EFFECT OF LIQUID SEAWEED EXTRACT ON POTTED GROWTH OF *Eustoma grandiflorum* (Raf.) Shinners) †

[EFECTO DEL EXTRACTO LÍQUIDO DE ALGAS MARINAS EN EL CRECIMIENTO EN MACETA DE *Eustoma grandiflorum* (Raf.) Shinners)]

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SUMMARY

Background. The use of liquid algae extract as a biostimulant when applied to leaves or roots, improves crop development and consequently yield, since by stimulating natural processes they benefit from the use of nutrients and increases resistance to conditions of biotic and / or abiotic stress. **Objective.** This investigation evaluated the response of two varieties of *Eustoma grandiflorum* to foliar and root application of algae extracts, grown in pots under nursery conditions. **Methodology.** Macroalgae extract was applied in a foliar way (20 mL spray) and directly to the substrate (50 mL), while drinking water was applied as the control. **Results.** The parameters obtained indicate that the foliar application has noticeably effects on var. Rosie for the variables root length, number of shoots, and number of leaves; while for var. Florida, the variables total plant length and number of shoots obtained greater significance. The method of application via substrate generates positive effects on the total length and stem length for var. Rosie, although the variables with the greatest noticeably were obtained from var. Florida for root length, number of leaves, and flower diameter. **Implications.** The low doses of the algae extracts, application form and the handling of cultural tasks during the growth of *E. grandiflorum*, contribute to a better absorption and/or efficiency of the fertilizers in the crop. **Conclusion.** The application of algae extracts favors the production of *E. grandiflorum* under nursery conditions, making their use a viable option to minimize the application of conventional fertilizers, thereby attenuating the effects on the environment.
Keywords: Horticulture; Lisianthus; mexican algae; seaweed extract.

RESUMEN

Antecedentes. El uso de extracto liquido de algas como bioestimulante cuando se aplican a las hojas o en las raices, mejora el desarrollo del cultivo y consecuentemente el rendimiento, ya que mediante la estimulación de procesos naturales benefician el aprovechamiento de nutrientes e incrementa la resistencia a condiciones de estrés biótico y/o abiótico. **Objetivo.** Esta investigación evaluó la respuesta de dos variedades de *Eustoma grandiflorum* a la aplicación foliar y radicular de extractos de algas, cultivadas en macetas en condiciones de vivero. **Metodología.** El extracto de macroalgas se aplicó de forma foliar (20 mL) y directamente al sustrato (50 mL), mientras que el agua potable se aplicó como control. **Resultados**. Los parametros obtenidos indican que la aplicación foliar tiene efectos notables sobre la var. Rosie para las variables longitud de la raíz, número de brotes y número de hojas; mientras que para la var. Florida, las variables longitud total de la planta y número de brotes obtuvieron mayor importancia. El método de aplicación a través del sustrato genera efectos positivos sobre la longitud total y la longitud del tallo para las var. Rosie, aunque las variables con mayor notoriedad se obtuvieron de var. Florida para la longitud de la raíz, el número de hojas y el diámetro de la flor. **Implicaciones.** Las bajas dosis de los extractos de algas, forma de aplicación y el manejo de labores culturales durante el cremiento de *E. grandiflorum*, contribuyen a una mejor absorción y/o eficiencia de los fertilizantes en el cultivo. **Conclusión**. La aplicación de extractos de algas favorece la producción de *E. grandiflorum* en condiciones de vivero, lo que hace que su uso sea una opción viable para minimizar la aplicación de fertilizantes convencionales, atenuando así los efectos sobre el medio ambiente.
Palabras clave: Horticultura; Lisianthus; algas mexicanas; extracto de algas.

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INTRODUCTION

Mexico occupies the third place in the global ornamental industry, with more than 20,000 ha for its production. Morelos, Jalisco, State of Mexico, and Puebla lead 90% of the production that generates 1.72 million pesos in the domestic consumption of flowers and plants, and generate $77 million from export to the USA, with 96.7%, and 3.3% to Canada. The national market is a well-established industry, with high-quality production of high-value species, such as *Rosa* spp., *Lilium* spp., *Gerbera* spp., *Chrysanthemum* spp., *Strelitzia reginae*, *Dianthus caryophyllus*, *Limonium sinuatum*, *Bellis perennis*, and representative flowers from Mexico, like *Euphorbia pulcherrima* and *Tagetes erecta* (Ramírez and Camacho, 2017).

Eustoma grandiflorum (known as lisianthus in Mexico) is an ornamental plant of increasing demand. This species is native to the arid areas of the southern USA and northern Mexico (Gómez-Pérez *et al.,* 2014). It is an herbaceous plant of the Gentianaceae family, which form a rosette of leaves on a rigid stem that reaches heights ranging between 0.50 and 0.90 m. The native plants have blue to purple flowers, but the hybrids have different shades (Camargo *et al.*, 2004).

Eustoma grandiflorum has become a favorite species for consumers in the flower market, as a cut flower, due to its exceptional flowers and its long life in a vase (Shimizu-Yumoto and Ichimura 2010). There are several cultures of *E. grandiflorum* with morphological variations in color, size, and shape of the flower (Ashrafi and Rezaei 2018). Despite its increasing demand in the national and international markets and broad scope as a crop (Cruz-Crespo *et al.,* 2006), there are still no statistics on its cultivation and commercialization (Rojas-Morales *et al.,* 2017).

This crop's nutrition is an important factor in maximizing its production. There is indiscriminate use of agrochemicals in several regions, and excessive cultural practices have created serious consequences for the environment. In addition, the high costs of fertilizers in Mexico have put them out of the reach of producers, resulting in low yields, low incomes, and low-quality produce (Basak *et al.,* 2018). The implementation of biological control and organic nutrition with biofertilizers could reduce the dependence on agrochemicals, making its production less hazardous to the environment and the consumer. Additionally, the application of these products could reduce the use of chemical fertilizers, increase the plants performance and resistance against water and temperature stress, and positively influence their growth and physiology (Shaheen *et al.,* 2013).

There is currently an increased interest in natural products that stimulate the growth of plants, particularly toward algal biomass as a source of plant growth biostimulants (Calvo *et al.,* 2014) for sustainable agriculture (Khan *et al.,* 2009). Algae have long been viewed a valuable source of food and traditional remedies. Over the centuries, various types of macroalgae, such as *Gelidium robustum* and *Chondracanthus* spp. were grown and harvested in coastal areas. Other important commercial applications of algae are in the production of healthcare products and cosmetics, as well as in the biochemical industry (Michalak *et al.,* 2016).

