



## Influence of Yeast, Moringa, and Their Combination on Growth, Yield and Fatty Acid of Chia, *Salvia Hispanica*, Plant

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### Abstract

Substitution of chemical fertilizers with biofertilizers is a key component of sustainable agricultural plans in crop production. This field experiment has been conducted during two successive seasons 2018/2019 and 2019/2020 to examine the effect of substitution of mineral NPK fertilizers with biostimulant (yeast and/or moringa extract). Three fertilization treatments have been conducted in this experiment in factorial design. Yeast, moringa extract and their combination have been used compared to control (100% NPK). Treatment with 100% NPK significantly increased growth and yield components of chia plants. Plant height, number of branches, leaf area, spike height, number of spikes, fresh and dry spike weight, seed yield, leaf greenness, and protein content gave highest values under 100% NPK dose, and decreased with biostimulant treatments; yeast and/or moringa extract. In contrast, treatment with yeast and/or moringa extract significantly increased the quality components of chia. Treatment with yeast and/or moringa gave the highest fatty acid content. It is concluded that completely substitution of mineral fertilizer with biostimulants had negative impact on growth and yield components. In contract, the quality significantly improved under biostimulant treatments. The results of this study suggest that partial substitution of mineral fertilization with yeast and/or moringa extract may maintain high yield with high quality.

**Keywords:** *Salvia hispanica*; Chemical fertilizers; Moringa extract; Yeast; Fatty acids.

### Introduction

Chia, *Salvia hispanica* L., is an annual herbaceous plant belonging to the mint family (Lamiaceae or Labiatae). In pre-Columbian times, it was one of the basic foods of several Central American civilizations. Chia seeds together with corn, amaranth and beans were important staple crops for the Aztecs (Ayerza and Coates. 2004). Chia is native to southern Mexico and northern Guatemala; it grows naturally in tropical and subtropical environments. Its cultivation is spreading rapidly from its center of origin America to new in Central areas of cultivation such as Africa, Australia, Europe and North America as the market for chia is profitable and favorable (Bochicchio. *et al.*, 2015). Chia seeds have been investigated and recommended due to their high levels of proteins (15–25%),

fats (30–33%), carbohydrates (26–41%), dietary fiber (18–30%), and ash (4–5%). The moisture content of the seed is approximately 6 to 8% (Ixtaina. *et al.*, 2010; Jiménez. *et al.*, 2013). It is also observed a high amount of vitamins, minerals, antioxidants and polyphenols (Ixtaina. *et al.*, 2008). Furthermore, it is a good source of B vitamins, calcium, phosphorus and potassium (Muñoz. *et al.*, 2013). Chia seeds contain up to 39% of oil, which has the highest known content of  $\alpha$ -linolenic acid (U-3), up to 68%, compared to other natural sources (Capitani. *et al.*, 2012; Guiotto. *et al.*, 2013).

The use of chemical fertilizers has a great importance for the world's food production as it works as a fast food for plants causing them

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DOI: <http://dx.doi.org/10.21746/aps.2022.11.01.16>

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to grow more rapidly and efficiently. While adverse effects are being noticed due to the excessive and imbalanced use of these synthetic inputs (Bansal. 2017). Moreover, persistent use of conventional chemical fertilizers subverts the soil ecology, disrupt environment, degrade soil fertility and consequently shows harmful effects on human health and contaminates ground water (Daneshmandi and Seyyedi. 2019; Tang. et al., 2019; Ostadi. et al., 2020). Therefore, the agrochemical corporations' strive to replace the chemical fertilizers with biofertilizers in sustainable agricultural plans to achieve applicable crop productiveness (Sharma. et al., 2013). There may be an urgent need to improve agricultural practices to make certain that agricultural production is balanced with environmental sustainability. There is also a whole lot interest in harnessing the blessings of plant-boom-selling microorganisms for sustainable crop production (Vessey. 2003). Microorganisms can sell plant boom and health via more than a few mechanisms, which include imparting vegetation with biologically fixed nitrogen, phytohormones, volatiles, protection compounds, and enzymes (Ryu. et al., 2003; Kuklinsky-Sobral. et al., 2004; Lugtenberg and Kamilova. 2009). Yeast is a price-effective biofertilizer that improves not only plant vitamins but also additionally plant energy during the early boom segment as well as rhizophagy contribution to plant nutrient acquisition (Lonhienne. et al., 2014). Yeast extract, that is environmentally pleasant, nutritious, and convenient to apply, has blessings over not unusual plant increase regulators and soil conditioners. It is well worth verifying the impact of the utility of yeast extract to afforestation in a semiarid soil location (Xi. et al., 2019). Yeast extract has come to be a hot subject matter as it's far secure, nutritious, and handy to use. Yeast extract is rich in powerful components, consisting of low-molecular-weight organic compound, amino acids, nucleotides, peptides, nitrogen, phosphorus, and trace elements. Moreover, yeast extract is freed from chemically synthesized hormones and toxic ingredients (Zhang. et al., 2000; Vieira. et al., 2016).

