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Effect of foliar application by growth bio-stimulants on the growth and productivity of lettuce plants under salinity stress

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ABSTRACT

Two greenhouse experiments were conducted during 2019/2020 and 2020/2021 at the Agriculture Test Station, Faculty of Agriculture, Fayoum University, Egypt, to study effect of plant growth promoters on morphological characteristics, membrane permeability, leaf photosynthetic pigments content and productivity and quality of lettuce plants (*Lactuca sativa* L.) to elucidate their potential to reduce the harmful effect of soil salinity (ECe = 7.00 ± 0.20 dS m^{-1}). Treatments comprised with twelve of plant growth promoters; palm pollen grains (100 and 200 mg L^{-1}), corn grain extract (100 and 200 mg L^{-1}), soybean seed extract (100 and 200 mg L^{-1}), mannitol (1500 and 3000 mg L^{-1}), sorbitol (1500 and 3000mg L^{-1}) and calcium phosphate (800 and 1000 $Ca_3(PO_4)_2$ mg L^{-1}), as well as, tap water. Three foliar applications of the thirteen treatments were made to run-off.; 25, 40 and 55 days after transplanting. The experimental layout was a randomized complete blocks design with five replications. All the foliar application of the twelve treatments (plant extracts, mannitol, sorbitol and calcium phosphate) were more effectively, with substantially higher mean values reported of morphological characteristics, membrane permeability and leaf photosynthetic pigments content comparing to control. Enhancement of the plant physio-biochemical components resulted from this., which reflected on improve productivity and quality of lettuce plants (cv. Big Bell) under the salinized soil stress of the Fayoum Governorate and other comparable areas. In general, foliar application of soybean seed extract at concentration of 200 mg L^{-1} or palm pollen grains extract at concentration of 100 mg L^{-1} were superior and particularly increased mean values were recorded for data recorded comparing to other treatments.

Keywords: *Lactuca sativa* L., Mannitol, Sorbitol, Leaf photosynthetic pigments, Yield and Quality.

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1. INTRODUCTION

Lettuce, a yearly leafy vegetable (*Lactuca sativa* L.) in the Astaraceae family, is the most widely grown salad crop worldwide **(Funk *et al*. 2005).** Regarding nutritious value, lettuce comes in at number 26th among fruits and vegetables, and 4th when it comes to intake. The outer leaves, which are a darker green color, have the most nutrients **(FAO, 2009).**

One of the main vegetable crops grown in Egypt is lettuce. It is widely grown throughout the majority of Egypt's various environments **(Lindqvist, 1960).**

n Egypt, lettuce is frequently grown on recently reclaimed land. However, salinity affects the majority of recently recovered soils. One of the main issues with agriculture, especially in arid and semiarid areas, is saline soil, which inhibits plant development and productivity **(Seday *et al*., 2014; Bargaz *et al*., 2016; Rady *et al*., 2016; Hemida *et al*., 2017).** Plant extracts are rich sources of bioactive stimuli and using them is one of the most important strategies according to several studies **(Desoky *et al*., 2018 and Rehman *et al*., 2018)**. Plant extracts are an amazing environmentally friendly creation that enhances tolerance to a variety of abiotic stresses and improves blooming, plant growth, fruit development, crop output, and nutrient utilization efficiency because of the biostimulants they contain **(Desoky *et al*., 2021 and Yaseen and Takacs-Hajos, 2022).** Maize grain embryos extract (MEE) is a biostimulant due to its abundance in cytokinins (CKs), particularly zeatin-type cytokinin (Z-CK), auxins, notably indole-3-acetic acid, gibberellins, antioxidants, and vital nutrients to improve morpho-physio-biochemical traits for promote plant tolerance regarding unfavorable conditions **(Rehman *et al*., 2018).**

Because salinized soil is thought to pose a risk to agricultural output. The primary aim of this study was how to lessen the negative impacts of salt stress on the quality, productivity, and growth of lettuce plants produced in two locations with varying salinity levels (ECe = 7.00 ± 0.20 dS m-1) using foliar application of plant extracts (date palm pollen grains, maize and soybean seeds) and mannitol, sorbitol and calcium phosphate to elucidate their potential to modulate plant responses to saline soil stress.

2. MATRIALS AND METHODS

two greenhouse experiments were conducted during two winter seasons of 2019/2020 and 2020/2021 at the Agriculture Test Station, Faculty of Agriculture, Fayoum University, Egypt (29º 17'N; 30º 53'E).

2.1. Sources of search materials used

2.1.1. Preparation of maize, soybean date palm pollen grains extracts

The extracts was prepared from maize, soybean date palm pollen grains using the procedure reported by **Rady *et al*. (2019)** Chemical characteristics of maize, soybean date palm pollen grains extracts, which were ascertained and recognized by GC/MS in a dedicated laboratory at the National Research Center, are shown in **Table 1**.

