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Effect of *Pterocladia capillacea* Seaweed Extracts on Growth Parameters and Biochemical Constituents of Jew's Mallow

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Abstract: We performed field experiments to evaluate the influence of two extraction treatments, seaweed (*Pterocladia capillacea* S.G. Gmelin) water extraction (WE) and ultrasound-assisted water extraction (USWE) at three concentrations (5%, 10%, and 15%), as well as control NPK traditional mineral fertilizer on the growth, yield, minerals, and antioxidants of Jew's Mallow (*Corchorus olitorius* L.) during the two seasons of 2016 and 2017 in Egypt. Plant height, number of leaves, and fresh weight of WE10 treatment were the highest ($p < 0.05$) as 59.67 cm, 10.67 and 2.41 kg m⁻² in 2016, respectively, and 57.33 cm, 11.00 and 2.32 kg m⁻² in 2017, respectively. WE10 and USWE5 treatments produced the highest dry matter (17.07%) in 2016 and (16.97%) in 2017, respectively. WE10 plants had an increased water productivity of 41.2% relative to control plants in both seasons. The highest chlorophyll 'a' was recorded after the WE10 treatment in 2016 and 2017 (17.79 $\mu\text{g g}^{-1}$ and 17.84 $\mu\text{g g}^{-1}$, respectively). The highest levels of total antioxidant capacity, total phenolics, and total flavonoids were also recorded after the WE10 treatment. Application of WE10 boosted growth, yield, minerals, and antioxidants of Jew's Mallow. The CROPWAT model was used to estimate the evapotranspiration, irrigation water requirements, and yield response to irrigation scheduling. Our data showed a yield reduction in the initial growth stage if a limited amount of water was provided. Therefore, irrigation water should be provided during the most important stages of crop development with the choice of effective irrigation practices to avoid water losses, as this helps to maximize yield.

Keywords: seaweed extract; ultrasound-assisted water; foliar spray; *Pterocladia capillacea*; bio-fertilizer; growth parameters; antioxidants; Jew's Mallow; CROPWAT model

1. Introduction

Vegetables and their products contain non-enzymatic antioxidants and micro-nutrients that stabilize free radicals and in turn, increase the capacity of the plant to fight against pathogens that may affect humans and animals [1–4]. For example, the antioxidant compounds in vegetables and fruits could help prevent oxidative stress, diabetes, neurodegenerative disorders, cardiovascular disease, and cancer [5,6]. Jute (*Corchorus olitorius* L.), or Jew's mallow, belongs to the *Tiliaceae* family. *C. olitorius* thought to have originated from South China, from where it was introduced to India and Pakistan. However, a wild variety has been discovered in many areas in India, China, Australia, and Africa, particularly in Southeastern Nigeria. Jute leafy vegetable is commonly used in the preparation of soup [7]. The young shoot tips can be consumed raw or cooked and contain elevated concentrations of protein and vitamin C [8,9]. Jute is generally suggested for pregnant and nursing women because it is thought to be rich in iron [10].

Chemical fertilizers have been used in large quantities to compensate the nutrients deficiency in the soil. It has been observed that this use affects soil, plants, and human health. Their potential carcinogenicity and toxicity have been demonstrated, particularly after the reduction of nitrate to nitrite, or just reacting with amines and/or amides in the formation of N-nitroso compounds, N-nitrosamines, and other nitrogen compounds with high levels of nitrate [11]. Screening of native algal species must be considered to achieve a successful commercial and biotechnological potential of native algal species [12]. Seaweeds are the most promising plants from marine ecosystems and are used as a source of food and medicine. The coast of Egypt has a wide range of wild seaweed available throughout the year, even the Mediterranean coast [13] or the Red Sea coast [14]. Along the Egyptian Mediterranean coast, especially near Alexandria, red algae (*Pterocladia capillacea*; Rhodophyta) are the most dominant native seaweeds. Khairy and El-Shafay [13] studied the seasonal variations (spring, summer and autumn 2010) of biochemical composition of *P. capillacea* collected from Abu Qir Bay, Mediterranean Coast of Alexandria, Egypt. In 2010 spring season, *P. capillacea* achieved the highest significant protein (23.72%) and lipid (2.71%), while in 2010 summer season, *P. capillacea* achieved the highest significant carbohydrate (50.96%), ash (15.81%), and moisture (10.19%). Total fatty acids (248–515 µg/g), total saturated fatty acids (189–360 µg/g), total mono-unsaturated fatty acids (29–77 µg/g), total poly-unsaturated fatty acids (30–78 µg/g), total amino acids (2836–3924 µg/g), total essential amino acids (1136–1445 µg/g), and total non-essential amino acids (1700–2445 µg/g) of seasonally collected *P. capillacea* species were observed. Moreover, Khairy and El-Sheikh [15] observed the mineral composition and antioxidant activities of *P. capillacea* species collected seasonally (spring, Summer and autumn 2010) from Abu-Qir Bay, Mediterranean coast of Alexandria, and they concluded that this species is a rich in carotenoids, phenolic compounds, DPPH free radicals and minerals, therefore, this species can be used as potential source of health food in human diets and may be of use to food industry. In general, marine algae are a rich source of protein, lipids, carbohydrates, polysaccharides, minerals, antioxidants, and other bioactive compounds that can serve in multiple biological activities related to different industries [15,16].

Seaweed extracts can be used as fertilizer for flowering plants, vegetables, and grain crops [17–19]. Furthermore, they have been marketed as fertilizer additives, which are better than other fertilizers [20,21]. Using such extracts (bio-fertilizers) in cultivation many protect the soil and improve crop quality. Therefore, applying them to seeds or adding them to the soil stimulates plant growth [22]. Liquid extracts obtained from seaweeds have gained popularity as foliar sprays for many crops. These extracts contain cytokines, growth promoting hormones, elements, vitamins, and amino acids [23,24]. Some unknown bioactive component in seaweed acts to illicit the plant's own production of plant hormones through internal metabolic pathways [25].

Booth [20] reported that the efficacy of seaweeds as extracts was due to the presence of several metabolites and trace elements. The green seaweed *Enteromorpha* has a high potential for commercial exploitation because of its abundant and varied chemical composition, quality, and concentration of basic nutrients [26]. *Enteromorpha* sp. contains 28 times more calcium than spinach, 26 times more than nopal, and 13 times more than quelite [27]. *Ulva lactuca* and *Enteromorpha intestinalis* are used as seaweed liquid

extract for many crops [21,28]. Rama Rao [29] reported good yields of *Zizyphus rugosa* fruits, when leaves sprayed with seaweed liquid extracts obtained from *Sargassum*. Seaweed extracts are now available commercially as Maxicrop (Sea-Born), Algifert (Marinure), Goemar GA14, Kelpak 66, Seaspray, Seasol, Cytex, and Seacrop. It has been reported that seaweed extracts are better than other extracts [21,23].

Traditional methods employed for extracting bioactive compounds are time consuming and have low extraction efficiencies. To overcome these disadvantages, novel technologies for extraction of bioactive compounds from marine algae have been investigated including the use of microwaves [30], enzymes [31], and super-critical fluids [32]. Recently, ultrasonic technologies have been used to enhance the extraction efficiencies of bioactive compounds (total phenolics, fucose, and uronic acid) from brown seaweed *Ascophyllum nodosum* [33–35] and starch from microalgae *Chlamydomonas fasciata* Ettl NIES-437 [36]. Moreover, ultrasonic assisted extraction was utilized in various industrial fields including phenolic compounds from citrus peel [37], lycopene from tomatoes [38], and anthocyanins from raspberries [39]. Ultrasound-assisted extraction is a simple and employed to improve extraction of bioactive compounds from seaweed [34–36,40].