Natural growth stimulators play an important role in promoting growth and development of crops. For decades, such plant growth stimulators have been used extensively in crop production, whether by soil application or foliar spraying. Plant stimulators have positive effects not only on plant growth and nutrition, but also on abiotic and biotic stress tolerance (du Jardin. 2015). The term biostimulant is relatively new and its use in the scientific community is still nebulous (du Jardin. 2012). One broad definition was introduced by (du Jardin. 2012): "Plant biostimulants are substances or materials, with the exception of nutrients and pesticides, which, when applied to plants, seeds or growing substrates in specific formulations have the capacity to modify physiological processes in plants in a way that provides potential benefits to growth, development, or stress response". There are several studies that have reviewed the role of bio-stimulants in relation to promoting growth and nutrient availability (Colla and Rouphael 2015; du Jardin 2015). In addition to numerous studies that have reviewed broadly defined categories of bio-stimulants such as protein hydrolysates, seaweed extracts, silicon, chitosan, humic acid, and fulvic acid (Van Oosten. et al., 2017). On the other hand, moringa leaves extract positively improve seeds germination, plant growth and yield, nutrient use efficiency, crop quality characters, as well as tolerance to environmental stresses (Abdel-Rahman and Abdel-Kader. 2020; Zulfikar. et al., 2020; Hassanein. et al., 2021).

The main objective of this study was to investigate the effect of yeast and/or moringa extract on growth, yield and fatty acids of chia (*Salvia hispanica* L.) plants.

## Materials and methods

### Plant material and growth conditions

A field experiment was conducted during the two successive seasons of 2018/2019 and 2019/2020 at the Agricultural Experimental Farm of Faculty of Agriculture and Natural Resources, Aswan University, Aswan, Egypt. Seeds of chia (*Salvia hispanica*) were obtained

from National Research Centre, Cairo, Egypt. The experiment was laid out in randomized complete block in factorial design incorporated three replicates. Plot size was 3×1.5 m comprising two rows. In the beginning of November for both seasons, Chia seeds were directly sown at distant of 30 cm in row and 70 cm between rows. Soil samples were obtained from a depth of 30 cm from the used soil surface in this study and some physical and chemical properties have been analysis according to the methods described by Jackson (1973) and Black. *et al.* (1982) as shown in Table (1).

**Table 1:** Physical and chemical analysis of soil

Soil constituents	
Sand	13.66
Silt	46.19
Clay	40.15
Texture grade	silt clay
EC (m.mhos/cm)	0.60
Organic matters %	1.30
Available N %	0.08
Available P% (ppm)	8.00
Available K% (mg/100 g.soil)	0.62

The experiment was conducted to examine yeast (0.6%), moringa extract (10%) and their combination compared to 100% NPK recommended dose as control (Ammonia nitrate 33.5%, superphosphate 19.5% and solo potassium 49.5%). The treatments were applied as soil application three times starting 45 days after planting and repeated at two week interval for both seasons. Drip irrigation system was used for irrigation purpose and all other agricultural practices were performed as recommended during both seasons.

