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Standardization of green synthesized silver nanoparticles through *Ocimum sanctum* for seed quality traits of wheat (*Triticum aestivum* L.)

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Abstract

The present experiment was conducted during 2021-22 at Green Nanotechnology Lab, UAS, Dharwad, Karnataka. This study involved standardization of priming of wheat seeds with green synthesized silver nanoparticles (AgNPs) through *Ocimum sanctum* leaf extract. The lab experiment comprised of ten different treatments with three replications in completely randomized design. Seed priming with green synthesized AgNPs at three different concentrations of (250 ppm, 500 ppm and 750 ppm) along with two soaking durations (4 and 8 hours). The results revealed that, AgNPs 750 ppm 4h reported significantly higher germination (92.67%), root length (11.76 cm), shoot length (18.80 cm) and increased seedling vigour index (2832) followed by AgNPs at 500 ppm for 4h and AgNPs at 250 ppm for 4h compared to control (78.00 %), (10.67 cm), (9.33 cm) and (1560) in bread wheat variety UAS-304.

Keywords: Green synthesis, nanoparticles, silver, standardization, *Ocimum sanctum*, wheat

Introduction

Wheat (*Triticum* spp.) is ranked as number one staple food crop of several global countries including South Asia, America and Europe. Wheat and wheat-based products meet nutritional needs. Utilising contemporary technologies to increase wheat output, nutritional value is necessary due to the importance of wheat as a staple crop. In more than 40 nations it offers over 20% of the calories and proteins required for human nutrition which serves more than one third of the global population to being a major source of starch and energy Asseng *et al.*, (2015) [1].

Nanotechnology with considerable potential in different discipline has been recently applied in genetic enhancement, soil texture, pest and pathogen management, nutrient delivery, environmental conservation all of which affect plant growth and development cycle (Zadeh *et al.*, 2020) [10]. A new era of green synthesis approaches gaining greater importance in agriculture mainly because of cost effectiveness and eco-friendly action (Dangi and Verma, 2021) [2]. *Ocimum sanctum* is an important aromatic plant belongs to family *Lamiaceae*. The constituent of *Ocimum sanctum* leaf rich in alkaloids, glycosides, tannins, saponins and aromatic compounds and also minerals like Ca, Mn, P, K, Zn, Na, Mg. *Ocimum sanctum* is source of bio reduction and stabilizers. Different nano particles can be synthesized through *Ocimum sanctum* leaves.

Silver nanoparticles (AgNPs) are commonly utilized nanoparticles and have attracted much study interest due to their striking distinctive and elemental properties. Ag is toxic to micro-organisms such as bacteria and fungi. They are widely used in emerging biomedical and industrial applications. AgNPs exhibits completely different characteristics as compared to bulk materials derived from the same material due to their elevated surface to volume ratio. In recent times, the synthesis of silver NPs through bio-organisms containing phytochemical agents has become an important tool for researchers. Various unique secondary metabolites derived from plant extracts such as sugars, alkaloids, phenolic acids, flavonoids and terpenoids are responsible for bio-reduction of ionic silver metal into silver nanoparticles (Sharma *et al.*, 2014) [8].

Nano-priming induces the formation of nanopores on the surface of seed and through those nanopores helps in more uptake of water and nutrient and forms hydroxyl radicals to loosen the walls of the cells and acts as an inducer for rapid hydrolysis of starch to developing embryo. This induces breaking seed dormancy, promotion of seed germination, and their

impact on primary and secondary metabolite production (Nile *et al.*, 2022) [5].

Material and Methods

The present laboratory studies were carried out during 2021-22 for green synthesis and standardization of silver nanoparticle at Green Nanotechnology Laboratory and seed quality investigations were carried out in the Department Seed Science and Technology, University of Agricultural Sciences, Dharwad Karnataka.

Treatment details

This experiment consists of 10 treatments with 3 replications viz., T₁-Seed soaking in AgNPs at 250 ppm 4 hours, T₂- Seed soaking in AgNPs at 500 ppm 4 hours, T₃- Seed soaking in AgNPs at 750 ppm 4 hours, T₄- Hydropriming 4 hours, T₅- Control 4 hours, T₆- Seed soaking in AgNPs at 250 ppm 8 hours, T₇- Seed soaking in AgNPs at 500 ppm 8 hours, T₈- Seed soaking in AgNPs at 750 ppm 8 hours, T₉-Hydropriming 8 hours, T₁₀- Control 8 hours. The germination per cent count was recorded on 8 th day of germination. Germination per cent was calculated by taking the ratio of number of seeds germinated to the total number of seeds and expressed as percentage with the statistical tool completely randomized design (CRD). Wheat seeds of variety UAS 304 were soaked with synthesized silver nanoparticle (AgNPs) solution at 250, 500 and 750 ppm concentration along with hydropriming with distilled water and control for a period of 8 and 4 hours, respectively. Bread wheat seeds of the variety UAS - 304 were collected from All India Co-ordinated Bread wheat and Barley Improvement Project, Main Agricultural Research Station, UAS, Dharwad Karnataka.

