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Research Article Effect of Two Seaweed Products and Equivalent Mineral Treatments on Lettuce (*Lactuca sativa* L.) Growth

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Abstract

Background and Objective: Seaweeds and seaweed products have been applied in vegetable production systems for many years. Seaweeds and their extracts or by-products may have beneficial effects on vegetable production through increased growth. Possible mechanisms include the nutrient concentration of the seaweed product, the presence of organic compounds such as plant growth regulators (PGR), or through effects on soil processes. **Materials and Methods:** The effect of application rates and whether mineral nutrients alone can account for plant growth responses, was assessed by using two commercial seaweed products (Maxicrop[®] and Seasol[®]) applied at four rates (0, 1, 2, 4 × recommended rate) as well as ashed product. **Results:** The results show that both Maxicrop and Seasol can significantly increase crop performance. Maxicrop increased shoot biomass, root biomass and leaf chlorophyll content above that of the mineral treatments by 66, 47 and 9%, respectively, while Seasol increased root biomass only (by 50%). By ashing the seaweed product or preparing a mineral-only nutrient solution, we have confirmed that seaweed products can improve plant growth beyond that of mineral nutrients alone. However, seaweed fertilizer products with very low nutrient content is adequate, growth may be greater than equivalent mineral nutrient applications. **Conclusion:** These pot trials demonstrate the potential value of some seaweed fertilizer products for nursery production and other containerised plant systems. Further research is required to clarify the role of various plant growth regulators, biostimulants and soil conditioning compounds.

Key words: Biofertilizer, mineral content, ash, biomass, nutrient solution, chlorophyll

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

An adequate supply of essential plant nutrients, solar energy, carbon dioxide, water and heat influence plant growth^{1,2}. Chemical fertilisers have been successfully used to increase food production, however, excessive use of these to address nutrient deficiencies of the soil has created several environmental problems, leading to increased pollution, acidification and production of greenhouse gases³. A major strategy to counteract the decline in environmental quality is to promote practices that support sustainable agriculture. Productivity can only be achieved by use of ecologically sustainable inputs⁴.

There is global interest in the use of seaweed concentrates in agriculture and horticulture⁵ and there are a number of seaweed products on the market^{6,7}. These products may cause differing effects on plant growth and development because there are a range of possible factors present in these products⁸ and the various settings in which they used such as field-scale usage and applications in containerised plant systems^{9,10}.

Marine seaweeds are high in PGRs and their application to crops are reported to have beneficial effects on plant growth and yield which seems to be unrelated to the nutrient content. Seaweed products have therefore been used both as supplements or substitutes for inorganic fertilisers, because seaweeds have high levels of macro- and micro nutrients as well as PGR^{11,12}. Inorganic amendments are likely to diminish in availability as global energy demands and costs increase and finite nutrient resources are depleted. Unlike inorganic amendments, organic amendments such as seaweed products can also improve the health of soil by increasing the organic carbon content, the availability of plant nutrients, microbial biomass and activity and by enhancing soil structural stability¹³.

Reports indicate that seaweed extracts can improve the growth rate and yields of crop plants¹⁴, including vegetables, trees, cut flower crops and grain crops. This may be due to the effect of PGRs in the extract or the effect on soil micro-organisms, soil structure and soil physics and macro- and micro-nutrient availability¹⁵. Owing to the wide range of elicited physiological responses, it is probable that more than one group of PGRs is involved with cytokinins^{16,17}, auxins¹⁸ and gibberellins¹⁹ all reported to be present in seaweed extracts. Despite these reports, there is some doubt about the efficacy of seaweed products beyond their known nutrient effect^{20,21}.

To evaluate the effects of two seaweed products (Maxicrop[®] and Seasol[®]), a greenhouse pot trial was

conducted using lettuce (*Lactuca sativa* L.), a widely-grown salad vegetable²². Results from bioassays²³ indicate that Seasol was effective in promoting initial seedling shoot and root elongation in field pea seedlings. However, it is uncertain whether the effects observed in the bioassay are also manifested at the whole plant level.

The aims of this research were, therefore, to (a) investigate the effect of increasing application rates of Seasol and Maxicrop seaweed products and (b) to evaluate a range of allied treatments that included or excluded the corresponding PGRs and mineral nutrients, on the growth and development of lettuce growing under glasshouse conditions. We hypothesised that the effect of the seaweed products was not exclusively due to the nutrient content. We tested this by ashing the products to remove organic compounds (e.g., PGRs) and by preparing mineral fertiliser blends with identical rates of N, P and K to the recommended rates of the seaweed products.