#### Vegetative growth parameters

In the end of March, Plant height (cm), number of lateral branches per plant, leaf area (mm<sup>2</sup>), spike height (cm), spike number per plant, spike fresh and dry weight/plant, seeds yield/plant, and 100 seeds weight have been recorded. Leaf area was measured using a Portable Leaf Area meter AM350 (Eijkelkamp company, EM Giesbeek, The Netherlands).

#### The relative leaf greenness:

Chlorophyll Meter reading (SPAD) was used for measuring the relative leaf greenness (level of chlorophyll) in chia leaves using SPAD chlorophyll meter (SPAD-502plus, Konica Minolta, INC., Osaka, Japan). The SPAD values of chia were measured on the middle part of the leaf blade.

#### Protein content

Nutrient elements were extracted from known weight of dried plant samples (0.2 g) of chia. Nitrogen (N) content was measured using semi-micro kjeldahl method (Black. *et al.*, 1965). Protein content was calculated by multiplying nitrogen content × 6.25.

#### Total fatty acids content

Fatty acid content in oil was determined by AOAC Method 996.01 (Satchithanandam. *et al.*, 2001).

#### Statistical analysis

Data obtained were subjected to statistical analysis using “F” Test (Snedecor and Cochran. 1989). Statistical analysis was carried out using JMP software (version 4.0; SAS institute, Cary, NC, USA). Differences among treatments were detected using Tukey’s Honest Significant Difference (HSD) test.

#### Results

Table (2) represented the analysis of variance (ANOVA) resulted from the application of yeast, moringa extract or their combination on the growth, yield, and chemical characterizations of chia plant in both 2018-2019 and 2019-2020 seasons. Plant height, number of branches, leaf area significantly decreased when plants treated with yeast, moringa or their combination compared to control (100% NPK recommended dose) in the first and second season. There were no significant differences between yeast, moringa and their combination in the studied parameters (Figure 1). In the same line, the spike characters including spike height, number of spikes per plant, and spike fresh and dry weight significantly decreased under treatment with yeast, moringa or their combination compared to the recommended NPK dose in both seasons. Also there were no significant differences between yeast, moringa

and their combination in the spike characters (Figure 2). On the other side, seeds weight per plant significantly decreased with treatment of yeast, moringa or their combination compared to the recommended mineral fertilization in both in the first and second season. In contract, there was no significant differences in the trait of 100 seeds weight when plants treated with yeast, moringa extract or combination in both seasons (Figure 3).