In the context of agricultural applications, the beneficial properties of algae result from their permanent exposure to abiotic and biotic stress. Due to the demanding living conditions, these organisms have developed mechanisms that protect them from drought, salinity, changing the light intensity, frost, and colonization by BACTERIA or fungi. As a result, algal cells contain a wide variety of plant growth-promoting substances, such as auxins, cytokinins, betaines, gibberellins, and organic substances, including amino acids, macronutrients, and trace elements (Abd *et al.,* 2008; Sathya *et al.,* 2013). These cell extracts are natural bioactive materials soluble in water. As organic fertilizers, that promotes the germination of seeds and increase the development and yield of crops, particularly if they are used in the production of the ornamental sector (Norrie and Keathler, 2006; Khan *et al.,* 2009; Ahmed *et al*., 2012). When the aqueous extract of *Sargassum* spp. was applied to the soil and foliar pathway in a vine plantation, it enhanced fruit yield and increased the chlorophyll content (Zermeño *et al.,* 2015). The application of the extracts of the species of seaweed *Padina gymnospora* and *Sargassum liebmannii* yielded significant results in percentages of germination in seeds and optimum growth in plants of *Solanum lycopersicum* L. (Hernández-Herrera *et al.,* 2014). García *et al.,* (2014) demonstrated that the application of seaweed extracts in the cultivation of *Gerbera jamesonii* was significant in yield and production. Anbuchezhian *et al.,* (2015) mentioned that the great biofertilizers are derived from marine algae, and improve soil quality and yields considerably.

Marine algal species *Ascophyllum*, *Ecklonia*, and *Fucus,* are commonly utilized as fertilizers containing amounts of nitrogen and potassium comparable to animal manure and organic fertilizers, but with a low phosphorus content. Application rates, frequency, and timing of the treatments vary with species, season, geographical location, and local environmental variables. Important ancillary benefits of seaweed products for crop production include the improvement of damage caused by insects and bacterial or fungal diseases (Trejo *et al.,* 2018).

Given the above, and based on the importance that is being given to the ornamental sector in Mexico, mainly in the state of Jalisco, this research evaluated the effect of *Macrocystis pyrifera* seaweed extracts on two varieties of *E. grandiflorum*, grown in pots under nursery conditions.

MATERIALS AND METHODS

Preparation of liquid seaweed extract (LSE)

The seaweed *M. pyrifera*, used in this study, was collected from the intertidal zone at low tide, in April and May 2017, from the coast of Ensenada Baja California in Mexico (31° 52' 4'' LN, 116° 36' 5'' LO). The seaweed was collected by hand and washed with seawater to remove debris, shells, and sand. Samples were transported to the laboratory in plastic bags, washed with tap water to remove surface salt, oven-dried at 60 °C for 72 h, and then ground in an electric mill (IKA-M 20) to less than 0.50 mm. This milled material (100 g) of each sample was subjected to acid digestion and analyzed by atomic absorption spectrophotometry for mineral analysis of Na, K, Ca and P (by colorimetry) based on standard procedures (Association of Official Analytical Chemists [AOAC] 1990). Then, 100 g of each sample was added to 1 L of distilled water with constant stirring for 15 min, followed by autoclaving at 121 °C for 1 h at 1.21 kg^{-1} cm^{-2}. The hot extracts were filtered through Whatman No. 40 filter paper and stored. The liquid seaweed extracts (LSEs) were designated as stock solution. Furthermore, the pH and electrical conductivity (EC, dS m^{-1}) of the LSEs were measured using a pH meter and conductivity meter, respectively. Finally, the color of the seaweed extracts was observed visually. All determinations were performed in triplicate.

Establishment of the experiment inside the nursery garden

The experiment was carried out during the year 2018, in the nursery garden of the University Center of Biological and Agricultural Sciences of the University of Guadalajara in Jalisco, Mexico (20°45′LN, 103° 31′LW; 1650 masl).

In the experiments, two varieties of *E. grandiflorum* seedlings (Florida and Rosie; Figure 1) were used. Both varieties are long day (less early) because of their tendency toward erect growth, producing an abundance of flowers. These varieties were transplanted in February 2018, when they had an average height of 15.24 cm for var. Florida, and 11.14 cm for var. Rosie, placing them in pots of 20.5 cm in circumference and 16 cm in height. A substrate mixture (leaf soil, volcanic rock, corn stubble without corn and peat moss in 3:5:1:1 ratio, p/p) was homogenized, and 4.5 L added to the pots. The filled pots were placed in a double row inside the nursery, which was constituted by a roof covered with milky white plastic with 25% opacity, in addition to monofilament shade mesh and black polyethylene raffia, stabilizing the retractable ultraviolet rays, and anti-aphids mesh on the periphery of the nursery, with black ground cover on the floor.

During the first seven days following the transplant of *E. grandiflorum* var. Florida and var. Rosie, the substrate was only moistened with 300 mL of drinking water every 72 h, to maintain the turgidity and viability of the seedling.

After this acclimatization period, all the plants were subjected to the same nutritional treatment and agronomic management, which consisted of three liquid fertilizations of monoammonium phosphate (12-52-00, 3 g L^{-1}) at a dosage of 150 mL $plant^{-1}$, with 72-h intervals between applications. Then, a granulated application of Nitrofoska® (21-08-11) at 1.5 g per plant was applied at the foot of the plant. After 15 days, the granulated fertilization was repeated, adding 2 g $plant^{-1}$ of urea (46-00-00). Additionally, a Tradecorp® AZ microelement fertilizer (0.5 g L^{-1}) was added at a dosage of 200 mL per plant as a nutritional supplement in the vegetative stage.

The nutrient solution was modified at the time of the flowering stage, which consisted of four liquid fertilizations of potassium nitrate (12-00-46) and two applications of calcium nitrate (12-00-00 + 23Ca) prepared at 3 g L^{-1}, applied at a dosage of 150 mL per plant, with 72-h intervals between applications.

Three different treatments were carried out: *M. pyrifera* seaweed extract (LSE) applied at 1% (20 mL foliar spray); *M. pyrifera* (LSE) applied at 1% directly to the substrate (50 mL); drinking water applied (control). The cultivation was carried out along with the nutritional applications. The control of pests and diseases as most important and this involved scheduled explorations every 48 h in random plants, observing possible alterations in the morphology and normal functioning of physiological activities, roots, stems, leaves, and flower buds, as well as the programming of activities, such as defoliation or elimination of apical meristems for the development of an optimal canopy.

