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Response of Yield, Quality, and Bioactive Constituents of Green Onion to Foliar Spraying with *Moringa oleifera* Leaf Extract and Yeast Extract

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THE APPLICATION of biostimulants to improve plant growth, quality, and productivity is an effective and eco-friendly field. Green onion is a widely consumed vegetable due to its nutritional content and distinctive flavor. The response of green onion growth, yield, bioactive compounds accumulation, and antioxidant activity to foliar application of moringa (*Moringa oleifera*) leaf extract (MLE) and yeast extract (YE), as biostimulants was investigated in the present study during 2021/2022 and 2022/2023 seasons under open field conditions. MLE (2%, 4%, and 6%) and YE (1, 2, and 3 g/L) were applied as foliar solutions. The results showed that plant height, fresh weight (FW), dry matter, yield, nutrients content, photosynthetic pigments (chlorophyll and carotenoids), bioactive compounds [ascorbic acid, total phenolics content (TPC), and total flavonoids content (TFC)], and antioxidant properties [total antioxidant capacity (TAC) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging activity (IC_{50})] exhibited a better response to specific levels of YE and MLE. When compared to chemical fertilizers, the increase in green onion yield achieved by YE and MLE was accompanied by a decrease in nitrate content and an increase in total soluble solids (TSS), pyruvic acid, and ascorbic acid. TPC was maximized with chemical fertilizers and 2 g/L of YE. However, no statistical differences were observed in the TFC, TAC, and IC_{50} among treatments involving chemical fertilizers, moderate, and higher levels of MLE and YE. According to Pearson's correlation analysis, a positive correlation existed between yield and each of ascorbic acid, TPC, TFC, and TAC. The nitrate content correlated negatively with pseudo-stem length, TSS, pyruvic acid, and bioactive compounds. Conclusively, YE and MLE have the potential to make green onion farming more sustainable and resilient with high quality and high nutraceutical content, thus mitigating the need for chemical fertilizers.

Keywords: *Allium cepa*, biostimulants, nitrate, phenolics, antioxidants, DPPH.

1. Introduction

The *Allium* genus, belonging to the family Alliaceae, encompasses a diverse array of species primarily cultivated for their bulbous structures. Some species, however, are cultivated for their fresh aboveground portions, such as green onions (*A. cepa* L. and *A. fistulosum* L.), since they have excellent nutritional value and are a popular seasoning around the world (Gao et al., 2021; Papoui et al., 2022). Green onion (*A. cepa*), also known as bunching onion and spring onion, is a short-lived vegetable crop that is very popular in Egypt, Greece, Iraq, Turkey, the USA and is commercially grown for immediate consumption in spring (Kapoulas et al., 2017; Seymen et al., 2023). Global green onion production in 2020 amounted to 4,452,347 tons, cultivated on a total land area of 208,347 hectares (Seymen et al., 2023). According to available data, Egypt's green onion production amounts to 43,981 tons cultivated in an area of

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2,528 hectares (Agricultural Statistics Bulletin, 2019). Green onions can be ingested in several forms, including fresh, dried, or as ingredients in processed or therapeutic products (Gao et al., 2021). The leaves, pseudo-stems (which consist of leaf sheaths and young leaves), and immature bulbs of green onions are commonly consumed due to their significant nutraceutical composition. Additionally, their distinct flavor profile and capacity to enhance the taste of various dishes contribute to their popularity (Papoui et al., 2022; Qiao et al., 2024).

Green onion (*A. cepa*) is highly valued for its nutritional value in supplying essential minerals (potassium, calcium, magnesium, selenium, and iron) that are important for a healthy body, as well as vitamins such as ß-carotene, vitamin A, vitamin C, and folate (Kapoulas et al., 2017). Moreover, it has been found that green onions possess a high content of flavonoids, including quercetin, kaempferol, and luteolin (Miean and Mohamed, 2001). The distinguiareing feature of green onions is their distinct flavor derived from sulfur-containing molecules (Imai et al., 2002). The pungency of onions can be attributed to various volatile organosulfur compounds (Wang et al., 2020). In the past few years, both customers and scientists have become more interested in plant-based antioxidants (Gao et al., 2022; Pan et al., 2023). The biopharmacological studies conducted on the phytochemicals present in green onion have demonstrated their effectiveness as antioxidants, anticancer agents, antimicrobials, anti-platelet, anti-inflammatory, anti-thrombotic, anti-hyperlipidemic, anti-hypertensives, anti-diabetics, and anti-asthmatics, as well as cardiovascular benefits (Xiao and Parkin, 2007; Nicastro et al., 2015).

Chemical fertilizers serve as a predominant and expeditious means of supplying essential macro- and micro-nutrients to various crops (Obiadalla-Ali et al., 2022). Although the use of synthetic fertilizers is a prevalent practice in conventional agriculture, the resulting decline in soil biodiversity and subsequent environmental degradation pose significant challenges to agricultural sustainability (Gamage et al., 2023). The rise in popularity of organic foods has effectively mitigated adverse environmental effects (Youssef et al., 2020). However, it was observed that organic farming yields are lower compared to conventional farming (Gamage et al., 2023), primarily due to reduced nutrient availability and challenges related to pest and disease management (Kapoulas et al., 2017; Fahrurrozi et al., 2022). The inevitability of high consumer demand for organically grown green onions is expected to align with the overall surge in demand for organic vegetables (Fahrurrozi et al., 2022).

The pursuit of environmentally sustainable applications in the field of agriculture is of utmost importance and necessity. Accordingly, the employment of biostimulants in crop production is a prominent field of applied agricultural research for promoting sustainable development, mainly when it is associated with augmenting crop quality and accumulating desired substances (Hegazi et al., 2023). Using biostimulants to enhance plant growth, quality, and productivity has proven to be an effective and eco-friendly field (Abou Elhassan et al., 2023). On the other hand, the potential health risks correlated with consuming vegetables containing high concentrations of nitrate necessitate reevaluating the use of synthetic N fertilizers. Unfortunately, chemical fertilizers are extensively used in the conventional growing of green onions. Biostimulants are natural products that can reduce chemical fertilizer requirements while simultaneously promoting plant growth, improving the absorption and utilization of nutrients, and increasing tolerance to both biotic and abiotic stress (Arif et al., 2023; Gholami et al., 2023; Rakkammal et al., 2023; Shalaby, 2024). According to Toscano et al. (2023), the application of biostimulants in vegetable farming has the potential to reduce the nitrate levels found in the consumable portions.

Plant-derived biostimulants have garnered significant interest and are being more frequently included in high-value production systems, resulting in enhanced productivity and improved quality in an environmentally sustainable manner (Zulfiqar et al., 2020). Moringa leaf extracts (MLE) (*Moringa oleifera*; family Moringaceae) in particular have shown significant improvements in various aspects of plant biology. These include the facilitation of seed germination, promotion of plant growth and yield, optimization of nutrient utilization efficiency, improvement of quality attributes, and augmentation of tolerance towards different stressors (Zulfiqar et al., 2020; Awad et al., 2022; Admane et al., 2023). MLE is a highly abundant source of phytohormones, polyphenols, sterols, ascorbates, vitamins, amino acids, and macro- and micro-elements (Latif and Mohamed, 2016; Admane et al., 2023; Arif et al., 2023). The chemical composition of moringa leaves exhibits variability based on geographic origin and growing system. However, it has been shown that the extract derived from *M. oleifera* leaf possesses properties that make it a cost-effective and eco-friendly enhancer that has the potential to enhance sustainable agricultural practices and crop production (Arif et al., 2023). In addition, lettuce plants treated with MLE exhibited higher yield and lower nitrate content (Shalaby, 2024).

Table 1. Chemical analysis of the experimental soil as an average of the two growing seasons (2021/2022 and 2022/2023).

EC ($dS\ m^{-1}$)	pH	Available nutrients ($mg\ kg^{-1}$)			Soluble cation ($meq\ L^{-1}$)			Soluble anion ($meq\ L^{-1}$)		
		N	P	K	Ca^{++}	Mg^{++}	Na^{+}	HCO_3^{-}	Cl^{-}	SO_4^{--}
0.37	7.44	13.4	1.6	192	1.35	0.88	1.44	1.18	1.24	1.25

Yeast, a naturally occurring bio-substance, has been proposed as a potentially beneficial biofertilizer for vegetable crops. Yeast extract (YE) has been found to have a stimulating effect on various plant processes, including cell division and the synthesis of chlorophyll, protein, and nucleic acid (Gholami et al., 2023). This is attributed to the presence of protective agents and essential components such as cytokinins, thiamine, pyridoxine, riboflavin, and vitamins B1, B2, and B12, in addition to sugars, amino acids, proteins, minerals, and other nutrients (Barnett et al., 1990; Fawzy et al., 2012; Abdelaal et al., 2021; El-Serafy et al., 2021). Treated garlic and bulb onion plants with YE had notable impacts on growth, yield, and quality parameters (Ali, 2017; Marey and El-Masry, 2024). Furthermore, biotic elicitation with YE has been found to play an important role in enhancing the accumulation of biomass and diverse by-products in medicinal crops (Bosila et al., 2016; Elshahawy et al., 2022).