Result and discussion

The data on various seed characteristics were significantly influenced by priming treatments with green synthesized silver nanoparticles are presented in the Table 1 and Plate 1. Among the various priming treatments, seed primed with green synthesized AgNPs at 750 ppm for 4 hours (T₃)

recorded significantly higher germination per cent (92.67%), root length (11.76 cm), shoot length (18.80 cm) and seedling vigour index (2832) followed by Ag NPs at 500 ppm for 4h (T₂) (92.00, 14.91, 10.90, 2374.52), Ag NPs at 250 ppm for 4h (T₁) (90.00, 14.18, 10.70, 2239.20), Ag NPs at 750 ppm for 8h T₈ (88.00, 13.05, 10.36, 2060.08), Ag NPs at 500 ppm for 8h T₇ (86.00, 12.10, 10.02, 1902.32) and Ag NPs at 500 ppm for 8h T₆ (83.00, 11.85, 9.80, 1796.95). whereas, seeds without any priming shown significantly lower germination percent, root length, shoot length and seedling vigour index (78.00, 10.67, 9.33, 1560.0, respectively) in bread wheat variety UAS-304.

The results obtained for seed germination, shoot length, root length and seedling vigour index clearly indicate that when compared to soaking durations of 4 and 8 hours, seeds primed for 4 hours showed superior results as compared to those primed for 8 hour and also in comparison to the control. This is because seed primed with nanoparticles facilitates better water penetration through the seed coat, resulting in increased water absorption, which in turn accelerates seed germination and leads to better establishment and higher emergence rates. Whereas, 8 hour primed may have led to loss of seed leachates as soaking for longer duration and lesser cell membrane permeability (Rawat *et al.*, 2018) [7].

Among different concentrations, it was observed that concentrations of 250 ppm, 500 ppm, and 750 ppm for silver nanoparticles 750 ppm recorded higher percentages of germination, shoot length, and root length compared to the other concentrations. Due to increased germination per cent and mean seedling length leads to increase in seedling vigour index. Reactivation of reactive oxygen species (ROS) and a significant increase in the activity of antioxidant enzymes, resulting in substantial changes to the antioxidant metabolism. Furthermore, it's worth noting that seeds undergo various physiological and biological transformations during germination. Organic materials like starch, fats, and proteins undergo a series of metabolic changes, including physiological hydrolysis, transportation and reconstruction, as described by Mahakham *et al.* (2016) [4].

Table 1: Effect of silver nanoparticles on germination, Root length, shoot length and seedling vigour index of wheat as influenced by different priming hours at different concentrations.

Treatments	Germination %	Root length (cm)	Shoot length (cm)	Seedling vigour index
T ₁ - Ag NPs 250 ppm 4h	90.00 ^a	14.18 ^a	10.70 ^a	2239.20 ^a
T ₂ - Ag NPs 500 ppm 4h	92.00 ^a	14.91 ^b	10.90 ^b	2374.52 ^b
T ₃ - Ag NPs 750 ppm 4h	92.67 ^{ab}	18.80 ^b	11.76 ^b	2831.99 ^c
T ₄ - Hydropriming 4h	82.00 ^{abc}	11.26 ^c	9.50 ^{bc}	1702.32 ^d
T ₅ - Control 4h	79.00 ^{abcd}	10.74 ^{cd}	9.35 ^{cd}	1587.11 ^e
T ₆ - Ag NPs 250 ppm 8h	83.00 ^{bcd}	11.85 ^d	9.80 ^{cde}	1796.95 ^f
T ₇ - Ag NPs 500 ppm 8h	86.00 ^{cde}	12.10 ^{de}	10.02 ^{de}	1902.32 ^g
T ₈ - Ag NPs 750 ppm 8h	88.00 ^{cde}	13.05 ^{de}	10.36 ^e	2060.08 ^g
T ₉ - Hydropriming 8h	82.00 ^{de}	11.17 ^e	9.43 ^e	1689.20 ^h
T ₁₀ - Control 8h	78.00 ^e	10.67 ^e	9.33 ^e	1560.00 ^h
C.D. (p=0.01)	7.31	1.00	0.59	83.52
SE(m)±	2.46	0.34	0.20	28.11
CV	4.80	4.52	3.39	2.47