Table 1. Chemical chracteristics of the tested maize, soybean and date palm grains extract on a dry weight basis identified by GC/MS during the seasons of 2019/2020 and 2020/2021

Properties	Maize	Soybean	Date palm pollens
Osmoprotectants (mg g^{-1} dry weight)			
Soluble sugars	70.4	9.05	60.50
Proline	4.89	2.37	2.80
Mineral nutrients (mg g^{-1} dry weight)			
Nitrogen	25.5	56.0	54.1
Phosphorus	3.16	7.04	6.60
Potassium	27.1	17.97	17.5
Magnesium	2.52	2.80	3.18
Calcium	3.27	2.70	5.60
Iron	1.22	0.15	2.41
Manganese	0.76	0.25	2.84
Zinc	0.48	0.48	2.81
Copper	0.24	0.16	3.19
Antioxidants and vitamins (mM g^{-1} dry weight)			
Total B-group vitamins	128	53.0	119
Ascorbic acid; vitamin C	1.57	2.93	1.97
Glutathione	0.84	0.92	0.23
Phytohormones (µg g^{-1} dry weight)			
Total indoles	3.20	3.20	9.00
Indole-3-acetic acid	1.73	3.40	4.92
Gibberellic acid	1.89	2.29	6.74
Trans-Zeatin and trans-Zeatin riboside	2.81	1.80	2.33

2.1.2. Mannitol, sorbitol and calcium phosphate monobasic

Mannitol and sorbitol $C_6H_{14}O_6$ produced by Techno Pharmchem, Bahadurarh, Haryana (India) AN ISO 9001: 2008 Certified Company. Hopkin & Williams Ltd., chemical producers (St Davids Court, Union Street, Wolverhampton, West Midlands, United Kingdom, WV1 3JE), produces calcium phosphate monobasic $Ca(H_2PO_4)_2$.

2.2. Physical and chemical properties of soil

Soil samples were taken down to a depth of 25 cm in order to determine certain physical and chemical characteristics of the experimental location before each season's treatment got underway. Following established published techniques **(Wilde *et al.*, 1985)** soil samples were examined at the Soil Testing Laboratory, Faculty of Agriculture, Fayoum University, and the findings were reported in **Table 2**.

Table 2. Some physical and chemical characteristics of the experimental site during the seasons of 2019 /2020 and 2020/2021

Properties	2019/2020	2020/2021
Physical Properties		
Clay (%)	**12.59**	**12.21**
Silt (%)	**12.01**	**12.53**
Coars Sand (%)	**49.79**	**50.33**
Fine Sand	**25.61**	**24.93**
Soil Texture	**Sand loamy**	**Sand loamy**
Chemical Properties		
PH	**7.70**	**7.59**
ECe (dS m^{-1})	**7.12**	**7.08**
Organic matter (%)	**0.84**	**0.92**
$CaCo_3$ (%)	**14.01**	**13.97**
N (%)	**0.09**	**.11**
Availbe elements (mg kg^{-1} soil)		
P	**65.3**	**66.7**
K	**168.5**	**171.3**
Exchangeable Cations ($meqL^{-1}$)		
Ca^{+2}	**9.87**	**9.97**
Na^{+}	**27.5**	**26.57**
Mg^{+2}	**7.11**	**7.21**

2.3. Experimental work

2.3.1. Greenhouse experiments

Imported lettuce hybrid seeds cv. Big Bell (Agent MECCA Trade; Produced by EC-System St. Naktuinbouw 21, China) were hand sown on October 25th, 2019 and October 20th, 2020. Every year, thirty days after the seeds were sown, the seedlings were moved onto the field in rows on both sides of the ridges, with a 25-cm gap between each row. Transplanting date was November 28 and 23 in both seasons 2019 and 2020, respectively. Each experimental unit had five rows that would each be 2.4 meters long and 1.5 meters broad, for a total area of 18 meters. Every two neighboring experimental units were separated by a 0.5 m alley to guard against border effects.

Treatments comprised with twelve of plant growth promoters; Palm pollen grains (100 and 200 mg L^{-1}), Corn grain extract (100 and 200 mg L^{-1}), Soybean seed extract (100 and 200 mg L^{-1}), Mannitol (1500 and 3000 mg L^{-1}), Sorbitol (1500 and 3000 mg L^{-1}) and Calcium phosphate (800 and 1000 $Ca_3(PO_4)_2$ mg L^{-1}), as well as, tap water. Following transplanting, the thirteen treatments were applied foliarly to run-off three times, at 25, 40, and 55 days. To the spraying solution, a small amount of salient film was added as a wetting agent. Both conducted experiments were a randomized complete blocks design, with five replications, each replicate included 13 treatments. Every experimental unit received recommended doses according to MLAR (Market- Led Agrarian Reform) shown in **Table 3**

Table 3. Chemical fertilization program as a ratio among N: P_2O_5: K_2O

Fertilization period	% From total amount	N	P_2O_5	K_2O
Frist month	25	3.0	1.0	2.0
Second month	35	3.0	1.5	3.0
Thread month	25	3.0	1.5	3.0

2.3.2. Plant sampling

For morphological traits, membrane permeability measures (RWC and MSI), and leaf photosynthetic pigments, plants in the first row were assigned to each experimental unit. But in order to compute yield and its components, the second row was selected.

2.4. Data Recorded

2.4.1. Morphological characteristics

Sixty- five days following transplanting date of lettuce, five plants were haphazardly selected, in every test unit. The following morphological characteristics were measured:

2.3.1.1. Plant height (cm); measured starting from the ground level to the highest peak of the canopy.

2.3.1.2. Stem diameter (cm); measured by using Sealy So707-Digital Electronic Vernier Caliper 0-150 mm/0-6″ at ground level.

2.4.1.3. Stem length (cm); evaluated from the level of the growth medium to the main stem's apical meristem.

2.4.1.4. Leaves and stems dry weights plant^{-1} (g); gained by drying at 70°C in a forced-air oven till the weight became constant.

2.4.1.5. Number of leaves plant^{-1}

2.4.1.6. Leaf area plant^{-1} (cm); measured using leaf-blades weight relationship as illustrated by **Taha and Osman (2018);** and was calculated using the following formula:

Leaf area plant^{-1} = $\frac{(\mathrm{LDW})}{(\mathrm{DDW})} \times \mathrm{DA}$

where LDW is the total leaf dry weight (g), DDW is the discs dry weight and DA is the discs area.

