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Influence of Botanical Seed Priming and Foliar Application of Zinc Oxide Nanoparticle on Physiological Studies for Yield Enhancement in Finger Millet+ Greengram Intercropping System

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ABSTRACT: Finger millet is extremely suitable for cultivation under severe conditions while maintaining average yield. Seed priming is the pre-treatment of seeds using various ways in order to increase seed germination rate, germination percentage and seedling emergence uniformity by regulating the amount of water accessible in the seed. With limited arable land and water resources, agricultural sector development can only be achieved by improving resource utilization efficiency while, nanotechnology has the ability to completely alter the agricultural sector. An experiment has been undertaken to investigate the impact of nanoscale zinc oxide particles on plant growth and development. A field experiment was conducted at Tamil Nadu Agricultural University, Coimbatore during *rabi* season to evaluate the response of effective farming practice for sole finger millet and finger millet + greengram intercropping system under rainfed conditions to varied levels of bio-seed priming and foliar application of nanoparticles. However, there is a need to study the effect of finger millet + greengram intercropping along with seed priming and nanoparticle applications on crop growth and productivity. Twelve treatment combinations involving with two factors, sole finger millet and finger millet intercropped with greengram system were replicated thrice in Factorial Randomized Block Design (FRBD). The results of the experiment revealed that finger millet intercropped with greengram (2:1) had a significant increase in growth and yield parameter of finger millet compare to sole finger millet. Among the treatments, botanicals *viz.*, Application of *Prosopis* leaf extract 1 per cent alone + Foliar ZnO nanoparticle @ 500 ppm showed a significant increase in growth and yield parameter and onpar with pungam leaf extract 1% alone + Foliar ZnO nanoparticle @ 500 ppm. Seed priming with botanical leaf extract with foliar nanoparticle of ZnO is an effective way and eco-friendly technique which improve the faster emergence, with more uniform plant population and yield with increased nutritional content in grains. The recent study primarily focused on cropping system, bio seed priming, and foliar application of nano zinc oxide included under rainfed situations to maximize uniform germination, drought tolerance and crop production while also improving nutritional quality in seeds.

Keywords: Fingermillet, botanical priming, nanoparticles, intercropping and sustainability.

INTRODUCTION

Finger millet (*Eleusine coracana* L.) is one of the predominant millet crops generally known as ragi belongs to the *Poaceae* family and enormously cultivated in India, next to sorghum and pearl millet (Dass *et al.,* 2013). The lower productivity in finger millet due to some factors, such as marginal and poor soils or inadequate moisture and poor management practices. Among these factors, one of the important reasons for poor yield is inadequate moisture availability. Robin *et al.,* (2003) reported that drought is the most prevalent abiotic constraint that causes for widespread yield reduction in millets. Nutrients and PGRs to improve physiological efficiency of finger millet as one of the management options to exploit higher yield potential under drought condition.

India is the world's leading producer of finger millet, accounting for approximately 60 percent of worldwide production. Finger millet contains 9.2 percent protein, 76.3 percent carbohydrate, 2.2 percent minerals, 1.3 percent fat and 3.9 percent ash as well vitamin A and B. The grains are rich in phosphorus, potassium and amino acid and has the richest source of energy calcium (410 mg/100g grain), which is especially important supplements for growing children and aged people (Tomar *et al.,* 2011). Intercropping is a highly useful system that has a significant yield advantage over sole cropping and reduction in risk (Singh & Singh, 1996).

Additionally, an intercropping system can utilize the environment and physical resources more efficiently, resulting in a more productive and economically viable system with less exploitation of land resources or even improving intercropping can optimize production and productivity by making better use of existing land resources, reducing risk and bringing stability under rainfed situations.

Seed priming is a technique of controlled hydration (soaking in water) and drying that result in more rapid germination when the seeds are reimbibed (Balaji & Narayana, 2019; Callan *et al.,* 1997).

