



# Effect of Biosynthesized Iron Nanoparticles on Wheat Production (*Triticum aestivum* L.)

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## ABSTRACT

**Background:** Iron deficiency is a common nutritional disorder affecting crop production worldwide, leading to stunted growth, reduced yields and decreased nutritional value. Traditional iron fertilizers have limitations, such as low solubility, poor mobility and potential environmental toxicity. Biosynthesized iron nanoparticles have emerged as a promising alternative to address these challenges. These nanoparticles are synthesized using microorganisms offering a sustainable and eco-friendly approach.

**Methods:** The iron nanoparticles were biosynthesized through *Pseudomonas* and actinobacteria and characterized through UV-visible spectroscopy, particle size analyzer (PSA), scanning electron microscope (SEM), (EDX), X-Ray diffraction (XRD) and fourier transform infrared spectroscopy (FTIR). The Biosynthesis and characterization of iron nanoparticles were done in Green Nanotechnology Laboratory, University of Agricultural Sciences, Dharwad.

**Result:** Wheat seed priming at 250 ppm and foliar spraying at 500 ppm at panicle initiation stage with iron nanoparticles biosynthesized through actinobacteria (T<sub>9</sub>) recorded significantly higher plant height (96.28 cm), number of tillers per meter row length (173.00), leaf area (73.45 dm<sup>2</sup> m row length<sup>-1</sup>), leaf area index (3.26), total dry matter production (368.34 g m row length<sup>-1</sup>), productive spikes per square meter (253.67 m<sup>-2</sup>), number of grains per spike (48.23), grain weight per spike (1.80 g), 1,000 grain weight (42.19 g), grain yield (4563 kg ha<sup>-1</sup>) and straw yield (6357 kg ha<sup>-1</sup>), which was on par with seed priming at 250 ppm and foliar spraying at 500 ppm at panicle initiation stage with iron nanoparticles biosynthesized through *Pseudomonas* (T<sub>3</sub>) and commercial iron nanoparticles (T<sub>9</sub>).

**Key words:** Biosynthesis, Iron, Nanoparticles, Seed priming, Wheat.

## INTRODUCTION

Wheat is a major food crop cultivated globally, providing food for 35 per cent of the world's population Mohammadijoo *et al.* (2015). The most of wheat that is grown on a worldwide is hexaploid and extensively utilised to produce a variety of baked food products including bread, there is a substantial impact on human health based on the composition and nutritional quality of the wheat. Iron (Fe), an essential nutrient for the crop growth and development of crops. It is essential for the formation of chlorophyll and is involved in the electron transport system and activation of several enzymatic functions. Chlorosis in young plant leaves is the main sign of Fe deficiency, which affects both physiological function and nutritional quality (Kasote *et al.*, 2019 and Ali *et al.*, 2021).

Iron absorption by plants is frequently limited because ferric oxide (Fe<sub>2</sub>O<sub>3</sub>), also known as hematite, is the most prevalent form of Fe and is highly insoluble in soils. In the food chain, Fe shortage affects not only plant growth and development but also causes Fe insufficiency in both animals and humans. Therefore, it's critical to increase the effectiveness of Fe fertiliser use (Rout and Sahoo, 2015 and Dhage and Vidyashree, 2022).

Nanotechnology may help bring about a new technological revolution in agriculture. Several problems with conventional biofortification could potentially be resolved by nanotechnology Shakiba *et al.* (2020). It is possible to produce nanofertilizers using nanomaterials because of their high surface-to-volume ratio, gradual and controlled release at target places and other characteristics Feregrino-

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Perez *et al.* (2018). The encapsulation of nutrients with nanomaterials results in efficient nutrient absorption by plants, due to the gradual or controlled release of nanoparticles and simple passage through biological barriers by nanoparticles entering the plant vascular system De La Torre-Roche *et al.* (2020). In comparison to conventional fertilisers, long-term delivery of plants via

nanofertilizers enables enhanced crop growth. As nanofertilizers are added in small amounts, these also prevent soil from becoming burdened with the by-products of chemical fertilisers and reduce the environmental hazards Leon-Silva *et al.* (2018).

In order to increase productivity and the quality of the food produce, seed priming has been used to synchronise and speed up germination, boost seedling vigour and increase plant resistance to biotic and abiotic stresses Acharya *et al.* (2019). Using nanotechnology for seed priming is a relatively new field of study; it can be used to target seed biofortification to reduce malnutrition Nile *et al.* (2022).