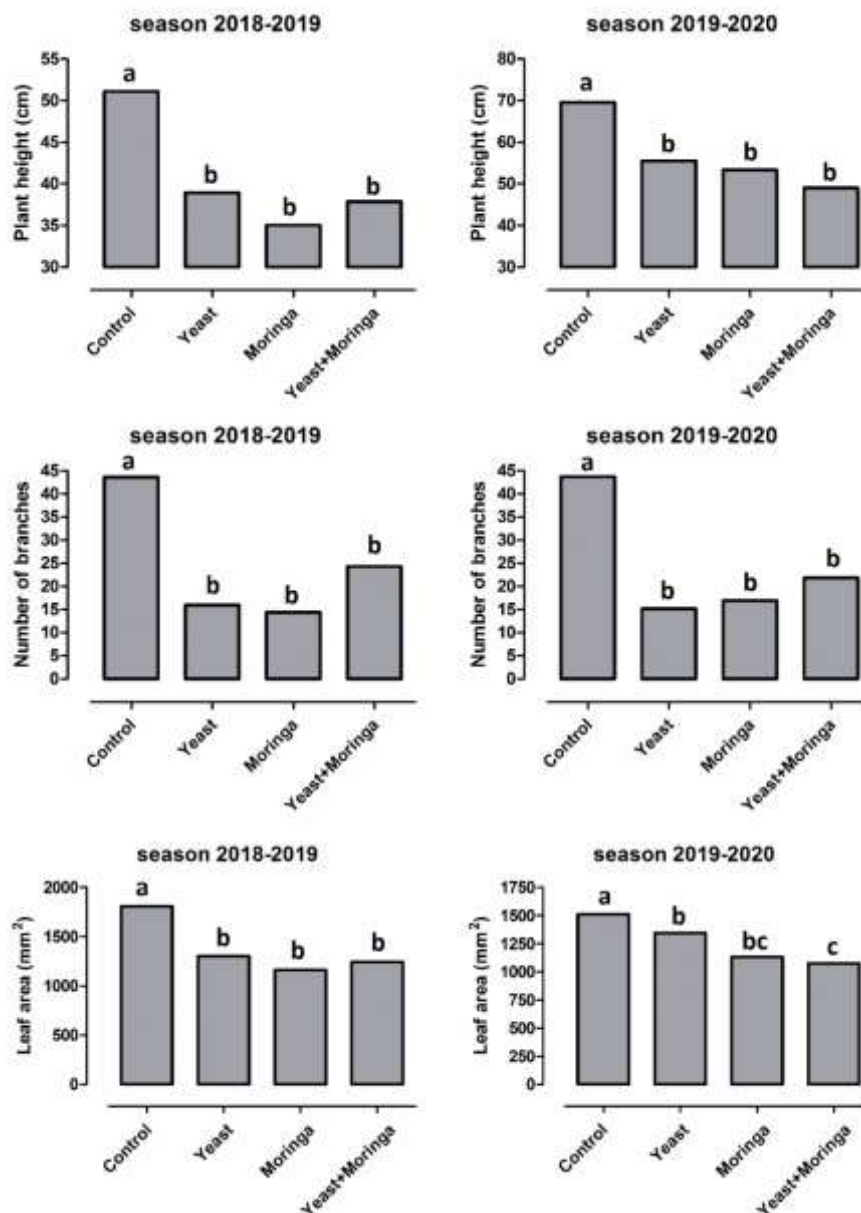
The relative leaf greenness (total chlorophyll content) significantly decreased under zero NPK level treatment in the both seasons. Moringa extract showed much higher relative leaf greenness compared to yeast or their

combination, but not significant (Figure 3). Protein content also was significantly higher under control (100% NPK recommended dose) compared to yeast, moringa, and yeast plus moringa treatments in the first and second seasons (Figure 4). In contrast, the quality of chia plant represented by crude fatty acids content increased under treatment with yeast, moringa extract, and their combination compared to the recommended mineral NPK fertilization in both season. In addition, there was no significant difference between yeast, moringa, and their combination in term of fatty acids content (Figure 4).

