



## Carbon Nanoparticles Synthesized from Glucose and Different Amino acids: A Novel Foliar Stimulant for Improving Growth Performance and Productivity of Garlic Plant



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**D**UE TO THE increasing environmental and economic challenges, there is an urgent need to conduct research into areas that will contribute to creating new strategies that improve the efficiency of the agricultural sector and maintain sustainability. Currently, carbon nanoparticles (CNP) have emerged as a promising technology for enhancing the effectiveness of amino acids and other bioactive compounds. So, a field research work was executed aiming to evaluate the response of garlic plant to the foliar application of the studied natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids under 9 treatments [T<sub>1</sub>: Control (tap water), T<sub>2</sub>: β-Alanine (at rate of 80 mg L<sup>-1</sup>), T<sub>3</sub>: Citrulline (at rate of 80 mg L<sup>-1</sup>), T<sub>4</sub>: Proline (at rate of 80 mg L<sup>-1</sup>), T<sub>5</sub>: Taurine (at rate of 80 mg L<sup>-1</sup>), T<sub>6</sub>: β-Alanine CNP (at rate of 80 mg L<sup>-1</sup>), T<sub>7</sub>: Citrulline CNP (at rate of 80 mg L<sup>-1</sup>), T<sub>8</sub>: Proline CNP (at rate of 80 mg L<sup>-1</sup>), T<sub>9</sub>: Taurine CNP (at rate of 80 mg L<sup>-1</sup>)] under completely randomized design. All treatments of natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids significantly increased the values of all studied traits related with growth criteria (e.g., leaves fresh and dry weights and leaf area), photosynthetic pigments (e.g., Chlorophyll a & b), leaf nutritional status (e.g., nitrogen, phosphorus and potassium), bulb yield (e.g., No. of cloves bulb<sup>-1</sup>, bulb yield and marketable yield) and bulb quality (e.g., carbohydrates, total dissolved solid, dry matter and vitamin C). All CNP treatments outperformed the natural amino acid treatments. Regarding the natural amino acid treatments, the superior amino acid was taurine followed by proline then citrulline and lately β-Alanine. Concerning the CNP treatments, the superior treatment was taurine CNP followed by proline CNP then citrulline CNP and lately β-Alanine CNP. For example, the increasing rate in the marketable yield values due to the studied treatments compared to control treatment was 5.02, 8.21, 13.47, 16.89, 22.14, 26.48, 30.13 and 34.70 % with T<sub>2</sub>, T<sub>3</sub>, T<sub>4</sub>, T<sub>5</sub>, T<sub>6</sub>, T<sub>7</sub>, T<sub>8</sub> and T<sub>9</sub>, respectively. Overall, it can be concluded that the sustainability of agriculture can be improved by applying the carbon nanoparticles (CNP) in the agricultural sector.

**Keywords:** β-Alanine, Citrulline, Proline, Taurine, Carbon nanoparticles (CNP)

### 1. Introduction

Due to the increasing environmental and economic challenges, there is an urgent need to conduct research into areas that will contribute to creating new strategies that improve the efficiency of the agricultural sector and maintain sustainability. Therefore, it has become imperative to find innovative solutions based on effective and safe inputs that support higher plant growth while reducing reliance on synthetic fertilizers and pesticides (Li *et al.* 2025). Amino acids are important bioactive compounds that play a vital role in the growth of higher plants. They significantly help regulate physiological and enzymatic processes, thereby improving the higher plant's ability to adapt to any difficult environmental circumstances. Furthermore, they have special properties that enable them to have a significant impact on plant growth and agricultural productivity. Among these amino acids are β-Alanine, citrulline, proline and taurine. β-Alanine and taurine amino acids are known for their effective role in raising the resistance of higher plants to oxidative stress, while citrulline and proline amino acids are known for their roles in improving higher plant tolerance to salt stress (Karunaratne *et al.* 2025).

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Currently, carbon nanoparticles (CNP) have emerged as a promising approach for improving the effectiveness of bioactive compounds, including amino acids. It is a novel member of the renowned carbon-based nano materials family, have gained tremendous attention in various fields (Shojaei *et al.* 2019). It has high water solubility, stability and intense fluorescent characteristics. Additionally, it is characterized by high surface activity. CNP possess the ability to enhance the compounds' uptake by higher plants, opening a novel horizon in sustainable and smart agriculture (Salama *et al.* 2021).

Garlic (*Allium sativum* L.) is one of the most important vegetable crop which compels scientific researchers to focus on maximizing its productivity, given its economic, nutritional, and medicinal importance. Furthermore, it is a good indicator of the effectiveness of any treatments (Thakur, 2024). Therefore, this research aims to evaluate the impact of natural amino acids and CNP on the growth performance and productivity of garlic.

## 2. Materials and Methods

### 2.1. Materials for Carbon nanoparticles Synthesis

Four types of amino acids were used in this study, as they were obtained from Purita chemical company, Tower C Haijing International E&T Develop Hg Zone, Xi'an, Shaanxi, China. While glucose was purchased from Egyptian commercial market. The characteristics of the studied amino acids are shown in Table 1.

**Table 1. Characteristics of selected amino acids used in the study.**

Amino acid	Chemical name	Structural type	Functional group
<b><math>\beta</math>-Alanine</b>	3-Aminopropionic acid	Beta-amino acid	$\beta$ -Amino group (-NH <sub>2</sub> on $\beta$ -carbon)
<b>L-Citrulline</b>	2-Amino-5-(ureido)pentanoic acid	Non-proteinogenic amino acid	Ureido group (-NH-C(=O)-NH <sub>2</sub> )
<b>L-Proline</b>	Pyrrolidine-2-carboxylic acid	Heterocyclic amino acid	Secondary amine (cyclic structure)
<b>Taurine</b>	Beta-amino sulfonic acid	Sulfur-containing amino acid	Sulfonic acid (-SO <sub>3</sub> H)

### 2.2. Carbon nanoparticles Synthesis

Carbon nanoparticles (CNP) synthesis was done *via* Maillard reaction. Each amino acid (200 g) and glucose (200 g) were dissolved in 100 ml of deionized water, then heated at 120°C in the autoclave for 12 hours, then the obtained product (about 150 g) was naturally cooled to room temperature (Nguyen *et al.* 2024). Fig 1 illustrates the photo of natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids.



**Fig. 1. Natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids.**

### 2.3. CNP Characterization

The characterization of the synthesized carbon nanoparticles (CNP) derived from the studied amino acids (  $\beta$ -alanine, citrulline, proline, and taurine ) was done *via* various analytical techniques which were carried out at the Electron Microscope Unit, Faculty of Agriculture, Mansoura University, EL-Mansoura, Egypt to determine the morphological features, surface charge, particle size distribution and elemental composition of the studied CNP, including the following analytical techniques.