Following the application of specific biostimulants, there is a possibility of alterations in many characteristics of green onions, including but not limited to their productivity, aroma, and accumulation of bioactive ingredients. As green onion is a highly valuable horticultural crop, it is imperative to attain economic productivity accompanied by desirable quality attributes such as low content of nitrate, increase in pungency, total soluble solids (TSS), nutraceuticals, and antioxidant activity. However, investigations of the growth, productivity, and quality characteristics of green onion (*A. cepa*) in response to MLE and YE are scarcely available in the literature. Therefore, this paper studied the changes in the growth, yield, quality parameters, and bioactive compounds accumulation of green onion (*A. cepa*) under different applications of biostimulants (MLE and YE). The aim was to evaluate the potential of biostimulants as an environmentally friendly alternative to chemical fertilizers.

2. Materials and Methods

2.1. Experiment site and design

A field experiment was carried out at the Horticulture Department Farm, Faculty of Agriculture, Al-Azhar University, Nasr City, Cairo, Egypt (30°03′13.2″ N, 31°19′11.1″ E), during the consecutive winter seasons of 2021/2022 and 2022/2023. The soil used was classified as sandy loam (23.74% silt, 64.52% sand, and 11.74% clay). The chemical analysis of the experimental soil is outlined in Table 1 according to Page et al. (1982) and Klute (1986).

The seeds of green onion (*A. cepa*) cultivar ‘Giza 20’ were obtained from the Agricultural Research Center, Giza, Egypt. In the 2021/2022 and 2022/2023 seasons, seeds were sowed in nursery beds during the first week of October. Following 45 days, the seedlings were transplanted to the open field, specifically in plots (3 × 1.5 m) of three rows at 50 cm between rows. The seedlings were planted on both row sides at 6 cm between plants. Each plot consisted of 300 plants. The design of the experiment was a complete randomized block design, with three replicates for each treatment.

2.2. Treatments

The leaves of *M. oleifera* were obtained from Al-Sadat City, Menoufia, Egypt. Fresh green leaves were collected and cleaned. Then, using a blender and distilled water (100 mL), the leaves (100 g FW) underwent extraction at room temperature. The resulting mixture was filtered, then kept at 4 °C until use. The final concentrations of 2%, 4%, and 6% (MLE1, MLE2, and MLE3, respectively) were prepared using the crude extract (Bashir et al., 2014).

Active dry yeast (*Saccharomyces cerevisiae*) was procured from the Egyptian Sugar and Integrated Industries Company, Giza, Egypt. The dried yeast was first dissolved in distilled water, then glucose was added in a 1:1 ratio. The mixture was left at room temperature overnight for activation and reproduction of the yeast (Shalaby and El-Ramady, 2014), and was then applied directly. The yeast extract (YE) was applied through spraying at 1, 2, and 3 g L^{-1}, denoted as YE1, YE2, and YE3, respectively.

The chemical fertilizers (positive control) used in this study included recommended NPK (R-NPK). Here, calcium superphosphate (15.5% P_2O_5) was added to the soil at a rate of 475 kg ha^{-1} before transplantation. Ammonium sulfate (20.5% N) and potassium sulfate (48% K_2O) were added to the soil at a rate of 165 kg ha^{-1} and 120 kg ha^{-1}, respectively, after 20 days following transplanting.

The solutions of MLE and YE were sprayed onto onion leaves with Tween-20 as a surfactant (0.1% v/v) until running off point three times: 15, 25, and 35 days after cultivation. Prior to planting, animal and plant residues compost (1.36% total N, 0.15% total P, 0.22% total K, 59.67% organic matter, and

34.69 organic carbon) was applied as basal fertilization at a rate of 48 m^3 ha^{-1} during the soil preparation. The soil moisture was maintained at field capacity. The treatments and measurements considered in this study are shown in Figure 1.

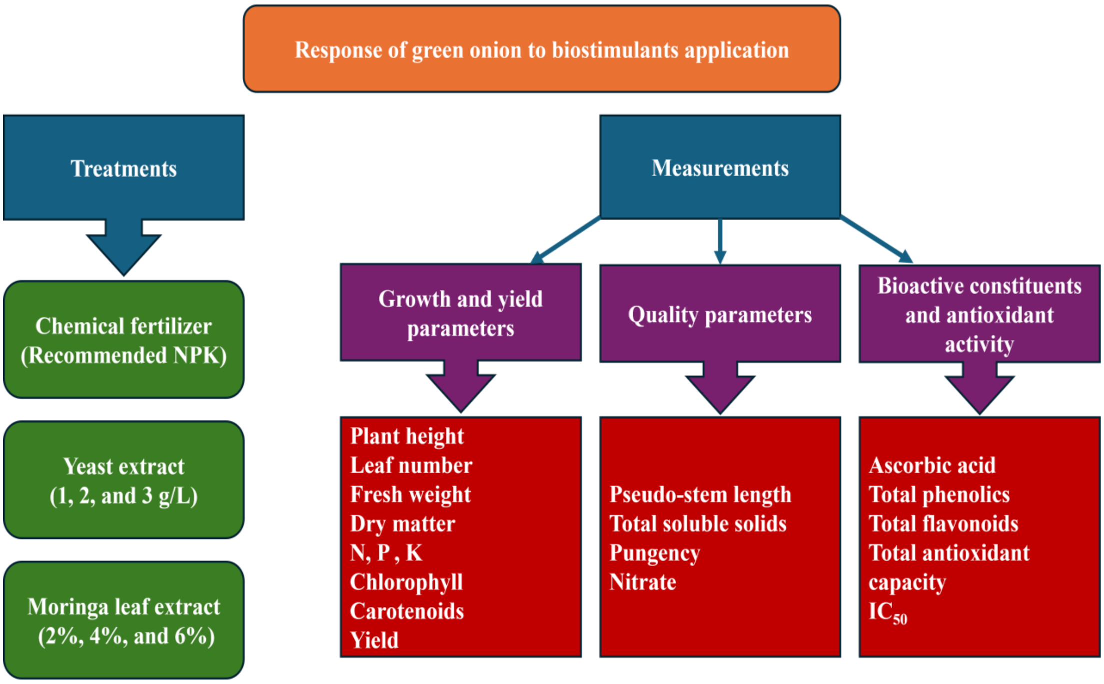

Fig. 1. The treatments and measurements considered in the current study.

2.3. Measurements

2.3.1. Growth and yield attributes

Green onion was harvested 50 days after transplanting. A sample of ten plants was chosen randomly from each treatment's experimental unit, and the clinging soil around the bulbs was removed. The growth and yield parameters were plant height (cm), leaf number per plant, fresh weight (FW) (g $plant^{-1}$), dry matter (%), and yield (ton ha^{-1}). The estimation of nitrogen (N) and phosphorus (P) percentages was conducted by following the method outlined in the AOAC (1995) publication. Meanwhile, the potassium (K) percentages assessed according to the methodology described by Dewis and Freitas (1970).

2.3.2. Photosynthetic pigments

The quantification of photosynthetic pigments (chlorophyll and carotenoids; mg g^{-1} FW) was conducted following the methodology outlined by Lichtenthaler (1987). Acetone 80% (15 mL) was added to 0.2 g of freshly harvested leaf. After filtration, the volume was adjusted to 15 mL with acetone (80%). Subsequently, the absorption was determined using a Jenway 6800 UV/Vis spectrophotometer (Bibby Scientific Ltd., Staffordshire, UK) at wavelengths of 663.2 nm, 646.8 nm, and 470 nm against a blank of 80% acetone. Chlorophyll (Chl.) and carotenoids (Cart) concentrations were calculated using the following formula:

Chl. a (µg mL^{-1}) = $12.25A_{663.2} - 2.79A_{646.8}$

Chl. b (µg mL^{-1}) = $21.50A_{646.8} - 5.1A_{663.2}$

Total Chl (µg mL^{-1}) = $7.15A_{663.2} + 18.71A_{646.8}$

Cart (µg mL^{-1}) = ($1000A_{470}$ - 1.8 Chl. a - 85.02 Chl. b) / 198

2.3.3. Quality parameters

The quality criteria of green onion were evaluated, including the length of the pseudo-stems (cm), which consisted of leaf sheaths and young leaves as the preferred edible organs, total soluble solids (TSS), pungency (pyruvic acid), and nitrate content. TSS (%) was measured with a refractometer (Carl Zeiss, Jena, Germany) calibrated with distilled water. The quantification of pyruvic acid content (μM g^{-1} FW) was performed by measuring the colour development spectrophotometrically at 515 nm using 2,4-dinitrophenylhydrazine reagent in the presence of NaOH, following the method described by Ketter and Randle (1998) and Anthon and Barrett (2003). Ascorbic acid (mg 100 g^{-1} FW) was measured by titration of plant extracts with 2,6-dichlorophenolindophenol dye (AOAC, 2000).