Full irrigation should be practiced to maximize water productivity [41]. Weekly skipping of irrigation during seed filling may substantially reduce seed yield and water productivity. Skipping during seed germination may be a viable option when water is scarce and land is not limiting. Economic evaluation will provide guidance to policy makers at basin scales for formulating improved and efficient water management plans under all varying weather conditions. CROPWAT is a software for irrigation planning and management [42,43]. Its main functions are: To calculate reference evapotranspiration (ET_o), crop water requirements, and crop irrigation requirements, which may be used to develop irrigation schedules under multiple management conditions and water supply schemes, to estimate rain-fed production and drought effects, and to evaluate the efficiency of irrigation practices. The CROPWAT model has been validated in previous studies for estimating the dynamics main components of soil water balance [44–47]. We undertook this study to investigate the effect of *P. capillacea* seaweed liquid extracts on the growth, yield, minerals, and antioxidants of Jew's Mallow (*C. olitorius* L.).

2. Materials and Methods

2.1. Seaweed

2.1.1. Sampling

P. capillacea seaweed was collected in spring 2016 from the submerged rocky site near Boughaz El-Maadya, Abu-Qir Bay, Alexandria (31.3000° N and 30.1667° E) in Egypt. Harvested samples were transferred to the Microalgae and Invertebrates Aquaculture Laboratories, National Institute of Oceanography and Fisheries (NIIOF), Alexandria, Egypt. Epiphytes were removed from samples, and the seaweed samples were cleaned, washed, and air-dried in shadow. Dried seaweed samples were powdered and stored at room temperature in plastic bag for further analysis and utilization.

2.1.2. Biochemical Composition

Protein, lipid, carbohydrates, and ash of identified seaweed *P. capillacea*, collected in spring 2016, were determined. Total proteins were extracted according to Rauch [48] and determined according to Hartree [49]. Total carbohydrates were extracted according to Myklestad and Haug [50] and determined according to Dubois et al. [51]. Total lipid was calculated according to Bligh and Dyer [52]. Fatty acids and amino acids were extracted and estimated as described by El-Shenody et al. [14].

2.1.3. Seaweed Liquid Extracts Preparation

In this study, seaweed crude liquid extracts of *P. capillacea* were prepared using two extraction methods: three treatments using water extract (WE) and the other three treatments using

Ultrasound-Assisted Water Extraction (USWE), as shown in Figure 1. For WE, 100 g seaweed powder was soaked in 1 L distilled water in a 60 °C water bath for 60 min (extraction phase I). The residual filtrate was filtered and soaked in 1 L distilled water (1:10, w/v) in a 60 °C water bath for 60 min (extraction phase II) and this process was repeated a third time (extraction phase III). Each extraction phase was filtered through Whatman No. 3 filter paper and the supernatants of the three phases (I, II, and III) were combined to the final WE volume of 3 L and stored at −20 °C. For USWE, the three extraction phases were prepared as described above for WE, but after each phase the mixture was subjected to ultrasonication. The USWE extraction was performed at 60 °C for 5 min and 99% amplitude of 20 kHz; these conditions were adjusted and stable for all three extraction phases. After three USWE extraction phases, the supernatants were combined to a final volume of 3 L, and stored at −20 °C. The final combined supernatants, both WE and USWE, were considered to be a 100% crude extract that was utilized as seaweed foliar spray.

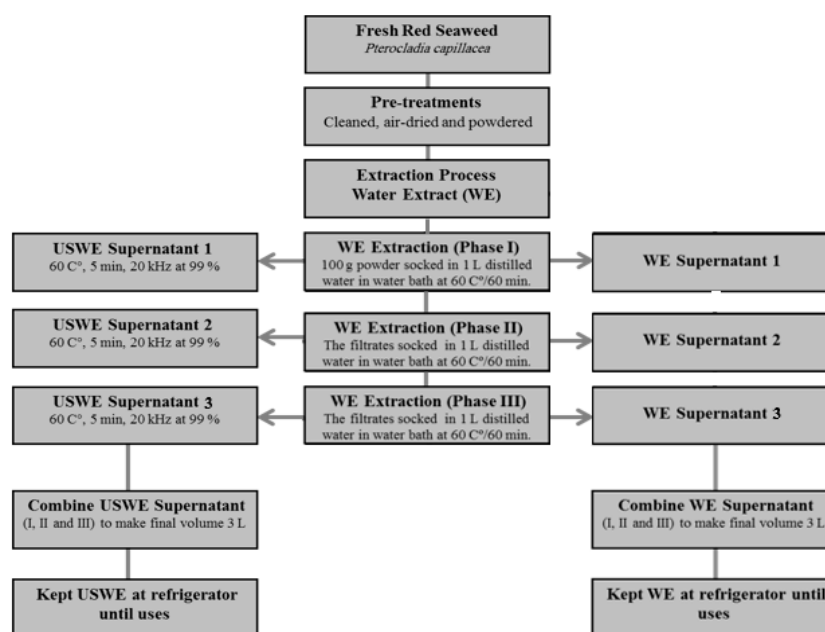


Figure 1. Procedures for water extraction (WE) and ultrasound-assisted water extraction (USWE) of pre-treated *Pterocladia capillacea*.

2.2. Experimental Design

The field experiment with Jew's Mallow (*C. olerius* cv. Balady) was conducted for two successive growing seasons (2016–2017) at Abeis Experimental Farm, Alexandria University, Alexandria (31.2001° N and 29.9187° E) in Egypt. Before sowing, soil samples were collected (0–30 cm depth) to determine physical and chemical properties following Page [53] (Table 1). Climatic data, such as maximum and minimum air temperature (T_{\max} and T_{\min}), relative humidity (RH), wind speed (u_2), and rainfall (P), were collected at a meteorological station near the experimental field location (Figure 2) to calculate daily ET_0 using the Penman–Monteith FAO-56 equation [54]. Evapotranspiration was estimated during growth using crop coefficient (Kc) values [54] multiplied by ET_0 . The experimental area of 220.5 m² was divided into three replicate blocks separated by 2-m buffer zones. Each block consisted of seven plots including one traditional fertilizer and six seaweed extract treatments. Each plot covered an area of 10.5 m² (3 × 3.5 m). A randomized complete block design (RCBD) was used. Commercial seeds were sown on March 20, 2016 and March 22, 2017 at the rate of 28 kg ha⁻¹ [55]. The site was irrigated five times during the first 20 days after sowing to allow germination and establishment before the application of extract treatments. After that, irrigation was carried out every six to seven days for all treatments. The first dose (0.5 m³ ha⁻¹) of growth fertilizer or seaweed extract was applied 10 days

after sowing (DAS), the second one ($0.75 \text{ m}^3 \text{ ha}^{-1}$) was applied 18 DAS, and the third dose ($1 \text{ m}^3 \text{ ha}^{-1}$) was adding 26 DAS. Harvesting included two cuttings at 45 and 70 DAS.

Table 1. Soil physical and chemical properties.