Figure 1. Establishment of the experiment in pots inside the nursery A) *Eustoma grandiflorum* var. Florida, B) *Eustoma grandiflorum* var. Rosie.

For the phytosanitary control, two preventive applications of propamocarb (1 ml L^{-1}) were carried out at a dosage of 150 mL per plant, with a biweekly interval between applications after transplantation to avoid the complex of fungi causing damping off. For the epigeal part of the plant, CAPTAN® (1 g L^{-1}) was applied, and cypermethrin (2 ml L^{-1}) was sprinkled using a flat cone nozzle until the plant was saturated.

Evaluated variables

A randomized complete block experimental design was used with three repetitions for each treatment. Variable measurements were made in nine randomly selected plants per experimental unit (nine plants per treatment). Stem length was measured by means of a tape measure, using the basal area of the plant until the presence of the first floral button as the reference (From the base of the stem to the formation of the flower calyx). Total length was measured by the use of a tape measure, using the apical meristem to the length of the main root as the reference (From the apical meristem to the main root). For number of leaves, those branches that presented apical meristems were counted, as these gave indications of generating floral buttons. Days to flowering were evaluated at the end of the vegetative stage and the beginning of the flotation stage. The number of buttons per plant was determined when the plant had three open buttons. Flower diameter was measured using a digital Vernier (Once the button opened and the flower was ready, measurements were taken from the tip of a petal to the terminal part of the opposite petal). For root length, a measuring tape was used, and the base of the stem to the maximum length of the root was used as the reference. Days until flowering were measured in chronological order by selecting fully formed floral buttons about to open, until senescence.

Statistical analysis

Variance analyzes were performed to detect significant differences in effects between treatments. The treatments were compared using Tukey's test (p < 0.05). The analyzes were performed using the Statgraphics® Centurion XV statistical package for Windows.

RESULT AND DISCUSSION

Regarding the morphological and growth parameters of the two varieties of *E. grandiflorum* plants, in this investigation, the influence of the different forms of application of the organic fertilizer on stem length, total length, the number of leaves, size of the floral button, flower diameter, and root length were determined (Figure 2).

Macroelements in seaweeds and physicochemical content of LSE

The results obtained from the nutrient analysis showed the presence of Na, K, Ca and P macroelements (g·100 g^{-1}, dry weight) at concentrations of 1.72 ± 0.40, 5.56 ± 2.0, 3.20 ± 0.40 and 0.17 ± 0.05, respectively (Values are average ± standard error; $n = 3$). The pH values for the LSE were alkaline, and the EC was 1–1.3 dS cm^{-1}, and the color was brownish red.

Previous research reported that *M. pyrifera* has a high content of organic material, in addition to minerals, vitamins, carbohydrates, proteins, lipids, and phytohormones, compounds that make algae rich in nutrients (Rodríguez-Montesinos and Hernández-Carmona 1991). Hernández-Carmona *et al.*, (2012) mentioned that the most abundant minerals in *M. pyrifera* are Mg, Ca, P, K, Cl, and I. The mineral content of *M. pyrifera* used in this research was generally consistent with the typical values for this seaweed from other countries (Beratto-Ramos *et al.*, 2019; García *et al.*, 2018; Astorga-España *et al.*, 2017) and in Mexico (Canales, 2000; Ondarza and Ricones, 2008) Quantitative ranges for the various components in algae may vary according to season, location, and analytical methods (Castro-González *et al.*, 1996; Khotimchenko *et al.*, 2002).

Stem length

The extract of the seaweed *M. pyrifera* showed no significant effects on the stem length of *E. grandiflorum* var. Florida relative to the control plants. The average maximum stem lengths were, respectively, 23.80 cm in the plants that received the foliar application, 20.8 cm in those that received the extract directly to the substrate, and just 18.4 cm in the control plants (Figure 2A). Similarly, *E. grandiflorum* var. Rosie also showed no significant results relative to the control. The average maximum stem lengths were, respectively, 24.5 cm in plants that received the application directly to the substrate, 23.5 cm in plants that received foliar application, and 22 cm in the control plants (Figure 2B). Despite not having significant effects on any of the varieties, it can be said that the length of stems obtained in this experiment with the application of both treatments is of good quality, considering that they are within the intervals reported by various authors. Camargo *et al.*, (2004) observed a length of 30.5 cm with *E. grandiflorum* var. Echo Blue. Backes *et al.*, (2007) obtained stems of 47.19, 42.22, 51.19, and 52.22 cm in length in the varieties Echo Champagne, Mariachi, Puer White, Balboa Yellow, and Ávila Blue Rim, respectively. Hernández-Herrera *et al.*, (2014) observed that the application of the seaweed extracts of *Ulva lactuca* and *Padina gymnoespora* via substrate in *S. lycopersicum* plants responded to a high germination percentage effect and, consequently, increased vigor of the stem length. Macroalgae are highly promising as a source of bioactive compounds with potential to promote plant growth, vigor, and yield (Zheng *et al.*, 2016; Fan *et al.*, 2013).

Total length of the plant

The extract of the algae *M. pyrifera* did show significant effects ($p < 0.05$) on the total length of *E. grandiflorum* var. Florida that were treated with foliar and substrate applications. The plants that received foliar algae extract application were longer, with an average value of 51 cm, and the plants that received the extract directly to the substrate had an average value less than 50 cm compared with the control plants which had an average total plant length of 38.7 cm (Figure 2C). The plants of *E. grandiflorum* var. Rosie that received the application of the extract on the substrate averaged 50.19 cm in total length, compared with the average of 49.53 cm for the plants exposed to foliar application, and 37 cm for the control plants (Figure 2D). The results obtained in this investigation demonstrate that considerable lengths can be obtained even when the root system is confined to a container. Bajpai (2016) indicated that algae have active organic compounds as growth regulators. Di Filippo-Herrera *et al*., (2019) affirmed that extracts based on *M. pyrifera* were effective as a growth biostimulant in mung beans. Sunarpi *et al.*, (2019) noticed that the liquid fertilizer of the algae *Sargassum crassifolium* and *Sargassum aquifolium* significantly induced the height of the *Oryza sativa* plant.