- TEM: Transmission electron microscopy
- Zeta potential analysis

### 2.4. Field experimental of garlic plant

A field experiment was executed in a private farm located at Meit-Anter Village, Talkha District, El-Dakahlia Governorate, Egypt during the winter season of 2024/25 aiming to evaluate the response of garlic plant to the foliar application of the studied natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids under 9 treatments [T<sub>1</sub>: Control (tap water), T<sub>2</sub>:  $\beta$ -Alanine (at rate of 80 mg L<sup>-1</sup>), T<sub>3</sub>: Citrulline (at rate of 80 mg L<sup>-1</sup>), T<sub>4</sub>: Proline (at rate of 80 mg L<sup>-1</sup>), T<sub>5</sub>: Taurine (at rate of 80 mg L<sup>-1</sup>), T<sub>6</sub>:  $\beta$ -Alanine CNP (at rate of 80 mg L<sup>-1</sup>), T<sub>7</sub>: Citrulline CNP (at rate of 80 mg L<sup>-1</sup>), T<sub>8</sub>: Proline CNP (at rate of 80 mg L<sup>-1</sup>), T<sub>9</sub>: Taurine CNP (at rate of 80 mg L<sup>-1</sup>)] under completely randomized design with three replicates for each treatment. Fig 2 shows the flowchart of the field experiment. Before executing the planting, the soil of the experimental location was taken at the depth of 0-30 cm and analyzed as routine work according to the standard methods described by Dewis & Freitas, (1970), Hesse, (1971), Gee & Bauder, (1986) and Haluschak, (2006), as their properties are shown in Table 2.

**Table 2. Attributes of the initial soil.**

Properties and unit	Values
Sand, %	20.5
Silt, %	30.0
Clay, %	49.5
Textural class is clay	
Available-N, mgKg <sup>-1</sup>	23.6
Available- P, mgKg <sup>-1</sup>	7.20
Available- K, mgKg <sup>-1</sup>	201.3
pH	8.000
EC, dSm <sup>-1</sup>	3.420
WHC, %	36
O.M %	1.29

Garlic bulbs (Seds 40 variety) as seeds were obtained from the Egyptian Ministry of Agriculture and soil Reclamation (MASR). The cloves were sown on November 1<sup>st</sup>, at a rate of 450 kg of garlic (including foliage). The soil was thoroughly plowed before planting, adding 20 m<sup>3</sup> of compost, 400 kg of calcium superphosphate, 100 kg of agricultural sulfur, and 50 kg of ammonium sulfate fed<sup>-1</sup> then the soil was leveled and lined with rows.

The soil was irrigated, and after draining the water, the cloves were sown when the soil was moderately moist. The cloves were placed upright, 10 cm apart, on both sides of the row. The cloves were placed upright, with the base facing down and the top facing up. All the guidelines of the MASR to improve garlic productivity, including irrigation, fertilization, feeding, pest control, fungal diseases, insects and weeds, were implemented. The studied amino acids were sprayed in their nine treatments 5 times, starting from the 50<sup>th</sup> day of sowing, with a 15-day interval between each time. Harvesting took place 176 days after sowing. Table 3 illustrates the measurements during two studied stages (after 120 and 176 days from sowing) and data statistical analysis method.

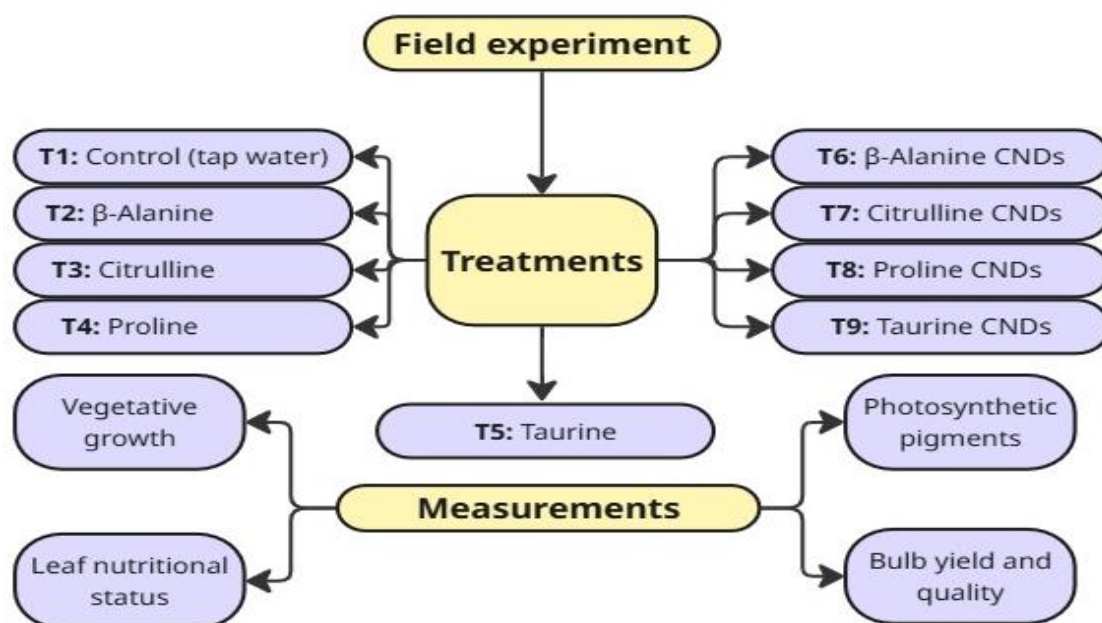


Fig. 2. The flowchart of the field experiment.

Table 3. Measurements during the studied stages and data statistical analysis method.

Parameters	The used methods	References
Plant height (cm), No. of leaves plant <sup>-1</sup> , leaves fresh and dry weights (g plant <sup>-1</sup> ) and leaf are, (cm <sup>2</sup> plant <sup>-1</sup> ) at 120 days from sowing	Manually	-----
Chlorophyll a & b and carotene pigments (mg g <sup>-1</sup> ) in fresh weight at 120 days from sowing	Spectrophotometrically (using acetone)	[1]
Digestion of the leaves at 120 days from sowing	HClO <sub>4</sub> + H <sub>2</sub> SO <sub>4</sub>	[2]
Leaf chemical constitutes (NPK)	Kjeldahl (N) Spectrophotometr (P) Flame photometer (K)	[3]
Average bulb weight (g), bulb diameter (cm), neck diameter (cm), bulbing ratio, No. of cloves bulb <sup>-1</sup> , bulb yield (ton fed <sup>-1</sup> ) and marketable yield (ton fed <sup>-1</sup> ) at 176 days from sowing	Manually Bulbing ratio = Bulb diameter / Pseudostem diameter	-----
Carbohydrates, total dissolved solid and dry mater (%), vitamin C(mg 100g <sup>-1</sup> ) and pungency (pervate content μmol ml <sup>-1</sup> ) at 176 days from sowing	Depending on the standard methods described by the AOAC Association of Official Analytical Chemists	[4]
Data statistical analysis	Using CoStat software at the 0.05 probability level (Version 6.303, Copyright ,1998-2004)	[5]

List of refs: [1] Wellburn (1994), [2] Peterburgski (1968), [3] Walinga *et al.* (2013), [4] AOAC (2000), [5] Gómez and Gómez, (1984)