Soil Physical Properties					
Season	2016	2017	Season	2016	2017
Sand (%)	43.3	42.8	Saturation moisture ($\text{m}^3 \text{ m}^{-3}$)	0.49	0.52
Silt (%)	25.5	23.5	Field capacity ($\text{m}^3 \text{ m}^{-3}$)	0.40	0.41
Clay (%)	31.2	33.7	Wilting point ($\text{m}^3 \text{ m}^{-3}$)	0.17	0.17
Soil texture	Clay loam	Clay loam	Total available moisture ($\text{m} \text{ m}^{-1}$)	0.22	0.24
Bulk density (g cm^{-3})	1.48	1.3	Infiltration rate (mm h^{-1})	3.44	3.20
Soil chemical properties					
pH	8.45	8.88	Total Nitrogen (%)	0.19	0.15
E.C. (dS m^{-1})	3.01	3.0	Phosphorus (ppm)	0.41	0.44
Soluble cations (meq L^{-1})			Soluble anions (meq L^{-1})		
Ca ⁺	2.08	1.97	CO ₃ ^{−−}	0.0	0.0
Mg ⁺⁺	1.98	1.88	HCO ₃ [−]	1.43	1.28
Na ⁺	2.47	2.39	Cl [−]	2.05	1.95
K ⁺	0.40	0.37	SO ₂ ^{−−}	3.46	3.37

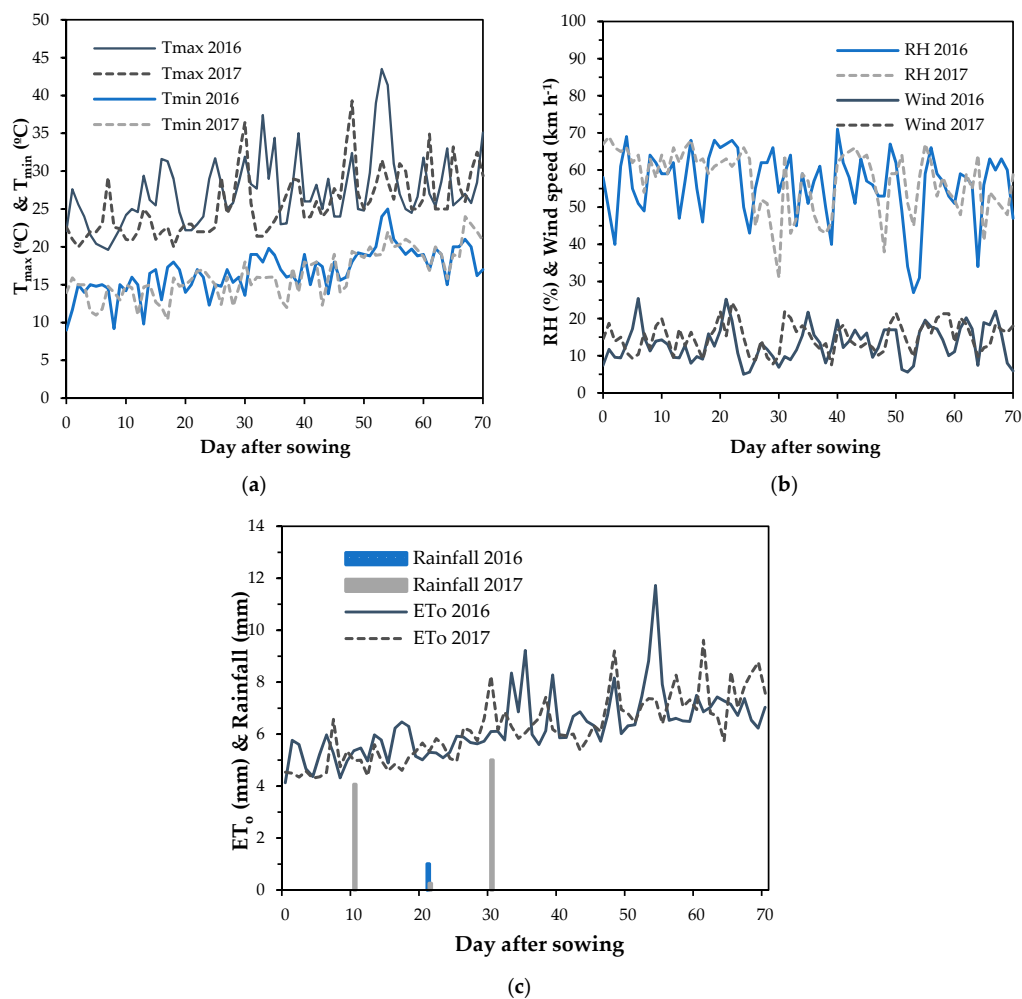


Figure 2. Daily climate parameters in the 2016 and 2017 experimental periods during the Jew's Mallow growing season. (a) daily maximum and minimum air temperature (T_{\max} and T_{\min} , °C), (b) relative humidity (RH, %) and wind speed (km h^{-1}), and (c) reference evapotranspiration (ET_0 , mm) and rainfall (mm).

2.3. Treatments

The following seven treatments were used: mineral NPK fertilizer (control); water extracted seaweed at 5%, 10%, and 15% (WE5, WE10, and WE15); and ultrasound-assisted water extraction seaweed at 5%, 10%, and 15% (USWE5, USWE10, and USWE15).

NPK fertilization was carried out according to the recommendations for commercial production of Jew's Mallow plant. The NPK treatment dose consisted of ammonium nitrate NH_4NO_3 (33%N) at the rate of 300 kg ha^{-1} , calcium superphosphate $(\text{Ca}(\text{H}_2\text{PO}_4)_2 \cdot \text{H}_2\text{O})$ (15% P_2O_5); 525 kg ha^{-1} ; and potassium sulphate (K_2SO_4) (48% K_2O); 125 kg ha^{-1} . Nitrogen fertilizer was applied thrice at 7, 15, and 21 DAS. Phosphorus fertilizer was mixed during soil preparation. Potassium fertilizer was applied at 15 DAS.

2.4. Measurements

2.4.1. Agronomic and Physiological

Plants were harvested (cut) twice, at 45 DAS and 70 DAS, from the center of each plot (treatment) per season to determine leaf and stem fresh weight (kg m^{-2}). Five plants were randomly chosen from each plot to measure plant height and; number of leaves. Ratios between dry leaf weight and fresh leaf weight were determined after drying 70°C in a forced-air oven until reaching a constant weight. Water productivity (WP, kg m^{-3}) was used to evaluate treatments, calculated by dividing total fresh weight (kg m^{-2}) at harvest by the amount of water applied (supplemental irrigation plus rainfall, $\text{m}^3 \text{ m}^{-2}$) to the crop. Chlorophyll 'a', Chlorophyll 'b', and total carotene ($\mu\text{g g}^{-1}$) as described by Dere, et al. [56].

2.4.2. Nutrient Contents

Plant nutrient content (N, P, and K) was analyzed and expressed as percentage on leaf dry weight basis. Total N and P contents were determined calorimetrically using a spectrophotometer at 662 and 650 nm, following the methods of Evenhuis [57]. K was quantified by atomic absorption spectrometry as described by Cottenie, et al. [58].