Number of sheets

The extract of *M. pyrifera* applied as a foliar spray to the plants of *E grandiflorum* var. Florida resulted in an average of 8 leaves, while the plants treated with the algae extract to the substrate presented an average of 10 leaves, and the control plants had an average of 3.7 leaves (Figure 3A). Following application of the extract of algae to the substrate, *E. grandiflorum* var. Rosie plants presented an average of 8 leaves, and the plants exposed to foliar treatment displayed a total of 7 leaves, whereas the control only obtained a total of 3 leaves (Figure 3B). This trend could be due to the biological activity of algae extracts, which are especially attractive as a natural source of bioactive molecules that promote plant growth (Khan *et al.*, 2009). When Díaz-Leguizamón *et al.,* (2016) compared foliar and substrate applications of algae extracts (*Ascophyllum nodosum*) to *Solanum quitoense,* foliar application was the most effective, obtaining a greater number of leaves and foliar area. The generation of the number of leaves with both applications of seaweed via foliar and substrate can be due to its composition, consisting of organic matter and NPK in the available forms, which led to the optimal nutritional status of *E. grandiflorum* plants (Yagoub *et al.*, 2012).

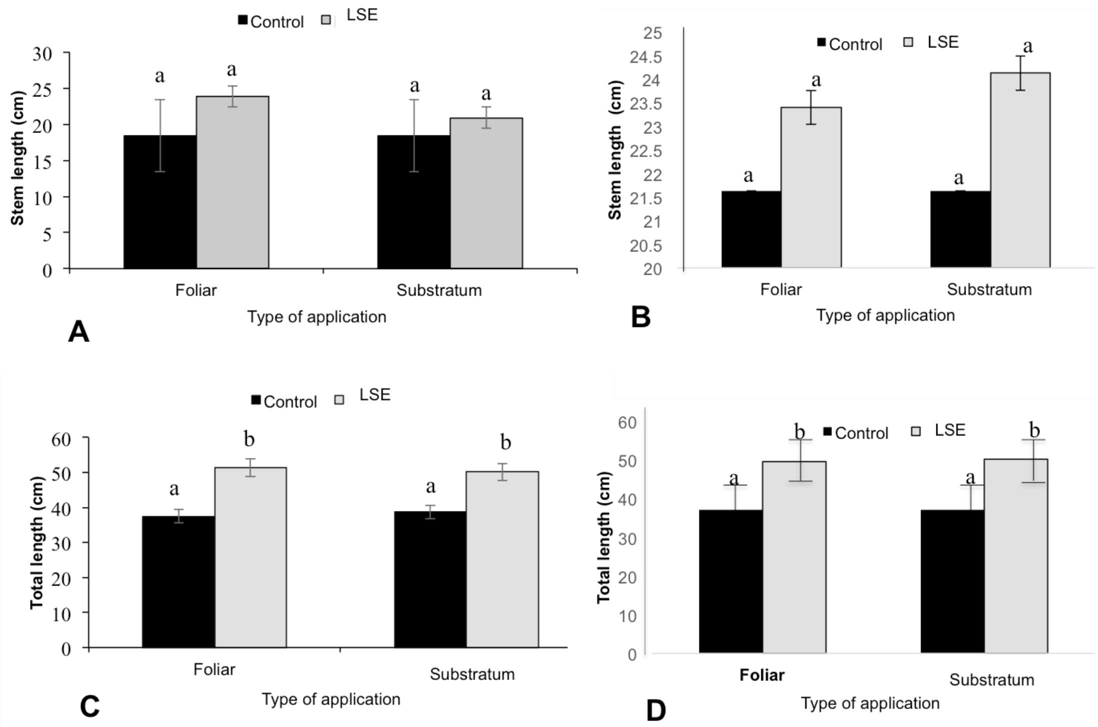

Figure 2. Effect of liquid seaweed extract treatment applications *Macrocystis piryfera* in substrate and foliar aplications in two varieties of *Eustoma grandiflorum*. A) Stem length var. Florida. B) Stem length var. Rosie. C) Total lenght of the plant var. Florida. D) Total lenght of the plant var. Rosie. For all treatments, values are the mean of 3 replicates (nine plants per treatment). Means with different letters represent statistically significant differences according to Tukey's test (p≤ 0.05).

Floral button size

The extract of *M. pyrifera* applied as a foliar spray to the plants of *E. grandiflorum* var. Florida had a significantly favorable effect on the formation of flower buds, with an average of 20 cm relative to the control plants (13 cm). In contrast, the plants treated with the extract applied directly on to the substrate did not show a significant effect on the formation of flower buds (Figure 3C). Similarly, there was a significant favorable effect of foliar application of LSE to *Eustoma grandiflorum* var. Rosie had a, significant favorable effect obtaining flower buds with an average of 18 cm relative to the control plants (13 cm). However, the plants treated with the extract applied directly to the substrate did not show a significant favorable effect on the formation of flower buds (Figure 3D). Martinez-Lozano *et al.*, (2003) reported an increase in the number of flower buds and size after two applications of seaweed extract to safflower. This finding is consistent with the results obtained, given the significant increase in the number of flowers when applying the algae extract at 1%, and perhaps that level is optimal to stimulate s growth and development, and at a higher amount could hamper the plant's metabolic process. Martinez (1995) improved the number of branches in rose bushes by applying 1% seaweed treatment, via initial foliar spray, followed by the treatment via the substrate.

It was observed in *E. grandiflorum* that the application of the algae extract accelerated its flowering, concluding that the algae augmented the metabolism of flowering and leaf growth.

Flower diameter

Treatment of *E. grandiflorum* var. Florida with seaweed extract dramatically increased the flower diameter, from a minimum of 35.7 mm (control) to averages of 62.45 (foliar spray) and 72.12 mm (applied directly to the substrate) (Figure 4A). Likewise, in *E. grandiflorum* var. Rosie, the corresponding values were 61 (foliar spray) and 72 mm (substrate application), respectively (Figure 4B). According to García *et al.*, (2014), the application of commercial seaweed in the cultivation of *Gerbera jamesonii* yielded significant results for the diameter of chapters and highly significant data for the number of leaves and stem size. Trejo *et al.*, (2018) mentioned that algal extracts increase the quality of flower and fruit to obtain the highest antioxidant capacity. The same authors further suggested the use of algal extracts as a viable option to minimize the application of conventional fertilizers, thereby attenuating the effects on the environment and improving the health of the population.