2.3.4. Nitrate content determination

The nitrate content was estimated according to Cataldo et al. (1975). One hundred mg of dried material was homogenized in distilled water (5 mL) for one hour, followed by 15 min centrifugation at 10,000× g. To 0.1 mL of supernatant, 0.4 mL of salicylic acid (5% in concentrated sulfuric acid) was added and mixed. After 20 min, 9.5 mL of NaOH 2N were combined with the solution. The absorbance was detected at 410 nm in the spectrophotometer against a blank. KNO_3 dilutions were prepared to create the standard curve. The content of nitrate was expressed as mg kg^{-1} dry weight (DW).

2.3.5. Total phenolics determination

Dried green onion powder (100 mg) was soaked in 5 mL of 95% ethanol and agitated for 5 h. The tubes containing the mixture were centrifuged for 10 min. Total phenolic content (TPC) was assayed using the Folin-Ciocalteu reagent (Singleton and Rossi, 1965). The supernatant (1 mL) was mixed with 95% ethanol (1 mL), distilled water (5 mL), and 50% Folin-Ciocalteu (0.5 mL). After 5 min, 5% Na_2CO_3 (1 mL) was added and thoroughly mixed. After incubation for an hour at 25 °C, the absorbance was read in the spectrophotometer against a blank at 725 nm. Gallic acid dilutions were measured to create the calibration curve. TPC was calculated as mg gallic acid equivalents (GAE) g^{-1} DW of green onion.

2.3.6. Total flavonoids determination

Green onion powder (100 mg DW) was combined with 5 mL of ethanol (95%). The mixture was shaken for 4 h, followed by a 24-h incubation period at 25 °C. After centrifuging the mixture for 10 min, total flavonoid content (TFC) was estimated by using the aluminum chloride ($AlCl_3$) colorimetric protocol (Chang et al., 2002). The supernatant (0.5 mL) was combined with ethanol 95% (1.5 mL), 10% $AlCl_3$ (0.1 mL), 1 M potassium acetate (0.1 mL), and distilled water (2.8 mL). After incubation (30 min at 25 °C), the absorption was detected in the spectrophotometer at 415 nm against a blank. The standard curve was constructed from quercetin dilutions. The values of TFC were displayed as mg quercetin equivalents (QE) g^{-1} DW of green onion.

2.3.7. Determination of total antioxidant capacity

The total antioxidant capacity (TAC) was assessed using the phosphor-molybdenum method (Prieto et al., 1999). The standard phosphomolybdate reagent was prepared by combining equal volumes of 4 mM ammonium molybdate, 28 mM sodium phosphate, and 0.6 M H_2SO_4. A volume of 3 mL of reagent solution was added to 0.3 mL of extract. The tubes holding the reaction solution were subjected to thermal treatment by immersion in a water bath set at 95 °C for 90 min. After cooling to room temperature, the absorption was determined at 695 nm in the spectrophotometer against a blank. The standard curve was established using different dilutions of ascorbic acid. TAC was expressed as mg ascorbic acid equivalents (AsAE) g^{-1} DW.

2.3.8. Determination of 50% inhibitory concentration (IC_{50})

The DPPH (2,2-diphenyl-1-picrylhydrazyl) assay was also used to measure the antioxidant activity of green onion extracts (Elateeq et al., 2022). One mL of serial dilutions of the green onion extracts was combined with 2 mL of a 150 µM DPPH ethanol solution. The mixture was agitated and placed in the dark for 30 min at 25 °C. The measurement of the absorbance was read at 517 nm using a spectrophotometer. The percentage of DPPH radical scavenging activity was obtained from the following formula:

DPPH activity (%) = $[A_{control} - A_{sample})/A_{control}] \times 100$

where A_{sample} represents the absorption of green onion extract mixed with DPPH solution, and $A_{control}$ stands for the absorption of the DPPH with 1 mL of ethanol 95% instead of the sample. The 50% inhibitory concentration (IC_{50}) of green onion was reported as the amount of sample needed to reduce the initial DPPH concentration by 50%. It was calculated by plotting the DPPH scavenging activity (%) versus sample concentrations.

2.4. The statistical analysis

The statistical analysis was carried out using the analysis of variance (ANOVA) method at $P \leq 0.05$ according to Snedecor and Cochran (1980). The statistical analysis was conducted by CoStat software, version 6.4 (CoHort Software, Monterey, CA, USA). Pearson's correlation and principal component analyses (PCA) were performed with OriginPro 2021 (OriginLab Corporation, Northampton, MA, USA).

3. Results

3.1. Growth parameters

Data recorded from the growing seasons (2021/2022 and 2022/2023) showed that the application of foliar spraying of biostimulants (MLE and YE) significantly affected the growth traits of green onion plants (Table 2). In the first season (2021/2022), YE3 (YE at 3 g L^{-1}) resulted in the highest significant value of plant height (71.40 cm), while no statistical differences were observed between R-NPK and the rest treatments (Table 2). However, in the second growing year, 2022/2023, both biostimulants (MLE and YE) were significantly similar to R-NPK except for low dose of MLE (MLE1; 2%), which recorded the lowest significant height compared to R-NPK (50.25 and 58.90 cm, respectively). The application of MLE and YE did not significantly enhance the leaf number compared to the R-NPK treatment (Table 2). In both seasons, the average number of green onion leaves ranged from 4.00 to 5.67 leaf $plant^{-1}$.

The FW of the green onion plant (bulb + pseudo-stem + leaf) was increased by increasing the dose of both biostimulants (Table 2). During the first season, the highest significant ($P \leq 0.05$) value of FW (36.09 and 35.31 g $plant^{-1}$) was recorded for YE3 and R-NPK treatments, respectively, followed by YE2 and MLE3 (32.37 and 29.17 g $plant^{-1}$, respectively). In the second season, significantly equal FW values were recorded for the R-NPK, MLE3, and YE3 (29.40, 27.66, and 28.93 g $plant^{-1}$, respectively). Moreover, there is no significant statistical variance between the biostimulants treatments at the lowest or moderate levels for FW values during the second season (Table 2). The dry matter percentage was also affected in response to MLE or YE, and the significance level varied according to the applied concentrations (Table 2).

The highest significant dry matter was noticed for green onion plants that received R-NPK (21.11% and 24.66%) and Y3 (20.96% and 22.22%), followed by MLE3 (19.49% and 21.65%) in both seasons, respectively. No significant variation was reported between the other applications.

Table 2. Effect of moringa leaf extract and yeast extract on growth and yield parameters (plant height, leaf number, fresh weight, dry matter, and yield) of green onion during seasons 2021/2022 and 2022/2023.

Treatments	Plant height (cm)		Leaf number plant^{-1}		Fresh weight (g plant^{-1})		Dry matter (%)		Yield (ton ha^{-1})	
	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
R-NPK	57.63bc	58.90a	5.00ab	4.67ab	35.31ab	29.40a	21.11a	24.66a	22.83ab	19.01a
MLE1	55.80c	50.25b	5.00ab	4.67ab	21.90e	19.54b	16.13d	19.01c	14.16e	12.63b
MLE2	62.50b	60.20a	4.67b	4.00b	27.43d	22.57b	17.82bcd	19.16c	17.74d	14.59b
MLE3	62.73b	60.47a	5.33ab	5.00ab	29.17cd	27.66a	19.49ab	21.65b	18.86cd	17.89a
YE1	62.60b	57.97a	5.33ab	4.67ab	25.77d	20.57b	17.09cd	20.09bc	16.66d	13.30b
YE2	63.60b	61.63a	5.33ab	5.33a	32.37bc	23.26b	18.60bc	20.94bc	20.93bc	15.04b
YE3	71.40a	62.63a	5.67a	5.67a	36.09a	28.93a	20.96a	22.22ab	23.34a	18.71a
F. Test	**	**	ns	ns	***	***	***	**	***	***

R-NPK: recommended dose of NPK chemical fertilizers (positive control). MLE1, MLE2, and MLE3 are moringa leaf extract at 2%, 4%, and 6%, respectively. YE1, YE2, and YE3 are yeast extract at 1, 2, and 3 g L^{-1}, respectively. Values followed by different letters in the same column are significantly different ($P≤0.05$). *: significance; ns: not significant.