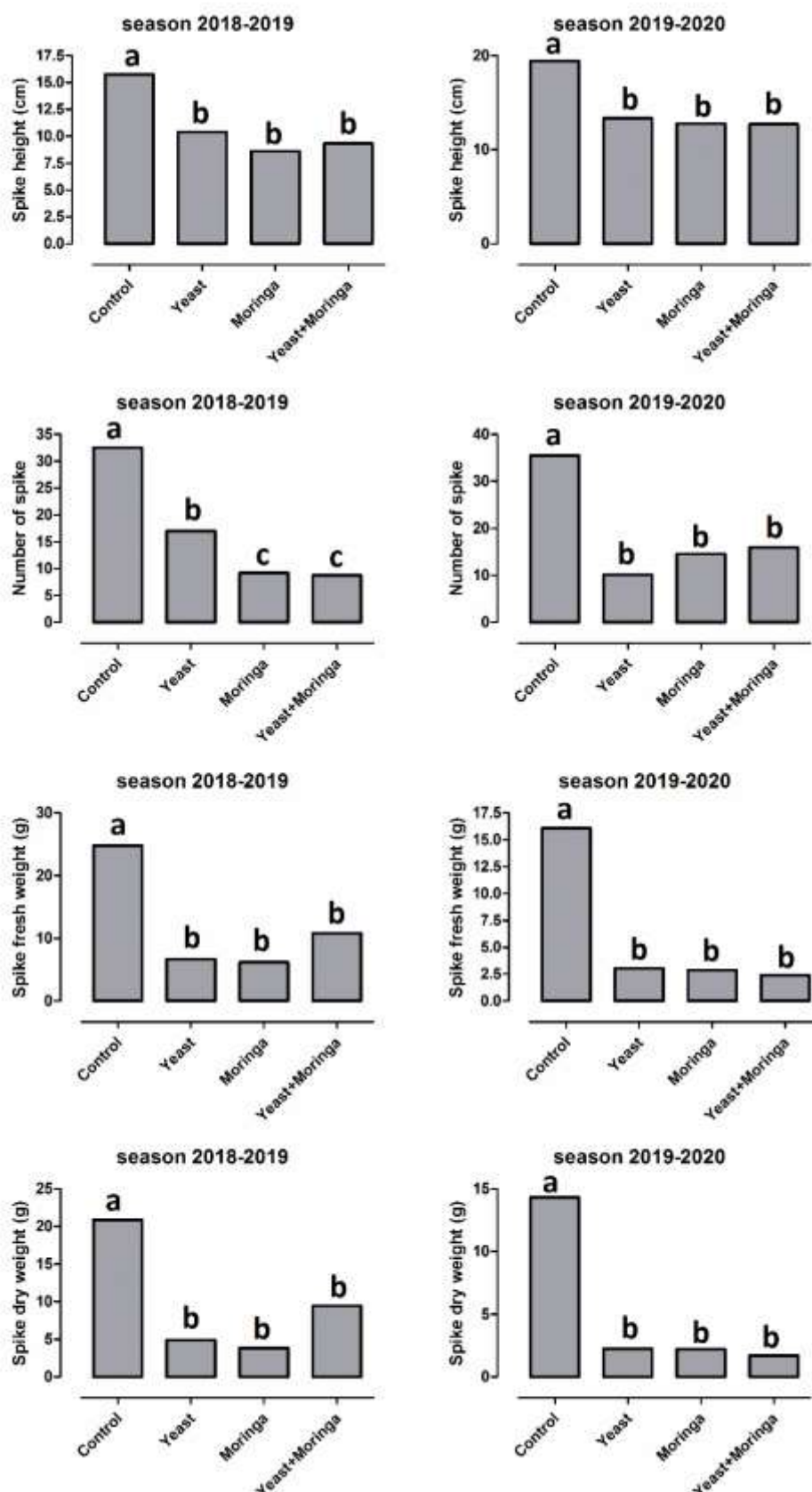
**Table 2:** Effect (*F* value) of yeast, moringa extract and their combination on vegetative and chemical characterizes of chia plant

Variables	Season 2018-2019	Season 2019-2020
Plant height	35.97***	40.55***
Number of branches	29.25***	93.04***
Leaf area	55.92***	18.83***
Spike height	19.03***	11.08**
Number of spikes	74.12***	86.02***
Spike fresh weight/plant	87.31***	360.2***
Spike dry weight/plant	88.73***	438.2***
Seeds weight/plant	67.22***	65.25***
100 seeds weight	1.60ns	1.74ns
Relative leaf greenness	6.58*	10.45**
Protein content	42.01***	19.90***
Crude fatty acids content	6.61*	38.13***