There are different methods of priming like hydropriming, halopriming, thermopriming, bio-priming, etc. Bio-priming is a process of biological seed treatment that refers to a combination of seed hydration and inoculation of the seeds with beneficial microorganisms. It improves seed viability, germination, vigour indices, plant growth and subsequent protection against diseases and finally enhances crop yield (Chauhan & Patel, 2017).

Bio-priming on biocontrol features, such as the administration of beneficial bacterial inoculum to seeds and their hydration, protects seeds from seed-borne infections. Seed biopriming is being emphasised because it ensures the entry of endophytic bacteria into the sidewalls while also avoiding the influence of high temperature (Reddy, 2013).

Nutrients have a key function in enhancing pulse seed production (Chandrasekhar & Bangarusamy, 2003). Foliar application is associated with the advantages of rapid and effective nutrient use, reduction of losses due to leaching and fixation and aids in controlling nutrient uptake by plants (Manonmani & Srimathi, 2009). Micronutrient deficiencies in humans and crop plants are hard to detect, therefore the problem is referred to as "hidden hunger" (Stein *et al.,* 2008).

While zinc (Zn) deficiency being the most prevalent nutritional deficiency next after iron and iodine. According to the world health organization, Zn deficiency is the fifth major cause of illness among juveniles and old age peoples in developing countries. Majority of the Indian soils are reported to be Zn deficient, therefore food crops cultivated in those soils contain minimum level of Zn nutrient. Crop species have shown significant genetic variability in sustaining growth and yield in Zn deficient conditions.

Nanotechnology has been regarded as the **"next great frontier of agricultural research"** and it plays an important role in revolutionizing agriculture and food production through effective soil nutrient management. Nanotechnology is a new area in agriculture that has the potential to change agriculture for long-term food grain production. Due to the fact that nanotechnology has been completely utilized in other areas, its application in agriculture has yet to be achieved. Normally the nanoparticle has a size of 10^{-9} m, nanotechnology is a unique field of research that deals with atom-by-atom manipulation to produce methods and products that have the potential to improve traditional farming into precision agriculture (Subramanian & Tarafdar, 2011).

MATERIALS AND METHOD

The laboratory analysis was conducted at the Agronomy Department, Tamil Nadu Agricultural university, Coimbatore with the foremost aim to prepare botanical leaf extracts for seed priming using *Neem, Prosopis* and *Pungam* leaves and synthesis of zinc oxide nanoparticles for foliar spray using chemical method. Thus, synthesized zinc oxide nanoparticle was characterized by using zeta potential and average size of zinc oxide nanoparticles was tested using Particle Size Analyzer, UV-Visible Spectroscopy, Fourier Transform Infrared Spectroscopy, X-Ray Diffraction, Scanning Electron Microscope and Transmission Electron Microscope and Energy Dispersive X-Ray Spectroscopy available at Department of Nano Science & Technology, Tamil Nadu Agricultural University, Coimbatore.

Standardization of soaking duration and concentration of botanical leaf extracts as priming agent for seed priming

Fresh leaves of Neem (*Azadirachta indica*), Prosopis (*Prosopis juliflora*), and Pungam (*Pongamia pinnata*) were picked individually. Then, using a weighing scale, precisely weigh one gram of leaves weighed and crushed using mortar and pestle and dissolve it in 100 ml of distilled water that has originally been measured in the beaker to generate a 1 percent extract. Finally, to eliminate undesired material and leaf debris, the leaf extract was filtered through muslin cloth (Sajjan *et al.,* 2017).

The seeds of finger millet and greengram were primed by adopting the following seed to solution ratio and seed soaking duration already standardized as per (CPG, 2012), Department of Agriculture, Government of Tamil Nadu given in the Table 1. The seeds were indeed air dried in the shade to return to their normal moisture content before being tested for seed quality characteristics.

