and its interaction on the seed quality traits

		T1	T2	T3	Mean (V)	S.Em ±	CD (5%)
Final plant stand m^{-2}	JS 20-116	25.18	26.87	31.25	27.77	T= 1.94	T= 5.60
	JS 20-34	51.06	57.31	65.12	57.83	V=1.58	V=4.57
	Mean (T)	38.12	42.09	48.18		V×T=2.75	V×T=NS
Germination %	JS 20-116	76	78.5	83	79.16	T= 1.46	T= 4.22
	JS 20-34	74.5	82	88.75	81.75	V= 1.19	V= NS
	Mean (T)	75.25	80.25	85.87		V×T=2.07	V×T=NS
Seedling shoot length (cm)	JS20-116	15.63	15.85	16.17	15.88	T= 0.20	T= 0.58
	JS 20-34	11.46	12.93	13.25	12.55	V=0.16	V=0.47
	Mean (T)	13.54	14.39	14.71		V×T=0.28	V×T=NS
Seedling root length (cm)	JS 20-116	11.57	12.12	13.11	12.27	T= 0.33	T= 0.96
	JS 20-34	10.8	12.29	12.41	11.83	V=0.27	V= NS
	Mean (T)	11.18	12.2	12.76		V×T=0.47	V×T=NS
Seedling dry weight (g)	JS 20-116	0.71	0.75	0.77	0.74	T= 0.02	T= NS
	JS 20-34	0.65	0.66	0.68	0.66	V= 0.02	V= 0.05
	Mean (T)	0.68	0.7	0.72		V×T=0.03	V×T=NS
Vigour index I	JS 20-116	2,066.22	2,196.91	2,431.72	2,231.61	T= 44.40	T=128.00
	JS 20-34	1,659.24	2,061.62	2,276.08	1,998.98	V=36.25	V=104.51
	Mean (T)	1,862.73	2,129.26	2,353.90		V×T=62.79	V×T=NS
Vigour index II	JS 20-116	54.77	58.82	64.72	59.44	T= 2.34	T= 6.76
	JS 20-34	48.58	54.54	60.795	54.642	V=1.91	V= NS
	Mean (T)	51.68	56.68	62.76		V×T=3.31	V×T=NS

The range of Seed Vigour Index I was found to be 1,659.24 to 2,431.72 (Table 4). The range of Seed Vigour Index II was found to be 48.58 to 64.72 (Table 4). The seed treatment with ZnO nano-particle @ 500 ppm leads to increment of 26.40% and 21.43% in Seed Vigour Index I and Seed Vigour Index II, respectively over control. In terms of seed treatment, seed treatment with ZnO nano-particle @ 500 mg L^{-1} was found to be superior over untreated control. As a result, using nano-ZnO based treatments in soybean has shown accelerated germination and reduced the amount of fertilizer used, minimizing production costs and reducing the risk of pollution. It also mitigates the negative effects of drought stress during soybean germination. ZnO-based treatments improved germination ratio, seedling root and shoot development, and represent a viable alternative to Zn supply for boosting soybean germination and seedling development [35,36]. Rawat et al., [37] revealed that seed treatment with nano-particles at 50 ppm concentration increases root length, shoot length, seedling length, shoot dry weight, dry seedling weight, seedling vigour index I, and seedling vigour index II. Singh et al., [38] reported that the ZnO NPs application improved seed germination, root/shoot growth, seedling vigor index, chlorophyll content, grain zinc concentration, and yield.

4. CONCLUSION

Growth analytical attributes studies indicated superior performance of seed treatment with nano-ZnO @ 500mg L^{-1} for LAI @ 60 DAS, @ 70 DAS, @ 80 DAS; LAD, CGR, RGR and BMD with respect to 24.26%, 21.39%, 28.50%, 12.09%, 21.42%, 20.17% and 20.56% enhancement over control respectively. Varietal studies revealed superior performance of variety JS 20-116 for LAI @ 60 DAS (5.62), @ 70 DAS (6.20), @ 80 DAS (5.62), LAD (20,313.30), CGR (0.0026), RGR (0.053) and BMD (103.28 g days). Biophysical parameters studies revealed superior performance of treatment T3 (seed treatment with nano –ZnO @ 500mg L^{-1}) for chlorophyll content index and energy interception with 5.19% and 26.51% enhancement over control respectively. With respect to variety superior performance of variety JS-20-116 for chlorophyll content index (46.37) and energy interception (0.513 cal cm^{-2} min^{-1}). Seed treatment with nano –ZnO @ 500mg L^{-1} for final plant stand (48.18/m2), germination (80.25%), and seed vigour index I (2353.90) and seed vigour index II (62.76). with respect to 29.16%, 14.11%, 26.40%, 21.43% enhancement over control. With respect to variety, superior performance of variety JS-20-116 for final Plant stand (27.77/m^{-2}), and seed vigour index I (2231.61) and seed vigour index II (59.44) and JS 20-34 for seed germination (81.75 %).

Zinc being a cofactor for large number of enzymes involved in hormone synthesis and plant metabolism, it was hypothesized that the nano formulation in the form of ZnO @ 500mg L^{-1} will stabilize seed yield and enhance seed quality of soybean. The present study justifies our hypothesis through improvement of growth analytical attributes and physiological efficiency by the seed treatment with ZnO @ 500mg L^{-1}. Seed quality attributes particularly germination percentage and field emergence was improved by seed treatment with rhizobium and vitavax @ 5g kg^{-1} seed. However, seed treatment with nano-ZnO @ 500mg L^{-1} lead to enhancement of seed vigour. Hence the seed treatment with nano-ZnO @ 500mg L^{-1} is recommended to farmers for maximum productivity, physiological efficiency and sowing seed quality.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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