2.4.3. Antioxidant Activities

Antioxidant activities of crude extracts of WE, USWE and Jew's Mallow were observed. Free radical scavenging activity against DPPH (2,2-diphenyl-1-picrylhydrazyl) was determined as described by Suresh Kumar, et al. [59]. The total antioxidant content (TAC; mg g^{-1}) was determined with a Phosphomolybdate assay using ascorbic acid as the standard [60]. The total phenolic content (TPC; mg g^{-1}) was determined by using the Folin-Ciocalteu method as modified by Suresh Kumar, et al. [59]. Total flavonoid content (TVC; $\mu\text{g g}^{-1}$) was determined according to the method of Chang, et al. [61] with Quercetin as the standard.

2.5. CROPWAT Model

Before Jew's Mallow cultivation, the water application depth and irrigation timing intervals were calculated using the CLIMWAT 2.0 and CROPWAT models. CLIMWAT 2.0 is climatic software [62] presenting the monthly agro-climatic data of over 5000 stations worldwide, including the Alexandria-Nouzha agroclimatic station, which was the nearest to the experimental site (4 km). The CROPWAT model was used for calculation of crop water requirements and the development of irrigation schedules using the option to irrigate at critical depletion and refill soil to field capacity. Irrigation times and the amounts were estimated based on the efficiency of the basin irrigation system and applied for both growth seasons (Figure 3). At the end of each season, the CROPWAT model with the options of user defined application depth and irrigation at user defined intervals were used to evaluate the irrigation schedule. The input data for the CROPWAT version 8.0 model [63] required the following data:

- The daily climatic (T_{max} , T_{min} , RH, daylight hours, and u_2) and P data for the seasons of 2016 and 2017 were accessed from the Meteorological Data of Central Laboratory for Agricultural Climate (Figure 1).
- A cropping pattern consisting of the crop type, planting date, growing stage (20, 20, 25, and 8 days for initial, development, mid-season, and late-season stages, respectively), K_c (0.7 for initial, 1.15 for mid-, and 0.95 for late-season stage) and critical depletion fraction; P (0.3 for initial and development, 0.45 for mid-season stages, and 0.5 for late season stage), rooting depth; Z_r (0.18 m for initial stage and 0.5 m for maximum (mid- and late-season)), and yield response factor; k_y (0.8 for initial, 0.4 for development, 1.2 for mid-, and 1 for late-season). The crop values were assumed as data for a small vegetable according to Allen, et al. [54].
- Soil type: Total available soil moisture, maximum infiltration rate and initial soil moisture depletion were obtained from measured data (Table 1).

The output of CROPWAT model consists of daily root zone depletion ($D_{r,i}$, Equation (1)), deep percolation (DP_i), actual water use by crop (ET_c)_{actual}, efficiency of the irrigation schedule (EIS , Equation (2)), deficiency of the irrigation schedule (DIS , Equation (3)) and yield reduction (Y_R , Equation (4)) were collected and analyzed using the following equation:

$$D_{r,i} = D_{r,i-1} + (ET_{c,i})_{actual} - P_i - I_i + RO_i + DP_i \tag{1}$$

where $D_{r,i}$, and $D_{r,i-1}$ are at days i and $i-1$, P_i is total rainfall over day i , I_i is net irrigation on day i , RO_i is water loss by runoff from the soil surface on day i , in our study the RO is equal to zero, and DP_i is water loss by deep percolation on day i .

$$EIS = \frac{\sum(I_i - DP_i)}{\sum I_i} \times 100 \tag{2}$$

$$DIS = \frac{Sesaonal (ET_c)_{potential} - Sesaonal (ET_c)_{actual}}{Sesaonal (ET_c)_{potential}} \times 100 \tag{3}$$

$$Y_R = K_y \left(1 - \frac{(ET_c)_{actual}}{(ET_c)_{potential}} \right) \tag{4}$$

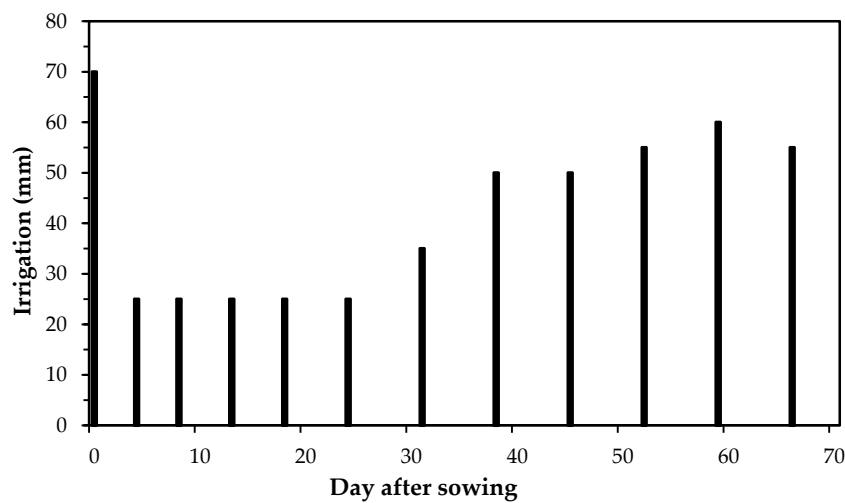


Figure 3. Amounts of irrigation water (mm) applied during the Jew’s Mallow growing season.

2.6. Statistical Analyses

Analysis of variance (ANOVA) with RCBD was performed on data obtained from both growing seasons (2016 and 2017) using the IBM SPSS Version 23 software to determine the significance of differences among treatments. Standard errors (SE) were presented for the mean of data from both growing seasons. Differences among means of replicates were measured using Duncan method at $p \leq 0.05$ [64].

3. Results

3.1. Biochemical Composition Seaweed *P. Capillacea*

Nutritional compositions of the red seaweed species *P. capillacea* collected during spring season of 2016 were investigated. Lipid, protein, carbohydrate, and ash percentages, based on dry weight, were 2.46%, 18.47%, 51.36%, and 13.71%, respectively. Moreover, total fatty acids, total saturated fatty acids, total mono-unsaturated fatty acids, total poly-unsaturated fatty acids, total amino acids, total essential amino acids, and total non-essential amino acids were 247.6, 188.6, 29.1, 29.9, 2836, 1136.3, and 1700 $\mu\text{g g}^{-1}$, respectively. The antioxidant activities result of WE and USWE crude extracts observed that no significant differences ($p < 0.05$) were found in total antioxidant content (22.48 and 22.33 mg g^{-1}) and total phenolic content (17.79 and 16.85 mg g^{-1}) in WE and USWE, respectively, while USWE achieved a significant difference ($p < 0.05$) in total flavonoid content (45.68 $\mu\text{g g}^{-1}$) and total carotene (2.03 $\mu\text{g g}^{-1}$) in comparing to WE (34.77 $\mu\text{g g}^{-1}$ and 1.29 $\mu\text{g g}^{-1}$, respectively).

3.2. Agronomic Traits

Table 2 shows the significant differences in in plant height ($p < 0.05$). WE10 treatment produced the tallest plants, followed by WE5 in both seasons. In WE10 and WE5 treated plants, height increased by 39.8% and 28.1%, respectively, compared with mineral fertilizer treated plants (control treatment) in 2016 while plant height increased by 28.3% and 23.2% in 2015. The USWE10 treatment had the smallest effect on height in both years, with increases of 4.6% and 0.7% in 2016 and 2017, respectively, relative to control. Statistically significant differences ($p < 0.05$) were found between treatments for leaf number, where WE5 and WE10 treatments had the highest values in both seasons.