2.41.7. Leaf area leaf^{-1} (cm); calculated using the following formula:

Leaf area leaf^{-1}= Leaf area plant^{-1}/Number of leaves plant^{-1}

2.4.2. Plant water status

In order to determine membrane permeability, four randomly chosen plants from each experimental unit provided leaf samples. Following the transplant date by 65 days, rinsed three times with distilled water and cleaned with tape water. The morphological traits listed below were assessed:

2.4.2.1.Relative water content (RWC%); The percentage of RWC was calculated using the formula introduced by **Hayat *et al*. (2007)** and modified by **Osman and Rady (2014)** as follows:

RWC (%) = [(Fw - Dw)/(Tw - Dw)] ×100

2.4.2.2. Membrane stability index (MSI %); The **Premchandra *et al*. (1990)** methodology was used to assess the leaf membrane stability index. Membrane stability index was calculated according to the following formula that mentioned by **Sairam (1994)**

MSI (%) = [1 - (EC_1/ EC_2)] × 100

2.4.3. Leaf photosynthetic pigments contents

2.4.3.1. Chlorophyll *a*, *b*, and carotenoid (mg g^{-1} fresh weight)

Sixty- five days from transplanting date of lettuce, 10 plants were haphazardly selected, in every test unit. Chlorophyll a, b and carotenoid contents were determined using the dimethyl formamide (DMF) method **(Moran and Porath, 1980; Wellburn, 1994)**. Chlorophyll a and b contents were measured by the absorption at wave lengths 647 nm and 664 nm, orderly using a spectrophotometer (UV-Visible Spectroscopy System, Hewlett Packard 95-98). The concentration of chlorophyll *a* and *b* was calculated according to the following formulas **(Wellburn, 1994):**

Chlorophyll *a* (Chl *a*) = 11.65 A664 - 2.69 A647 (x)

Chlorophyll *b* (Chl *b*) = 20.81 A647 - 4.53 A664 (xi)

Total chlorophyll = Chl *a* + Chl *b*

Total carotenoids content was measured by the absorption at wavelength 480 nm via a spectrophotometer (UV-Visible Spectroscopy System, Hewlett Packard 95-98). Total carotenoids concentration was calculated according to the following formula:

Total carotenoids = [1000A480 - 0.89 (Chl *a*) - 52.02 (Chl *b*)]/245 (xii)

2.4.3.2.Measurements of photosynthetic Efficiency

Seventy- five days from transplanting date of lettuce, 10 plants were haphazardly selected, in every test unit. The maximum quantum yield of PSII in a dark-adapted state (variable fluorescence by maximum fluorescence; F_v/F_m and photosynthetic performance index; PPI) was measured using a fluorimeter **(Handy PEA, Hansatech Instruments Ltd., Kings Lynn, UK)** as defined by **Maxwell and Johnson (2000) and Clark *et al*. (2000)**, respectively.

2.4.4. Yield

The lettuce yields were harvested and measured 90 days after transplanting from second row in each experimental unit. The following data were recorded:

2.4.4.1. Head weight plant^{-1} (kg); average weight of heads.

2.4.4.2. Heads yield per square meter (m^2 kg^{-1}) theoretically calculated 16 plants (every square meter has 4 rows, and inside each row there are 4 plants).

2.5 Statistical analysis

The homogeneity of error variances was tested using **InfoStat software estadistico (2016)** in accordance with the methodology described by **Gomez and Gomez (1984)** before all data were subjected to analysis of variance (ANOVA) using a randomized complete blocks design. Use of Duncan's multiple range test revealed significant differences across treatments at $p \leq 0.05$**.**

3. RESULTS AND DISCUSSION

The gained results of two greenhouse experiments were conducted during two winter seasons of 2019/2020 and 2020/2021 seasons to identify effects of foliar applications of plant extracts (date palm pollen, maize and soybean seeds) and mannitol, sorbitol and calcium phosphate as well as control (tap water) are presented in the following topics; morphological characteristics, membrane permeability, leaf photosynthetic pigments content, leaf N, P, K, Ca, and Na contents, enzymatic antioxidants assays, and yield of lettuce plants grown on saline soil.

3.1. Morphological characteristics

Generally, all the foliar application of the twelve treatments (plant extracts, mannitol, sorbitol and calcium phosphate) were superior and significantly recorded higher mean values of plant height, stem length and diameter, dry weight plant^{-1}, number of leaves plant^{-1} and leaf area plant^{-1} of lettuce plants comparing to control (tap water), and the trend was parallel in both years. In another direction, foliar application of tap water (control) gave the highest mean value of leaf area leaf^{-1} of lettuce plants, in two seasons shown in **Tables 4-6.**

Table 4. Effect of some plant growth promoters (PGP) applications on plant height, stem length and stem diameter of lettuce plants grown on saline soil during two winter seasons of 2019/2020 (1st) and 2020/2021 (2nd)