Root length

The extract of the algae *M. pyrifera* showed significant effects ($p < 0.05$) on the root length of *E. grandiflorum* var. Florida that were treated with foliar and substrate applications. It can be seen that the plants that received the application of the algae extract directly to the substrate showed a larger root length, averaging 29.2 cm compared with less than 27.5 cm for the plants that received the foliar extract, and 20.29 cm for the control plants (Figure 4C). Regarding *E. grandiflorum* var. Rosie, the average root lengths were 26 (substrate application), 26.13 (foliar spray), and 15.47 cm (control plants), respectively (Figure 4D). According to Anisimov *et al.,* (2014) the use of brown algae *Saccharina japónica* and *Sargassum pallidum* have a positive effect on the length of the roots of soybean seedlings in comparation with use algae green Metting *et al.,* (1990) described the physiological responses derived from the application of seaweed as increased nutrient mobilization, development of a vigorous root system, increased chlorophyll content and leaf area and delayed senescence of various fruits. Briceño-Domínguez *et al.,* (2014) explained the enhanced growth of *S. lycopersicum* roots was due to the amount of auxin in the applied *M. pyrifera* extracts.

Algae ingredients include macro and microelement nutrients, amino acids, vitamins, cytokinins, auxins, and abscisic acid, which affect cell metabolism in treated plants, leading to increased growth and crop yield (Wightman *et al.,* 1980; Stirk *et al.,* 2004; Nabti *et al.,* 2016) and contributing to the maintenance of plant health.

Macroalgae-based fertilizers used in different crop production systems have improved crop productivity. In addition, Sivasankari *et al.,* (2006) noted the potential of *Chaetomorpha antennina* and *Rosenvingea intricata* as an organic fertilizer to promote the growth of *Abelmoschus esculentus* and *Raphanus sativus*.

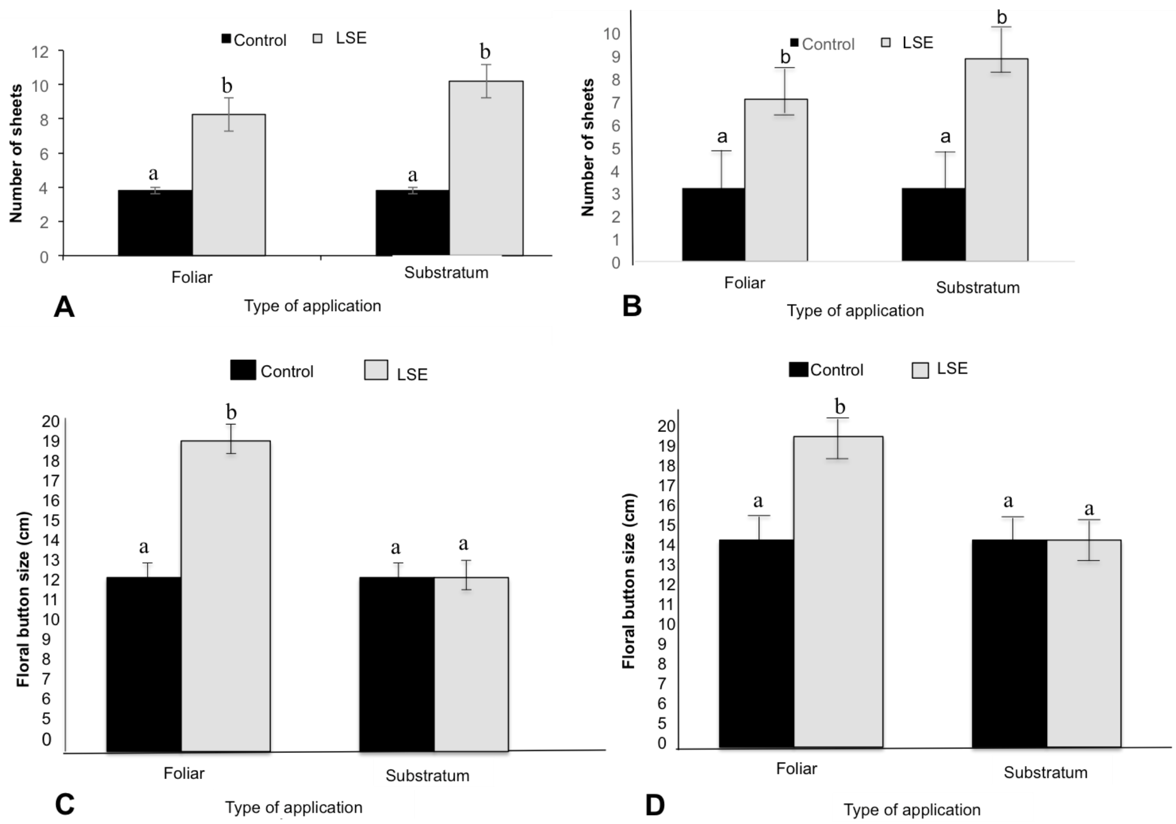

Figure 3. Effect of liquid seaweed extract treatment applications *Macrocystis piryfera* in substrate and foliar aplications in two varieties of *Eustoma grandiflorum*. A) Number of sheets var. Florida. B) Number of sheets var. Rosie. C) Floral button size var. Florida. D) Floral button size var. Rosie. For all treatments, values are the mean of 3 replicates (nine plants per treatment). Means with different letters represent statistically significant differences according to Tukey's test (p≤ 0.05).

It has been reported that the enzymes associated with seaweed extracts reinforce the treated plants' defenses, nutrition and physiology, providing enhanced resistance to stress (Biocentral México, 2017). Furthermore, when applied via substrate and foliar, algae extracts fix nitrogen to the air, which helps improve crop nutrition and vigor (Tuhy *et al.*, 2013). Khan *et al.* (2009) listed the physiological effects caused by macroalgae extracts and their possible bioactivity mechanisms applied via the substrate, as modulating root exudates, altering the root architecture, improving the absorption of water and nutrients, and functioning as suppressants of diseases and soil nematodes. Meanwhile, foliar application increases photosynthetic efficiency, modulates phytohormones, repels insects, reduces perspiration, and improves stomatal conductance (Khan *et al.*, 2009).

CONCLUSIONS

It is believed that solid fertilizers provide greater efficacy to promote growth and crop yield compared with liquid fertilizers; However, with this research, we prove that liquid organic fertilizers can offer sustainable agriculture, as specific compounds in the liquid organic fertilizers, such as chitin, humic and fulvic acids, and other biopolymers, could be biostimulants for plants if used in the correct concentrations. The application of *M. pyrifera* macroalgae extract was effective in the production of ornamental species, such as *E. grandiflorum*. This study provides information to encourage producers to use organic products in agricultural production, as well as the use of Mexican seaweed resources.

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Conflict of interest. The authors confirm that there are no known conflicts of interest associated with this publication.