The field experiments were carried out at field number '37 F' of Eastern Block Farm, Central farm unit, Tamil Nadu Agricultural University, Coimbatore during *rabi* seasons of 2020 to study the response of effective farming practice for sole finger millet and with greengram intercropping system under rainfed conditions to varied levels of bio-seed priming and zinc oxide nanoparticles. The farm is geographically situated in the North Western part of Tamil Nadu at 11°N latitude and 77°E longitude with an altitude of 426.72 m above mean sea level (MSL). Coimbatore is located in the Western Argo Climatic Zones of Tamil Nadu.

Table 1: Standardized seed to solution ratio and seed soaking duration as per Crop Production Guide (2012).

Crop	Seed to solution ratio	Seed soaking duration
Finger millet	1:1	6 hours
Greengram	1:0.3	3 hours

Finger millet variety CO 15 was taken as the main crop in this study. This variety was released by the Centre of Excellence on Millets, Athiyandal, TNAU during 2013. It is a popular high yielding and long duration variety rich in protein (11.8 percent) and non-lodging strain with a duration of 125 days. Greengram variety CO 8 was taken as the intercrop in this study. This variety was released by the Department of Pulses, TNAU, Coimbatore during 2013.

The following treatments schedules were used for conducting the field trail in *rabi* season of finger millet intercropping system will be test verified to optimizing the suitable treatment combinations of botanical seed priming and zinc oxide nanoparticles application for finger millet intercropping system. The field experiment was laid out in a factorial randomized block design (FRBD) with three replications. All the treatments and replications were randomized to reduce the experimental error. FACTOR – I (cropping system)M_1 – Sole finger millet and M_2 – Finger millet + Greengram (2:1) in main plot and FACTOR – II (Bio seed priming & Foliar ZnO nanoparticle) S_1 – Neem extract 1% alone, S_2 – Neem extract 1% + Foliar Nano ZnO @ 250 ppm, S_3 – Neem extract 1% + Foliar Nano ZnO @ 500 ppm, S_4 – Neem extract 1% + Foliar Nano ZnO @ 750 ppm, S_5 – Prosopis extract 1% alone, S_6 – Prosopis extract 1% + Foliar Nano ZnO @ 250 ppm, S_7– Prosopis extract 1% + Foliar Nano ZnO @ 500 ppm, S_8– Prosopis extract 1% + Foliar Nano ZnO @ 750 ppm, S_9 –Pongamia leaf 1% alone, S_{10}–Pongamia leaf extract 1% + Foliar Nano ZnO @ 250 ppm, S_{11}–Pongamia leaf extract 1% + Foliar Nano ZnO @ 500 ppm and S_{12}–Pongamia leaf extract 1% + Foliar Nano ZnO @ 750 ppm. The treatments were randomly allotted to the plots as per the experimental design. *Foliar spray will be done twice on 30 and 60 DAS.

RESULTS AND DISCUSSION

A. Growth Components

The experimental treatments were showed significant difference in main as well as sub plot treatments of observations presented in Table 2. All the growth components *viz.*, Plant height at harvest, LAI at harvest and number of tiller m^{-2} showed superior performance with finger millet intercropped with greengram as compared to sole crop of finger millet.

Table 2: Effect of Bio seed priming and Nano Zinc foliar spray on plant height (cm), Leaf area index (LAI) and number of tillers per m^{-2} of finger millet at harvest.