Treatment	(mg L^{-1})	Plant height (cm)		Stem length (cm)		Stem diameter (mm)	
		1st	2nd	1st	2nd	1st	2nd
Control (tap water)		14.9^{j}	17.8^{j}	4.9^{g}	5.8^{g}	14.9^{f}	15.5^{f}
Palm pollen grains extract	**100**	24.7^{a}	29.5^{a}	9.2^{bc}	11.0^{bc}	25.9^{a}	27.0^{a}
	200	21.9^{cd}	26.2^{cd}	9.0^{bcd}	11.8^{bcd}	26.8^{a}	27.9^{a}
Corn grain extract	**100**	20.2^{ef}	24.2^{de}	10.2^{a}	12.2^{a}	25.7^{a}	26.8^{a}
	200	22.3^{bcd}	26.7^{bcd}	10.0^{ab}	11.8^{ab}	20.5^{bc}	21.3^{bc}
Soybean seed extract	**100**	22.9^{bc}	27.3^{bc}	9.5^{ab}	11.3^{ab}	26.3^{a}	27.3^{a}
	200	23.7^{ab}	28.3^{ab}	7.9^{e}	9.3^{e}	22.3^{b}	23.2^{b}
Mannitol	**1500**	16.9^{hi}	20.2^{hi}	7.5^{ef}	9.0^{ef}	19.6^{cd}	20.4^{cd}
	3000	21.1^{de}	25.2^{de}	6.7^{f}	8.0^{f}	15.7^{ef}	16.4^{ef}
Sorbitol	**1500**	19.1^{fg}	22.8^{fg}	8.1^{e}	9.7^{e}	17.8^{de}	18.5^{de}
	3000	17.8^{gh}	21.3^{gh}	8.2^{de}	9.8^{de}	20.3^{bc}	21.1^{bc}
$Ca(H_2PO_4)_2$	**800**	15.8^{ij}	18.8^{ij}	9.2^{bc}	11.0^{bc}	20.3^{bc}	21.1^{bc}
	1000	18.0^{gh}	21.5^{gh}	8.4^{cde}	10.0^{cde}	20.4^{bc}	21.2^{bc}

Values marked with the same letter(s) within the effects are statistically similar using Duncan's multiple range test at $p \leq 0.05$.

The highest values recorded in morphological characteristics measurements via foliar application of plant growth promoters treatments were as follows:

Palm pollen grains extract; at 100 mg L^{-1} in plant height, whilst, at 200 mg L^{-1} in stem dry weight plant^{-1}. While, the concentration at 100 and 200 mg L^{-1} in stem diameter and number of leaves plant^{-1} of lettuce plants, in both experimental seasons.

Corn grain extract; at 100 mg L^{-1} in plant height stem length and stem diameter, in two seasons

Soybean seed extract; at 100 mg L^{-1} in stem diameter and total dry weight plant^{-1}, whilst, concentration at 100 and 200 mg L^{-1} in leaf area plant^{-1}. While, the concentration at 100 and 200 mg L^{-1} in leaves dry weight plant^{-1} of lettuce plants, in both year.

On other side, the foliar application of tap water (control) gave the highest mean value of leaf area leaf^{-1} of lettuce plants, in two seasons.

Table 5. Effect of some plant growth promoters (PGP) applications on dry weight plant^{-1} of lettuce plants grown on saline soil during two winter seasons of 2019/2020 (1st) and 2020/2021 (2nd)

		Dry weight plant^{-1} (g)					
		Leaves		Stem		Total plant	
Treatment	**(mg L^{-1})**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**
Control (tap water)		3.8^{f}	4.5^{f}	0.40^{f}	0.47^{f}	4.16^{f}	4.97^{f}
Palm pollen grains extract	**100**	8.6^{d}	10.3^{d}	1.40^{b}	1.67^{b}	10.0^{d}	11.98^{d}
	200	10.1^{bc}	12.1^{bc}	1.57^{a}	1.87^{a}	11.7^{c}	14.01^{c}
Corn grain extract	**100**	8.3^{d}	9.9^{d}	1.21^{cd}	1.44^{cd}	9.51^{d}	11.38^{d}
	200	8.2^{d}	9.8^{d}	1.10^{d}	1.32^{d}	9.32^{d}	11.14^{d}
Soybean seed extract	**100**	11.7^{a}	14.0^{a}	1.47^{ab}	1.75^{ab}	13.15^{a}	15.72^{a}
	200	11.5^{a}	13.7^{a}	1.46^{ab}	1.75^{ab}	12.95^{ab}	15.49^{ab}
Mannitol	**1500**	6.5^{e}	7.8^{e}	0.80^{e}	0.96^{e}	7.35^{e}	8.79^{e}
	3000	5.6^{e}	6.7^{e}	0.67^{e}	0.80^{e}	6.31^{e}	7.53^{e}
Sorbitol	**1500**	10.6^{ab}	12.7^{ab}	1.30^{bc}	1.56^{bc}	11.90b^{c}	14.22^{bc}
	3000	10.2^{bc}	12.1^{bc}	1.43^{ab}	1.71^{ab}	11.58^{c}	13.84^{c}
$Ca(H_2PO_4)_2$	**800**	9.2^{cd}	11.0^{cd}	0.79^{e}	0.94^{e}	10.00^{d}	11.92^{d}
	1000	6.3^{e}	7.6^{e}	1.11^{d}	1.32^{d}	7.44^{e}	8.89^{e}

Values marked with the same letter(s) within the effects are statistically similar using Duncan's multiple range test at $p \leq 0.05$.

Table 6. Effect of some plant growth promoters (PGP) applications on Number of leaves, leaf area and its segments of lettuce plants grown on saline soil during two winter seasons of 2019/2020 (1st) and 2020/2021 (2nd)