### 3. Results

#### 3.1. CNP Characterization

The TEM of β-Alanine CNP reveals that the particles have a quasi-spherical shape with a relatively fine distribution, exhibiting no obvious agglomeration (Fig. 3 a). The particle sizes range from 42.20 to 77.17 nm, with an average size of approximately 57 nm. On the other hand, the zeta potential of β-Alanine CNP was

approximately -18.5 mV (Fig. 3 b), indicating that the particle surface carries a moderate negative charge in aqueous media. TEM of citrulline CNP (Fig. 4a) shows that it is distributed in a spherical to oval shape, with some slight agglomeration, which may be due to intermolecular interactions. The particle sizes range from 50.20 nm to 99.02 nm, indicating a relatively larger average size than those prepared from  $\beta$ -Alanine. Furthermore, the citrulline CNP exhibited a zeta potential of -21.0 mV (Fig. 4b), an acceptable negative value indicating moderate to good colloidal stability in aqueous solution. The TEM of proline-CNP shows that the particles have a regular, almost spherical shape, with light, close clusters, (Fig. 5a). The particles' diameters range from 50.51 nm to 79.80 nm. Proline-CNP particles showed (as shown in Fig. 5 b) a zeta potential value of -13.1 mV, a relatively low negative value, indicating weak to moderate colloidal stability. The TEM of Taurine-CNP shows that it is distributed in a spherical to irregular shape, with some size variation. The particle diameters range between 50.46 nm and 103.17 nm, indicating a relatively broad size distribution compared to the other amino acids used (Fig. 6a). Despite this variation, there is no significant agglomeration, indicating good interparticle surface repulsive forces. Figure 6 b illustrates that the zeta potential value of Taurine-CNP was -32.8 mV, the highest negative value among the four types.

### 3.2. Growth Performance after 120 Days from Sowing

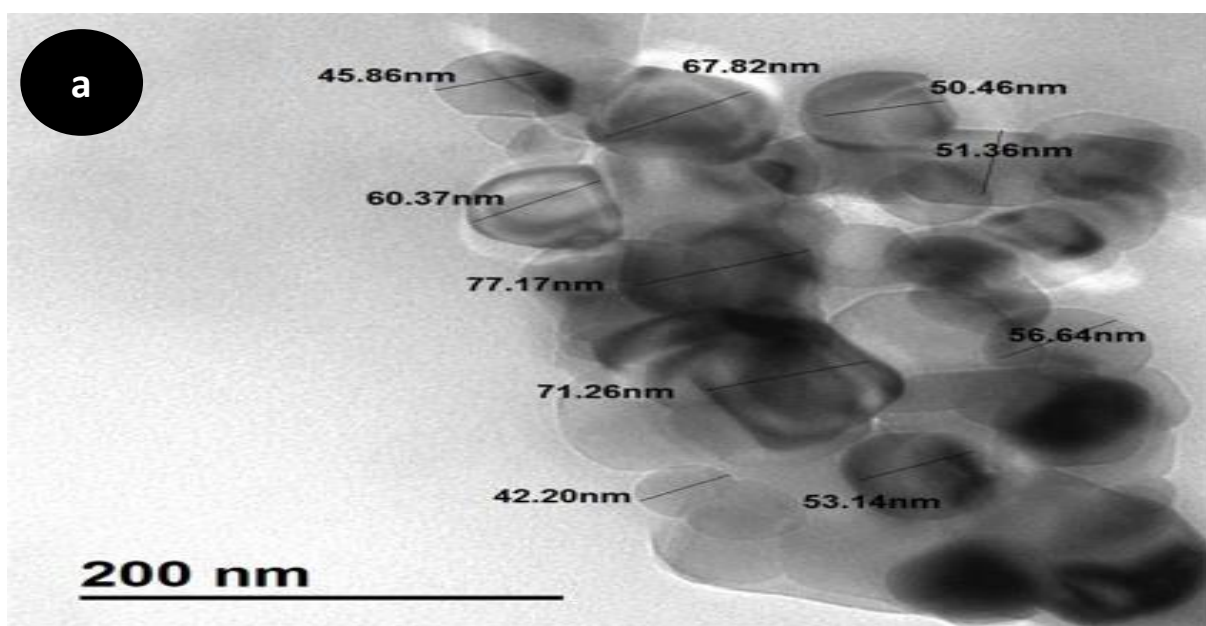
The impact of spraying natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids on growth criteria of garlic at 120 days from sowing was significant as shown in Table 4. These parameters included plant height (cm), No. of leaves plant<sup>-1</sup>, leaves fresh and dry weights (g plant<sup>-1</sup>) and leaf area, (cm<sup>2</sup> plant<sup>-1</sup>) at 120 days from sowing. In addition, the impact of the same treatments was significant on the photosynthetic pigments (Chlorophyll a & b and carotene pigments (mg g<sup>-1</sup>) in fresh weight) and leaf nutritional status (NPK, %) of garlic leaves at 120 days from sowing as shown in Table 5. All treatments of natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids significantly increased the values of all traits presented in both Tables. All CNP treatments outperformed the natural amino acid treatments. Regarding the natural amino acid treatments, the superior amino acid was taurine followed by proline then citrulline and lately  $\beta$ -Alanine. Concerning the CNP treatments, the superior treatment was taurine CNP followed by proline CNP then citrulline CNP and lately  $\beta$ -Alanine CNP. For example, the increasing rate in plant height values due to the studied treatments compared to control treatment was 4.06, 5.49, 8.05, 9.29, 10.28, 12.90, 14.10 and 14.49% with T<sub>2</sub> ( $\beta$ -Alanine), T<sub>3</sub> (Citrulline), T<sub>4</sub> (Proline), T<sub>5</sub> (Taurine), T<sub>6</sub> ( $\beta$ -Alanine CNP), T<sub>7</sub> (Citrulline CNP), T<sub>8</sub> (Proline CNP) and T<sub>9</sub> (Taurine CNP), respectively.

### 3.3. Bulb Yield and its Components at Harvest Stage (176 days from sowing)

Spraying natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids significantly affected bulb yield [Average bulb weight (g), bulb diameter (cm), neck diameter (cm), bulbing ratio, No. of cloves bulb<sup>-1</sup>, bulb yield (ton fed<sup>-1</sup>) and marketable yield (ton fed<sup>-1</sup>) (Fig 6) ] and quality [Carbohydrates, total dissolved solid, dry matter (%), vitamin C (mg 100g<sup>-1</sup>) and pungency (pungent content  $\mu$ mol ml<sup>-1</sup>) ] of garlic at 176 days from sowing (Table 6 and 7 ). The data of both Tables indicate the positive effect of natural amino acids and CNP treatments, as the sequence order from the superior treatment to less was as follows;

T<sub>9</sub>: Taurine CNP > T<sub>8</sub>: Proline CNP > T<sub>7</sub>: Citrulline CNP > T<sub>6</sub>:  $\beta$ -Alanine CNP > T<sub>5</sub>: Taurine > T<sub>4</sub>: Proline > T<sub>3</sub>: Citrulline > T<sub>2</sub>:  $\beta$ -Alanine > T<sub>1</sub>: Control (tap water)

For example, the increasing rate in the marketable yield values due to the studied treatments compared to control treatment was 5.02, 8.21, 13.47, 16.89, 22.14, 26.48, 30.13 and 34.70 % with T<sub>2</sub> ( $\beta$ -Alanine), T<sub>3</sub> (Citrulline), T<sub>4</sub> (Proline), T<sub>5</sub> (Taurine), T<sub>6</sub> ( $\beta$ -Alanine CNP), T<sub>7</sub> (Citrulline CNP), T<sub>8</sub> (Proline CNP) and T<sub>9</sub> (Taurine CNP), respectively.