3.2. Content of nitrogen, phosphorus, and potassium

Overall, the content of N, P, and K was higher in the second season than in the first season. The nutrient content was increased in the green onion tissues upon spraying YE compared to MLE. R-NPK- and YE1-treated plants accumulated the highest significant content of N; 2.17% and 2.02% in the first season, and 2.32% and 2.22% in the second season, respectively. This was significantly followed by YE2 and YE3 treatments (Figure 2a). Similarly, R-NPK and YE were superior to MLE in increasing the content of P and K (Figure 2b and c). In the first season, the same level of significance was recorded for P and K values between R-NPK, YE1, YE2, and YE3 treatments: 0.27%, 0.26%, 0.25%, and 0.24%, respectively, for P, while it was 1.59%, 1.62%, 1.61%, and 1.63%, respectively, for K. The lowest significant content of P and K was found for MLE applications.

3.3. Photosynthetic pigments

No statistical variations were detected between chlorophyll-a values for green onion plants that received YE and MLE compared to R-NPK plants in both seasons (Figure 3a). In the 1st season, chlorophyll-b reached the highest significant contents (1.60 to 1.80 mg g^{-1} FW) in the treatments of R-NPK, MLE3, YE1, YE2, and YE3 without significant differences between them, while the lowest significant value was found with MLE2 (1.36 mg g^{-1} FW). However, no significant difference was observed between the contents of chlorophyll-b for different treatments and R-NPK in the second season (Figure 3b). In the first season, the topmost significant contents of total chlorophyll (4.81, 4.69, 4.67, 4.60, and 4.41 mg g^{-1} FW) were recorded for YE3, YE2, MLE3, YE1, and R-NPK, respectively (Figure 3c). In contrast, in the second growing season, no significant variation was observed between total chlorophyll contents for different treatments. The results showed that applying YE and MLE did not significantly affect the carotenoids pigment when compared with R-NPK-treated plants during both seasons (Figure 3d).

3.4. Green onion yield

The yield of green onion increased by increasing the concentration of YE and MLE (Table 2). During the first season, the yield was significantly ($P \leq 0.05$) maximized by spraying YE3 and adding R-NPK (23.34 and 22.83, respectively) followed by YE2 (20.93 ton ha^{-1}). The lowest significant yield was found for the low concentration of MLE (MLE1). Data from the second season showed that higher concentrations of MLE (MLE3) and YE (YE3) significantly enhanced green onion yields, which were similar to R-NPK (17.89, 18.71, and 19.01 ton ha^{-1}, respectively). The rest of the treatments did not show significant differences between them (Table 2).

3.5. Quality parameters

The results in Table 3 show that different treatments significantly affected the quality parameters of green onion. The pseudo-stem length values ranged from 10.43 to 14.23 cm during the two growing seasons. The statistical analysis showed the same significance of the pseudo-stem length values in the R-NPK and both biostimulants treatments in the second season, except for the low level of MLE and YE, which recorded the lowest length of the pseudo-stem. However, no significant differences were observed between lengths of pseudo-stem of R-NPK and different doses of biostimulants in the first year.

Compared to R-NPK treatment, plants sprayed with YE and MLE had moreTSS and pyruvic acid content in both growing seasons (Table 3). The content of these two quality characteristics in green onion increased with increasing concentration of the MLE and YE spray solution. The TSS reached the highest significant percentage with applying MLE3, YE2, and YE3; 1.48–1.73-fold higher than R-NPK in the first and second seasons. Likewise, MLE3 and YE3 recorded the highest significant values of pyruvic acid: 11.04 and 10.87 µM g^{-1} FW in the 1^{st} season, 12.07 and 12.66 µM g^{-1} FW in the 2^{nd} season, respectively, followed by YE2. These treatments represent a 1.38–1.74-fold increase compared to R-NPK.

Regarding nitrate content, the biostimulants (MLE and YE) positively reduced the nitrate content in green onion (Table 3). YE was more effective than MLE in reducing the nitrate content. The lowest significant content of nitrate, 184.87 and 201.37 mg kg^{-1} DW, was reported by YE2 treatment in both seasons, respectively. This decrement represents 34.73% and 38.00% compared to R-NPK. The nitrate content in R-NPK plants accounted for the highest significant value of nitrate accumulation during the two growing years, followed by MLE1 and MLE2.

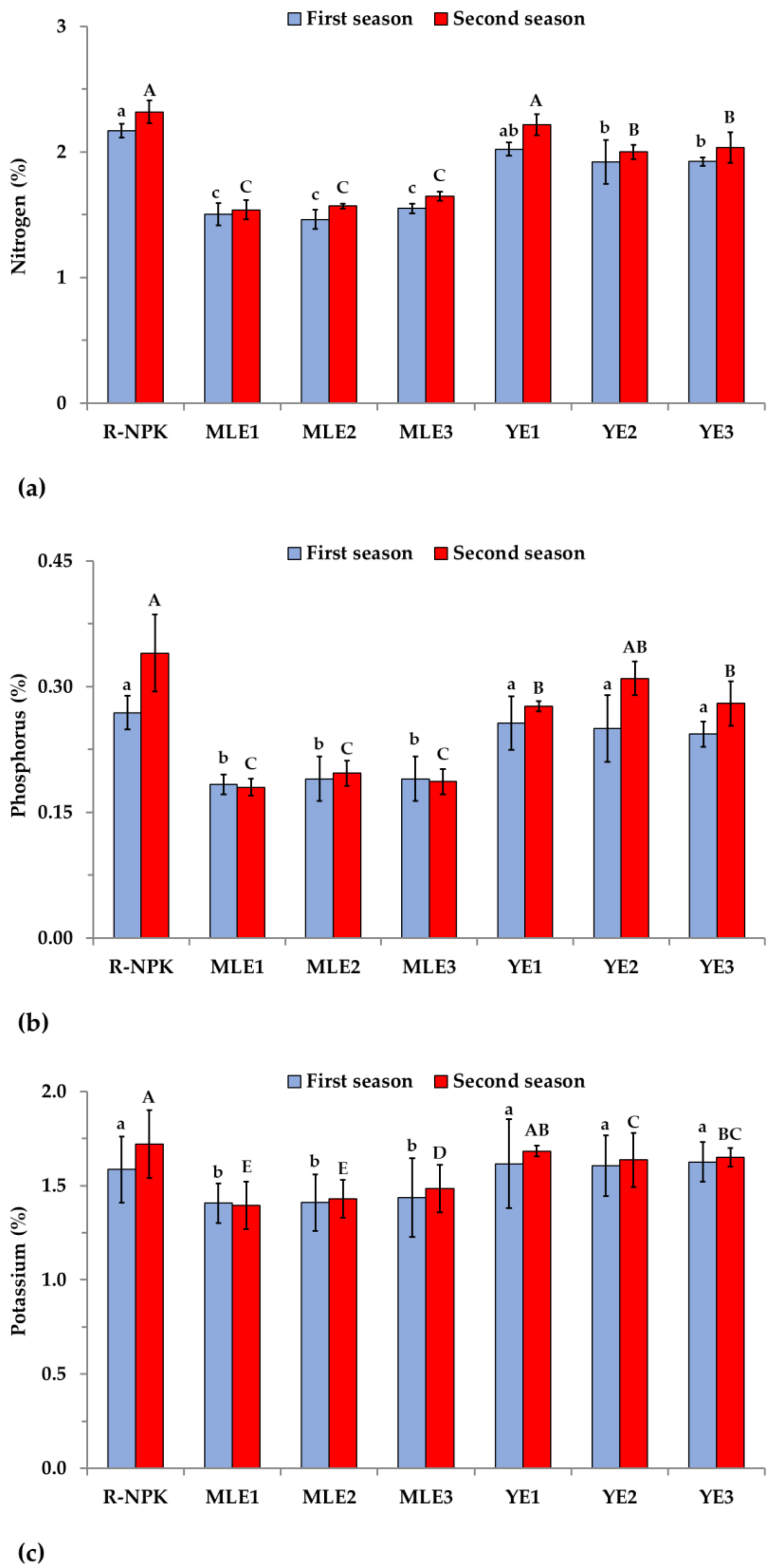

Fig. 2. Effect of moringa leaf extract and yeast extract on nitrogen (a), phosphorus (b), and potassium (c) percentage of green onion during seasons 2021/2022 and 2022/2023. Bars represent ±SD, (n=3). Different letters showed statistical differences (*P*≤0.05) (lowercase and uppercase letters refer to the first and second seasons, respectively). R-NPK: recommended dose of NPK chemical fertilizers (positive control). MLE1, MLE2, and MLE3 are moringa leaf extract at 2%, 4%, and 6%, respectively. YE1, YE2, and YE3 are yeast extract at 1, 2, and 3 g L^{-1}, respectively.