MATERIAL AND METHODS

Experimental conditions: The experiments were conducted in a greenhouse pot trial on the campus of the University of New England, Armidale, New South Wales, Australia. A greybrown granite-derived Chromosol with a sandy to sandy-loam texture used for the experiments was collected from the Kirby Research Farm, UNE (30.487° S and 151.638° E, elevation 1,050 m). Properties for the soil are given in Table 1.

Lettuce was germinated in petri dishes and after the radicle had reached 5 mm in length four seedlings were sown into plastic pots (175 mm deep \times 185 mm diameter) containing 3 kg of soil. After 1 week, the seedlings were thinned to two per pot and watered daily thereafter. The experiment was conducted under natural light with a 25/15°C day/night temperature regime.

Experimental design and treatments: This experiment used two commercial seaweed extract products. Maxicrop is made from *Ascophyllum nodosum*²⁴ and Seasol is made from *Durvillaea potatorum, Durvillaea antarctica* and *A. nodosum*²⁵. Both products are certified for use in organic farming in Australia^{24,25}.

Seasol (SS) was applied at the recommended dilution rate of 1:600 and Maxicrop (MC) at the recommended dilution rate of 1:375 as well as 2 and 4 times the recommended rates. The concentrations of N, P and K in Maxicrop and Seasol were determined (Table 2) and mineral nutrient solutions were prepared with the equivalent amounts of N, P and K to that found in Maxicrop (MC-NPK) and Seasol (SS-NPK). Laboratory-grade urea ($[NH_2]_2CO$), tripotassium phosphate (K_3PO_4) and potassium sulphate (K_2SO_4) in distilled water were used to make these solutions. Finally, Seasol and Maxicrop were heated in crucibles for 48 h at 450 °C to create SS-ash and MC-ash treatments, with presumably similar mineral contents as the unashed products but no organic compounds such as PGRs. A control treatment consisted of 100 mL deionized water while all other treatments consisted of a total of 100 mL, with the additives included in that volume. The eleven treatments are listed in Table 3. The solutions were applied as a soil drench. Each treatment was replicated four

Table 1: Soil properties for the soil used in the pot trial

Parameters	Units	Values
pH (1:5 water)	-	5.86
Organic carbon	%	0.65
Nitrogen	%	0.04
Sulphur	mg kg ⁻¹	10.00
Phosphorus ^c	mg kg ⁻¹	47.00
Potassium ^a	meq100 g^{-1}	0.38
Calcium ^a	meq100 g ⁻¹	19.00
Magnesium ^a	meq100 g^{-1}	19.00
Aluminium ^b	meq100 g^{-1}	0.11
Sodiumª	meq100 g^{-1}	0.26
Chloride	mg kg ⁻¹	15.00
CEC	meq100 g^{-1}	38.70
EC	dS m ⁻¹	0.06
EC (se)	$dS m^{-1}$	0.40

a: Ammonium acetate extraction, b: KCl extraction, c: Colwell

Table 2:	Fertiliser requirement (kg ha ⁻¹) for lettuce (Napier 2004) and amount of
	nitrogen, phosphorus and potassium (kg ha-1) in the recommended
	rate of Maxicrop [®] and Seasol [®]

Nutrients	Lettuce	Maxicrop®	Seasol®
Nitrogen	200	230	11
Phosphorus	100	61	2
Potassium	150	109	174

times in a completely randomised design. Treatments were applied one day after transplanting and were repeated every 14 days until the conclusion of experiment at harvest (60 days after sowing).

Measurements and analysis: Plant height was measured weekly. At harvest, leaf chlorophyll content was recorded using a Minolta SPAD-502 chlorophyll meter. The measurements were taken from top, middle and bottom leaves and the average of these leaves for each plant was calculated. Shoot and root biomass was collected, oven-dried to constant weight and the dry weights recorded.

Data analysis was carried out using²⁶ R version 3.4.1. Linear regression was used to assess the increase in height over time and ANOVA was used to evaluate treatment effects. Assumptions of heterogeneous variances and normal distributions were confirmed and significantly different means were separated using 95% confidence intervals $(CI = 1.96 \times standard error)^{27}$.