Treatments	Plant height at harvest			LAI at harvest			Number of tillers per m^{-2} at harvest		
	M_1	M_2	Mean	M_1	M_2	Mean	M_1	M_2	Mean
S_1	126.05	129.28	**127.67**	3.40	3.72	3.56	114.56	122.24	**118.40**
S_2	132.36	138.57	**135.47**	3.88	3.88	3.88	130.24	150.08	**140.16**
S_3	145.84	149.03	**147.44**	5.23	5.50	5.36	199.36	216.00	**207.68**
S_4	138.74	145.81	**142.28**	4.73	4.93	4.83	157.12	174.40	**165.76**
S_5	128.87	133.62	**131.25**	3.64	3.99	3.82	126.40	143.36	**134.88**
S_6	137.79	142.83	**140.31**	4.42	4.71	4.56	152.32	164.16	**158.24**
S_7	149.33	151.33	**150.33**	5.55	5.78	5.66	219.84	235.20	**227.52**
S_8	144.25	147.36	**145.81**	5.04	5.29	5.16	188.16	204.16	**196.16**
S_9	127.23	131.94	**129.59**	3.51	3.88	3.69	119.68	131.52	**125.60**
S_{10}	134.91	140.31	**137.61**	4.02	4.48	4.25	140.48	157.44	**148.96**
S_{11}	147.77	150.64	**149.21**	5.42	5.62	5.52	210.56	219.84	**215.20**
S_{12}	142.12	146.48	**144.30**	4.85	5.09	4.97	167.04	187.52	**177.28**
Mean	**137.94**	**142.27**		4.47	4.74		**160.48**	**175.49**	
	SEd	**CD (0.05)**		**SEd**	**CD (0.05)**		**SEd**	**CD (0.05)**	
M	2.111	4.248		0.079	0.160		2.591	5.216	
S	5.170	10.407		0.195	0.392		6.347	12.776	
M X S	7.311	NS		0.275	NS		8.976	NS	

Among the two-cropping system, finger millet + greengram (2:1) (M_2) recorded the highest mean of plant height at harvest (142.27 cm), LAI at harvest (4.74) and number of tiller m^{-2} at harvest (175.49) followed by sole finger millet (M_1) 137.94 cm, 4.47 and 160.4 at Plant height at harvest, LAI at harvest, number of tiller per m^{-2} at harvest, respectively.

With respect to bio seed priming and foliar zinc oxide nanoparticle spray, priming of Prosopis leaf extract 1 per cent along with 500 ppm of foliar ZnO nanoparticle (S_7) recorded higher plant height at harvest of (150.33 cm), LAI at harvest (5.66) and number of tillers per m^{-2} at harvest (227.52). This was on par with Pungam leaf extract 1 per cent alone + foliar ZnO nanoparticle @ 500 ppm (S_{11}) at all the stages of observation. The least growth components were obtained with Neem leaf extract 1 per cent alone (S_1), the plant height at harvest, LAI at harvest, number of tiller m^{-2} at harvest being 127.67 cm, 3.56 and 118.40 respectively. The interaction effect of cropping system and bio seed priming and foliar zinc nanoparticle spray was non-significant irrespective of the growth stages.

The increased growth of finger millet with greengram may be due to the compensating impact of greengram, which provided nitrogen to finger millet, as well as the greater usage of natural resources by the finger millet + greengram intercropping system. Tripathi & Kushwaha, (2013) also reported that plant height and number of leaves per plant of pearl millet under intercropping system were either higher or statistically similar to sole pearl millet, which could be attributed to effective utilisation of space and light interception, as well as nutrient contribution of leguminous crop to cereal crop.

Finger millet intercropped with greengram had a beneficial influence on LAI at all stages of crop growth. This might be due to a larger tiller number, which led in a greater number of leaves, resulting in greater LAI value.

Table 3: Effect of Bio seed priming and Nano Zinc foliar spray on grain yield, finger millet equivalent yield (FMEY) and straw yield(kg ha^{-1}) of finger millet.