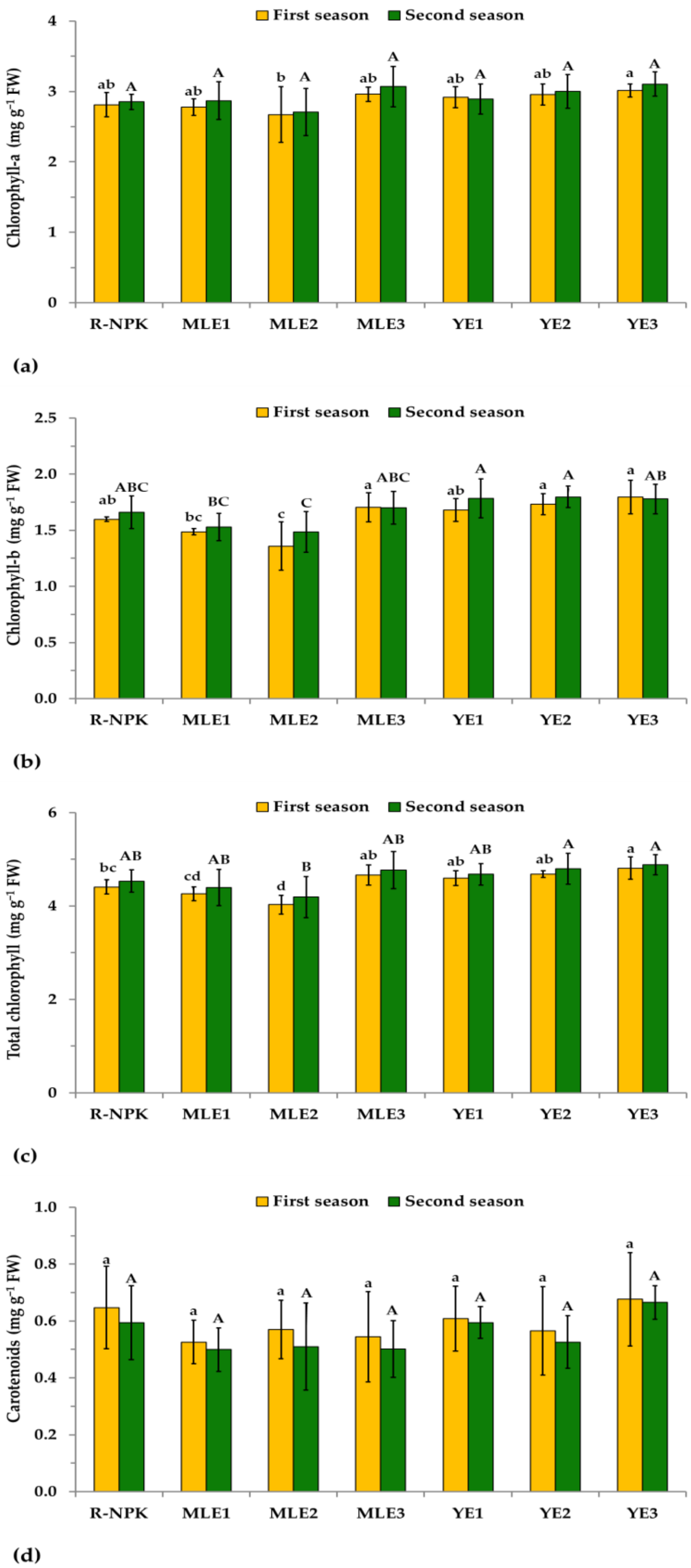

Fig. 3. Effect of moringa leaf extract and yeast extract on photosynthetic pigments [chlorophyll-a (a), chlorophyll-b (b), total chlorophyll (c), and carotenoids (d)] content (mg g^{-1} FW) of green onion during seasons 2021/2022 and 2022/2023. Bars represent ±SD, (n=3). Different letters showed statistical differences ($P \leq 0.05$) (lowercase and uppercase letters refer to the first and second seasons, respectively). R-NPK: recommended dose of NPK chemical fertilizers (positive control). MLE1, MLE2, and MLE3 are moringa leaf extract at 2%, 4%, and 6%, respectively. YE1, YE2, and YE3 are yeast extract at 1, 2, and 3 g L^{-1}, respectively.

Table 3. Effect of moringa leaf extract and yeast extract on quality parameters (pseudo-stem length, and content of TSS, pyruvic acid, and nitrate) of green onion during seasons 2021/2022 and 2022 /2023.

Treatments	Pseudo-stem length (cm)		TSS (%)		Pyruvic acid ($\mu M\ g^{-1}$ FW)		Nitrate ($mg\ kg^{-1}$ DW)	
	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
R-NPK	13.80ab	13.57a	3.64f	4.33d	7.08d	7.28d	532.23a	529.93a
MLE1	13.00abc	10.43c	5.21e	6.02c	8.48bc	8.90c	425.03b	430.45b
MLE2	12.28c	12.21ab	5.65cd	6.16c	9.40bc	9.90bc	419.93b	424.52b
MLE3	13.57abc	12.92a	6.00ab	6.41b	11.04a	12.07a	321.26d	316.18d
YE1	12.62bc	10.83bc	5.40de	6.15c	8.33cd	9.94bc	344.08c	335.18c
YE2	13.62abc	12.41a	5.83bc	6.51ab	9.80ab	10.75b	184.87f	201.37f
YE3	14.23a	13.06a	6.28a	6.70a	10.87a	12.66a	244.96e	270.12e
F. Test	ns	**	***	***	***	***	***	***

R-NPK: recommended dose of NPK chemical fertilizers (positive control). MLE1, MLE2, and MLE3 are moringa leaf extract at 2%, 4%, and 6%, respectively. YE1, YE2, and YE3 are yeast extract at 1, 2, and 3 g L^{-1}, respectively. Values followed by different letters in the same column are significantly different ($P \leq 0.05$). *: significance; ns: not significant.

3.6. Bioactive compounds content

The ascorbic acid content in the green onion ranged from 16.95 to 22.92 mg 100 g^{-1} FW (Table 4). The treatment of MLE2, MLE3, YE2, and YE3 led to a significant increase in ascorbic acid content in the second season compared to R-NPK, while no statistical differences were noticed between all levels of biostimulants and R-NPK in the first year. YE2 and YE3 maximized the content of ascorbic acid in the second season, 1.26 and 1.35-fold increase compared to R-NPK.

The highest significant value of TPC was observed for R-NPK-treated plants (8.59 and 8.68 mg GAE g^{-1} DW) as well as for YE2-sprayed plants (7.75 and 8.42 mg GAE g^{-1} DW) in both growing seasons, respectively (Table 4). MLE2 and YE3 were significantly similar in TPC values. The TFC reached the highest significant value in the treatments of R-NPK, YE2, and YE3 in the 1st season without significant differences between them (4.79, 4.81, and 4.06 mg QE g^{-1} DW, respectively). Moreover, in the second season, these treatments were significantly equal to MLE2 and MLE3 treatments. Applying lower concentrations of MLE and YE decreased the TPC and TFC when compared to R-NPK and other levels during the growing season.

3.7. Antioxidant activity

The statistical analysis revealed a non-significant difference ($P \leq 0.05$) between the antioxidant activities (TAC and IC_{50}) of R-NPK and both biostimulants at moderate and higher concentrations (Table 4). MLE1 and YE1 reduced the TAC by about 41.95% - 56.85% compared with R-NPK in both seasons. R-NPK, medium, and higher levels of both biostimulants showed similar significant responses, with TAC values ranging from 11.15 to 13.51 mg AsAE g^{-1} DW and 11.86 to 13.11 mg AsAE g^{-1} DW in both seasons, respectively. Moreover, the activity of antioxidants was expressed in terms of IC_{50} [the inhibitory concentration (mg mL^{-1}) required to scavenge 50% of free radicals (DPPH)]. The IC_{50} values ranged from 2.30 to 15.44 mg mL^{-1} of the green onion extract in both growing seasons. The highest antioxidant properties with the lowest values of IC_{50} were estimated in R-NPK, MLE2, MLE3, YE2, and YE3 in the 1st and 2nd seasons, with close significant differences ($P \leq 0.05$) between them.