### Results

	Mean (mV)	Area (%)	St Dev (mV)
<b>Zeta Potential (mV): -18.5</b>	<b>Peak 1: -18.5</b>	100.0	4.12
<b>Zeta Deviation (mV): 4.12</b>	<b>Peak 2: 0.00</b>	0.0	0.00
<b>Conductivity (mS/cm): 1.05</b>	<b>Peak 3: 0.00</b>	0.0	0.00
<b>Result quality : Good</b>			

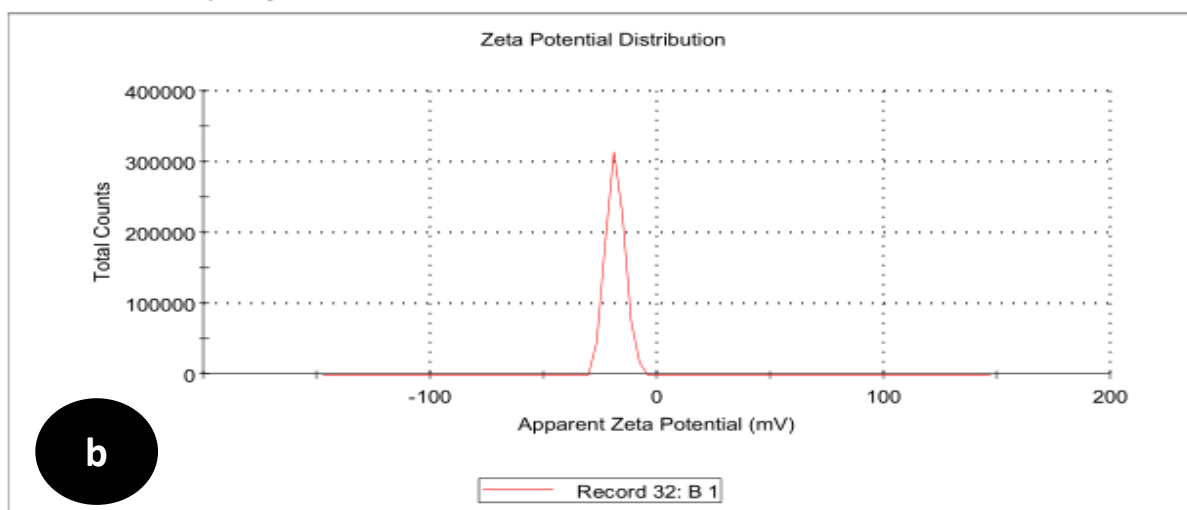


Fig. 3. Characterization of  $\beta$ -Alanine CNP. a, TEM. b, Zeta potential.

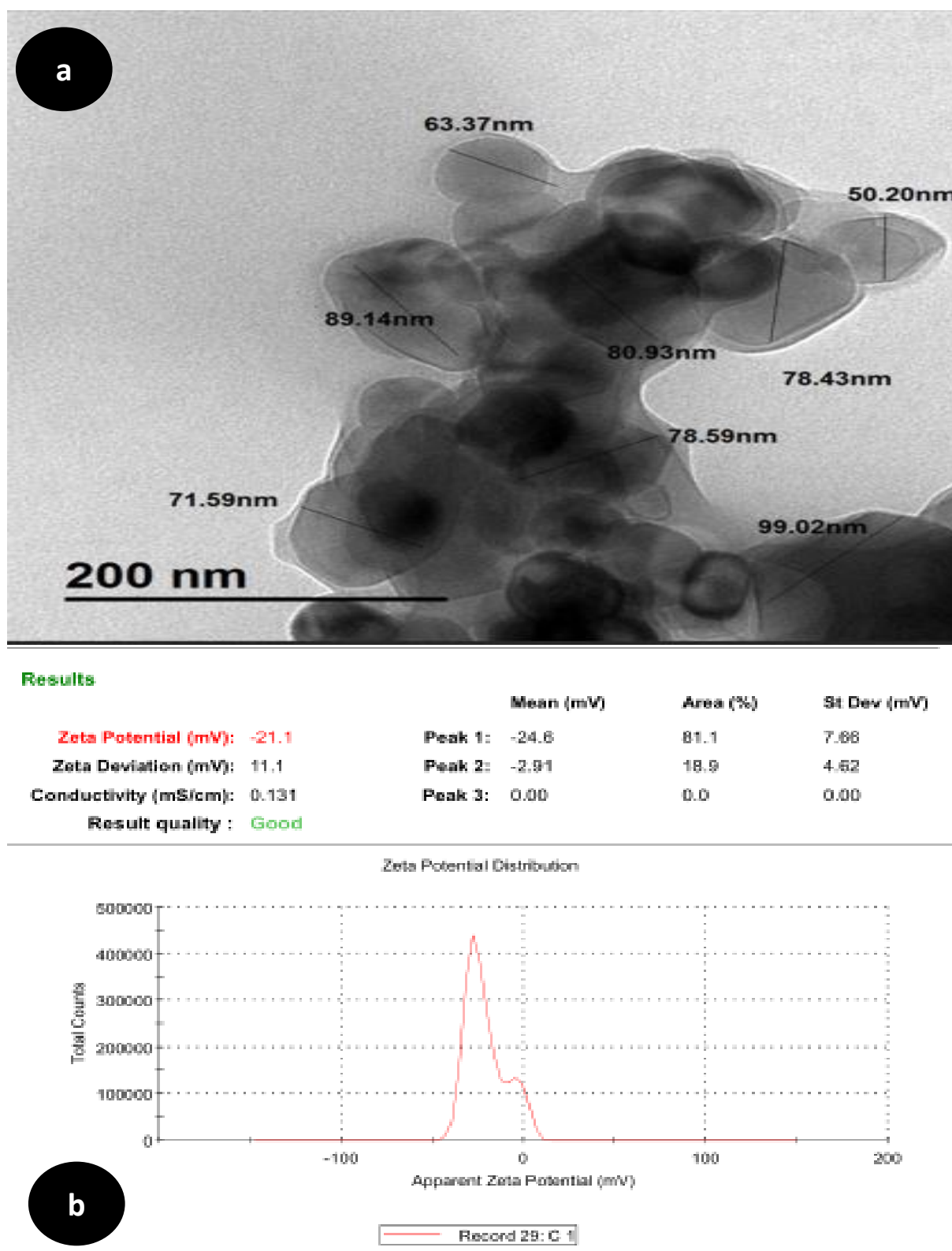
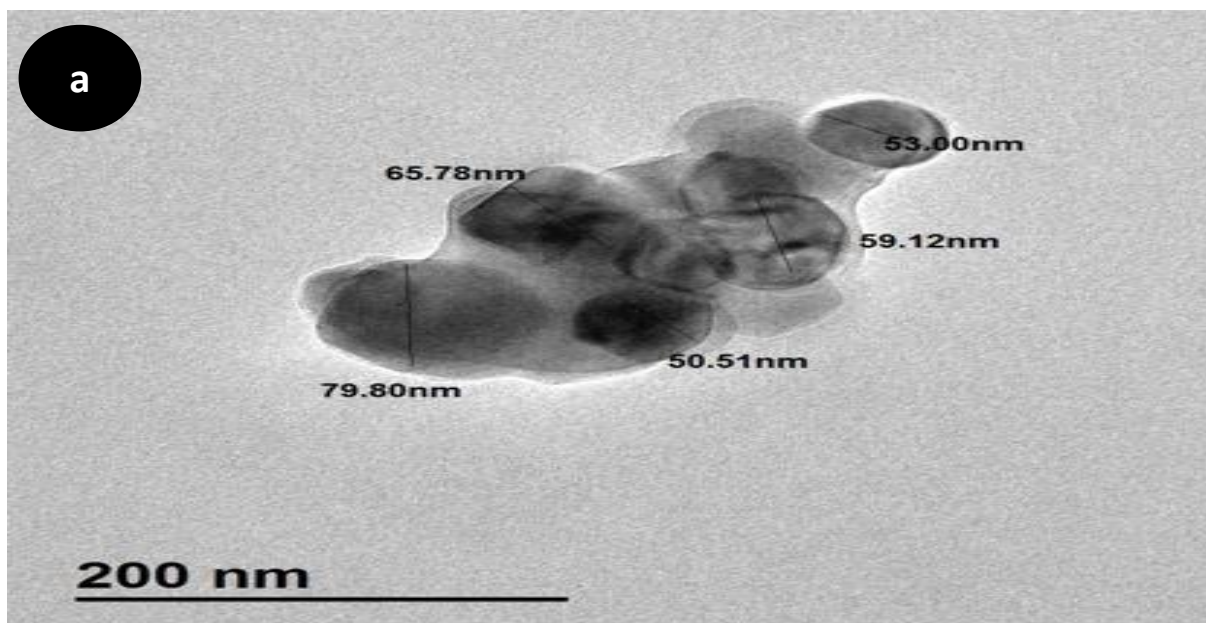