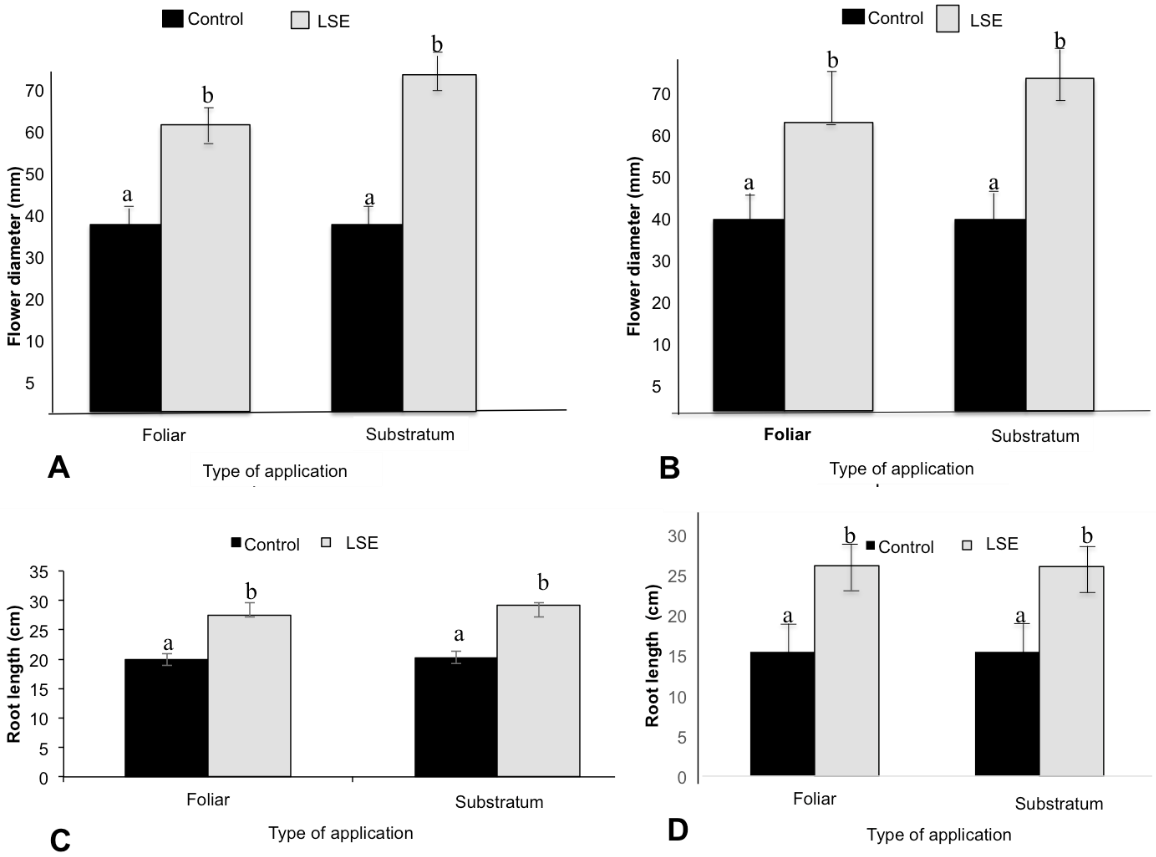

Figure 4. Effect of liquid seaweed extract treatment applications *Macrocystis piryfera* in substrate and foliar aplications in two varieties of *Eustoma grandiflorum*. A) Flower diameter var. Florida. B) Flower diameter var. Rosie. C) Root length var. Florida. D) Root lenght var. Rosie. For all treatments, values are the mean of 3 replicates (nine plants per treatment). Means with different letters represent statistically significant differences according to Tukey's test (p≤ 0.05).

Compliance with ethical standards. The research work did not involve human subjects. The work was carried out in accordance with the respective guidelines of the experimental fields of the Centro Universitario de Ciencias Biologicas y Agropecuarias (CUCBA).

Data availability. Data are available with the corresponding author (pauvela73@hotmail.com) upon reasonable request.

REFERENCES

Abd, E.l., Moniem, E.A. and Abd-Allah, A.S.E. 2008. Effect of Green Alga Cells Extract as Foliar Spray on Vegetative Growth, Yield and Berries Quality of Superior Grapevines. *Journal of Agriculture and Environmental Sciences,* 4, pp. 427–433.

Ahmed, Y.M. and Shalaby, E.A. 2012. Effect of different seaweed extracts and compost on vegetative growth, yield and fruit quality of cucumber. *Journal of Horticultural Science and Ornamental Plants,* 4, pp. 235–240.

Anbuchezhian, R., Karuppiah, V. and Li, Z. 2015. Prospect of Marine Algae for Production of Industrially Important Chemicals. In: Das D. (eds) *Algal Biorefinery: An Integrated Approach*. Springer, Cham.

Anisimov, M.M. and Chaikina, E.L. 2014. Effect of seaweed extracts on the growth of seedling roots of soybean (*Glycine max* (L.) Merr.) seasonal changes in the activity. *International Journal of Current Research and Academic Review,* 2, pp. 19–23.

AOAC (Association of Official Analytical Chemists). 1990. Official methods of analysis, AOAC 15th edn. (P) method 965.01, Washington, DC. p12. https://law.resource.org/pub/us/cfr/ibr/002/aoac.methods.1.1990.pdf. (Accessed on 15 July 2019).

Ashrafi, N. and Rezaei, N.A. 2018. Lisianthus response to salinity stress. *Photosynthetica,* 56 (2), pp. 487–494. http//:dx.doi 10.1007/s11099-017-0709-0

Astorga-España, M.S., Mansilla, J., Ojeda, J., Marambio, S., Rosenfel, F., Mendez, J.P., Rodriguez, P. and Ocaranza. 2017. Nutritional properties of dishes prepared with sub-Antarctic macroalgae an opportunity for healthy eating. *Journal of Applied Phycology,* 29(59), pp. 2399–2406. http//:dx.doi 10.1007/s10811-017-1131-5.

Bajpai, V. 2016. Antimicrobial bioactive compounds from marine algae: A mini review. *Indian Journal of Geo-Marine Sciences,* 45(9), pp. 1076-1085.

Backes, L.F.A.A., Barbosa, J.G., Cecon, P.R., Saraiva, G.J.A., Backes, R.L. and Finger, F.L. 2007. Cultivo hidropônico de lisianto para flor de corte em sistema de fluxo laminar de nutrientes. *Pesquisa Agropecuária Brasileira,* 42(11), pp. 1561-1566.