Treatment	**(mg L^{-1})**	**Number of leaves plant^{-1}**		**Leaf area plant^{-1} (dm^{2})**		**Leaf area leaf^{-1} (dm^{2})**	
		1st	**2nd**	**1st**	**2nd**	**1st**	**2nd**
Control (tap water)		9.6^{e}	11.5e	35.5^{h}	36.6^{h}	3.63^{a}	3.15^{a}
Palm pollen grains extract	**100**	29.0^{a}	34.7^{a}	58.8^{abc}	60.6^{abc}	1.91^{g}	1.66^{g}
	200	25.2^{abc}	30.0^{abc}	60.3^{ab}	62.2^{ab}	2.00^{fg}	1.73^{g}
Corn grain extract	**100**	23.1^{c}	27.7^{c}	47.1^{ef}	48.4^{ef}	2.03^{fg}	1.75^{fg}
	200	22.3^{c}	26.7^{c}	49.1^{e}	50.6^{e}	2.22^{defg}	1.92^{defg}
Soybean seed extract	**100**	25.4^{abc}	30.3^{abc}	57.2^{bc}	59.0^{bc}	2.22^{defg}	1.92^{defg}
	200	28.1^{ab}	33.7^{ab}	61.1^{a}	62.9^{a}	2.09^{efg}	1.81^{efg}
Mannitol	**1500**	14.2^{d}	17.9^{d}	43.6^{g}	45.0^{g}	2.94^{b}	2.55^{b}
	3000	16.9^{d}	20.2^{d}	45.2^{fg}	46.6^{fg}	2.57^{cd}	2.22^{cd}
Sorbitol	**1500**	23.2^{c}	28.3^{c}	56.3^{cd}	58.0^{cd}	2.34^{cdef}	2.03^{cdef}
	3000	24.3^{bc}	29.0^{bc}	60.5^{ab}	62.4^{ab}	2.41^{cde}	2.09^{cde}
$Ca(H_2PO_4)_2$	**800**	21.8^{c}	26.0^{c}	53.6^{d}	55.3^{d}	2.52^{cd}	2.18^{cd}
	1000	21.7^{c}	25.4^{c}	56.3^{cd}	58.0^{cd}	2.59^{bc}	2.24^{c}

Values marked with the same letter(s) within the effects are statistically similar using Duncan's multiple range test at $p \leq 0.05$.

3.2. Membrane permeability

In both seasons, foliar application of corn grain extract at 200 mg L^{-1}, mannitol at 3000 mg L^{-1} and calcium phosphate at 1000 mg L^{-1} significantly, attained higher values of MSI %, and the trend was parallel in both experimental seasons.

Foilar application of palm pollen grains extract at 200 mg L^{-1} and calcium phosphate at 1000 mg L^{-1} significantly, attained higher values of RWC % than the other treatments, in the two years according to **Table 7**.

Table 7. Effect of some plant growth promoters (PGP) applications on membrane permeability of lettuce plants grown on saline soil during two winter seasons of 2019/2020 (1st) and 2020/2021 (2nd)

Treatment	(mg L^{-1})	MSI (%)		RWC (%)	
		1st	2nd	1st	2nd
Control (tap water)		60.6^{e}	62.5^{ef}	61.5^{e}	63.4^{e}
Palm pollen grains extract	100	62.6cd^{e}	64.6^{def}	67.2^{d}	69.3^{d}
	200	69.0b	71.1b	84.9a	87.6a
Corn grain extract	100	62.1d^{e}	63.8^{def}	68.3^{d}	70.1^{d}
	200	73.7^{a}	75.9^{a}	80.6^{b}	83.1^{b}
Soybean seed extract	100	61.1^{e}	63.0^{ef}	67.1^{d}	69.2^{d}
	200	65.7^{c}	67.8^{c}	77.8^{bc}	80.2^{bc}
Mannitol	1500	63.5^{cde}	65.4^{cde}	65.8^{d}	67.8^{d}
	3000	71.1^{ab}	73.3^{ab}	76.0^{c}	78.3^{c}
Sorbitol	1500	62.0^{e}	63.9^{ef}	65.7^{d}	67.7^{d}
	3000	65.2^{cd}	67.2^{cd}	76.3c	78.7^{c}
$Ca(H_2PO_4)_2$	800	60.4^{e}	62.2^{f}	67.9^{d}	70.0^{d}
	1000	73.0^{a}	75.2^{a}	84.5^{a}	87.1^{a}

Values marked with the same letter(s) within the effects are statistically similar using Duncan's multiple range test at $p \leq 0.05$.

3.3. Leaf photosynthetic pigments

As usual, spraying via any twelve treatments of plant growth promoters applications with any level led to a significant increase in leaf chlorophyll a, a+b, carotenoids and SPAD chlorophyll contents of lettuce plants comparing to control, and the trend was parallel in the two years. Generally, foliar application of palm pollen extract at any levels (100 or 200 mg L^{-1}) were superior and significantly recorded higher mean values of the photosynthetic response of PPI of lettuce plants compared to the other plant growth promoters. Also, foliar application of palm pollen, corn and soybean seed extracts at concentrations 200 mg L^{-1} gave the highest significant values on leaf SPAD chlorophyll, in both years shown on **Tables 8 and 9.**

Table 8. Effect of some plant growth promoters (PGP) applications on Chlorophyll *a*, *b* and carotenoid of lettuce plants grown on saline soil during two winter seasons of 2019/2020 (1st) and 2020/2021 (2nd)