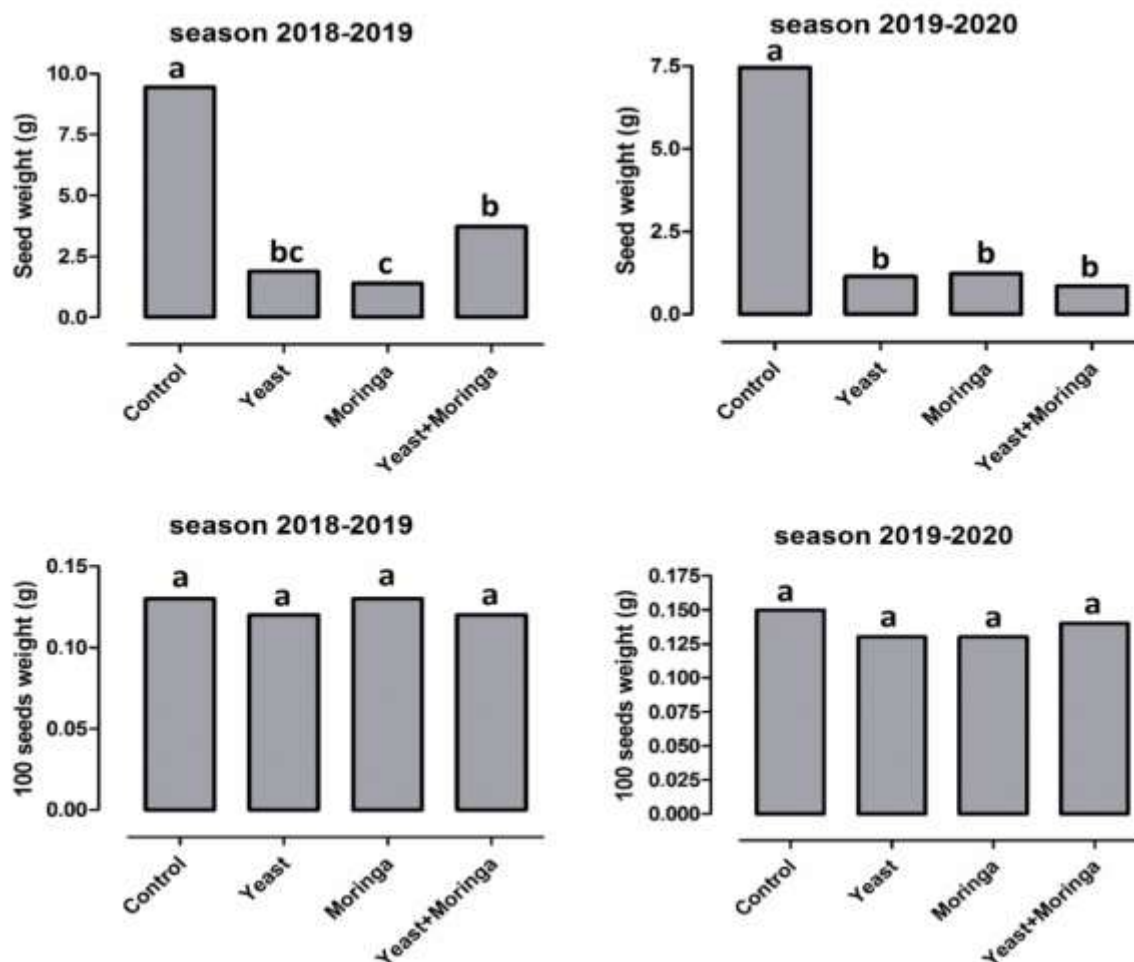
\*, \*\*, \*\*\* represented significance of differences at 0.05, 0.01 and 0.001, respectively. Ns represented non significance.