Table 2. Effects of water extract (WE) and ultrasound-assisted Water Extraction (USWE) on growth characteristics of Jew's Mallow during 2016 and 2017 growing seasons.

Treatment	Plant Height (cm)		Leaf Number (Plant ⁻¹)		Fresh Weight (kg m ⁻²)		Dry Matter (%)	
	2016	2017	2016	2017	2016	2017	2016	2017
Control	42.7 ± 0.5 ^d	44.7 ± 0.6 ^d	9.3 ± 1.1 ^{ab}	9.0 ± 1.0 ^b	1.64 ± 0.1 ^d	1.65 ± 0.1 ^e	13.9 ± 1.1 ^c	14.1 ± 1.6 ^b
WE5	54.7 ± 2.0 ^b	55.0 ± 2.3 ^{ab}	10.7 ± 0.5 ^a	11.0 ± 0.2 ^a	2.01 ± 0.1 ^c	2.07 ± 0.1 ^{bc}	16.4 ± 0.5 ^{ab}	16.3 ± 0.4 ^a
WE10	59.7 ± 1.5 ^a	57.3 ± 3.2 ^a	10.6 ± 1.1 ^a	11.0 ± 1.0 ^a	2.41 ± 0.1 ^a	2.32 ± 0.1 ^a	17.1 ± 1.0 ^a	16.6 ± 0.4 ^a
WE15	52.0 ± 2.0 ^c	52.5 ± 2.2 ^{abc}	9.0 ± 0.1 ^b	9.0 ± 0.1 ^b	1.91 ± 0.1 ^c	1.84 ± 0.1 ^{de}	16.7 ± 0.9 ^{ab}	16.2 ± 0.4
USWE5	44.7 ± 0.5 ^d	47.7 ± 1.8 ^{cd}	9.3 ± 0.6 ^{ab}	9.3 ± 0.5 ^b	2.09 ± 0.0 ^{bc}	2.15 ± 0.2 ^{ab}	16.1 ± 1.0 ^{ab}	16.9 ± 1.1 ^a
USWE10	44.6 ± 1.5 ^d	45.0 ± 2.2 ^d	9.0 ± 0.2 ^b	9.0 ± 0.1 ^b	2.22 ± 0.1 ^{ab}	2.16 ± 0.1 ^{ab}	16.4 ± 0.2 ^{ab}	16.6 ± 0.9 ^a
USWE15	50.0 ± 1.0 ^c	49.9 ± 0.4 ^{bcd}	9.7 ± 0.6 ^{ab}	9.6 ± 0.6 ^b	1.88 ± 0.2 ^c	1.90 ± 0.1 ^{cd}	14.9 ± 1.5 ^{bc}	15.1 ± 1.5 ^b

Control: NPK fertilization; WE5, WE10, and WE15: water extracted seaweed at 5%, 10%, and 15%, respectively; USWE5, USWE10, and USWE15: ultrasound-assisted water extraction seaweed at 5%, 10%, and 15%, respectively. Data are means ± SE. Different superscript letters in each column indicate significant differences ($p \leq 0.05$).

There was a significant difference ($p < 0.05$) in fresh weight and dry matter between the treatments in 2016 and 2017. Fresh weight was the highest in WE10 treated plants in both seasons, followed by the USWE10 treatment. For WE10 and USWE10 treatments, the fresh weight increased by 47% and 35.4% in 2016, respectively; then 40.6% and 30.9% in 2017, compared to control treatment. Across all treatments, WE15 and USWE15 treatments reduced the fresh weight in both seasons. There were significant differences ($p < 0.05$) between bio- and mineral fertilizer-treated plants dry matter in 2016 and 2017 (Table 2). The highest dry matter value was recorded in plants that received the WE10 and USWE5 treatments, which was 23% and 19.9% higher, respectively, than control in 2016.

3.3. Water Productivity

Water requirement varied from 3.8 to 9.1 and 3.5 to 8.5 mm day⁻¹ from the early stage to the peak demand period (mid-season) for 2016 and 2017, respectively. Water productivity (WP) values determined for treatments in 2016 and 2017 are shown in Figure 4. In both seasons, there were significant differences ($p < 0.05$) between WP values. The highest WP values were recorded with the WE10 treatment 41.2% higher than control. Among extract treatments, USWE15 and WE15 had the lowest WP in 2016 and 2017, which was lower by 23% and 20.7% than WE10, respectively.

3.4. Physiological Traits

Water extraction treatments had the highest content of chlorophyll 'a' in both seasons (average, 17.49 $\mu\text{g g}^{-1}$), while the lowest chlorophyll 'a' content was observed with the control treatment (average, 9.4 $\mu\text{g g}^{-1}$, Table 3). WE10 and WE15 treated plants showed significant increases in chlorophyll 'b' content compared to control treatment in both seasons. The chlorophyll 'b' content for the WE10 and WE15 treated plants (average, 13 $\mu\text{g g}^{-1}$ and 13.3 $\mu\text{g g}^{-1}$, respectively) was two-fold higher than the content in the control treatment in 2016 and 2017. Conversely, USWE5 and USWE10 application resulted in the lowest chlorophyll 'b' content in both growing seasons, 16.8% and 26.8% lower than control, respectively. The lowest carotene content was achieved by control treatment in 2016 and 2017 (2.9 and 2.8 $\mu\text{g g}^{-1}$, respectively; Table 3). The highest carotene content in 2016 and 2017 was measured with the USWE10 treatment (71.2% and 72% higher than control, respectively), followed by the USWE15 treatment (53.7% and 54.3% higher than control, respectively).

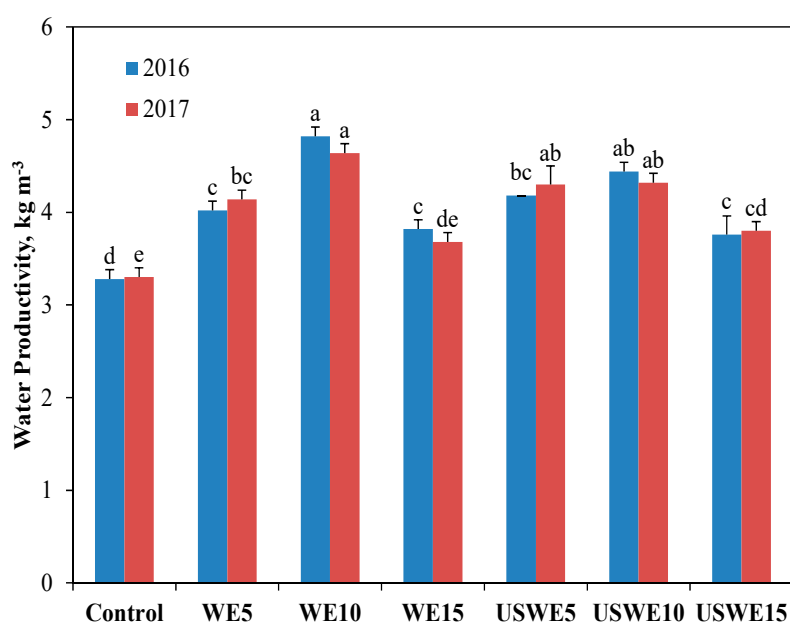


Figure 4. Water productivity (kg m^{-3}) of Jew's Mallow as a function of water extract (WE) and ultrasound-assisted water extract (USWE) treatments in the 2016 and 2017 growing seasons. Different letters (a, b, etc.) above bars indicate a significant difference among treatments in each season.