RESULTS

Plant height over time: Increasing the application rate significantly increased lettuce plant height for Maxicrop ($p_{slope} < 0.001$) but not Seasol (Fig. 1). The slopes for the control and all SS treatments were similar, whereas the slopes for MC treatments were significantly greater than the control. The overall order of responses for the Maxicrop treatments was MC4 > MC2 > MC1 > MC-ash > MC-NPK.

Plant biomass and chlorophyll: Application of Maxicrop resulted in a significant increase in shoot dry weight of lettuce with the effect increasing with concentration. However,

Treatments	Water (mL)	Seaweed product (mL)	Amount of minera
Control	100.00	0	0
SS1	99.84	0.16 Seasol [®]	0
SS2	99.68	0.32 Seasol [®]	0
SS4	99.36	0.64 Seasol [®]	0
SS-ash	100.00	0.16 Seasol [®]	0
SS-NPK	100.00	0	Urea = 0.17 g
			$KH_2PO_4 = 0.025 g$
			$K_2 SO_4 = 6.6 g$
MC1	99.74	0.26 Maxicrop®	0
MC2	99.48	0.52 Maxicrop®	0
MC4	98.96	1.04 Maxicrop®	0
MC-ash	100.00	0.26 Maxicrop®	0
MC-NPK	100.00	0	Urea = 4 g
			$KH_2PO_4 = 0.87 g$
			$K_2SO_4 = 5 q$



Fig. 1: Effect of two seaweed products on plant height (cm)

MC: Maxicrop treatments, SS: Seasol treatments, b: Slope coefficient and its standard error (n = 4)

Table 4: Effect of Maxicrop* and Seasol* seaweed products on shoot and root dry weight (g pot⁻¹) and leaf chlorophyll content of lettuce

Treatments	Shoot dry weight (g pot ⁻¹)	Root dry weight (g pot ⁻¹)	Chlorophyll content
Control	0.09±0.008	0.03±0.011	30.60±0.608
MC1	0.29±0.051	0.11±0.017	33.86±1.343
MC2	0.82±0.059	0.32±0.010	35.26±0.586
MC4	3.07±0.120	1.17±0.098	38.55±1.375
MC-ash	0.19±0.017	0.08±0.013	31.50±0.386
MC-NPK	0.16±0.015	0.07±0.009	30.81±1.676
SS1	0.11±0.008	0.06±0.007	31.21±0.777
SS2	0.12±0.015	0.06±0.005	32.79±1.133
SS4	0.13±0.023	0.07±0.012	32.85±0.297
SS-ash	0.10±0.006	0.03±0.005	30.88±1.277
SS-NPK	0.11±0.013	0.05±0.006	31.16±0.933

Means \pm 95% confidence intervals are presented

increasing the application rate of Seasol did not significantly increase shoot dry weight (Table 4). MC1 was significantly more effective than both MC-ash and MC-NPK (by about 65%) which were themselves equivalent in nutrient content. Lettuce shoot dry weight for SS1, SS-ash and SS-NPK were not significantly different.

Lettuce root dry weight showed a similar pattern to shoot dry weight for all treatments (Table 4). One exception was that of SS1, which was higher than SS-ash by 50%. Overall, Maxicrop had a significant positive effect on root growth compared with the control and had a 46% increase in root growth compared with MC-ash and MC-NPK, whereas the Seasol showed little or no benefit for root growth. Lettuce leaf chlorophyll content showed a similar pattern to shoot dry weight for all treatments (Table 4). Chlorophyll in MC1 was 9% higher than for the mineral treatments (MC-ash and MC-NPK).

DISCUSSION

Application rates of seaweed products: The highest concentration of Maxicrop (MC4) had the greatest effect of the various treatments. In general, Maxicrop had a significant effect on lettuce for all measured parameters. In contrast, Seasol had only a minor significant effect on root growth. Most of the literature reporting the beneficial effects of seaweed products have indicated that plant growth and chlorophyll of

treated plants was significantly increased^{19,28,29}. In general, plants treated with increasing rates of the Maxicrop had increased lettuce height and biomass accumulation. This trend was not observed for Seasol, even at $4 \times$ the recommended rate. A study on the effect of seaweed liquid fertiliser on okra (*Abelmoschus esculentus*), showed that a moderate concentration (20%), resulted in the greatest shoot length, root length and yield, when compared to other concentrations ranging¹² from 10-100%. This contradictory result may be because the beneficial effect of seaweed extract is dependent on the type of seaweed fertiliser product and the crop species under study as well as the local environmental conditions³⁰.