Treatments	Grain yield (kg ha^{-1})			Finger millet equivalent yield			Straw yield (kg ha^{-1})		
	M_1	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**	**M_1**	**M_2**	**Mean**
S_1	2663.03	2501.62	**2582.33**	2663.03	3003.55	**2833.29**	5260.30	4455.25	**4857.77**
S_2	3106.65	2721.59	**2914.12**	3106.65	3261.09	**3183.87**	6343.16	4969.67	**5656.42**
S_3	3475.42	3161.19	**3318.31**	3475.42	3740.96	**3608.19**	7767.74	6060.42	**6914.08**
S_4	3274.27	2853.75	**3064.01**	3274.27	3416.52	**3345.39**	7275.10	5380.19	**6327.64**
S_5	2828.31	2657.27	**2742.79**	2828.31	3183.81	**3006.06**	5954.61	4906.47	**5430.54**
S_6	3215.09	2815.25	**3015.17**	3215.09	3367.73	**3291.41**	6498.21	5294.78	**5896.50**
S_7	3593.27	3282.91	**3438.09**	3593.27	3884.14	**3738.71**	7565.43	6289.03	**6927.23**
S_8	3450.52	3036.28	**3243.40**	3450.52	3612.02	**3531.27**	7891.62	5798.45	**6845.04**
S_9	2716.10	2595.51	**2655.80**	2716.10	3118.02	**2917.06**	5906.95	4876.52	**5391.74**
S_{10}	3160.33	2769.21	**2964.77**	3160.33	3314.98	**3237.65**	6788.04	5035.74	**5911.89**
S_{11}	3510.82	3213.53	**3362.18**	3510.82	3803.58	**3657.20**	7115.95	6129.50	**6622.73**
S_{12}	3389.54	2998.92	**3194.23**	3389.54	3563.92	**3476.73**	6997.40	5415.51	**6206.46**
Mean	**3198.61**	**2883.92**		**3198.61**	**3439.19**		**6780.38**	**5384.29**	
	SEd	**CD (0.05)**		**SEd**	**CD (0.05)**		**SEd**	**CD (0.05)**	
M	42.630	85.809		51.934	104.538		101.357	204.021	
S	104.421	210.189		127.213	256.066		248.273	499.747	
M X S	147.674	NS		179.906	NS		351.110	NS	

Fawusi *et al.,* (1982) indicate that maize-based intercropping systems have a greater leaf area index than solo crops.

Kumar *et al.,* (2008) observed that both little millet and pigeonpea sole crops produced greater total dry matter. They also found that a 6:2 row ratio led to an increased total dry matter production of little millet and pigeonpea. This might be attributed to the increased dry matter accumulation in the leaf, stem, and reproductive sections. Kaushik & Sharma, (2017) reported similar findings in a wheat-based intercropping system.

Prosopis leaf extract 1 percent alone with 500 ppm of foliar ZnO nanoparticle (S8) recorded higher growth parameters compared to higher concentration (1000 ppm) due to enhance in plant height and photosynthetically active leaf area due to nano ZnO might have been the reason for increased dry matter accumulation and could be due to the complementary effect of other innate nutrients like magnesium, iron, and sulphur with zinc (Koti *et al.,* 2009; Poornima & Koti, 2019).

Zinc acts as an enzyme activator in plants and is directly involved in the biosynthesis of auxin, which generates more cells and dry matter that could then be stored in seeds as Rehman *et al.,* (2002) Slaton *et al.,* (2001) in rice, (Genc *et al.,* 2006; Ozkutlu *et al.,* 2006) in bread wheat, and Anand *et al.,* (2008) in rabi sorghum found similar increase in total Higher concentrations of 1000 ppm, on the other hand, had a negative impact on the growth of finger millet.

Prasad *et al.,* (2012) demonstrated that nano ZnO increased seed germination, seedling vigour, early flowering, and leaf chlorophyll content. They also found beneficial effects of NPs in enhancing plant growth, development, and yield in peanut at low concentrations, but at larger concentrations, ZnO NPs were negative, exactly like the bulk nutrients.

Yield

Grain yield, finger millet equivalent yield and straw yield

The present investigation showed significant difference in main as well as sub treatments of observations given in Table 3. The grain yield, finger millet equivalent yield and straw yield showed superior performance with finger millet intercropped with greengram as compared to sole crop of finger millet (M_1) alone.