Treatment	**(mg L^{-1})**	**Chlorophyll (mg/mm^2 fresh weight)**						**Carotenoid (mg/mm^2 fresh weight)**	
		a		***b***		***a +b***			
		1st	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**
Control (tap water)		0.378^{f}	0.390^{f}	0.257^{h}	0.265^{h}	0.635^{i}	0.655^{i}	0.244^{g}	0.252^{g}
Palm pollen grains extract	**100**	0.505^{cd}	0.521^{cd}	0.534^{de}	0.551^{de}	1.039^{def}	1.071^{def}	0.494^{e}	0.510^{e}
	200	0.671^{a}	0.691^{a}	0.693^{a}	0.714^{a}	1.364a	1.406a	0.655a	0.675a
Corn grain extract	**100**	0.483^{cde}	0.496^{cde}	0.502^{ef}	0.516^{ef}	0.985^{fg}	1.012^{fg}	0.477^{e}	0.490^{e}
	200	0.456^{de}	0.470^{de}	0.489^{f}	0.504^{f}	0.945^{g}	0.974^{g}	0.468^{e}	0.482^{e}
Soybean seed extract	**100**	0.432^{ef}	0.445^{ef}	0.433^{g}	0.447^{g}	0.865^{h}	0.892^{h}	0.411^{f}	0.424^{f}
	200	0.501^{cd}	0.517^{cd}	0.504^{ef}	0.520^{ef}	1.006^{efg}	1.037^{efg}	0.486^{e}	0.501^{e}
Mannitol	**1500**	0.486^{cde}	0.501^{cde}	0.594^{c}	0.613^{c}	1.080^{cde}	1.114^{cde}	0.565^{bcd}	0.582^{bcd}
	3000	0.502^{cd}	0.517^{cd}	0.616^{bc}	0.635^{bc}	1.118^{c}	1.152^{c}	0.585^{b}	0.603^{b}
Sorbitol	**1500**	0.497^{cd}	0.512^{cd}	0.580^{c}	0.599^{c}	1.078^{cde}	1.111^{cde}	0.542^{d}	0.559^{d}
	3000	0.521^{bc}	0.537^{bc}	0.574^{cd}	0.592^{cd}	1.095^{cd}	1.129^{cd}	0.549^{cd}	0.566^{cd}
$Ca(H_2PO_4)_2$	**800**	0.536^{bc}	0.553^{bc}	0.604^{c}	0.623^{c}	1.140^{c}	1.176^{c}	0.577^{bc}	0.595^{bc}
	1000	0.572^{b}	0.590^{b}	0.658^{ab}	0.678^{ab}	1.230^{b}	1.268^{b}	0.628^{a}	0.647^{a}

Values marked with the same letter(s) within the effects are statistically similar using Duncan's multiple range test at $p \leq 0.05$.

Table 9. Effect of some plant growth promoters (PGP) applications on leaf SPAD chlorophyll and photosynthetic responses (Fv/Fm and PPI) of lettuce plants grown on saline soil during two winter seasons of 2019/2020 (1st) and 2020/2021 (2nd)

Treatment	($mg\ L^{-1}$)	SPAD		Fv\Fm		PPI	
		1st	2nd	1st	2nd	1st	2nd
Control (tap water)		33.9^{f}	35.0^{f}	0.826^{a}	0.852^{ab}	4.16^{ab}	4.29^{ab}
Palm pollen grains extract	**100**	38.0^{abc}	39.2^{abc}	0.829^{a}	0.855^{ab}	4.43^{a}	4.57^{a}
	200	40.0^{a}	41.3^{a}	0.832^{a}	0.857^{ab}	4.54^{a}	4.68^{a}
Corn grain extract	**100**	38.3^{abc}	39.4^{abc}	0.833^{a}	0.855^{ab}	3.42^{cd}	3.52^{cd}
	200	39.6^{a}	40.8^{a}	0.831^{a}	0.857^{ab}	2.81^{e}	2.89^{e}
Soybean seed extract	**100**	36.9^{bcd}	38.0^{bcd}	0.827^{a}	0.853^{ab}	3.74^{bc}	3.86^{bc}
	200	39.0^{ab}	40.2^{ab}	0.829^{a}	0.855^{ab}	3.00^{de}	3.09^{de}
Mannitol	**1500**	35.4^{def}	36.5^{def}	0.823^{a}	0.849^{b}	3.60^{c}	3.72^{c}
	3000	36.9^{bcd}	38.0^{cde}	0.832^{a}	0.858^{a}	3.59^{c}	3.70^{c}
Sorbitol	**1500**	36.4^{cde}	37.5^{cde}	0.805^{b}	0.830^{c}	3.78^{bc}	3.90^{bc}
	3000	38.3^{abc}	39.5^{abc}	0.825^{a}	0.850^{ab}	3.84^{bc}	3.96^{bc}
$Ca(H_2PO_4)_2$	**800**	34.6^{ef}	35.7^{ef}	0.827^{a}	0.853^{ab}	3.79^{bc}	3.91^{bc}
	1000	36.6^{cde}	37.7^{cde}	0.827^{a}	0.852^{ab}	3.89^{bc}	4.01^{bc}

Values marked with the same letter(s) within the effects are statistically similar using Duncan's multiple range test at $p \leq 0.05$.

3.4. Yield

Generally, foliar application of plant growth promoters was superior and significantly recorded higher mean values of head weight and heads yield per 1 m^2 of lettuce plants comparing to control, in both experimental seasons.

Foliar spraying with soybean seed extract at concentration of 200 mg L^{-1} gave higher values in head weight and heads yield per 1 m^2, however, there was no discernible significant increase between it and the foliar application with palm pollen grains extract at concentration of 100 mg L^{-1}, in both seasons shown in **Table 10.**

Table 10. Effect of some plant growth promoters (PGP) applications on Head weight and Total heads yield of lettuce plants grown on saline soil during two winter seasons of 2019/2020 (1st) and 2020/2021 (2nd)