The results presented in this study are based on lettuce, a crop with high N and P requirements (200 and 100 kg ha⁻¹, respectively)³¹. The very low concentration of N and P (11 and 2 kg ha⁻¹, respectively) in the recommended rate of Seasol is the most likely reason that it did not improve biomass production, a conclusion confirmed by the lack of response to the equivalent NPK treatment. In contrast, the recommended rate of Maxicrop has a relatively high concentration of N and P (230 and 61 kg ha⁻¹, respectively) compared with the standard mineral fertiliser recommendations in the region.

While adequate level of macro- and micro-nutrients are critical for maximising plant growth^{7,32}, some of published reports found that the concentration of mineral nutrients present in the seaweed products could not directly account for the observed growth responses^{21,33}. The beneficial effect may therefore relate to the various plant growth regulators present in these seaweed extract^{7,34}. Increases in plant growth have been attributed to seaweed components such as macro- and micro-element nutrients, amino acids, vitamins, cytokinins, auxins and gibberellins and others PGRs substances^{7,35,36}. All these compounds can affect plant metabolism leading to enhanced growth, development and stress tolerance^{37,38}.

Comparison of responses to seaweed compared to equivalent ash and NPK applications: By (a) ashing the products to remove organic compounds (e.g. PGRs) and applying the ash as a mineral fertiliser and (b) preparing a mineral fertiliser blend with identical rates of N, P and K as the recommended rates of the seaweed content. While there were almost no differences among the Seasol treatments (due to the low overall nutrient content), Maxicrop showed a clear benefit in plant growth above that observed for both the equivalent ash treatment and the equivalent NPK treatment, increasing shoot biomass, root biomass and leaf chlorophyll content by 66, 47 and 9%, respectively.

The beneficial effects of seaweed application in this investigation might be due to organic compounds such as

PGRs present in Maxicrop and are responsible for some of the increase in crop performance^{7,39}. Maxicrop could also improve the fertility of soil as the algin, the hydrophilic, colloidal substances found in seaweed extracts, may help in conditioning the soil, facilitating aeration, moisture retention and adsorption of nutrient elements⁴⁰. Other mechanisms which are involved include increased root proliferation as the root dry weight was increased by the seaweed treatment. This may have then allowed plant to obtain more nutrients by exploring a greater soil volume¹⁰. The increase root weight dry of seaweed treated-plants may also related to better canopy establishment and better interception of light, increasing photosynthesis and translocation of assimilates⁴¹.

Plant height, shoot and root dry biomass and leaf chlorophyll were unaffected by the ash and NPK treatments but were affected by the Maxicrop treatments. This proved that NPK which applied to plant test suited to amount mineral consist in seaweed products and also NPK could be the only nutrient needed by the test plant³⁵.

CONCLUSION

By ashing the seaweed product or preparing a mineralonly nutrient solution, we have confirmed that seaweed products can improve plant growth beyond that of mineral nutrients alone. However, the nutrient content of Seasol was very low and was therefore ineffective. Seaweed fertiliser products with very low nutrient analyses are unlikely to improve plant growth without supplementary nutrient additions from other sources. Subject to further testing, it would therefore need to be used in high fertility soils or for pasture or crops with lower nutrient requirements. In contrast, where the nutrient content of the seaweed product is adequate or high, growth has been demonstrated to be greater than equivalent mineral nutrient applications. A range of plant growth regulators, biostimulants and soil conditioning compounds may be responsible, alone or in combination for the observed plant responses. Further research is required to clarify the role of these compounds in seaweed extracts in stimulating plant growth. Field testing is also warranted to confirm these results on a broader scale, although these pot trials demonstrate the potential value of some seaweed fertiliser products for nursery production and other containerised plant systems.

SIGNIFICANCE STATEMENT

This study discovered the potential effect of seaweeds on the growth of lettuce that can be beneficial for researchers and growers. This study will help the researchers to uncover the critical areas of horticulture that many researchers were not able to explore. Thus a new theory on using seaweed to replace chemical fertilizer may be arrived at.

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