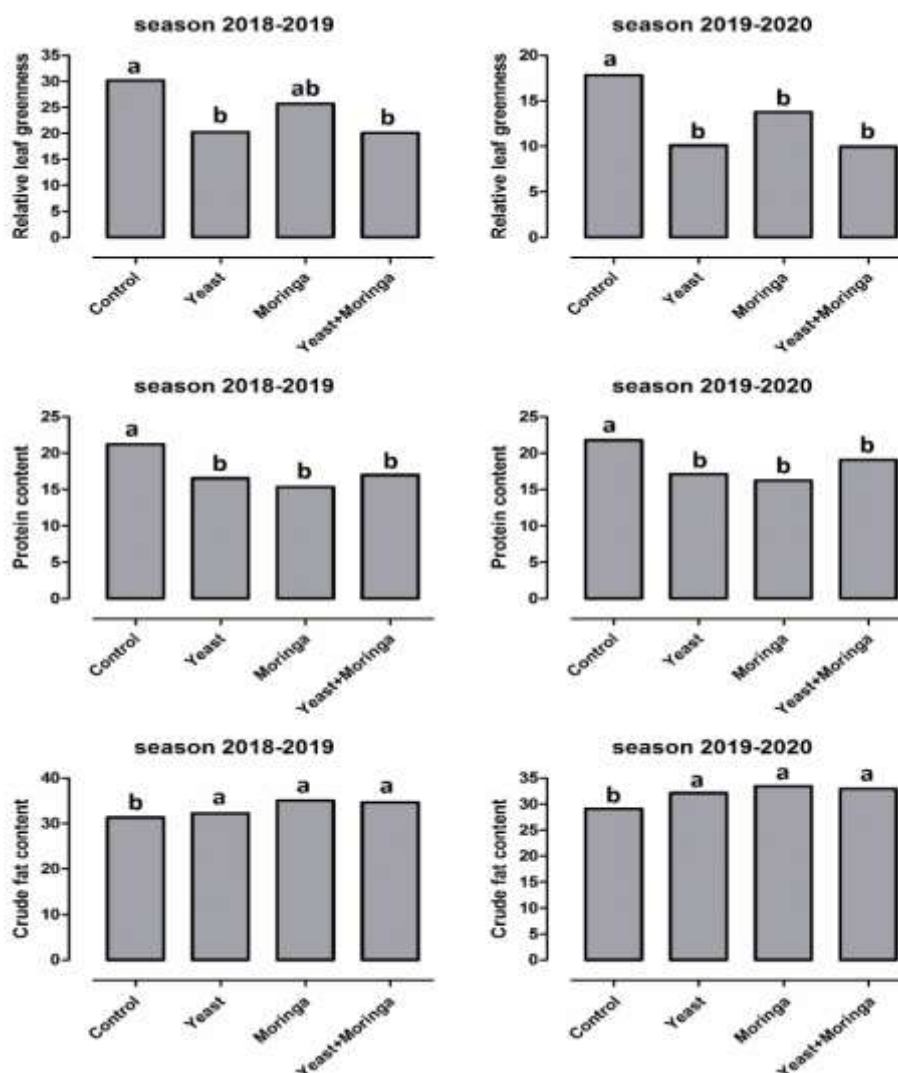
**Figure 1.** Effect of yeast, moringa extract and their combination compared to 100% NPK on plant height, number of branches, and leaf area of chia in the two successive seasons 2018/2019 and 2019/2020.



**Figure 2.** Effect of yeast, moringa extract and their combination compared to 100% NPK on spike height, number of spikes, and spike fresh and dry weight of chia in the two successive seasons 2018/2019 and 2019/2020



**Figure 3.** Effect of yeast, moringa extract and their combination compared to 100% NPK on seeds weight per plant and 100 seeds weight of chia in the two successive seasons 2018/2019 and 2019/2020.



**Figure 4.** Effect of yeast, moringa extract and their combination compared to 100% NPK on relative leaf greenness, protein content and fatty acids content of chia plant in the two successive seasons 2018/2019 and 2019/2020

## Discussion

Growing conditions, such as fertilization, largely determine raw material yield and quality. Nutrition plays a key role in the growth and development of all crop plants. In the case of medicinal plants that synthesize secondary metabolites, nutrients can effectively increase active substances such as oil yield and quality (Aziz. *et al.*, 2010; Jabbari. *et al.*, 2011; Sharafzadeh. *et al.*, 2011; Zheljaskov. *et al.*, 2011). The significance of the mineral micronutrients N-P-K in improving the plant's vegetative development characteristics have been extensively described to examine the major functions in plant growth, development, and quality

improvement. Nitrogen is a major component of amino acids, enzymes, and energy transmission elements like chlorophyll, ADP, and ATP (Bidwell. 1974). Phosphorus also plays a key role in cell division, carbohydrate conversion, photosynthesis, and biological oxidation (Lambers. *et al.*, 2010). Potassium interacts synergistically with carbs when it serves as an osmotic agent (Mengel and Kirkby. 1987). On the other hand, natural plant bio-stimulants are important in sustainable horticulture production systems. Dry yeast extract for example is a bio-substance advised having stimulating, dietary and protecting capabilities. Yeast is a source of various plant development chemicals such