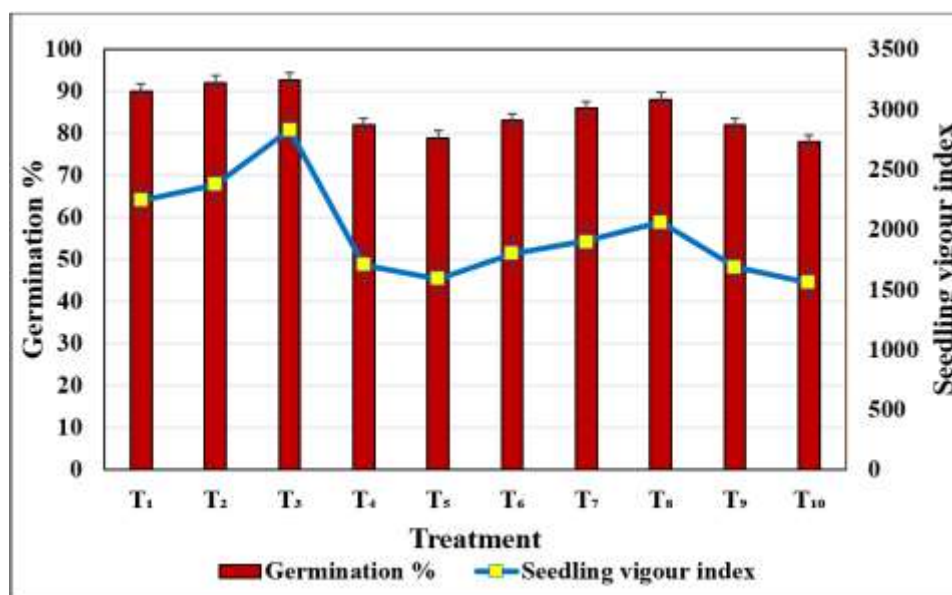


Fig 1: Effect of silver nanoparticles on germination and seedling vigour index of wheat as influenced by different priming hours at different concentrations.

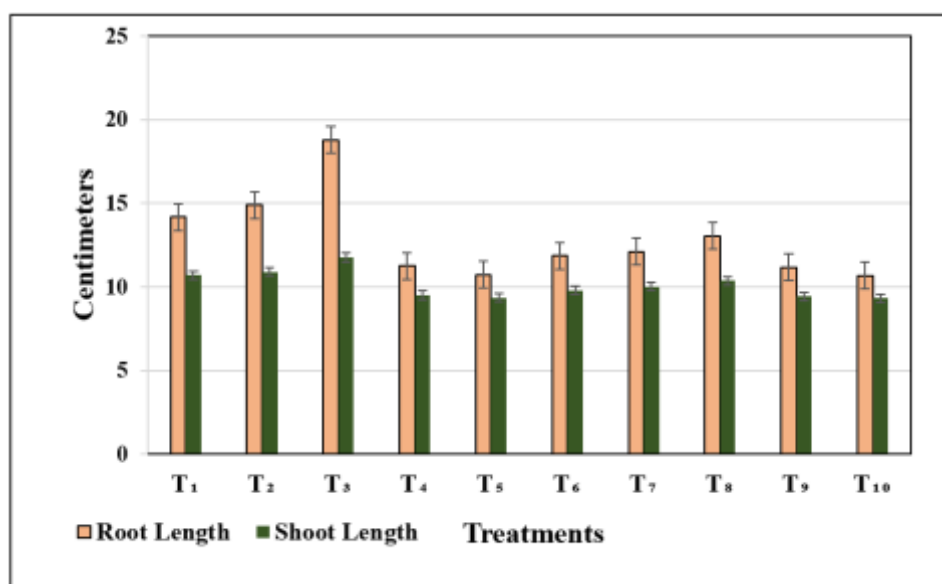


Fig 2: Effect of silver nanoparticles on Root length and shoot length of wheat as influenced by different priming hours at different concentrations.

Conclusion

Priming of wheat (UAS-304) seeds at 750 ppm concentration with green-synthesized silver nanoparticles for a duration of 4 hours is an effective method to enhance several key seed quality characteristics. Priming seeds with silver nanoparticles have been demonstrated to accelerate the rate of seed emergence, facilitate quicker seedling establishment and promote vigorous seedling growth. This is primarily attributed to the nanoparticles serving as catalysts that enhance the activity of various enzymes, expediting the breakdown of stored food reserves to nourish the developing embryo. Moreover, priming also helps to mitigate the negative effects of environmental stress factors during the germination phase, ultimately leading to improved seedling emergence and robust seedling establishment.

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