Although applying nutrients to the soil is the most popular method, it has significant drawbacks in terms of the nutrients availability to the plants, due to the insoluble forms of the inorganic nutrients are fixed in the soil and also prone to leaching by irrigation or rainfall (Alshaal and El-Ramady, 2017). Foliar feeding has demonstrated to be the quickest way to rectify nutrient shortages, increase crop production and improve crop product quality. It also minimises environmental pollution and optimises nutrient utilisation by using less amount of fertiliser to the soil Morab *et al.* (2021).

## MATERIALS AND METHODS

### Experimental site

Biosynthesis, characterization and standardization of iron nanoparticles and lab experiments were done in Green Nanotechnology Laboratory, University of Agricultural Sciences, Dharwad. At the Microbial Genetics Laboratory, Department of Agricultural Microbiology, UAS, Dharwad, *Pseudomonas* and actinobacterial isolates were collected and screened. The field study was carried out during the *rabi* season of 2022-23 at the Main Agricultural Research Station, UAS, Dharwad.

### Physio-chemical properties of the soil

The textural class of experimental soil was clayey and pH- 7.79; EC-0.28 dS m<sup>-1</sup>; organic carbon- 0.51%; available nitrogen- 268.45 kg ha<sup>-1</sup>; phosphorus- 35.04 kg ha<sup>-1</sup>; potassium -342.26 kg ha<sup>-1</sup>; zinc- 0.56 ppm and iron- 7.12 ppm.

### Experimental procedure

Wheat seeds of the UAS 334 variety were collected from the Main Agricultural Research Station in Dharwad. The seeds were sown at a rate of 150 kg per hectare, evenly distributed in furrows spaced 20.0 cm apart using a wooden marker and subsequently covered with soil manually. The sowing was taken up on November 21, 2022. Seeds were primed with biosynthesized zinc nanoparticles solution at 500 ppm, for a period of six hours for respective treatments.

### Treatment details

The study was carried out using a randomized complete block design (RCBD), twelve treatments replicated three times. The experimental details was T<sub>1</sub>- seed priming with

BS [Bacterial (*Pseudomonas*) synthesized] FeNPs @ 250 ppm; T<sub>2</sub>- foliar spraying with BS FeNPs @ 500 ppm; T<sub>3</sub>- seed priming @ 250 ppm + foliar spraying @ 500 ppm with BS FeNPs; T<sub>4</sub>- seed priming with ABS (actinobacterial synthesized) FeNPs @ 250 ppm; T<sub>5</sub>- foliar spraying with ABS FeNPs @ 500 ppm; T<sub>6</sub>- seed priming @ 250 ppm + foliar spraying @ 500 ppm with ABS FeNPs; T<sub>7</sub>- seed priming with commercial FeNPs @ 250 ppm; T<sub>8</sub>- foliar spraying with commercial FeNPs @ 500 ppm; T<sub>9</sub>- seed priming @ 250 ppm + foliar spraying @ 500 ppm with commercial FeNPs; T<sub>10</sub>- foliar spraying with FeSO<sub>4</sub> @ 0.5%; T<sub>11</sub>- RDF (recommended dose of fertilizers-100:75:50, N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg ha<sup>-1</sup>, respectively) and T<sub>12</sub>- control (without any fertilizer application). Foliar spraying at panicle initiation stage of the crop is common for all the foliar applied treatments. RDF- 100:75:50, N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg ha<sup>-1</sup> common for all the treatments.

### Leaf area

Leaf area is computed by length and width method. It was multiplied by the factor 0.65. Data on leaf area were recorded at 30, 60 and 90 DAS, the leaf area at harvest could not be measured due to complete drying of leaves. It was expressed in dm<sup>2</sup> by following procedure given by Gomez (1972).

$$\text{Leaf area (dm}^2\text{) of each leaf} = L \times W \times K$$

Where,

L = Maximum length of leaf.

W = Maximum width of leaf.

K = Factor (0.65).

### Leaf area index

Leaf area index was calculated by using the formula as suggested by Sestak *et al.* (1971).

$$\text{Leaf area index} = \frac{\text{Leaf area (dm}^2\text{)}}{\text{Land area occupied by the plant (dm}^2\text{)}}$$

### Statistical analysis

The data collected from the experiment at various growth stages were subjected statistical analysis following the method given by Gomez and Gomez (1984). The significance level used in the 'F' test was P = 0.01 (1%) and P = 0.05 (5%). The critical difference (CD) at 1% and 5% levels was computed whenever the 'F' test was given significant results. The mean values of treatments were separately subjected to duncan multiple range test (DMRT) using the corresponding error mean sum of squares and degrees of freedom.