Fig. 4. Characterization of citrulline CNP. a, TEM. b, Zeta potential.



### Results

	Mean (mV)	Area (%)	St Dev (mV)
<b>Zeta Potential (mV): -13.1</b>	<b>Peak 1: -13.0</b>	98.4	4.02
<b>Zeta Deviation (mV): 4.30</b>	<b>Peak 2: -28.5</b>	1.6	1.88
<b>Conductivity (mS/cm): 0.469</b>	<b>Peak 3: 0.00</b>	0.0	0.00
<b>Result quality : Good</b>			

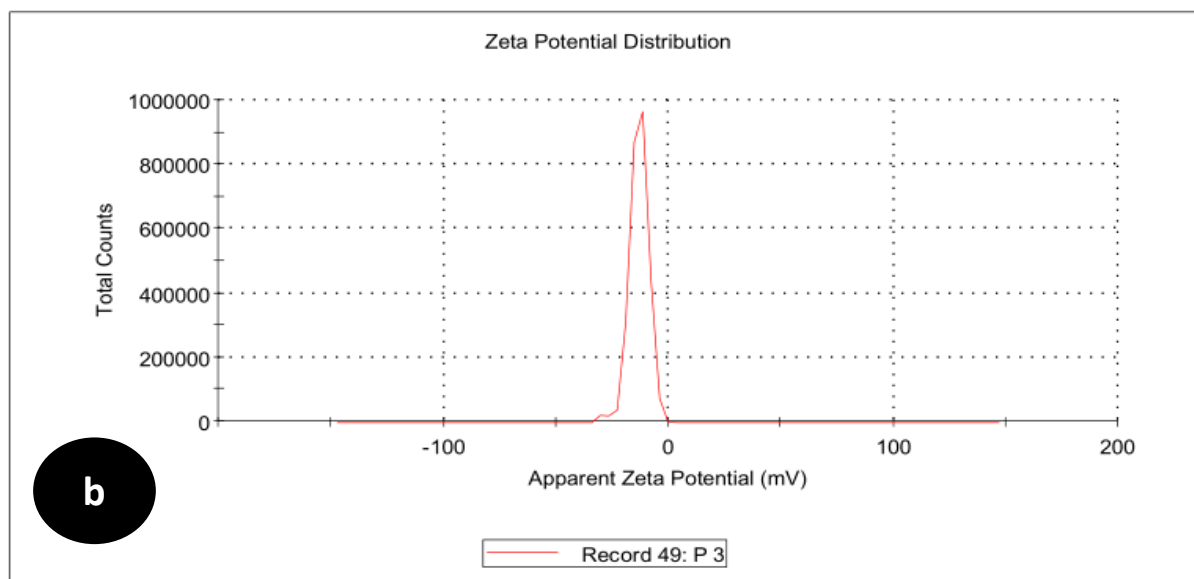
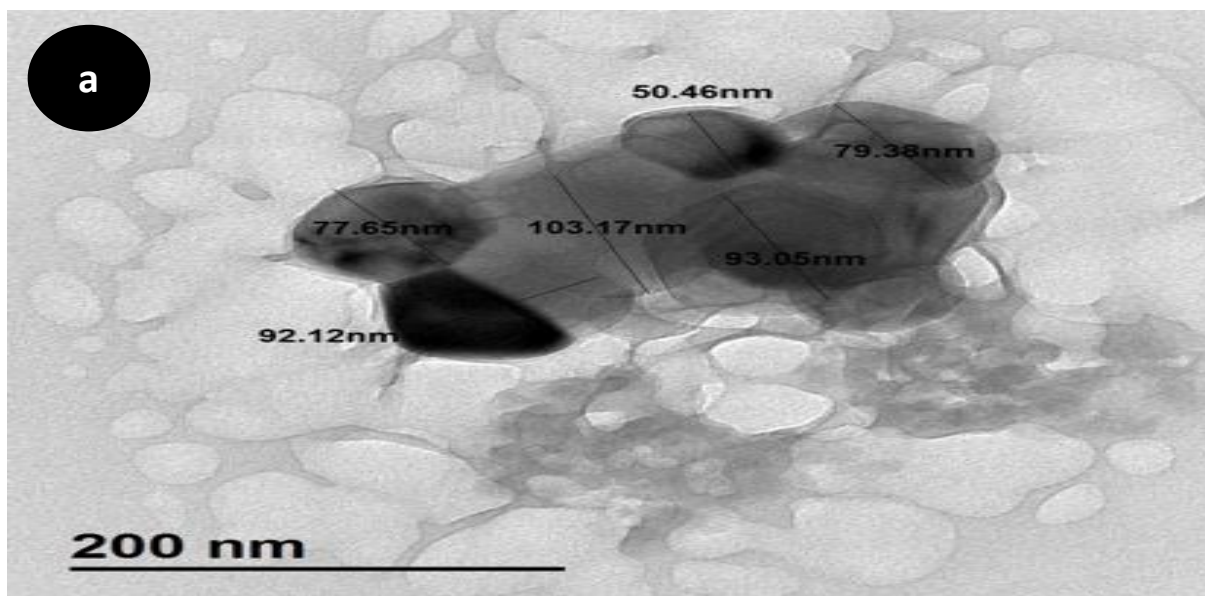


Fig. 5. Characterization of proline CNP. a, TEM. b, Zeta potential .