Treatments	($mg\ L^{-1}$)	Head weight (kg)		Heads yield ($m^2\ kg^{-1}$)	
		1st	2nd	1st	2nd
Control (tap water)		0.442^{h}	0.511^{h}	7.1^{g}	8.2^{g}
Palm pollen grains extract	**100**	0.835^{ab}	0.964^{ab}	13.4^{ab}	15.4^{ab}
	200	0.705^{c}	0.815^{c}	11.3^{c}	13.0^{c}
Corn grain extract	**100**	0.784^{b}	0.905^{b}	12.5^{b}	14.5^{b}
	200	0.553^{fg}	0.639^{fg}	8.9^{ef}	10.2^{ef}
Soybean seed extract	**100**	0.621^{ef}	0.718^{ef}	9.9^{de}	11.5^{de}
	200	0.864a	0.999a	13.8^{a}	16.0^{a}
Mannitol	**1500**	0.626^{def}	0.723^{def}	10.0^{de}	11.6^{de}
	3000	0.527^{g}	0.609^{g}	8.4^{f}	9.7^{f}
Sorbitol	**1500**	0.695^{cd}	0.803^{cd}	11.1^{cd}	12.8^{cd}
	3000	0.666^{cde}	0.770^{cde}	10.7^{cd}	12.3^{cd}
$Ca(H_2PO_4)_2$	**800**	0.517^{gh}	0.597^{g}	8.3^{f}	9.6^{f}
	1000	0.536^{g}	0.619^{g}	8.6^{f}	9.9^{f}

Values marked with the same letter(s) within the effects are statistically similar using Duncan's multiple range test at $p \leq 0.05$.

Plant extracts can be associated with the amelioration of salinity stress as they are the sources of prominent phytochemicals like vitamins, carotenoids, amino acids, phytohormones, mineral nutrients, phenolics, and antioxidants **(Latif and**

Mohamed, 2016). There are several reports where these compounds, used either individually or in mixtures, were found to be effective against salinity stress (**Calvo *et al*., 2014; Drobek *et al*., 2019; Iqbal *et al*., 2014; Bulgari *et al*., 2015; Shukla *et al*., 2019; Zulfiqar *et al*., 2020).** The enhancing effect of plant growth promoters applications on morphological characteristics of lettuce plants **(Tables 4-6)** might be due to the plant extracts containing a lot of osmoprotectants, mineral nutrients, antioxidants and vitamins (antioxidants and vitamins, ascorbic acid and glutathione) and phytohormones as shown in **Table 1**. Perhaps because the best plant growth promoters are more easily accessible and can be applied in larger doses, plants may absorb them better and convert them into metabolic products, which in turn lead to increased plant development.

Spraying plant growth promoters also contributed to an increase MSI, RWC, chlorophyll a+b, DPPH and decrease Na under any levels of plant growth promoters applications **(Tables 7-9)**. Moreover, rising levels of leaf carotenoids **(Table 8)** allow tissues to deal with free radicals, particularly peroxyl radicals and singlet oxygen, thanks to their antioxidant properties **(Sies and Stahl, 1995)**. **Türkmen and Su (2019)** confirmed that the extracts promoted photosynthesis, activated the respiratory cycle, and delaying the aging of plants. Plant extracts also contain cytokinins, which stimulate carbohydrate metabolism, promote cell division, elongation, and chlorophyll biosynthesis; auxins, which improve cell elongation, stem growth, and lateral root formation; mineral nutrients, such as Ca, Mg, P, K, Fe, Mn, Zn, I, and Cu; osmoprotectants, such as soluble sugars and proline; and vitamins thus, extracts have the potential to stimulate plant growth externally **(Rady *et al*., 2019)**. Many other reports support on lettuce our obtained results such as **Abdulkadhim (2019), Byan (2020), Taha *et al*. (2020), Nessem *et al*. (2023), and EL-Sayed and Darwish (2023)** with extract of date palm pollen grains**, Semida and Rady (2014), ur Rehman *et al*. (2018), Rady *et al*. (2019), El-Yazal *et al*. (2021), and Alharby *et al*. (2021)** with extract of corn seed extract.

Bielski (2005) explained that due to the chemical composition, alcoholic sugars carbohydrates that are crucial to photosynthesis move freely and readily inside plants. Mannitol and sorbitol facilitate the transport of microelements inside the phloem tubes in a complex forms (**Jyolsna, 2005**). Therefore, mannitol may move both major and minor nutrients from the leaves to the active parts of the growing tops, flowers, and fruits, this property has a good effect on the yield and vegetative growth of plants **(Brown and Hu, 1996),** as shown in **Table 10**. Sugar alcohols (mannitol, sorbitol, and inositol) have been shown in numerous investigations to play an osmoprotective effect during salt and drought stress tolerance **(Williamson et al., 2002)**. Many other reports support on lettuce our obtained results such as **Mosleh and Rasool (2019), Habiba *et al*. (2019), Issa *et al*. (2020), Al-Abtan *et al*. (2021), Saleh and Hamzah, (2021), Rashid and Mosleh (2022), and Samy (2022)** with foliar spraying of sorbitol and mannitol.

P fertilizer application is required to provide ideal vegetative growth and quality of plants **(Zapata and Zaharah, 2002)**, as well as for obtaining, storing, and utilizing energy **(Epstein and Bloom, 2004)**. Thus, P foliar application and plant vegetative development were shown to be positively correlated in the current study, which is consistent with earlier research showing that P application increases all morphological features when compared to control **(Tables 4–6)**. It is well known that high P concentrations in the tissue are necessary for leaf development since P is essential for photosynthesis, which produces starch and sucrose and raises the dry weight of plants. (**Cakmak *et al*., 1994**). Through a rise in root and shoot photosynthetic product, enough P tries to improve dry matter accumulation (**Rady *et al*., 2018a**). According to the information above, it's possible that the foliar spray of calcium phosphate offset the drop in phosphorous absorption through the roots **(Table 10)**, which is caused by low temperatures below 13°C and promotes the growth of lettuce plants. Our results are supported by numerous additional reports, including **Rab and Haq (2012), Chondraki *et al*. (2012), Rady *et al*. (2018a), Sajid *et al*. (2020), and El-Masry *et al*. (2021).**