Basak, R. 2018. Benefits and Costs of Nitrogen Fertilizer Management for Climate Change Mitigation: Focus on India and Mexico. CCAFS Working Paper No. 161. CGIAR Research Program on Climate Change, Agriculture and Food Security (CCAFS): Copenhagen, Denmark. Available online: www.ccafs.cgiar.org (Accessed on 20 May 2018).

Beratto-Ramos, R.P., Castillo-Felices, N.A., Troncoso-Leon, A. and Agurto-Muñoz, C. 2019. Selection criteria for high-value biomass: seasonal and morphological variation of polyphenolic content and antioxidant capacity in two brown macroalgae. *Journal of Applied Phycology*, 31 (1), pp. 653–664.

Bio Central, México. 2017. https://biocentralconference.com/las-algas-marinas-y-su-efecto-bioestimunlate-en-plantas/. (Accessed on 22 may 2019).

Briceño-Domínguez, D., Hernández-Carmona, G., Moyo, M., Stirk, W. and van Staden, J. 2014. Plant growth promoting activity of seaweed liquid extracts produced from Macrocystis pyrifera under different pH and temperature conditions. *Journal of Applied Phycology*, 26 (5), pp. 2203–2210. http//:dx.doi 10.1007/s10811-014-0237-2.

Calvo, P., Nelson, L. and Kloepper, J.W. 2014. Agricultural uses of plant biostimulants. *Plant Soil,* 383, pp. 3–41.

Camargo, M.S., Shimizu, L.K., Saito, M.A., Kameoka, C.H., Mello, S.C. and Carmello, Q.A.C. 2004. Crescimento e absorção de nutrientes pelo Lisianthus (*Eustoma grandiflorum*) cultivado em solo. *Horticultura Brasileira*, 22, pp.143-146. http//:dx.doi 10.1590/S010205362004000100030.

Canales, L.B. 2000. Enzimas-algas: posibilidades de su uso para estimular la producción agrícola y mejorar los suelos. *Terra,* 17(3), pp. 271-276.

Castro-González, M.I., Pérez-Gil, R., Pérez-Estrella, S. and Carrillo-Domínguez, S. 1996. Chemical composition of the green alga *Ulva lactuca*. *Ciencias Marinas,* 22, pp. 205–213.

Cruz-Crespo, E., Arévalo-Galarza, L., Cano-Medrano, R. and Gaytán-Acuña, E.A.

2006. Soluciones pulso en la calidad postcosecha de lisianthus (*Eustoma grandiflorum* Raf.) cv. ‘Echo Blue’. *Agricultura Técnica en México,* (32), pp. 191-200.

Di Filippo-Herrera, D.A., Muñoz-Ochoa, M., Hernández-Herrera, R.M. and Hernández-Carmona, G. 2019. Biostimulant activity of individual and blended seaweed extracts on the germination and growth of the mung vean. *Journal of Applied Phycology*, 31, pp. 2025–2037. http//:dx.doi 10.1007/s10811-018-1680-2.

Díaz-Leguizamón, J.J., Chingaté-Cruz, O.F., SánchezReinoso, A.D. and Restrepo-Díaz, H. 2016. The effect of foliar applications of a bio-stimulant derived from algae extract on the physiological behavior of lulo seedlings (*Solanum quitoense* cv. *Septentrionale*). *Ciencia e Investigación Agraria,* 43(1), pp. 25-37.

Fan, D., Hodges, D.M., Critchley, A.T. and Prithiviraj, B. 2013. A commercial extract of brown macroalga (*Ascophyllum nodosum*) affects yield and the nutritional quality of spinach in vitro. *Communications in Soil Science and Plant Analysis*, 44, pp. 1873-1884. http//:dx.doi 10.1080/00103624.2013.790404.

García, S.M.L., De Luna Vega, A., Zúñiga, C.C., Bañuelos Gutiérrez, O.A. and Silva Echeverría, M. 2014. Efecto de algas marinas en el desarrollo de *Gerbera jamesonii* (Asteraceae). *e-CUCBA Universidad de Guadalajara, Centro Universitarios de Ciencias Biologicas y Agropecuarias,* 2, pp. 39–45.

García, F.E., Plaza-Cazónb, J.V., Montesinos, N.E., Donatib, R. and Littercd, M.I. 2018. Combined strategy for removal of Reactive Black 5 by biomass sorption on *Macrocystis pyrifera* and zerovalent iron nanoparticles. *Journal of Environmental Management,* 207(1), pp. 70-79. http//:dx.doi 10.1016/j.jenvman.2017.11.002.

Gómez-Pérez, L., Valdez-Aguilar, L.A., Sandoval-Rangel, A., Benavides-Mendoza, A., Mendoza-Villarreal, R., and Castillo-González, A.M. 2014. Calcium ameliorates the tolerance of lisianthus (*Eustoma grandiflorum* (Raf.) Shinn.) to alkalinity in irrigation water. *Horticulture Science,* 49, pp. 807-811.

Hernández-Carmona, G., Rodríguez-Montesinos, Y., Arvizu-Higuera, D., Reyes-Tisnado, R., Murillo-Álvarez, I. and Muñoz-Ochoa, M. 2012. Avances tecnológicos en la producción de alginatos en México. *Ingenieria Investigación y Tecnología,* 13(2), pp. 155-168.

Hernández-Herrera, M., Santacruz-Ruvalcaba, F., Ruiz-López, M., Norrie, J. and Hernández-Carmona, G. 2014. Effect of liquid seaweed extracts on growth of tomato seedlings (*Solanum lycopersicum* L.). *Journal of Applied Phycology*, 26, pp. 619–628. http//:dx.doi 10.1007/s10811-013-0078-4.

Khan, W., Rayirath, U.P., Subramanian, U.P., Jitesh, M.N., Rayorath, P., Hodges, D.M., Critchley, D.M., Craigie, J.S., Norrie, J. and Prithiviraj, B. 2009. Seaweed extracts as biostimulants of plant growth and development. *Journal of Plant Growth Regulation*, 28, pp. 386-399. http//:dx.doi 10.1007/s00344-009-9103-x.

Khotimchenko, S.V., Vaskovsky, V.E. and Titlyanova, T.V. 2002. Fatty Acids of Marine Algae from the Pacific Coast of North California. *Botanica Marina,* 45, pp. 17-22.

Martinez, L.S. 1995. Efecto de un extracto de algas y varios fitorreguladores sobre el cultivo de papa *Solanum tuberosum*. *Instituto Tecnológico y de Estudios Superiores de Monterrey* 104 pp.