## RESULTS AND DISCUSSION

### Effect of biosynthesized iron nanoparticles on wheat growth

Seed priming @ 250 ppm and foliar spraying @ 500 ppm with iron nanoparticles (FeNPs) biosynthesized by actinobacteria resulted in significantly higher plant height

(96.28 cm), number of tillers per meter row length (173.00), leaf area (73.45 dm<sup>2</sup> m row length<sup>-1</sup>), leaf area index (3.26) and total dry matter production (368.34 g m row length<sup>-1</sup>) and found on par with seed priming @ 250 ppm and foliar spraying @ 500 ppm with FeNPs biosynthesized by *Pseudomonas* (95.50 cm, 170.00, 72.92 dm<sup>2</sup> m row length<sup>-1</sup>, 3.24 and 365.78 g m row length<sup>-1</sup>, respectively) and commercial iron nanoparticles (95.23 cm, 168.67, 72.89 dm<sup>2</sup> m row length<sup>-1</sup>, 3.24 and 362.74 g m row length<sup>-1</sup>, respectively) (Table 1, 2, 3, 4 and 5). Iron is an important component of cell metabolism and is involved in respiration, photosynthesis, enzyme activity and other processes, due to its limited solubility, plants cannot readily access it. Therefore, applying iron to plant as a foliar spray may be an excellent way to deal with the issue. As opposed to the usual techniques of applying Fe to the soil, foliar application of Fe is a more ecologically friendly option by Bakhtiari *et al.*

(2015). Incesu *et al.* (2015) revealed that iron helps to build key components of plant cells like cytochromes, phytoferritin and ferredoxins that transport electrons during photosynthesis, stimulating growth and increasing plant height and leaf area. According to Aslani *et al.* (2014) the advantage of nanofertilizers over chelated fertilizers is due to the fact that their nanoparticles have a large surface area, which increases the activity of enzymes and biochemical processes. Due to their ease of solubility and proliferation, which promotes increased enzymatic activity, reactions and cellular divisions, as well as the fact that they travel directly to their intended destination, nanoparticles also inhibit the formation of reactive oxygen species, which lessens oxidative damage, delays senescence and promotes vegetative growth in plants. Marzouk *et al.* (2019) reported nanofertilizers may be readily absorbed by the epidermis of leaves and transported to

**Table 1:** Plant height of wheat as affected by seed priming and foliar spraying with biosynthesized iron nanoparticles.

| Treatment details                                                                                           | Plant height (cm)  |                      |                    |                    |
|-------------------------------------------------------------------------------------------------------------|--------------------|----------------------|--------------------|--------------------|
|                                                                                                             | 30 DAS             | 60 DAS               | 90 DAS             | At harvest         |
| T <sub>1</sub> : SP with BS FeNPs                                                                           | 37.73 <sup>a</sup> | 75.13 <sup>bc</sup>  | 84.97 <sup>b</sup> | 87.43 <sup>b</sup> |
| T <sub>2</sub> : FS with BS FeNPs                                                                           | 33.67 <sup>b</sup> | 71.40 <sup>cd</sup>  | 86.63 <sup>b</sup> | 89.67 <sup>b</sup> |
| T <sub>3</sub> : SP + FS with BS FeNPs                                                                      | 38.23 <sup>a</sup> | 81.17 <sup>a</sup>   | 93.90 <sup>a</sup> | 95.50 <sup>a</sup> |
| T <sub>4</sub> : SP with ABS FeNPs                                                                          | 38.40 <sup>a</sup> | 76.13 <sup>b</sup>   | 85.23 <sup>b</sup> | 88.13 <sup>b</sup> |
| T <sub>5</sub> : FS with ABS FeNPs                                                                          | 34.17 <sup>b</sup> | 73.40 <sup>b-d</sup> | 87.60 <sup>b</sup> | 90.37 <sup>b</sup> |
| T <sub>6</sub> : SP + FS with ABS FeNPs                                                                     | 38.53 <sup>a</sup> | 82.73 <sup>a</sup>   | 94.43 <sup>a</sup> | 96.28 <sup>a</sup> |
| T <sub>7</sub> : SP with commercial FeNPs                                                                   | 37.43 <sup>a</sup> | 74.23 <sup>bcd</sup> | 84.83 <sup>b</sup> | 87.20 <sup>b</sup> |
| T <sub>8</sub> : FS with commercial FeNPs                                                                   | 32.93 <sup>b</sup> | 70.50 <sup>d</sup>   | 85.13 <sup>b</sup> | 89.53 <sup>b</sup> |
| T <sub>9</sub> : SP + FS with commercial FeNPs                                                              | 37.57 <sup>a</sup> | 80.27 <sup>a</sup>   | 92.47 <sup>a</sup> | 95.23 <sup>a</sup> |
| T <sub>10</sub> : FS with FeSO <sub>4</sub> @ 0.5%                                                          | 32.90 <sup>b</sup> | 66.30 <sup>e</sup>   | 80.23 <sup>c</sup> | 82.37 <sup>c</sup> |
| T <sub>11</sub> : RDF (100:75:50, N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg ha <sup>-1</sup> ) | 32.10 <sup>b</sup> | 62.13 <sup>f</sup>   | 75.47 <sup>d</sup> | 77.13 <sup>d</sup> |
| T <sub>12</sub> : Control                                                                                   | 27.83 <sup>c</sup> | 57.43 <sup>g</sup>   | 69.51 <sup>e</sup> | 71.87 <sup>e</sup> |
| S.Em. ±                                                                                                     | 1.10               | 1.37                 | 1.54               | 1.59               |