When plant growth stimulants were administered to lettuce plants, higher RWC and MSI values were seen compared to control **(Table 7)**, this indicates that the plant growth promoters reflected positive effect on increasing water uptake or reduced water loss, which causes increase in leaf water potential. Plant extracts can be associated with the amelioration of salinity stress as they are the sources of prominent phytochemicals like amino acids, mineral nutrients and phenolics **(Latif and Mohamed, 2016),** which may lead to maintaining the water content of the cells. This led to an improvement in enzymatic antioxidants assays especially; total soluble in leaves, free proline, flavonoids, ascorbate, glutathione, DPPH and phynolics as an osmoticum, helps to maintain the turgor of plant cells **(Osman and Rady, 2012; Osman and El-Shatoury, 2014)**. All that were reflected in an enhancing the permeability of leaves cell membranes of lettuce plants favoring those physiological processes that enable plants to overcome the adverse conditions in saline soils. Similar findings were documented by **Hammad and Ali (2014), Abd El-Mageed *et al*. (2017), ur Rehman *et al*. (2018), Taha *et al*. (2020), and Alharby *et al*. (2021).** This was also supported by **Stoop and Pharr (1994)** reported that the accumulation of mannitol and sorbitol in celery cells grown in saline soil increased relative water content and membrane stability index.

A higher RWC and MSI suggest that the foliar application of calcium phosphate **(Table 7)** likely had a beneficial effect on water intake or decreased water loss, which in turn raised the leaf water potential. Thus, it is possible to draw the conclusion that calcium phosphate has a positive impact on the growth characteristics of lettuce plants **(Tables 4-6)**. This is because phosphorus has an impact on a number of essential plant processes, such as the regulation of certain enzymes, the conversion of sugars and starches, the movement of nutrients within the plant, and the transport of carbohydrates **(Taiz *et al*., 2015)**. Furthermore, to preserve the integrity and selectivity of the cell membrane, an appropriate concentration of Ca^{+2} must be present in the external medium. **(Upadhyaya, 2017). El-Masry *et al*. (2021)** documented similar findings.

Using plant extracts (date palm pollem, maize and soybean seeds) to enhance vegetative growth and boost plant production because these extracts include essential nutrients and growth regulators, such as vitamins and organic acids **(Table 1)** because may be led to significantly increases the levels of chlorophyll and SPAD in leaves. As a result, those extracts promoted photosynthesis, activated the respiratory cycle, and delayed the aging of plants **Türkmen and Su (2019)**. On the other side, the mannitol and sorbitol the chemical composition, which are crucial to photosynthesis, move freely and readily inside plants **Bielski (2005)**. When lettuce plants were exposed to salt stress, foliar spraying with an aqueous of growth stimulant (plant extracts, mannitol, sorbitol and calcium phosphate) leaves enhanced cytokinin, which encourages the manufacturing of chlorophyll and inhibited the absorption of Na^+, potentially mitigating the negative impact of this toxic ion on chlorophyll synthesis **(Rady *et al*., 2019)**. This promoting membrane permeability; RWC and MSI **(Table 7)** this may be led to an increase in chlorophyll a, b, a+b, carotenoids and leaf SPAD chlorophyll contents in **Tables 8 and 9** which, reflected in the increased performance and yield of lettuce plants under saline soils conditions **(Table 10)**. Similar findings were documented by **ur Rehman *et al*. (2018), Abdulkadhim (2019), Taha *et al*. (2020), ALharby *et al*. (2021), and Nessem *et al*. (2023).**

Mineral nutrition determines the amounts of photosynthetic pigments in leaves, including chlorophyll *a*, *b*, *a*+*b*, carotenoids, and leaf SPAD chlorophyll **(Daughtry *et al*., 2000)**. Phosphorus content affects the pigments used in leaf photosynthetic processes because it helps plants stay stable in adverse environments **(Bojovic and Stojanovic, 2006)**. However, the intake of ideal phosphorus levels is necessary for the facilitation of metabolic features and the production of leaf pigment molecules **(Waraich *et al*., 2015; Shubhra *et al*., 2004)**. It has been demonstrated that optimal phosphorus conditions in apricot seedlings boost plant growth and chlorophyll a+b levels **(Dutt *et al*., 2013)**. Additionally, prior research has shown that adding phosphorus to a blue-green alga *Spirulina platensis* boosts its biomass and total carotenoid synthesis. **(Celekli *et al*., 2009)**, Conversely, low levels of phosphorus result in lower levels of protein and total chlorophyll **(Liang *et al*., 2005). Kazemi (2014)** discovered that after applying 15 mM Ca and 30 ppm humic acid topically, tomato plants' leaf chlorophyll content rose from 13.12 in the control group to 22.41 and 21.14 (SPAD). Furthermore, he stated that when compared to the control plants, the interaction between HA and Ca (30 ppm humic acid + 15 mM Ca) produced the highest value (25.14) in the respect. **Ilias *et al*. (2007)** noticed that adding calcium increased the amount of chlorophyll. **El-Masry *et al*. (2021)** documented similar findings.

Typically, foliar administration of soybean seed extract at concentration of 200 mg L^{-1} or palm pollen grains extract at concentration of 100 mg L^{-1} act to enhancing the plant morphological characteristics, membrane permeability, leaf photosynthetic pigments content and leaf photosynthetic efficiency (Fv/Fm and PI) which reflected on improve productivity and quality of lettuce plants (*Lactuca sativa* cv. Big Bell) under saline soil stress in conditions of Fayoum Governorate and other similar regions.

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