as cytokinins, vitamin B, and microelements (P, K, S, Na, Ca, and Mg). It also is rich with organic compounds such as protein, carbohydrates, DNA, fats as well as auxins, cytokinins, and other endogenous plant hormone (Mahdi. 2010). Khalil and Ismail (2010) noted that yeast could be useful in increasing the availability of nutrients for crops production. Using yeast extract became discovered to growth increase, yield and quality of many vegetable plants (Hussain and Khalaf. 2007; Ahmed. *et al.*, 2011). On this regard, yeasts have been advised to be enriched source of phytohormones (especially cytokinins), nutrients, enzymes, amino acids and minerals (Barnett. *et al.*, 1990). It changed into additionally announced about its stimulatory impact on mobile department and growth, protein and nucleic acid synthesis as well as chlorophyll formation (Castelfranco and Beale. 1983). It is a bio-substance includes many nutrient factors and effective compounds of semi increase regulator compound like auxins, gibberellins and cytokinins.

On the other hand, previous studies have demonstrated that moringa leaf extracts improve seed germination, plant growth, yield, nutrient utilization efficiency, quality characteristics of crops, as well as tolerance to abiotic stresses (Abdel-Rahman and Abdel-Kader. 2020; Zulfiqar. *et al.*, 2020; Hassanein. *et al.*, 2021). Moringa extracts are considered an suitable alternative source of inorganic fertilizers due to their high content of micro and macro mineral nutrients, protein and essential amino acids, which contribute to supplement the nutritional requirements of crops (Yasmeen. *et al.*, 2014). Latif and Mohamed (2016) attributed growth and yield responses when treated with moringa to the presence of growth-promoting hormones in Moringa, as analyzes showed that Moringa extract contains antioxidants, gibberellins, IAA and ABA (Rady and Mohamed. 2015; Azam. *et al.*, 2020).

In this study, fertilization treatments showed significant impacts on the growth and yield parameters; plant height, number of branches,

leaf area, spike height, number of spike, spike fresh and dry weights, and seeds weight, but not for 100 seeds weight (Figure 1-3). In general, all growth parameters decreased when plants treated yeast, moringa extract, or their combination without mineral fertilizer (NPK). In addition, there were no significant differences between yeast and/or moringa extract in their effect on the growth and spike characters. This is consistent with recently previous finding (Mahmoud, *et al.*, 2021). In addition, this does not diminish the importance role of biostimulants, but it expresses the negative impact of the total replacement of mineral fertilizers, especially under newly reclaimed lands.

Analysis of variance also showed that completely substitution of mineral fertilizer with biostimulants significantly affected on chemical and quality of chia plants (Table 2 and Figure 4). Relative chlorophyll content (relative leaf greenness) and protein content decreased significantly when plants treated with zero mineral fertilization N-P-K replaced with biostimulants. This is obviously due to the decreased of nitrogen and other mineral which important in chlorophyll and protein synthesis (Bidwell. 1974; Mengel and Kirkby. 1987; Lambers. *et al.*, 2010). In contrast to the previous results of growth, spike character as well as chemical contents, the quality of chia seeds improved under zero mineral fertilizations. Total crude fatty acids content increased under treatments of yeast and/or moringa extract compared to the recommended mineral NPK fertilization (Figure 4). These results are due to richness of yeast and moringa with organic compounds and hormone-like contents (Barnett. *et al.*, 1990; Mahdi. 2010; Khalil and Ismail. 2010; Abdel-Rahman and Abdel-Kader. 2020; Zulfiqar. *et al.*, 2020; Hassanein. *et al.*, 2021).

### Conclusion

It is concluded that completely substitution of mineral fertilizer with biostimulants had significantly negative impact on growth and yield components. In contract, the quality significantly improved under biostimulant treatments. The results of this study suggest partial substitution of mineral fertilization

with yeast and/or moringa extract to maintain higher yield as possible with higher quality.

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**Source of support:** Nil; **Conflict of interest:** Nil.

**Cite this article as:**

Esmail, S.M., Saber, F. H., Sabri, S. and Wagdi, S.S. "Influence of Yeast, *Moringa*, and Their Combination on Growth, Yield and Fatty Acid of Chia, *Salvia Hispanica*, Plant." *Annals of Plant Sciences*.11.1 (2022): pp. 4679-4691

DOI: <http://dx.doi.org/10.21746/aps.2022.11.1.16>