SP-Seed priming; FS-Foliar spraying; BS-Bacterial (*Pseudomonas*) synthesized; ABS-actinobacterial synthesized; Seed priming @ 250 ppm and foliar spraying @ 500 ppm are common for all nano treated treatments; RDF common for all treatments except control. Note: Means followed by the same letter (s) did not differ significantly by DMRT (p= 0.05).

**Table 2:** Number of tillers in wheat affected by seed priming and foliar spraying with biosynthesized iron nanoparticles.

| Treatment details                                                                                           | Number of tillers per meter row length |                     |                     |                     |
|-------------------------------------------------------------------------------------------------------------|----------------------------------------|---------------------|---------------------|---------------------|
|                                                                                                             | 30 DAS                                 | 60 DAS              | 90 DAS              | At harvest          |
| T <sub>1</sub> : SP with BS FeNPs                                                                           | 79.33 <sup>a</sup>                     | 164.67 <sup>b</sup> | 157.33 <sup>b</sup> | 154.33 <sup>b</sup> |
| T <sub>2</sub> : FS with BS FeNPs                                                                           | 68.67 <sup>b</sup>                     | 161.33 <sup>b</sup> | 154.00 <sup>b</sup> | 151.67 <sup>b</sup> |
| T <sub>3</sub> : SP + FS with BS FeNPs                                                                      | 80.33 <sup>a</sup>                     | 181.00 <sup>a</sup> | 174.67 <sup>a</sup> | 170.00 <sup>a</sup> |
| T <sub>4</sub> : SP with ABS FeNPs                                                                          | 81.00 <sup>a</sup>                     | 166.33 <sup>b</sup> | 160.00 <sup>b</sup> | 157.33 <sup>b</sup> |
| T <sub>5</sub> : FS with ABS FeNPs                                                                          | 70.67 <sup>b</sup>                     | 162.00 <sup>b</sup> | 156.67 <sup>b</sup> | 154.33 <sup>b</sup> |
| T <sub>6</sub> : SP + FS with ABS FeNPs                                                                     | 82.33 <sup>a</sup>                     | 183.67 <sup>a</sup> | 176.33 <sup>a</sup> | 173.00 <sup>a</sup> |
| T <sub>7</sub> : SP with commercial FeNPs                                                                   | 78.67 <sup>a</sup>                     | 162.67 <sup>b</sup> | 155.67 <sup>b</sup> | 152.33 <sup>b</sup> |
| T <sub>8</sub> : FS with commercial FeNPs                                                                   | 71.33 <sup>b</sup>                     | 160.33 <sup>b</sup> | 153.67 <sup>b</sup> | 150.67 <sup>b</sup> |
| T <sub>9</sub> : SP + FS with commercial FeNPs                                                              | 80.00 <sup>a</sup>                     | 178.67 <sup>a</sup> | 171.33 <sup>a</sup> | 168.67 <sup>a</sup> |
| T <sub>10</sub> : FS with FeSO <sub>4</sub> @ 0.5%                                                          | 69.33 <sup>b</sup>                     | 147.00 <sup>c</sup> | 142.00 <sup>c</sup> | 139.33 <sup>c</sup> |
| T <sub>11</sub> : RDF (100:75:50, N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg ha <sup>-1</sup> ) | 71.00 <sup>b</sup>                     | 134.33 <sup>d</sup> | 130.67 <sup>d</sup> | 127.00 <sup>d</sup> |
| T <sub>12</sub> : Control                                                                                   | 32.67 <sup>c</sup>                     | 102.67 <sup>e</sup> | 97.33 <sup>e</sup>  | 94.67 <sup>e</sup>  |
| S.Em. ±                                                                                                     | 2.46                                   | 4.14                | 3.77                | 3.50                |