### Results

	Mean (mV)	Area (%)	St Dev (mV)
<b>Zeta Potential (mV): -32.8</b>	<b>Peak 1: -32.8</b>	100.0	6.12
<b>Zeta Deviation (mV): 6.12</b>	<b>Peak 2: 0.00</b>	0.0	0.00
<b>Conductivity (mS/cm): 0.865</b>	<b>Peak 3: 0.00</b>	0.0	0.00
<b>Result quality : Good</b>			

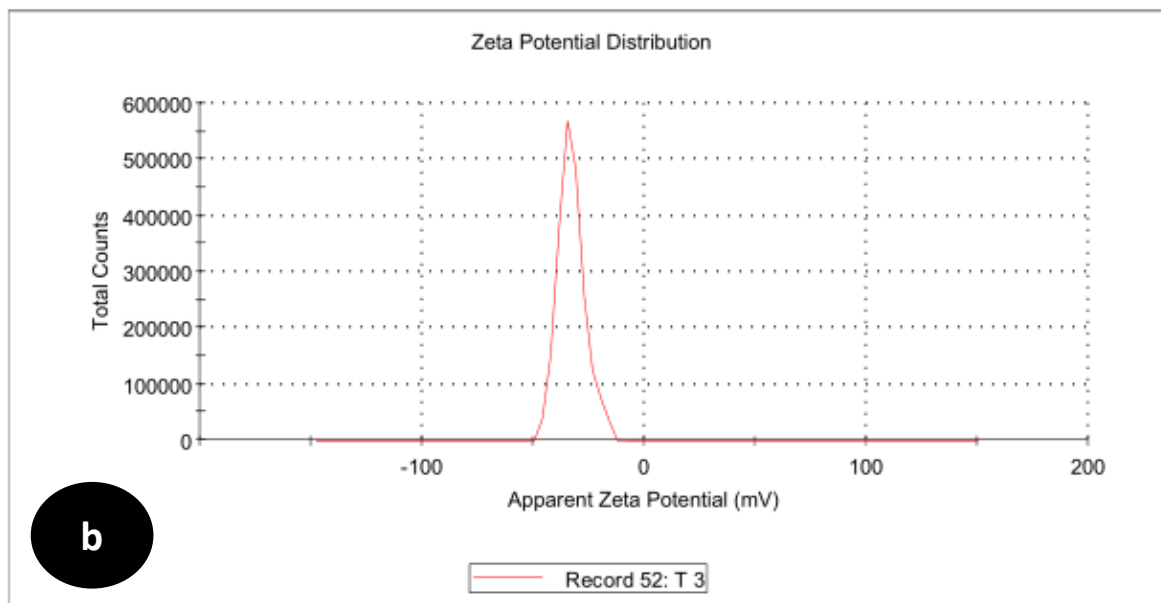


Fig 6. Characterization of Taurine CNP. a, TEM. b, Zeta potential.

**Table 4. Impact of spraying natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids on growth criteria of garlic at 120 days from sowing .**

Treatments	Plant height, cm	No. of leaves plant <sup>-1</sup>	Fresh weight, g plant <sup>-1</sup>	Dry weight, g plant <sup>-1</sup>	Leaf area, cm <sup>2</sup> plant <sup>-1</sup>
T <sub>1</sub>	70.07c	4.33c	59.92h	11.63g	228.04i
T <sub>2</sub>	72.92bc	4.67c	61.41g	12.49fg	235.85h
T <sub>3</sub>	73.92bc	5.00bc	63.21f	12.61f	249.98g
T <sub>4</sub>	75.71b	5.33bc	66.27e	12.89f	278.67f
T <sub>5</sub>	76.58b	5.67bc	68.35d	13.01e	284.44e
T <sub>6</sub>	77.28ab	6.33b	72.62c	13.51d	292.61d
T <sub>7</sub>	79.11ab	6.67ab	74.22bc	15.54c	298.25c
T <sub>8</sub>	79.95a	7.00a	75.67b	16.32b	305.35b
T <sub>9</sub>	80.23a	7.33a	78.76a	17.05a	312.28a
<b>F test</b>	**	**	***	***	***

Means within a row followed by a different letter (s) are statistically different at a 0.05 level

T<sub>1</sub>: Control (tap water), T<sub>2</sub>: β-Alanine, T<sub>3</sub>: Citrulline, T<sub>4</sub>: Proline, T<sub>5</sub>: Taurine, T<sub>6</sub>: β-Alanine CNP, T<sub>7</sub>: Citrulline CNP, T<sub>8</sub>: Proline CNP, T<sub>9</sub>: Taurine CNP

**Table 5. Impact of spraying natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids on photosynthetic pigments and chemical constituents of garlic leaves at 120 days from sowing.**

Treatments	Chlorophyll a, mg g <sup>-1</sup> F.W	Chlorophyll b, mg g <sup>-1</sup> F.W	Carotene, mg g <sup>-1</sup> F.W	N, %	P, %	K, %
T <sub>1</sub>	0.786i	0.555i	0.320i	2.73g	0.318h	2.18
T <sub>2</sub>	0.793h	0.558h	0.331h	2.78f	0.320g	2.23
T <sub>3</sub>	0.807g	0.571g	0.341g	2.80ef	0.324f	2.27
T <sub>4</sub>	0.821f	0.575f	0.356f	2.83e	0.328e	2.35
T <sub>5</sub>	0.842e	0.599e	0.363e	2.92de	0.332d	2.38
T <sub>6</sub>	0.853d	0.605d	0.371d	3.00d	0.355c	2.46d
T <sub>7</sub>	0.864c	0.614c	0.379c	3.09c	0.356c	2.51c
T <sub>8</sub>	0.878b	0.621b	0.390b	3.11b	0.363b	2.55b
T <sub>9</sub>	0.894a	0.634a	0.401a	3.15a	0.367a	2.58a
<b>F test</b>	***	***	***	***	***	***

Means within a row followed by a different letter (s) are statistically different at a 0.05 level

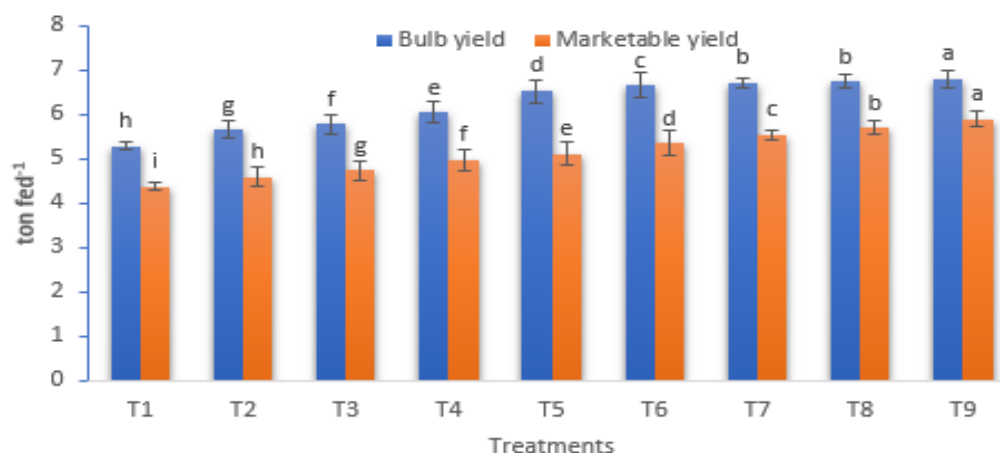
T<sub>1</sub>: Control (tap water), T<sub>2</sub>: β-Alanine, T<sub>3</sub>: Citrulline, T<sub>4</sub>: Proline, T<sub>5</sub>: Taurine, T<sub>6</sub>: β-Alanine CNP, T<sub>7</sub>: Citrulline CNP, T<sub>8</sub>: Proline CNP, T<sub>9</sub>: Taurine CNP