Table 4. Effect of moringa leaf extract and yeast extract on bioactive compounds content (ascorbic acid, total phenolics, and total flavonoids) and antioxidant activity (total antioxidant capacity and IC_{50}) of green onion during seasons 2021/2022 and 2022 /2023.

Treatments	Ascorbic acid (mg 100 g^{-1} FW)		Total phenolics (mg GAE g^{-1} DW)		Total flavonoids (mg QE g^{-1} DW)		Total antioxidant capacity (mg AsAE g^{-1} DW)		IC_{50} (mg mL^{-1})	
	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
R-NPK	17.97ab	16.95d	8.59a	8.68a	4.79a	4.70a	13.51a	12.11a	3.55c	3.22c
MLE1	17.95ab	18.11cd	2.07e	2.53d	2.48d	2.59b	5.83c	6.00b	15.44a	14.22a
MLE2	19.28ab	19.70bc	6.70c	6.60b	3.83b	4.04a	12.56ab	13.11a	2.50c	2.44c
MLE3	19.94ab	20.67b	5.31d	5.40c	3.53bc	3.86a	11.15b	11.88a	2.30c	2.34c
YE1	16.994b	18.71cd	2.73e	2.44d	2.85cd	2.77b	7.19c	7.03b	8.84b	8.33b
YE2	20.26a	21.42ab	7.75ab	8.42a	4.81a	4.64a	12.04ab	11.86a	3.00c	2.96c
YE3	20.92a	22.92a	7.00bc	7.16b	4.06ab	3.98a	11.94ab	12.10a	3.20c	3.15c
F. Test	ns	***	***	***	***	**	***	***	***	***

R-NPK: recommended dose of NPK chemical fertilizers (positive control). MLE1, MLE2, and MLE3 are moringa leaf extract at 2%, 4%, and 6%, respectively. YE1, YE2, and YE3 are yeast extract at 1, 2, and 3 g L^{-1}, respectively. Values followed by different letters in the same column are significantly different ($P \leq 0.05$). *: significance; ns: not significant.

3.8. Pearson's correlations analysis

The Pearson's correlations analysis showed significant correlations (positive or negative) between different attributes of green onion (Figure 4) under R-NPK and different treatments of biostimulants. The average data of the two growing seasons (2021/2022 and 2022/2023) were analyzed. In Figure 4, positive correlations could be observed with red circles, while negative correlations were shown with blue circles among the tested parameters. A positive correlation (P<0.05 and 0.01) was observed between green onion yield and growth parameters, including plant height, leaf number, FW, dry matter, N, P, K, and photosynthetic pigments. Moreover, the green onion yield was positively correlated with quality parameters, including pseudo-stem length and pungency (pyruvic acid). Regarding bioactive compounds, a positive correlation has existed between yield and each of ascorbic acid, TPC (P<0.01), TFC (P<0.01), and TAC (P<0.01). Interestingly, the nitrate content in green onion correlated negatively with growth parameters, yield, P, K, total chlorophyll (P<0.01), pseudo-stem length, TSS (P<0.01), pungency (P<0.01), and bioactive compounds. TPC, TFC, and TAC were positively correlated (P<0.01) with each other as well as with ascorbic acid, nutrients content, growth, yield (P<0.01), and quality attributes (except for TSS and nitrate content).

3.9. Principal component analysis

The principal component analysis (PCA) was performed to evaluate the association between treatments and different attributes in green onion. The PCA showed noticeable variations among growth, yield, quality, and biochemical parameters under different treatments, including R-NPK, MLE1, MLE2, MLE3, YE1, YE2, and YE3 (Figure 5). The data for different traits were displayed in a two-dimensional PCA-produced diagram. The data showed two different variability percentages of the principal component (PC), PC1 and PC2, which represent 43.5% and 23.2%, respectively. The first and second PCs explained 66.7% of the total data variability. Among the studied parameters, TSS and nitrate content are in PC2 but negatively correlated with each other, while the rest of the parameters are in PC1. The nitrate content negatively correlated with most of the studied parameters, while TSS positively correlated with plant height, leaf number, total chlorophyll, pungency, and ascorbic acid. Nutrients content (N, P, and K), dry matter, FW, yield, pseudo-stem length, and bioactive compounds content were positively correlated with each other while negatively correlated with other parameters in the same plot. R-NPK was the treatment mostly related to high yield, nutrients content, and accumulation of bioactive compounds; however, the nitrate content was maximized in R-NPK-treated plants.

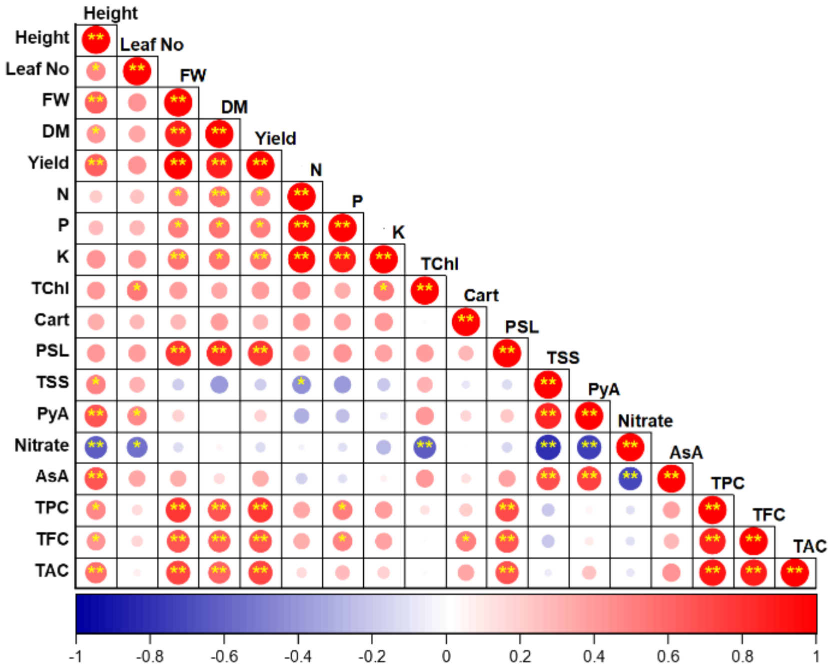

Fig. 4. Pearson's correlation analysis of growth, yield, quality, and biochemical parameters of green onion plants. Height: plant height, Leaf No: leaf number, FW: fresh weight, DM: dry matter, yield: yield ton ha^{-1}, N: nitrogen, P: phosphorus, K: potassium, TChl: total chlorophyll, Cart: carotenoids, PSL: pseudo-stem length, TSS: total soluble solids, PyA: pyruvic acid, AsA: ascorbic acid, TPC: total phenolic content, TFC: total flavonoid content, TAC: total antioxidant capacity. The red colour represents a positive correlation, and the blue colour represents a negative correlation. Significant correlations were shown by * at *P*<0.05 and ** at *P*<0.01.

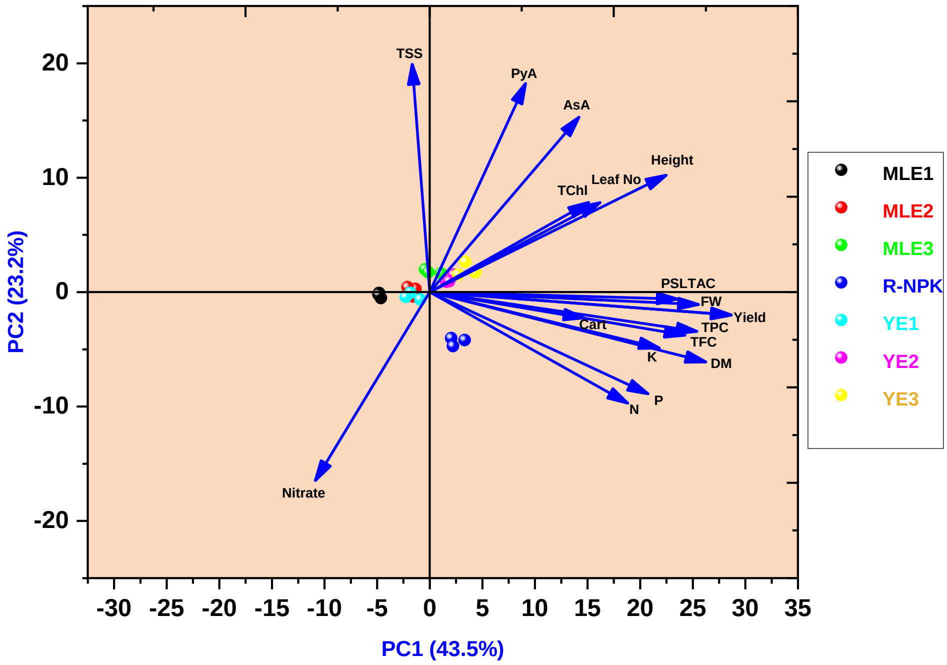

Fig. 5. Principal component analysis (PCA) of the most important growth, yield, quality, and biochemical parameters under R-NPK and biostimulants applications in green onion. Colored dots represent treatments: R-NPK are the recommended dose of NPK chemical fertilizers (positive control); MLE1, MLE2, and MLE3 are moringa leaf extract at 2%, 4%, and 6%, respectively; YE1, YE2, and YE3 are yeast extract at 1, 2, and 3 g L^{-1}, respectively. Height: plant height, Leaf No: leaf number, FW: fresh weight, DM: dry matter, yield: yield ton ha^{-1}, N: nitrogen, P: phosphorus, K: potassium, TChl: total chlorophyll, Cart: carotenoids, PSL: pseudo-stem length, TSS: total soluble solids, PyA: pyruvic acid, AsA: ascorbic acid, TPC: total phenolic content, TFC: total flavonoid content, TAC: total antioxidant capacity.