Table 3. Effects of water extract (WE) and Ultrasound-Assisted Water Extraction (USWE) on chlorophyll 'a', chlorophyll 'b', and carotene concentrations in Jew's Mallow during the 2016 and 2017 growing seasons.

Treatment	Chlorophyll 'a' ($\mu\text{g g}^{-1}$)		Chlorophyll 'b' ($\mu\text{g g}^{-1}$)		Carotene ($\mu\text{g g}^{-1}$)	
	2016	2017	2016	2017	2016	2017
Control	9.4 ± 1.9 ^d	9.4 ± 1.9 ^d	6.5 ± 0.2 ^{bc}	6.5 ± 0.2 ^{bc}	2.9 ± 0.2 ^d	2.8 ± 0.2 ^d
WE5	17.0 ± 0.7 ^a	17.1 ± 0.7 ^a	6.2 ± 0.0 ^{bc}	6.2 ± 0.0 ^c	3.1 ± 0.3 ^{cd}	3.1 ± 0.3 ^{cd}
WE10	17.8 ± 0.2 ^a	17.8 ± 0.3 ^a	13.0 ± 1.2 ^a	12.9 ± 1.1 ^a	3.0 ± 0.2 ^d	3.1 ± 0.2 ^{cd}
WE15	17.7 ± 1.7 ^a	17.5 ± 1.6 ^a	13.3 ± 0.8 ^a	13.3 ± 0.8 ^a	3.1 ± 0.2 ^{cd}	3.0 ± 0.2 ^{cd}
USWE5	12.3 ± 0.1 ^c	12.4 ± 0.0 ^c	5.4 ± 2.3 ^c	5.4 ± 2.3 ^c	3.9 ± 0.3 ^{bc}	3.8 ± 0.2 ^{bc}
USWE10	10.3 ± 0.7 ^{cd}	10.3 ± 0.7 ^{cd}	4.8 ± 0.6 ^c	4.7 ± 0.7 ^c	4.9 ± 0.8 ^a	4.9 ± 0.8 ^a
USWE15	14.7 ± 1.7 ^b	14.8 ± 1.7 ^b	8.5 ± 1.9 ^b	8.5 ± 1.8 ^b	4.4 ± 0.6 ^{ab}	4.4 ± 0.6 ^{ab}

Control: NPK fertilization; WE5, WE10, and WE15: water extracted seaweed at 5%, 10%, and 15%, respectively; USWE5, USWE10, and USWE15: ultrasound-assisted water extraction seaweed at 5%, 10%, and 15%, respectively. Data are means ± SE. Different superscript letters in each column indicate significant differences ($p \leq 0.05$).

3.5. N, P, and K

Nutrient content (i.e., N, P, and K) of Jew's Mallow plant treated with different seaweed extracts in comparison to control treatment are presented in Table 4. Control treatment had the highest N content in 2016 and 2017 (1.78% and 1.71%, respectively), while WE10 had the lowest N content (1.20% and 1.33%), respectively. USWE10 treatment had the highest P content in 2016 and 2017 (0.74% and 0.77%, respectively). The highest K content (1.90%) was achieved by WE15 treatment in both seasons.

Table 4. Effects of water extract (WE) and ultrasound-assisted Water Extraction (USWE) on N, P, and K content in Jew's Mallow during the 2016 and 2017 growing seasons.

Treatment	N (%)		P (%)		K (%)	
	2016	2017	2016	2017	2016	2017
Control	1.78 ± 0.20 ^a	1.71 ± 0.25 ^a	0.67 ± 0.06 ^{ab}	0.70 ± 0.07 ^{ab}	1.40 ± 0.01 ^c	1.40 ± 0.02 ^b
WE5	1.33 ± 0.13 ^b	1.40 ± 0.04 ^b	0.64 ± 0.01 ^b	0.66 ± 0.04 ^b	1.80 ± 0.10 ^b	1.90 ± 0.04 ^a
WE10	1.20 ± 0.10 ^b	1.33 ± 0.12 ^b	0.70 ± 0.06 ^{ab}	0.77 ± 0.01 ^a	1.80 ± 0.02 ^b	1.80 ± 0.01 ^a
WE15	1.41 ± 0.04 ^b	1.49 ± 0.23 ^{ab}	0.71 ± 0.05 ^{ab}	0.73 ± 0.06 ^{ab}	1.90 ± 0.03 ^a	1.90 ± 0.01 ^a
USWE5	1.39 ± 0.10 ^b	1.37 ± 0.02 ^b	0.66 ± 0.06 ^{ab}	0.69 ± 0.07 ^{ab}	1.80 ± 0.04 ^b	1.80 ± 0.02 ^a
USWE10	1.24 ± 0.12 ^b	1.32 ± 0.11 ^b	0.74 ± 0.03 ^a	0.77 ± 0.02 ^a	1.20 ± 0.10 ^d	1.30 ± 0.03 ^b
USWE15	1.41 ± 0.04 ^b	1.39 ± 0.06 ^b	0.68 ± 0.03 ^{ab}	0.74 ± 0.03 ^{ab}	1.20 ± 0.11 ^d	1.30 ± 0.02 ^b

Control: NPK fertilization; WE5, WE10, and WE15: water extracted seaweed at 5%, 10%, and 15%, respectively; USWE5, USWE10, and USWE15: ultrasound-assisted water extraction seaweed at 5%, 10%, and 15%, respectively. Data are means ± SE. Different superscript letters in each column indicate significant differences ($p \leq 0.05$).

3.6. Antioxidant Activity

The highest DPPH percentage was achieved by WE10 in 2016 and 2017 (40.78% and 40.74%, respectively). The lowest DPPH percentage was recorded in USWE15 in 2016 and 2017 (8.75% and 8.74%, respectively; Figure 5). The highest TAC was recorded in WE10 in both seasons (43.97 and 44.22 mg g^{-1} , respectively), followed by the USWE10 treatment ((35.69 and 36.38 mg g^{-1} ; Table 5). The lowest TAC was recorded with control (26.30 mg g^{-1}) in 2017 and USWE15 treatment (26.00 mg g^{-1}) in 2018. In both seasons, the highest significant TPC was obtained with WE10 treatment (116.28 and 115.81 mg g^{-1} , respectively), while the lowest TPC was obtained with the USWE15 treatment (49.62 and 49.61 mg g^{-1} , respectively). Although USWE5 had a higher TVC value, significant differences between extracts treatments were not observed, except for USWE15 treatment.