**Table 6. Impact of spraying natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids on bulb yield of garlic at 176 days from sowing .**

Treatments	Average bulb weight, g	Bulb diameter (cm)	Neck diameter (cm)	Bulbing ratio	No. of cloves bulb <sup>-1</sup>	Bulb yield (ton fed <sup>-1</sup> )	Marketable yield (ton fed <sup>-1</sup> )
T <sub>1</sub>	33.04f	4.00e	0.50d	0.12c	26.67d	5.29h	4.38i
T <sub>2</sub>	35.34ef	4.07e	0.57d	0.14c	28.67 CNP	5.65g	4.60h
T <sub>3</sub>	36.13e	4.20d	0.70c	0.17b	29.00c	5.78f	4.74g
T <sub>4</sub>	37.77d	4.37c	0.77c	0.18b	29.33c	6.04e	4.97f
T <sub>5</sub>	40.84c	4.47c	0.80b	0.18b	29.67bc	6.53d	5.12e
T <sub>6</sub>	41.58bc	4.70b	0.80b	0.17b	29.67bc	6.65c	5.35d
T <sub>7</sub>	41.95b	4.80a	0.90b	0.19b	30.33b	6.71b	5.54c
T <sub>8</sub>	42.14b	4.90a	0.90b	0.18b	30.67ab	6.74b	5.70b
T <sub>9</sub>	42.48a	4.90a	1.20a	0.24a	31.67a	6.80a	5.90a
<b>F test</b>	***	*	**	**	***	***	***

Means within a row followed by a different letter (s) are statistically different at a 0.05 level

T<sub>1</sub>: Control (tap water), T<sub>2</sub>: β-Alanine, T<sub>3</sub>: Citrulline, T<sub>4</sub>: Proline, T<sub>5</sub>: Taurine, T<sub>6</sub>: β-Alanine CNP, T<sub>7</sub>: Citrulline CNP, T<sub>8</sub>: Proline CNP, T<sub>9</sub>: Taurine CNP



**Fig. 6. Impact of spraying natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids on bulb yield of garlic at 176 days from sowing.**

T<sub>1</sub>: Control (tap water), T<sub>2</sub>:  $\beta$ -Alanine, T<sub>3</sub>: Citrulline, T<sub>4</sub>: Proline, T<sub>5</sub>: Taurine, T<sub>6</sub>:  $\beta$ -Alanine CNP, T<sub>7</sub>: Citrulline CNP, T<sub>8</sub>: Proline CNP, T<sub>9</sub>: Taurine CNP

**Table 7. Impact of spraying natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids on bulb quality of garlic at 176 days from sowing .**

Treatments	Carbohydrates, %	Total Dissolved Solid, %	Vitamin C, mg 100g <sup>-1</sup>	Dry matter, %	Pungency, purvate content $\mu\text{mol ml}^{-1}$
T <sub>1</sub>	19.16e	21.17c	11.84d	17.91e	8.06e
T <sub>2</sub>	19.18e	21.28c	11.91 CNP	17.95e	8.19de
T <sub>3</sub>	19.26d	21.43b	12.04c	18.42d	8.30d
T <sub>4</sub>	19.25d	21.49b	12.16bc	18.69 CNP	8.57 CNP
T <sub>5</sub>	19.61c	21.76b	12.17bc	18.73c	8.61c
T <sub>6</sub>	19.62c	22.08ab	12.27b	18.96bc	8.79bc
T <sub>7</sub>	19.90b	22.10ab	12.38b	19.11b	8.93b
T <sub>8</sub>	19.89b	22.39a	12.42a	19.64ab	9.03ab
T <sub>9</sub>	20.39a	22.41a	12.62a	19.87a	9.21a
<b>F test</b>	***	*	**	**	***

Means within a row followed by a different letter (s) are statistically different at a 0.05 level

T<sub>1</sub>: Control (tap water), T<sub>2</sub>:  $\beta$ -Alanine, T<sub>3</sub>: Citrulline, T<sub>4</sub>: Proline, T<sub>5</sub>: Taurine, T<sub>6</sub>:  $\beta$ -Alanine CNP, T<sub>7</sub>: Citrulline CNP, T<sub>8</sub>: Proline CNP, T<sub>9</sub>: Taurine CNP

#### 4. Discussion

The results of this research demonstrated that foliar spraying of amino acids in their natural form and in the form of carbon nanoparticles (CNP) resulted in significant improvements in all growth, yield, and quality parameters of garlic plants. This can be explained as follows:

##### 4.1. CNP Characterization

The particles of  $\beta$ -Alanine CNP are observed to be amorphous to semi-crystalline, lacking clear lattice fringes, indicating that the carbonization process successfully formed nanoparticles, but without high crystalline regularity. This size and relatively homogeneous distribution indicate that  $\beta$ -Alanine is a suitable starting material for the formation of stable carbon nanoparticles. On the other hand, the zeta potential of  $\beta$ -Alanine CNP was approximately -18.5 mV, indicating that the particle surface carries a moderate negative charge in aqueous media. This is likely due to the presence of functional groups such as carboxyl (-COOH) or amino derivatives associated with the  $\beta$ -Alanine structure. Although the ideal value for complete colloidal stability exceeds  $\pm 30$  mV, the recorded value (-18.5 mV) indicates an acceptable degree of colloidal stability, confirmed by the quality of the result, which is classified as "good." The narrow peak of the distribution and the relatively low standard deviation ( $\pm 4.12$  mV) also indicate the homogeneity of the surface charge among the different particles.

The particle sizes of TEM citrulline CNP indicate a relatively larger average size than those prepared from  $\beta$ -Alanine. This may be due to the different nature of the functional groups in citrulline and their effect on the particle growth rate during preparation. Regarding zeta potential, an acceptable negative value indicating moderate to good colloidal stability in aqueous solution. This indicates that the particles possess sufficient negative surface charge to prevent excessive agglomeration due to intermolecular electrostatic repulsion.

The particles' diameters of proline-CNP indicate a relatively small to medium size, suitable for biological and plant applications in terms of the particles' ability to penetrate tissues. Regarding zeta potential, a relatively low negative value indicates weak to moderate colloidal stability. The low surface potential may be due to the nature of the amino acid proline, which contains a closed pyrrolidine ring, which reduces the density of negative charges on the surface compared to other amino acids containing more free carboxylic or amino groups.

Concerning taurine CNP, there is no significant agglomeration, indicating good interparticle surface repulsive forces. This stability is likely due to the unique chemical properties of Taurine, which contains a sulfonic group ( $-\text{SO}_3\text{H}$ ) that imparts a strong negative charge to the surface of the particles. The highest negative value among the four types, indicating excellent colloidal stability. This means that the nanoparticles have a high ability to remain dispersed in a liquid medium without agglomerating, a property that is very important for biological applications such as intracellular transport or effective foliar spraying in plants. The obtained results are in harmony with those of Nguyen *et al.* (2024).