4. Discussion

Biostimulants have attained considerable interest in recent years as a natural resource innovation aimed at enhancing the productivity and quality of valuable commodities through sustainable and organic farming practices (Farooq et al., 2023; Mehdawe et al., 2023; Yuniati et al., 2023). Investigating the utilization of biostimulants or natural extracts to enhance vegetable production is a current demand as an alternative to synthetic chemical nutrients. In the present investigation, plants in the R-NPK treatment were provided with the recommended dosage of chemical NPK fertilizers. Therefore, MLE and YE treatments with results significantly close to or exceeding those obtained with R-NPK are believed to have a substantial favorable effect on green onion cultivation. The results of the present study indicate that foliar spraying of MLE and YE at higher concentrations on green onion plants resulted in significant improvements in some growth and quality attributes. Moreover, the nitrate content exhibited a notable decrease under different foliar application treatments compared to R-NPK. In general, the use of YE (1, 2, and 3 g L^{-1}) showed greater effectiveness compared to MLE (2%, 4%, and 6%) in improving different traits of green onion. The noticeable differences in some parameters between the first and second growing seasons may be due to the difference in climatic conditions between the two years.

MLE has been applied as a natural growth stimulator in different vegetable and medicinal plants such as garlic (Hegazi et al., 2016), bulb onion (Yaseen and Takacs-Hajos, 2020), fennel (El-Serafy and El-Sheshtawy, 2020), stevia (Sardar et al., 2021), lettuce (Admane et al., 2023), and pepper (Mehdawe et al., 2023). Additionally, MLE exhibits considerable efficacy in mitigating the deleterious effects of various stressors on plants, owing to its rich content of phytohormones, phytonutrients, and antioxidant compounds (Rady and Mohamed, 2015; Latif and Mohamed, 2016; Admane et al., 2023). Likewise, several studies have documented the stimulatory effects of YE on the growth, yield, and accumulation of bioactive metabolites in various species such as bulb onion (Hefzy et al., 2020), garlic (Abdelaal et al., 2021), fennel (El-Serafy et al., 2021), and echinacea (Elshahawy et al., 2022). Moreover, YE has been recognized for its ability to alleviate stress damage and sustain plant growth and productivity in stressful environments (Hefzy et al., 2020; Abdelaal et al., 2021; Gholami et al., 2023).

Applying MLE or YE on green onion plants resulted in significantly greater or equivalent values of plant height and leaf number to those obtained with chemical fertilizers (Table 2). Similar observations were found by Hefzy et al. (2020) on bulb onion. The presence of cytokinin in YE indicates its role in regulating cell division and stem elongation (Barnett et al., 1990). Moreover, YE is a biofertilizer containing several biologically active components, including hormones, carbohydrates, vitamins, and amino acids. These components could influence the fundamental metabolic processes of plants and stimulate their growth (Haider et al., 2021). The application of MLE has been found to have an advantageous effect on plant height in many crops, such as common beans (Latif and Mohamed, 2016), barley (Tahir et al., 2022), and green chili pepper (Yuniati et al., 2023). This increment in plant height could be attributed to the role of MLE, which is enriched in auxin, cytokinin, and gibberellin (Mehdawe et al., 2023). The primary mechanism by which these hormones impact metabolic activities is through the processes of cell division and elongation (Tahir et al., 2022). Moreover, cytokinin is known to stimulate cell division and leaf bud initiation, eventually enhancing leaf number (El-Serafy and El-Sheshtawy, 2020).

Increasing the content of mineral elements (N, P, and K) due to spraying YE is also observed in garlic (Ali, 2017) and fennel (El-Serafy et al., 2021). In other reports, compared with non-fertilized plants, MLE increased the content of N, P, K, and Mg in *Pelargonium graveolens*, which subsequently led to improvements in plant height, branch number, leaf area, essential oil content, and yield features (Ali et al., 2018). However, in the current study, MLE at all levels did not increase the content of mineral elements compared to R-NPK and YE. Moreover, spraying YE generally exhibited a higher accumulation of total chlorophyll than MLE. These may be one of the reasons why YE is superior to MLE in influencing the various tested characteristics (Figure 5).

Applying MLE and YE to green onion plants did not affect carotenoid content (Figure 3). However, its content did not decrease significantly when compared with the recommended chemical fertilizers. A slight increase in total chlorophyll was noticed for MLE3 and all levels of YE, especially in the first season. In other reports, under drought stress, spraying garlic plants with YE significantly increased chlorophyll-a and -b levels compared to stressed unsprayed plants (Abdelaal et al., 2021). Spraying MLE on radish plants increased chlorophyll and carotenoids, while there was no significant increase in turnip green plants (Toscano et al., 2023). MLE and YE stimulate earlier synthesis of cytokinin, preventing premature senescence and leading to larger levels of photosynthetic pigments (Latif and Mohamed, 2016; Haider et al., 2021). Cytokinin in MLE and YE activates isopentenyl transferase, a key enzyme in cytokinin formation, and leads to increased chlorophyll content (Rady and Mohamed, 2015; Abdelaal et al., 2021). Moreover, MLE and YE are high in Fe and Mg, which are essential for chlorophyll production and play a role in the conversion process of proporphyrine to chlorophyllide (Farooq and Koul, 2020).

The leaf extract of *M. oleifera* contains significant amounts of gibberellin, auxin, cytokinin, phenolics, flavonoids, sterols, ascorbates, vitamins, and several mineral elements, including N, P, K, Mg, Zn, Fe, Ca, and essential amino acids (Latif and Mohamed, 2016; El-Serafy and El-Sheshtawy, 2020; Admane et al., 2023; Arif et al., 2023; Yuniati et al., 2023) which makes MLE a potent natural growth promoter. Ascorbic acid and phenolics, two allelochemicals present in MLE, may also influence a plant’s endogenous hormone, enhancing plant growth and yield (Farhat et al., 2023). Comparison of MLE3 (6%) and YE3 (3 g L^{-1}) with R-NPK showed non-significant differences between them regarding green onion yield. Under these treatments, the green onion yield ranged from 17.89 to 23.34 ton ha^{-1}. According to the Agricultural Statistics Bulletin (2019), the average yield of green onion in Egypt is 17.40 ton ha^{-1}. Yaseen and Takacs-Hajos (2020) found that 4% MLE produced bulb onions with higher weight and yield. Moreover, MLE and YE function as nutritional increase factors, which can enhance plant growth and development and the production of many crucial components, such as amino acids, vitamins, enzymes, and essential elements that boost the photosynthetic rate and the metabolism of protein and hormones, thereby enhancing FW and yield (Abdelaal et al., 2021; Arif et al., 2023). A positive correlation (P<0.05 and 0.01) was observed between green onion yield and growth parameters, including plant height, leaf number, FW, dry matter, N, P, K, and photosynthetic pigments. This indicates that these growth parameters significantly affect productivity.

The quality parameters, including TSS, pyruvic acid, and nitrate, exhibited improvements in green onion treated with MLE and YE compared to those received R-NPK (Table 3). Farooq et al. (2023) also noted an enhancement in the quality traits of lettuce cultivars when treated with MLE (2–8%). It was reported that foliar spray of MLE enhanced the formation of phytohormones and increased yield components as well as quality attributes such as ascorbic acid, total soluble sugars, and TSS content in various crops (Latif and Mohamed, 2016; Nasir et al., 2016; Sardar et al., 2021; Toscano et al., 2021). In addition, under salt stress, lettuce plants treated with MLE showed high antioxidant activity including ascorbic acid, CAT, and APX (Shalaby, 2024).