Among the two-cropping system, sole finger millet (M_1) gave the highest mean grain and straw yield of 3198.61 kg ha^{-1} and 6780.38 kg ha^{-1} as compared to finger millet intercropping with greengram. Finger millet intercropping yield (M_2) is converted into finger millet equivalent yield (3439.19 kg ha^{-1}) obtain highest grain yield compare to sole finger millet (3198.61 kg ha^{-1}) respectively.

With respect to bio seed priming and foliar zinc oxide nanoparticle spray, Prosopis leaf extract 1 per cent alone + Foliar ZnO nanoparticle @ 500 ppm (S_7)recorded higher grain yield (3438.09 kg ha^{-1}), finger millet equivalent yield (3738.71 kg ha^{-1}), straw yield(6927.23 kg ha^{-1}). This was on par with Pungam leaf extract 1% alone + foliar ZnO nanoparticle @ 500 ppm (S_{11}). The least yield components were obtained with Neem leaf extract 1% alone (S_1).The interaction effect of cropping system and bio seed priming and foliar zinc nanoparticle spray was non-significant in irrespective of all the stages.

Finger millet intercropped with greengram yield is comparable to yield of finger millet grown as a sole crop. Tripathi & Kushwaha (2013) reported that yield attributes of pearl millet with intercrop *viz.,* seed per panicle and 100 seed weight were significantly higher than that of sole pearl.

Dass & Sudhishri, (2010) recorded higher system yield of finger millet in intercropped stand with pulses due to efficient use of nutrients, moisture, light and space in intercropped situation.

An experimental study suggests that use of nano zinc oxide in a foliar spray can result in higher grain yields than without the use of nanoparticles. Nanoparticles with a large surface area and small size Mazaherinia *et al.,* (2010) are expected to be the best material for use as zinc fertilizer in plants. As a result, when materials are transformed to a nanoscale, they change their physical, chemical, and biological properties, as well as catalytic properties and even become more active chemically and biologically. Prasad *et al.,* (2012) investigated the impacts of nanoscale zinc oxide on peanut germination, growth, and yield and found dramatically increased growth and yield. Reynolds, (2002) proved that micronutrients in the form of nanoparticles may be utilized to boost output in agricultural production.

Because of its nano size, high surface-to-volume ratio, and high reactivity, ZnO is rapidly absorbed by the leaf surface and metabolised faster than bulk form. Similar to our findings, nano ZnO showed significantly higher crop improvement by improving initial crop establishment, chlorophyll content, and ultimately crop growth and yield in the studies of Pandey *et al.,* (2010) in *Cicer arietinum*, Poornima & Koti (2019) in sorghum, Boonyanitipong *et al.,* (2011) in rice, (Prasad *et al.,* 2012) in peanuts, Sedghi *et al.,* (2013) in soybean, Jayarambabu *et al.,* (2014) in mungbean and Yang *et al.,* (2015) in maize and rice.

Prasad *et al.,* (2012) stated that foliar application of zinc oxide nano particle is more effective than soil application, and that zinc oxide foliar treatment improved pod yield and zinc content in peanut.

CONCLUSION

The study found that finger millet intercropped with greengram (2:1) performs better when combined with priming of Prosopis leaf extract 1% alone together along with 500 ppm of foliar ZnO nanoparticle recorded greater growth parameters, resulting in an increase in finger millet crop yield. As a result, utilizing a small amount of foliar application fertiliser may minimise fertiliser application dosages, fertiliser waste, environmental dangers, and boost nutrient usage efficiency. There is a need to explore the standardizing of nano fertiliser dosages for different crops and the ideal stage of crop development in order to produce improved crop output. There is also a need to understand the cellular mechanisms involved in nano particle absorption and translocation. Finally, these results confirmed their conclusion that foliar application of ZnO NP-based fertilisers increases crop product quality and that this is the most efficient technique for future agricultural sector management in arid areas where this crop grows. As a result, the above treatment might be advised to rainfed millets farmers in order to boost productivity and obtain significant economic benefits from enhanced soil fertility. The outcome of this research would be beneficial for other studies involving the application of nanotechnology in the field of agriculture.

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