**Table 3:** Leaf area in wheat is affected by seed priming and foliar spraying with biosynthesized iron nanoparticles.

| Treatment details                                                                                           | Leaf area (dm <sup>2</sup> m row length <sup>-1</sup> ) |                    |                    |
|-------------------------------------------------------------------------------------------------------------|---------------------------------------------------------|--------------------|--------------------|
|                                                                                                             | 30 DAS                                                  | 60 DAS             | 90 DAS             |
| T <sub>1</sub> : SP with BS FeNPs                                                                           | 23.94 <sup>a</sup>                                      | 64.57 <sup>b</sup> | 67.13 <sup>b</sup> |
| T <sub>2</sub> : FS with BS FeNPs                                                                           | 21.24 <sup>b</sup>                                      | 62.62 <sup>b</sup> | 68.17 <sup>b</sup> |
| T <sub>3</sub> : SP + FS with BS FeNPs                                                                      | 24.46 <sup>a</sup>                                      | 68.12 <sup>a</sup> | 72.92 <sup>a</sup> |
| T <sub>4</sub> : SP with ABS FeNPs                                                                          | 24.67 <sup>a</sup>                                      | 64.70 <sup>b</sup> | 67.83 <sup>b</sup> |
| T <sub>5</sub> : FS with ABS FeNPs                                                                          | 20.43 <sup>b</sup>                                      | 62.87 <sup>b</sup> | 68.21 <sup>b</sup> |
| T <sub>6</sub> : SP + FS with ABS FeNPs                                                                     | 24.57 <sup>a</sup>                                      | 68.34 <sup>a</sup> | 73.45 <sup>a</sup> |
| T <sub>7</sub> : SP with commercial FeNPs                                                                   | 23.78 <sup>a</sup>                                      | 63.86 <sup>b</sup> | 66.54 <sup>b</sup> |
| T <sub>8</sub> : FS with commercial FeNPs                                                                   | 20.36 <sup>b</sup>                                      | 61.93 <sup>b</sup> | 67.35 <sup>b</sup> |
| T <sub>9</sub> : SP + FS with commercial FeNPs                                                              | 23.82 <sup>a</sup>                                      | 67.85 <sup>a</sup> | 72.89 <sup>a</sup> |
| T <sub>10</sub> : FS with FeSO <sub>4</sub> @ 0.5%                                                          | 21.27 <sup>b</sup>                                      | 58.76 <sup>c</sup> | 63.05 <sup>c</sup> |
| T <sub>11</sub> : RDF (100:75:50, N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg ha <sup>-1</sup> ) | 20.42 <sup>b</sup>                                      | 55.60 <sup>d</sup> | 59.25 <sup>d</sup> |
| T <sub>12</sub> : Control                                                                                   | 17.35 <sup>c</sup>                                      | 37.85 <sup>e</sup> | 40.67 <sup>e</sup> |
| S.Em. ±                                                                                                     | 0.83                                                    | 1.07               | 1.04               |

**Table 4:** Leaf area index in wheat affected by seed priming and foliar spraying with biosynthesized iron nanoparticles.

| Treatmental details                                                                                         | Leaf area index   |                   |                   |
|-------------------------------------------------------------------------------------------------------------|-------------------|-------------------|-------------------|
|                                                                                                             | 30 DAS            | 60 DAS            | 90 DAS            |
| T <sub>1</sub> : SP with BS FeNPs                                                                           | 1.06 <sup>a</sup> | 2.87 <sup>b</sup> | 2.98 <sup>b</sup> |
| T <sub>2</sub> : FS with BS FeNPs                                                                           | 0.94 <sup>b</sup> | 2.78 <sup>b</sup> | 3.03 <sup>b</sup> |
| T <sub>3</sub> : SP + FS with BS FeNPs                                                                      | 1.09 <sup>a</sup> | 3.03 <sup>a</sup> | 3.24 <sup>a</sup> |
| T <sub>4</sub> : SP with ABS FeNPs                                                                          | 1.10 <sup>a</sup> | 2.88 <sup>b</sup> | 3.01 <sup>b</sup> |
| T <sub>5</sub> : FS with ABS FeNPs                                                                          | 0.91 <sup>b</sup> | 2.79 <sup>b</sup> | 3.03 <sup>b</sup> |
| T <sub>6</sub> : SP + FS with ABS FeNPs                                                                     | 1.09 <sup>a</sup> | 3.04 <sup>a</sup> | 3.26 <sup>a</sup> |
| T <sub>7</sub> : SP with commercial FeNPs                                                                   | 1.06 <sup>a</sup> | 2.84 <sup>b</sup> | 2.96 <sup>b</sup> |
| T <sub>8</sub> : FS with commercial FeNPs                                                                   | 0.90 <sup>b</sup> | 2.75 <sup>b</sup> | 2.99 <sup>b</sup> |
| T <sub>9</sub> : SP + FS with commercial FeNPs                                                              | 1.06 <sup>a</sup> | 3.02 <sup>a</sup> | 3.24 <sup>a</sup> |
| T <sub>10</sub> : FS with FeSO <sub>4</sub> @ 0.5%                                                          | 0.95 <sup>b</sup> | 2.61 <sup>c</sup> | 2.80 <sup>c</sup> |
| T <sub>11</sub> : RDF (100:75:50, N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg ha <sup>-1</sup> ) | 0.91 <sup>b</sup> | 2.47 <sup>d</sup> | 2.63 <sup>d</sup> |
| T <sub>12</sub> : Control                                                                                   | 0.77 <sup>c</sup> | 1.68 <sup>e</sup> | 1.81 <sup>e</sup> |
| S.Em. ±                                                                                                     | 0.03              | 0.04              | 0.05              |