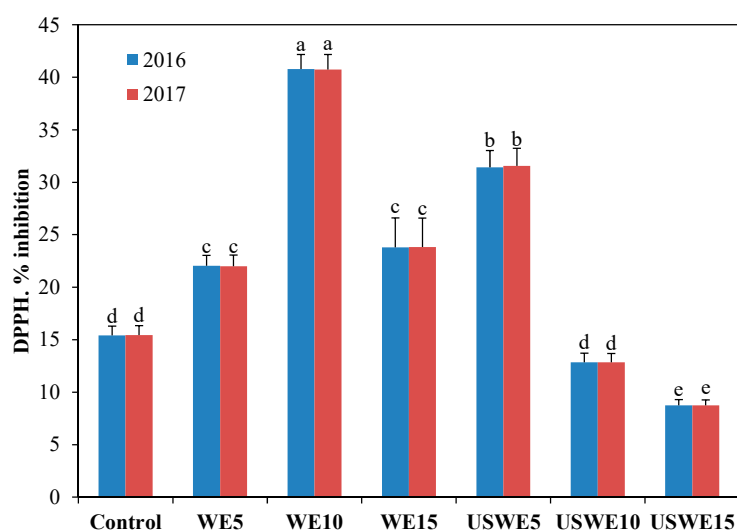


Figure 5. DPPH inhibition (% inhibition) of Jew's Mallow with water extract (WE) and ultrasound-assisted water extract (USWE) in the 2016 and 2017 growing seasons. Different letters (a, b, etc.) above bars indicate a significant difference among treatments in each season.

Table 5. Effects of water extract (WE) and ultrasound-assisted Water Extraction (USWE) on total antioxidant content (TAC), total phenolic content (TPC), and total flavonoid content (TVC) of Jew's Mallow during the 2016 and 2017 growing seasons.

Treatment	TAC (mg g ⁻¹)		TPC (mg g ⁻¹)		TVC (μg g ⁻¹)	
	2016	2017	2016	2017	2016	2017
Control	26.30 ± 3.43 ^c	26.22 ± 0.40 ^c	97.15 ± 11.78 ^b	98.15 ± 10.92 ^b	1168 ± 47.4 ^{ab}	1170 ± 49.2 ^{ab}
WE5	34.65 ± 3.36 ^b	32.69 ± 1.27 ^b	75.22 ± 2.19 ^{cd}	75.40 ± 2.43 ^{cd}	1193 ± 23.9 ^a	1194 ± 20.7 ^a
WE10	43.97 ± 1.04 ^a	44.22 ± 2.40 ^a	116.28 ± 6.59 ^a	115.81 ± 7.77 ^a	1208 ± 26.4 ^a	1204 ± 14.7 ^a
WE15	27.28 ± 1.37 ^c	28.13 ± 2.53 ^c	77.92 ± 11.12 ^{cd}	78.33 ± 10.70 ^{cd}	1191 ± 20.9 ^a	1187 ± 29.7 ^a
USWE5	35.35 ± 1.55 ^b	35.52 ± 1.32 ^b	66.56 ± 1.69 ^d	65.46 ± 1.47 ^d	1244 ± 3.1 ^a	1232 ± 7.3 ^a
USWE10	35.69 ± 2.41 ^b	36.38 ± 2.53 ^b	49.62 ± 6.54 ^e	49.61 ± 7.51 ^e	1205 ± 80.8 ^a	1209 ± 75.1 ^a
USWE15	26.78 ± 2.28 ^c	26.00 ± 2.54 ^c	80.20 ± 0.83 ^c	79.69 ± 0.56 ^c	1113 ± 31.1 ^b	1105 ± 30.1 ^b

Control: NPK fertilization; WE5, WE10, and WE15: water extracted seaweed at 5%, 10%, and 15%, respectively; USWE5, USWE10, and USWE15: ultrasound-assisted water extraction seaweed at 5%, 10%, and 15%, respectively. Data are means ± SE. Different superscript letters in each column indicate significant differences ($p \leq 0.05$).

3.7. CROPWAT Model

Figure 6 shows the depletion curve before and after each irrigation event during the 2016 and 2017 growth seasons. The highest values of depletion were 52 and 48 mm in the mid-season for each year, respectively. The depletion values were between those of field capacity and readily available moisture except for the initial 20 days of both seasons. Thus, there was no water stress. In the initial stage, there was a maximum DP of 28 mm at the first irrigation event in both seasons and after that it decreased to 2.5 and 8.5 mm, respectively, in the 2016 and 2017 growing seasons. The effective rainfall means modeled for 2016 and 2017 were 9.4 and 1 mm, respectively leaving deficits of 387 and 401.7 mm to be made up from irrigation. Thus, effective rainfall showed an ineffective pattern across the growth stages. By applying the basin irrigation system, the application water efficiencies were 78% and 79%, respectively, in 2016 and 2017. Hence, the values of EIS were 95.1% and 98.6%, respectively, in 2016 and 2017. The values of $(ET_c)_{actual}$ were 393.2 and 399.8 mm, respectively in 2016 and 2017; the values of DIS were 0.8% and 0.7%. On the other hand, there were no yield reductions across the growth stages, with the maximum values of 3.7% and 3.1% in the initial stage, respectively in 2016 and 2017.

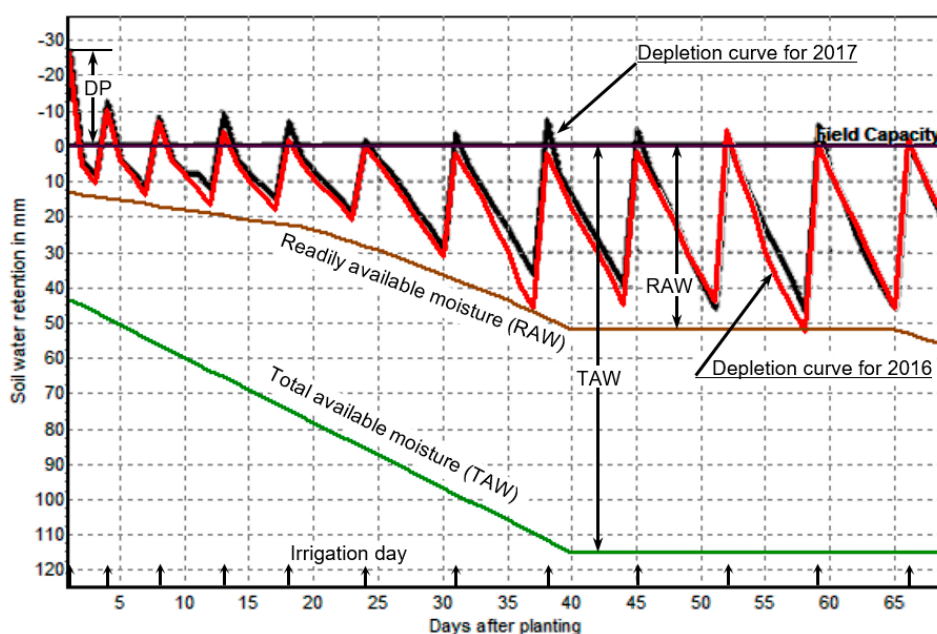


Figure 6. Soil water depletion during the 2016 and 2017 growing seasons.

4. Discussion

Marine algae are considered very important bioindicator for the marine ecosystem [13–15]. Many studies have reported that the constituents, diversity, and communities of marine algae are affected by variations in environmental parameters and nutrient limitation [65–70]. During the last few years, the attention on scientific and commercial interest to biotechnological applications of algae as a sustainable source and global commercial for aquaculture [71–75], biofuel [12,76], extracts [77,78], food supplement, pharmaceuticals, and cosmetics were increased [12].

In current study, data of biochemical composition (protein, lipid and carbohydrate) of *P. capillacea* showed that the large component is carbohydrate (51.36%), followed by protein (21.49%) and lipid (2.06%). The presented data may be act as an indicator for related bioactive secondary metabolites of *P. capillacea* liquid extract. However, our data is in the same line of the results presented by Khairy and El-Shafay [13] who found that, during spring season of 2010, the highest component is carbohydrate (50.49%), followed by protein (23.72%) and lipid (2.71%). Many authors reported that the biochemical constituents of marine algae are affected by variations in environmental conditions and nutrient availability [65–70].