Martínez-Lozano, S.J., Verde-Star, J., Gámez-González, H., Moreno-Limón, S., Cárdenas-Ávila, M.L., Núñez-González, M.A. and Lara-Hernández, E.M. 2003. Efecto del producto comercial de algaenzimas en el crecimiento y desarrollo del rosal enano Rosa chinensis. *Phyton International Journal of Experimental Botany*, 2003, pp.183-188.

Metting, B., Zimmerman, W.J., Crouch, I. and Van Staden, J. 1990. Agronomic uses of seaweed and microalgae. In: Akatsuka I (ed) *Introduction to Applied Phycology*. SPB, The Hague 589–628 pp.

Michalak, I., Górka, B., Wieczorek, P.P., Rój, E., Lipok, J. and Łeska, B. 2016. Supercritical fluid extraction of algae enhances level sof biologically active compounds promoting plant growth. *European Journal Phycology,* 51, pp. 243–252.

Nabti, E., Jha, B. and Hartmann, A. 2016. Impact of seaweeds on agricultural crop production as biofertilizer. *International Journal of Environmental Science and Technology,* 14(5), pp. 1119-1134. https://doi.org/10.1007/s13762-016-1202-1.

Norrie, J. and Keathley, J.P. 2006. Benefits of Ascophyllum nodosum marine-plant extract applications to ‘Thompson seedless’ grape production. *Acta Horticulturae*, 727, pp. 243–247.

Ondarza, M.B. and Rincones, R.E. 2008. El cultivo de algas marinas: alternativa industrial en acuacultura sustentable a mediano y largo plazo. *Universidad Autónoma de Tamaulipas*, 3(2), pp. 68-73.

Ramírez, E. and Camacho, Z. 2017. México, líder mundial en la industria Ornamental. Revista Fortuna. https://revistafortuna.com.mx/contenido/2017/08/30/mexico-lider-mundial-la-industria-ornamental/ (Accessed on 14 august 2019).

Rodríguez-Montesinos, Y.E. and Hernández-Carmona, G. 1991. Variación estacional y geográfica de la composición química de *Macrocystis pyrifera* en la costa occidental de Baja California. *Ciencias Marinas*, 17(3), pp. 91-107.

Rojas-Morales, A., Belem, G., Morales-Rosales, E.J., Morales-Morales, E.J., Estrada-Campuzano, G., Franco-Mora, O., and López-Sandoval, J.A. 2017. Índice de verdor de lisianthus (*Eustoma grandiflorum* (raf.) shinners) en función de la concentración de 6-bencilaminopurina. *Revista Fitotecnia Mexicana*, 40(4), pp. 461-469.

Sathya, R., Kanaga, N., Sankar, P. and Jeeva, C. 2013. Antioxidant properties of phlorotannins from brown seaweed *Cystoseira trinodis* (Forsskål) C. Agardh. *Arabian Journal of Chemistry*, 10, pp. S2608–S2614.

Shaheen, M.A., Abd ElWahab, S.M., El-Morsy, F.M. and Ahmed, A.S.S. 2013. Effect of organic and bio-fertilizers as a partial substitute for NPK mineral fertilizer on vegetative growth, leaf mineral content, yield and fruit quality of superior grapevine. *Journal of Horticultural Science and Ornamental* Plants, 5, pp. 151–159.

Shimizu-Yumoto, H. and Ichimura, K. 2010. Postharvest Physiology and Technology of Cut Eustoma Flowers. *Journal of the Japanese Society for Horticultural Science*, 79(3), pp. 227-238. http//:dx.doi 10.2503/jjshs1.79.227

Sivasankari, S., Venkatesalu, V., Anantharaj, M. and Chandrasekaran, M. 2006. Effect of seaweed extracts on the growth and biochemical constituents of *Vigna sinensis*. *Bioresource. Technology*, 97, pp. 1745-1751. http//:dx.doi 10.1016/j.biortech.2005.06.016.

Stirk, W.A., Arthur, G.D., Lourens, A.F., NovákM, O., StrnadJ, M. and van Staden, J. 2004. Changes in cytokinin and auxin concentrations in seaweed concentrates when stored at an elevated temperature. *Journal of Applied Phycology*, 16, p. 3. http//:dx.doi 10.1023/B:JAPH.0000019057.45363.f5.

Sunarpi, H., Faisal, A., Fadhillah, E., Putri, S., Azmiati, N., Hidayatun, N., Suparman, S.W. and Eka, S.P. 2019. Effect of Indonesian Macroalgae Based Solid and Liquid Fertilizers on the Growth and Yield of Rice (*Oryza sativa*). *Asian Journal of Plant Sciences*, 18(1), pp. 15-20. http//:dx.doi:10.3923/ajps.2019.15.20.

Trejo, V.R., Sánchez, S.A., Fortis, H.M., Preciado, P.R., Gallegos, R.M.A., Cruz, R.C.A. and Vázquez, V.C. 2018. Effect of Seaweed Aqueous Extracts and Compost on Vegetative Growth, Yield, and Nutraceutical Quality of Cucumber (*Cucumis sativus* L.). *Fruit Horticultural Agronomy*, 8(11), p. 264. http//:dx.doi 10.3390/agronomy8110264.

Tuhy, L., Chowańska, J. and Chojnacka, K., 2013. Seaweed extracts as biostimulants of plant growth. *Baz Technology*, 67(7), pp. 636–641.

Wightman, F., Schneider, E.A. and Thimann, K.V. 1980. Hormonal factors controlling the initiation and development of lateral roots. *Physiologia Plantarum*, 49(3), pp. 304-314. http//:dx.doi 10.1111/j.1399-3054.1980.tb02669.x.

Yagoub, S.O., Ahmed, W.M.A. and Mariod, A.A. 2012. Effect of Urea, NPK and Compost on Growth and Yield of Soybean (*Glycine max L*.), in Semi-Arid Region of Sudan. *International Scholarly Research Network Agronomy*, 2012, pp. 1-6.

Zermeño, G.A., Rodríguez, L.B.R., Alvarez, M.A.I., Rodríguez, H.R., Cárdenas, P.J.O. and López, M.J.P. 2015. Extracto de alga marina y su relación con fotosíntesis y rendimiento de una plantación de vid. *Revista Mexicana de Ciencias Agrícolas*, 12, pp. 2437-2446.

Zheng, S., Jiang, J., He, M., Zou, S. and Wang, C. 2016. Effect of kelp waste extracts on the growth and development of Pakchoi (*Brassica chinensis* L.). *Scientific Reports*, 6(38683), pp. 1-9. http//:dx.doi:10.1038/srep38683.