**Table 5:** Total dry matter accumulation in wheat affected by seed priming and foliar spraying with biosynthesized iron nanoparticles.

| Treatmental details                                                                                         | Dry matter production (g m row length <sup>-1</sup> ) |                     |                     |                     |
|-------------------------------------------------------------------------------------------------------------|-------------------------------------------------------|---------------------|---------------------|---------------------|
|                                                                                                             | 30 DAS                                                | 60 DAS              | 90 DAS              | At harvest          |
| T <sub>1</sub> : SP with BS FeNPs                                                                           | 55.45 <sup>a</sup>                                    | 160.46 <sup>b</sup> | 257.65 <sup>b</sup> | 332.56 <sup>b</sup> |
| T <sub>2</sub> : FS with BS FeNPs                                                                           | 43.27 <sup>b</sup>                                    | 157.87 <sup>b</sup> | 260.14 <sup>b</sup> | 335.78 <sup>b</sup> |
| T <sub>3</sub> : SP + FS with BS FeNPs                                                                      | 55.32 <sup>a</sup>                                    | 177.25 <sup>a</sup> | 282.75 <sup>a</sup> | 365.78 <sup>a</sup> |
| T <sub>4</sub> : SP with ABS FeNPs                                                                          | 56.47 <sup>a</sup>                                    | 163.62 <sup>b</sup> | 259.24 <sup>b</sup> | 334.42 <sup>b</sup> |
| T <sub>5</sub> : FS with ABS FeNPs                                                                          | 42.91 <sup>b</sup>                                    | 159.23 <sup>b</sup> | 263.15 <sup>b</sup> | 337.27 <sup>b</sup> |
| T <sub>6</sub> : SP + FS with ABS FeNPs                                                                     | 57.43 <sup>a</sup>                                    | 179.14 <sup>a</sup> | 285.72 <sup>a</sup> | 368.34 <sup>a</sup> |
| T <sub>7</sub> : SP with commercial FeNPs                                                                   | 52.51 <sup>a</sup>                                    | 158.92 <sup>b</sup> | 255.45 <sup>b</sup> | 328.57 <sup>b</sup> |
| T <sub>8</sub> : FS with commercial FeNPs                                                                   | 42.51 <sup>b</sup>                                    | 155.28 <sup>b</sup> | 257.19 <sup>b</sup> | 332.23 <sup>b</sup> |
| T <sub>9</sub> : SP + FS with commercial FeNPs                                                              | 53.28 <sup>a</sup>                                    | 175.56 <sup>a</sup> | 280.67 <sup>a</sup> | 362.74 <sup>a</sup> |
| T <sub>10</sub> : FS with FeSO <sub>4</sub> @ 0.5%                                                          | 43.15 <sup>b</sup>                                    | 143.24 <sup>c</sup> | 237.15 <sup>c</sup> | 297.89 <sup>c</sup> |
| T <sub>11</sub> : RDF (100:75:50, N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg ha <sup>-1</sup> ) | 41.48 <sup>b</sup>                                    | 131.45 <sup>d</sup> | 219.62 <sup>d</sup> | 270.87 <sup>d</sup> |
| T <sub>12</sub> : Control                                                                                   | 23.47 <sup>c</sup>                                    | 102.93 <sup>e</sup> | 125.35 <sup>e</sup> | 165.52 <sup>e</sup> |
| S.Em. ±                                                                                                     | 2.74                                                  | 3.94                | 5.88                | 8.65                |