The nutrient contents of seaweed *P. capillacea* used in current study were investigated previously by Khairy and El-Sheikh [14], at the same collected location of current study too, who observed that mineral were potassium (50.9 mg/100g), calcium (68.4 mg/100g), magnesium (22.1 mg/100g), copper (0.5 mg/100g), ferrous (18.37 mg/100g), and zinc (0.19 mg/100g). In current study, although the applied seaweed extract is a rich source of nutrient, it not characterized as a nutrient fertilizer because of many consecrations like its constituent of bioactive compound which act as a plant growth promoting. Interestingly, the *P. capillacea* seaweed species is reported as a potential source for human healthy food because its constituent of bioactive compounds like protein, lipid, carbohydrate, fatty acids (saturated, mono-unsaturated and poly-unsaturated fatty acids), amino acids (essential and non-essential), carotenoids, phenolic compounds, and DPPH [13,14]. Hence, *P. capillacea*, collected from the same study location, is reported as a rich source of alkaloids, flavonoids, steroids, terpenoids, phlobatannins and many other phytochemicals and secondary metabolites [79].

Moreover, *P. capillacea* as a red alga is characterized as a rich source of different phytohormones [40,80,81]. It well known that some unknown bioactive component in seaweed acts to illicit the plant's own production of plant hormones through internal metabolic pathways [25].

Seaweeds and its extracts are becoming of increasing importance because of their bioactive compounds and their potential application in different industries. Liquid seaweed extract is commonly used as commercial agricultural biostimulants because of many considerations.

In current study, to enhance the efficiency of seaweed liquid extract, we evaluate two extraction methods; (1) water (WE); and (2) ultrasound-assisted water extraction (USWE). The effect of different seaweed extracts as a foliar spray on quantity (growth and yield) and quality (minerals and antioxidants activity) of Jew's Mallow (*C. olerius* L.), comparing to NPK traditional fertilizers were observed. In general, Jew's Mallow (*C. olerius* L.) treated with liquid seaweed extract (either WE or USWE) achieved the highest significant quantity (yield) and quality (antioxidant activity, P %, and K %), comparing to NPK traditional fertilizers, which only achieved the highest significant N %. Jew's Mallow (*C. olerius* L.) treated with WE10 and USWE10 were achieved the highest significant yield (fresh weight), and P %. The highest significant Chlorophyll a and b; total antioxidant activity and total phenolic compounds were achieved by WE10, while the highest significant carotene and total flavonoid compounds were achieved by USWE10. In general, in the present study, it was observed that the seaweed liquid extract prepared from *P. capillacea* presented to Jew's Mallow gave better results in all aspects of growth to yield when compared to NPK traditional fertilizers. Using ultrasound-assisted water extraction (USWE) method was significantly improved the total flavonoid and carotene content in *P. capillacea* USWE crude extract, which is positively reflected on these compounds of Jew's Mallow (*C. olerius* L.), when comparing to WE. Carotenes are indispensable to plants and act as precursors for the biosynthesis of phytohormones and strigolactones, improve the plant development and responses to unstable environmental, and serve as a source of pro-vitamin A [82].

In the present study, WE10-treated plants showed the best response in plant height and leaf number. Similarly, Stephenson [40] reported that seaweed liquid extract prepared from *Ascophyllum* and *Laminaria* accelerated maize growth. Blunden and Wildgoose [83] reported a marked increase in lateral root development in potato plants as a result of treatment with seaweed extract. Similar results were obtained with *Padina* biofertilizer, which induced maximum growth in *Cajanus cajan* [84]. Thirumaran, et al. [85] reported similar findings 20% seaweed liquid extract from brown algae *Rosenvingea intricate* had an increased growth of *Cyamopsis tetragonoloba*. Similarly, Whapham, et al. [86] observed that the application of seaweed *Ascophyllum nodosum* liquid extract increased the chlorophyll content in cucumber cotyledons, tomato, and guar plants [83].

Seaweed liquid extracts can be an effective way to some crop plants to increase both the nutrient content of the soil and crop yield. Hence, seaweeds play a vital role in agriculture, where irrational use of chemical fertilizer and pesticides is a cause of concern. Extensive regional trials with the product are needed to determine the environmental limitations of biological activity and to monitor the survival and dispersal of the inoculate [87]. Hence, use of modern agriculture in conjunction with traditional farming practices is the sustainable solution for the future. The expansion of nature source of other manures, seaweed extract application will be useful in enriching the production in the place of costly chemical fertilizer. The use of seaweed liquid extracts helps to avoid environmental pollution by high doses of chemical fertilizer. The beneficial effects of seaweed extract on terrestrial plants are improving the overall growth, yield and the ability to with stand adverse environmental conditions [88].

From the outputs of the CROPWAT model for 2016 and 2017 growing seasons, it appeared that additional irrigation was required to meet the daily crop water requirements as rainfall had minor effects or none. This high irrigation requirement may be attributed to the low rainfall during the growing seasons. Our data indicate that irrigation is crucial in the initial growth stage of Jew's Mallow due to high DP caused by basin irrigation system. To avoid yield reductions in Jew's Mallow cultivation, large quantities of water should be applied during the initial stage. In areas where water is a restricting factor in crop production, a well-designed irrigation schedule can improve water productivity when full irrigation is not plausible. However, a certain yield reduction should be expected due to the relationship between ET_c and yield of some crops [44,89–91].

5. Conclusions

Seaweeds are one of the most important marine resources for food, industrial raw materials, therapeutic and botanical applications. In current study, to enhance the efficiency of seaweed liquid extract, we evaluate two extraction methods; (1) water (WE); and (2) ultrasound-assisted water extraction (USWE). The effect of different seaweed extracts as a foliar spray on quantity (growth and yield) and quality (minerals and antioxidants activity) of Jew's Mallow *C. olitorious* L., comparing to NPK traditional fertilizers were observed. The present study observed that the seaweed liquid extract prepared from *P. capillacea* (either WE or USWE) presented to Jew's Mallow *C. olitorious* L. gave better results in all aspects of quantity and quality when compared to NPK traditional fertilizers. No significant differences of quantity (yield) of *C. olitorious* L. treated with WE10 and USWE10. Water extraction (WE) method improves the Chlorophyll 'a' and 'b'; total antioxidant activity and total phenolic compounds of Jew's Mallow *C. olitorious* L. While, using ultrasound-assisted water extraction (USWE) method improves the carotene and total flavonoid compounds of *P. capillacea* USWE crude extract which positively reflected on the contents of these compounds in Jew's Mallow *C. olitorious* L., when comparing to WE. Carotenes are indispensable to the plants and act as precursors for the biosynthesis of phytohormones and strigolactones, improve the plant development and responses to unstable environmental, and serve as a source of pro-vitamin A. Thus, USWE is an attractive novel technology enhancing the efficiency of seaweed liquid extract on Jew's Mallow. The CROPWAT model has shown that an adequate amount of water is vital, especially during the initial growth stage of Jew's Mallow, but also in other stages. Therefore, it is important to adopt efficient irrigation practices to maximize yields while reducing adverse effects on water resources.

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