#### 4.2. Plant performance

Amino acids in carbon nanoparticles form were more effective in improving the performance and quantitative and qualitative crop characteristics of garlic compared to the traditional form, as this is possibly due to the fact that this technology increases the efficiency of absorption and permeability through the leaves when sprayed through the leaves on garlic plants, which accelerates the delivery of amino acids to the sites of action within plant cells, positively impacting all growth parameters and crop characteristics. Furthermore, this technology may improve the stability of the compounds and reduce their loss through decomposition or leaching (Nguyen *et al.* 2024). The studied amino acids, except taurine, may have played a key role in protein synthesis. Therefore, an improvement was found in the garlic performance when sprayed with the studied amino acids. They may have activated several physiological and enzymatic pathways within the garlic plant. Consequently, cell division, elongation and improved growth may have been caused by these amino acids. Taurine and proline may have increased the garlic plant's resistance to environmental stresses and improved the osmotic balance within the growing garlic plant cells (Li *et al.* 2024). They may also have helped maintain water content within the leaves, which positively impacted photosynthesis efficiency. Perhaps the soil EC value ( $3.4 \text{ dSm}^{-1}$ ), although it does not indicate that the soil is saline, it may have caused salt stress on the growing garlic plant, and thus caused the role of these two amino acids (taurine and proline) to become prominent.

This explains their superiority over the other two amino acids (citrulline and  $\beta$ -alanine). Taurine may have outperformed proline due to its role in scavenging free radicals (ROS) from plant cells. Citrulline came after taurine and proline, possibly due to its role in removing excess ammonia within the garlic plant cell.  $\beta$ -alanine, which ranks last among the studied amino acids in its natural form, due to its role as an important mediator in plant energy pathways (Karunaratne *et al.* 2025). Carbon nanoparticles (CNP) were superior compared to natural amino acid treatments, possibly due to the advantages of this approach. Amino acids, in their natural form, may be less stable at high temperatures compared to their carbon nanoparticles form. Converting them to CNP form may have protected the molecules and reduced their degradation or loss, thus increasing their effectiveness. Furthermore, their CNP form may have given them a high ability to penetrate cell walls and penetrate the inside of garlic plant cells, thus ensuring that amino acids quickly reach their sites of action within the garlic plant. In other words, CNP can act as smart carriers (Shojaei *et al.* 2019; Salama *et al.* 2021).

Additionally, CNP may have been effective carriers of bioactive compounds, due to their small size as well as high surface area, which contributes to improved leaf absorption and intracellular transport of active compounds. They also may have played a vital role in enhancing chlorophyll activity and stimulating electron transfer. Taurine-CNP was the most effective treatment, possibly due to their dual role as an osmotic compound and sulfur donor, thus enhancing garlic plant resilience to any environmental stress and improving the metabolic efficiency. Proline-CNP came in the second order may be due to its association with garlic plant responses to abiotic stresses such as drought and salinity (Nguyen *et al.* 2024).

#### 5. Conclusion

The obtained results confirm that all natural amino acids and carbon nanoparticles synthesized from glucose and selected amino acids have a vital role in enhancing the quantitative and qualitative properties of the garlic plant. All CNP treatments outperform the natural amino acid treatments. Regarding the natural amino acid treatments, the superior amino acid was taurine. Concerning the CNP treatments, the superior treatment was taurine CNP.

Generally, it can be concluded that spraying carbon nanoparticles (CNP) synthesized from natural amino acids is an effective strategy for improving the performance of garlic plants, highlighting the great potential of this approach in enhancing the efficiency of bioactive compounds usage within garlic plants. Overall, it can be concluded that the sustainability of agriculture can be improved by applying the CNP in the agricultural sector.

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## 6. References

- AOAC (2000). "Official Methods of Analysis". 18<sup>th</sup> Ed. Association of Official Analytical Chemists, Inc., Gaithersburg, MD, Method 04.
- CoStat, Version 6.303, Copyright (1998-2004), CoHort Software, Monterey, CA, USA.
- Dewis, J., & Freitas, F. (1970). Physical and chemical methods of soil and water analysis. FAO soils Bulletin, (10).
- Gee, G.W., & Bauder, J.W. (1986). Particle-size Analysis. p 383-411 In A. Klute (ed.) Methods of Soil Analysis Part 1. Soil Science Society of America Book Series 5, Madison, Wisconsin, USA.
- Gomez, K. A., & Gomez, A. A. (1984). "Statistical Procedures for Agricultural Research". John Wiley and Sons, Inc., New York, pp:680.
- Haluschak, P. (2006). Laboratory methods of soil analysis. Canada-Manitoba soil survey, 3, 133.
- Hesse, P. R (1971). A textbook of soil chemical analysis. Cambridge university press.
- Karunaratne, J. M. G. M. T., Weeraratna, E. K. W. W., Hong, X. Y., Wen, T., Hao, G., & Kandegama, W. M. W. W. (2025). Biogenic amino acids facilitate the management of plant health and stress tolerance for a modern sustainable approach. ACS Agricultural Science & Technology, 5(3), 294-315.
- Li, L., Awada, T., Shi, Y., Jin, V. L., & Kaiser, M. (2025). Global greenhouse gas emissions from agriculture: pathways to sustainable reductions. Global Change Biology, 31(1), e70015.
- Li, P., Sun, S., Zhang, W., Ouyang, W., Li, X., & Yang, K. (2024). The effects of L-citrulline supplementation on the athletic performance, physiological and biochemical parameters, antioxidant capacity, and blood amino acid and polyamine levels in Speed-Racing Yili Horses. Animals, 14(16), 2438.
- Nguyen, D. H., Muthu, A., El-Ramady, H., Daróczy, L., Nagy, L., Kéki, S., ... & Prokisch, J. (2024). Optimization of extraction conditions to synthesize green carbon nanoparticles using the Maillard reaction. Materials Advances, 5(8), 3499-3505.
- Peterburgski, A. V. (1968). "Handbook of Agronomic Chemistry". Kolos Publishing House, Moscow, (in Russian, pp. 29-86).
- Prokisch, J., Törös, G., Nguyen, D. H., Muthu, A., Labidi, S., Sheta, M. H., & El-Ramady, H. (2025). Sustainable Approach of Carbon Dots: Agro-Applications for Soil Health. Egyptian Journal of Soil Science, 65(1), 639-657.
- Salama, D. M., Abd El-Aziz, M. E., El-Naggar, M. E., Shaaban, E. A., & Abd EL-Wahed, M. S. (2021). Synthesis of an eco-friendly nanocomposite fertilizer for common bean based on carbon nanoparticles from agricultural waste biochar. Pedosphere, 31(6), 923-933.
- Shojaei, T. R., Salleh, M. A. M., Tabatabaei, M., Mobli, H., Aghbashlo, M., Rashid, S. A., & Tan, T. (2019). Applications of nanotechnology and carbon nanoparticles in agriculture. In Synthesis, technology and applications of carbon nanomaterials (pp. 247-277). Elsevier.
- Thakur, P., Dhiman, A., Kumar, S., & Suhag, R. (2024). Garlic (*Allium sativum* L.): A review on bio-functionality, allicin's potency and drying methodologies. South African Journal of Botany, 171, 129-146.
- Walinga, I., Van Der Lee, J. J., Houba, V. J., Van Vark, W. and Novozamsky, I. (2013). Plant Analysis Manual. Springer Science & Business Media.
- Wellburn, A. R. (1994). The spectral determination of chlorophylls a and b, as well as total carotenoids, using various solvents with spectrophotometers of different resolution. Journal of plant physiology, 144(3), 307-313.