**Table 6:** Yield attributes in wheat affected by priming and foliar spraying with biosynthesized iron nanoparticles.

| Treatmental details                                                                                         | Productive spikes (m <sup>-2</sup> ) | No of grains per spike | Grain weight per spike (g) | Test weight (g)    |
|-------------------------------------------------------------------------------------------------------------|--------------------------------------|------------------------|----------------------------|--------------------|
| T <sub>1</sub> : SP with BS FeNPs                                                                           | 237.33 <sup>b</sup>                  | 42.63 <sup>c</sup>     | 1.60 <sup>b</sup>          | 40.46 <sup>b</sup> |
| T <sub>2</sub> : FS with BS FeNPs                                                                           | 240.00 <sup>b</sup>                  | 43.80 <sup>bc</sup>    | 1.65 <sup>b</sup>          | 40.54 <sup>b</sup> |
| T <sub>3</sub> : SP + FS with BS FeNPs                                                                      | 251.67 <sup>a</sup>                  | 47.73 <sup>a</sup>     | 1.79 <sup>a</sup>          | 42.05 <sup>a</sup> |
| T <sub>4</sub> : SP with ABS FeNPs                                                                          | 239.67 <sup>b</sup>                  | 43.27 <sup>bc</sup>    | 1.63 <sup>b</sup>          | 40.59 <sup>b</sup> |
| T <sub>5</sub> : FS with ABS FeNPs                                                                          | 242.33 <sup>b</sup>                  | 45.20 <sup>b</sup>     | 1.67 <sup>b</sup>          | 40.75 <sup>b</sup> |
| T <sub>6</sub> : SP + FS with ABS FeNPs                                                                     | 253.67 <sup>a</sup>                  | 48.23 <sup>a</sup>     | 1.80 <sup>a</sup>          | 42.19 <sup>a</sup> |
| T <sub>7</sub> : SP with commercial FeNPs                                                                   | 235.33 <sup>b</sup>                  | 42.60 <sup>c</sup>     | 1.60 <sup>b</sup>          | 40.28 <sup>b</sup> |
| T <sub>8</sub> : FS with commercial FeNPs                                                                   | 238.00 <sup>b</sup>                  | 43.50 <sup>bc</sup>    | 1.63 <sup>b</sup>          | 40.35 <sup>b</sup> |
| T <sub>9</sub> : SP + FS with commercial FeNPs                                                              | 249.67 <sup>a</sup>                  | 47.57 <sup>a</sup>     | 1.76 <sup>a</sup>          | 41.87 <sup>a</sup> |
| T <sub>10</sub> : FS with FeSO <sub>4</sub> @ 0.5%                                                          | 228.00 <sup>c</sup>                  | 40.30 <sup>d</sup>     | 1.49 <sup>c</sup>          | 39.21 <sup>c</sup> |
| T <sub>11</sub> : RDF (100:75:50, N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg ha <sup>-1</sup> ) | 219.33 <sup>d</sup>                  | 37.23 <sup>e</sup>     | 1.38 <sup>d</sup>          | 38.07 <sup>d</sup> |
| T <sub>12</sub> : Control                                                                                   | 194.67 <sup>e</sup>                  | 31.10 <sup>f</sup>     | 0.72 <sup>e</sup>          | 35.25 <sup>e</sup> |
| S.Em. ±                                                                                                     | 2.47                                 | 0.77                   | 0.03                       | 0.35               |

**Table 7:** Grain yield, straw yield and harvest index in wheat affected by seed priming and foliar spraying with biosynthesized iron nanoparticles.