Onion pungency is closely associated with pyruvic acid concentration (Gallina et al., 2012). In the current study, MLE and YE presented a positive influence on the content of pyruvic acid (pungency) in green onion compared to R-NPK. Many factors influence the buildup of organosulfur compounds and pungency degree in onions, including nutrient source, sulfur-based fertilizer, the environment, and cultivar (Yoo et al., 2006; Bolandnazar et al., 2012; Gallina et al., 2012; Wang et al., 2020). However, pungency is not always positively associated with sulfur-based fertilizers in onion, with contradictory findings (Randle, 1992; Randle and Bussard, 1993). Levine et al. (2005) reported that cultivar and growth conditions alter the pungency rate of green onion (*A. fistulosum*), i.e., risng light and CO_2 levels enhance the biosynthesis of aroma precursors and/or alliinase activity (pungency factors).

The accumulation of nitrates in edible parts is an important quality parameter. A high intake of nitrates is linked to an elevated risk of carcinogenic nitrosamine formation in the stomach (Kyriacou et al., 2019). The buildup of nitrates in plants is affected by many factors, such as fertilizer type, elevated levels of fertilizers, plant maturity, nitrate concentration in the soil, stress conditions, and the growing season (Ahmed, 2009; Kapoulas et al., 2017; Papoui et al., 2022; Mostafa et al., 2024). Nitrate content was sensibly reduced in green onion plants sprayed with MLE and YE (Table 3). MLE was reported to be beneficial in decreasing nitrates in radish microgreens (Toscano et al., 2023). The study of Ahmed (2009) showed a decrease in nitrate content in leaves and bulbs of bulb onion after a combination treatment of biofertilizer and mineral fertilizer. In addition, lettuce plants sprayed with MLE (5% and 10%) exhibited higher yield and lower nitrate content (Shalaby, 2024). R-NPK treatment significantly (P≤0.05) boosted the nitrate content in green onion, implying that the application of recommended NPK increased the nitrate content within the plant to a level that exceeded the ability of nitrate reductase, glutamine synthetase, and glutamine synthase to reduce NO_3^- to NH_4^+ and NH_4^+ to amino acids (Andersen et al., 1999), leading to excessive nitrate (NO_3^-) accumulation in plants (Víllora et al., 2002).

Appositive correlation was noticed between green onion yield and quality parameters (pseudo-stem length and pungency). Furthermore, the nitrate content correlated negatively with growth parameters, yield, P, K, total chlorophyll, pseudo-stem length, TSS, pungency. Therefore, these results indicate the possibility of achieving satisfactory yield of green onion with high quality characteristics and lower nitrate content.

Green onions are high in bioactive compounds like ascorbic acid, phenolics, and flavonoids, which contribute to their high nutritional value (Miean and Mohamed, 2001). In response to biostimulants, a remarkable accumulation of ascorbic acid (up to 22.92 mg 100 g^{-1} FW with YE3) was detected in the second season (Table 4). Previous reports on garlic indicated that YE stimulated ascorbic acid accumulation (Ali, 2017; Abdelaal et al., 2021).

The reduction of ascorbic acid at R-NPK treatment may be due to a dilution effect, as observed by Bolandnazar et al. (2012) in their work on bulb onionBolandnazar et al. (2012) observed in their bulb onion work. Phenolics and flavonoids are important classes of secondary metabolites, possessing notable bioactive properties mostly attributed to their antioxidant abilities (Pan et al., 2023; Salam et al., 2023). A significant reduction in TPC and TFC was recorded by applying lower levels of MLE (2%) and YE (1 g L^{-1}). This may be correlated with a deficiency in providing essential nutrients necessary for growth, development, and biochemical synthesis and accumulation. Plants that received R-NPK and a medium dose of YE (2 g L^{-1}) achieved the highest values of TPC without significant differences between the two treatments. Except for low dosages of MLE and YE, the remaining treatments and R-NPK plants recorded similar TFC accumulation. Similar findings were observed for TAC.

According to Sanchez-Sampedro et al. (2005), YE stimulated the biosynthesis of jasmonic acid, exerting an influence on the production of bioactive metabolites. Also, YE has been shown to increase the activity of catalase, peroxidase, and polyphenol oxidase enzymes, leading to bioactive constituents' accumulation (Elshahawy et al., 2022). Previous reports indicated that MLE boosted the flavonoid and phenolic content of stevia (Sardar et al., 2021) and sunflower (Farhat et al., 2023). Moreover, applying MLE increased total antioxidant content and TPC as well as catalase and superoxide dismutase enzyme activity in mandarin fruit (Nasir et al., 2016). In our study, a strong correlation was observed between TAC and each of TPC and TFC (Figure 4). A positive correlation between TPC and DPPH scavenging activity was also found in the study of Toscano et al. (2023) on radish and turnip microgreens. TPC and antioxidant activity increased in lettuce plants treated with MLE (Admane et al., 2023). Farhat et al. (2023) mentioned that 10% MLE improves the antioxidant scavenging capacity of salt-stressed sunflower genotypes by increasing carotenoids, proline, total soluble sugars, TPC, and TAC.

Plants treated with R-NPK and medium and higher levels of biostimulants had the most potent antioxidant capabilities with the lowest values of IC_{50}. The lower IC_{50} value indicates a high scavenging capacity, while a higher IC_{50} value indicates limited antioxidant activity. In radish, TPC and antioxidant activity (DPPH) were elevated by applying MLE (Toscano et al., 2023). It has been claimed that biostimulants may cause a stressful event by inducing the plant response, including the antioxidant system, resulting in an increase in the production of antioxidant molecules (Di Mola et al., 2019). On the other hand, the application of biostimulants, including MLE and YE, can alter both primary and secondary metabolism, resulting in an increase in bioactive compounds content (Admane et al., 2023). These results show that biostimulants' efficacy in boosting bioactive compounds accumulation and antioxidant activity is not unique; it varies substantially depending on the type of biostimulants and the plant species.

PCA allows the differences between treatments to be summarized, confirming the individual results obtained on individual traits. R-NPK was the treatment mostly related to high yield and high accumulation of bioactive constituents. However, the nitrate content was maximized in R-NPK-treated plants. Interestingly, application of MLE3, YE2, and YE3 recorded values of yield, dry matter, pseudo-stem length, TFC, TAC, and IC_{50} that were significantly equal or close to those observed for the chemical fertilizer (R-NPK), while these treatments were significantly better than chemical fertilizer in terms of TSS, pungency, and ascorbic acid content. They also had less content of nitrate.

5. Conclusions

This study presents empirical evidence supporting the beneficial effects of biostimulants, such as MLE and YE, on the growth, yield, and quality of field-grown green onion. Effective synthetic fertilizer exists; however, potentially advantageous biostimulants may be offered as an environmentally friendly alternative to chemical fertilizer. The presence of nitrate, a detrimental component, was found to increase upon the utilization of R-NPK. The growth, yield, antioxidant molecules, and antioxidant properties of green onion exhibited a better response to foliar application of specific levels of YE, followed by MLE, which were found to be equivalent to the response observed with R-NPK fertilizer. Furthermore, applying biostimulants increased the content of the desirable components, e.g., ascorbic acid, TSS, and pungency, while also reducing the nitrate content compared to chemical fertilizers (R-NPK). MLE at 6% and YE at 3 g L^{-1} were more effective in enhancing most traits observed. Therefore, they are considered appropriate treatments to maximize the potential application of both biostimulants in green onion production. In general, the application of YE and MLE for enhancing green onion yield has multiple advantages due to the non-toxic, cost-effective, easily prepared, and environmentally sustainable nature of these extracts. Hence, it can be inferred that using biostimulants, specifically yeast extract (YE) and moringa leaf extract (MLE), in green onion cultivation, along with the avoidance of synthetic chemical fertilizers, ensures satisfactory productivity with acceptable market quality, sufficient nutraceuticals, and a minimal presence of nitrate.

6. Author contribution: Conceptualization, A.H.A., A.H.M.A.E.-W., A.A.E. and T.A.M.A.B.; mehodology, A.H.A., A.H.M.A.E.-W., A.A.E., M.M.I.M., M.M.Z., A.S.A.N., S.A.S.H., and R.S.N.; investigation, A.H.A., M.M.I.M. and T.A.M.A.B.; data curation, M.M.I.M., S.U., M.M.Z., A.S.A.N., S.A.S.H., and R.S.N.; original draft preparation, A.H.M.A.E.-W., A.A.E. and S.U.; review and final editing, S.U., A.A.E. and T.A.M.A.B. All authors read and approved the final version of the manuscript.

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