| Treatmental details                                                                                         | Grain yield (kg ha <sup>-1</sup> ) | Straw yield (kg ha <sup>-1</sup> ) | Harvest index     |
|-------------------------------------------------------------------------------------------------------------|------------------------------------|------------------------------------|-------------------|
| T <sub>1</sub> : SP with BS FeNPs                                                                           | 4214 <sup>de</sup>                 | 5985 <sup>b</sup>                  | 0.41 <sup>a</sup> |
| T <sub>2</sub> : FS with BS FeNPs                                                                           | 4345 <sup>bc</sup>                 | 6062 <sup>b</sup>                  | 0.42 <sup>a</sup> |
| T <sub>3</sub> : SP + FS with BS FeNPs                                                                      | 4539 <sup>a</sup>                  | 6339 <sup>a</sup>                  | 0.42 <sup>a</sup> |
| T <sub>4</sub> : SP with ABS FeNPs                                                                          | 4228 <sup>c-e</sup>                | 6045 <sup>b</sup>                  | 0.41 <sup>a</sup> |
| T <sub>5</sub> : FS with ABS FeNPs                                                                          | 4359 <sup>b</sup>                  | 6112 <sup>b</sup>                  | 0.42 <sup>a</sup> |
| T <sub>6</sub> : SP + FS with ABS FeNPs                                                                     | 4563 <sup>a</sup>                  | 6357 <sup>a</sup>                  | 0.42 <sup>a</sup> |
| T <sub>7</sub> : SP with commercial FeNPs                                                                   | 4156 <sup>e</sup>                  | 5972 <sup>b</sup>                  | 0.41 <sup>a</sup> |
| T <sub>8</sub> : FS with commercial FeNPs                                                                   | 4295 <sup>b-d</sup>                | 6045 <sup>b</sup>                  | 0.42 <sup>a</sup> |
| T <sub>9</sub> : SP + FS with commercial FeNPs                                                              | 4497 <sup>a</sup>                  | 6287 <sup>a</sup>                  | 0.42 <sup>a</sup> |
| T <sub>10</sub> : FS with FeSO <sub>4</sub> @ 0.5%                                                          | 4025 <sup>f</sup>                  | 5792 <sup>c</sup>                  | 0.41 <sup>a</sup> |
| T <sub>11</sub> : RDF (100:75:50, N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg ha <sup>-1</sup> ) | 3871 <sup>g</sup>                  | 5618 <sup>d</sup>                  | 0.41 <sup>a</sup> |
| T <sub>12</sub> : Control                                                                                   | 1752 <sup>h</sup>                  | 3230 <sup>e</sup>                  | 0.35 <sup>b</sup> |
| S.Em. ±                                                                                                     | 66.45                              | 75.82                              | 0.01              |

stems, facilitating the intake of active molecules and promoting growth and production. Nano micronutrient stimulatory effects on the production of chlorophyll, photosynthesis, mitochondrial respiration and hormone biosynthesis, including gibberellic acid and jasmonic acid, may contribute to the improvement of vegetative growth.

#### Effect of biosynthesized iron nanoparticles on yield and yield components in wheat

Seed priming @ 250 ppm and foliar spraying @ 500 ppm with iron nanoparticles (FeNPs) biosynthesized by actinobacteria recorded significantly higher productive spikes per square meter (253.67 m<sup>-2</sup>), number of grains per spike (48.23), grain weight per spike (1.80 g) and test weight (42.19 g) and found on par with seed priming @ 250 ppm and foliar spraying @ 500 ppm with FeNPs biosynthesized by *Pseudomonas* (251.67 m<sup>-2</sup>, 47.73, 1.79 g and 42.05 g, respectively) and commercial iron nanoparticles (249.67 m<sup>-2</sup>, 47.57, 1.76 g and 41.87 g, respectively) (Table 6). Seed priming @ 250 ppm and foliar spraying @ 500 ppm at panicle initiation stage with FeNPs biosynthesized from

actinobacteria, *Pseudomonas* and commercial iron nanoparticles increased the wheat yield by 17.87, 17.25 and 16.17 per cent, respectively compared to control (Table 7). Harsini *et al.* (2014), using nano chelated iron fertiliser can increase nitrogen uptake by considerably affecting the number of spikes, 1000-grain weight, grains per spike, grain yield, biological yield and harvest index when compared to the control. Due to the ultra-small size and diffusible nature of nano nutrients, which boosted their effectiveness in penetrating through the leaf surface and releasing ions across the cuticle, the nano composite increased the absorption of Fe in plants. The foliar application of iron nanoparticles increased the photosynthetic and metabolic activity of the plant, considerably raised the nutritional content, growth and yield (Sekaran and Singaravel, 2022 and Prerna *et al.* 2021).

#### CONCLUSION

Biosynthesis of nanoparticles using microorganisms is considered to be an environmentally friendly approach.

Farmers can replace the conventional iron source with nano forms to obtain the higher yields, where biosynthesized nanoparticles could be alternative to chemical nanoparticles in terms of high cost and pollution hazards. Seed priming and foliar spraying with biosynthesized iron nanoparticles recorded significantly higher growth, yield and yield attributing characters. In wheat seed priming and foliar spraying at panicle initiation stage with iron nanoparticles biosynthesized through actinobacteria recorded significantly higher plant height, number of tillers per meter row length, leaf area, leaf area index, total dry matter production, productive spikes per square meter, number of grains per spike, grain weight per spike, 1,000 grain weight, grain yield and straw yield, which was on par with seed priming and foliar spraying with iron nanoparticles biosynthesized through *Pseudomonas* and commercial iron nanoparticles.

### Conflict of interest

All authors declared that there is no conflict of interest.

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