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Effect of applications of seaweed extract on sugarcane yield in Australia

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- Abstract Sugarcane growers are seeking innovations that will increase their productivity and profitability and enable sustainable farming practices. The aim of this research was to evaluate the effect of applying seaweed extract on sugarcane production and economics in Australia under commercial conditions and over an extended time frame (2014 to 2019). Seaweed extract significantly improved sugarcane productivity by an average of 17% (cane yield, sugar yield), and increased grower returns by 18% or AU\$1,158/ha. This is the first publication to demonstrate the efficacy of seaweed extracts applied by sub-surface drip irrigation across such an extended number of cropping seasons.
- Key words Biostimulants, Saccharum, productivity, agriculture, revenue, soil biology

INTRODUCTION

Sugarcane is a global agricultural crop grown in tropical and subtropical regions for sucrose, which is consumed or converted into a renewable bioenergy source (ethanol). The Australian sugarcane industry contributes \$2.5 billion to the national economy and exports 80% of the sugar produced (Sugar Research Australia 2018). Most Australian sugarcane is grown along the tropical Queensland coast. Maintaining productivity and profitability is becoming more challenging for growers because of increasing climate volatility and fluctuating global sugar prices and international currencies.

There are many reports of improved agricultural productivity and crop quality due to the application of seaweed extracts (Arioli *et al.* 2015; Shukla *et al.* 2019). Interest in using seaweed extracts in agriculture worldwide is growing rapidly particularly because of their attributes in improving the efficiency of plant-nutrient use and tolerance of abiotic and biotic stresses (Brown and Saa 2015). For example, field studies have demonstrated that the use of seaweed extracts increased yield and quality in crops as diverse as wine grapes, tomatoes, broccoli and strawberries (Khan *et al.* 2009; Mattner *et al.* 2013; 2018; Shukla *et al.* 2019; Hussain *et al.* 2021). Other studies using tomato, soybean and *Arabidopsis* have reported that the application of seaweed extract enhanced plant tolerance of abiotic stresses such as drought, salt and freezing (Nair *et al.* 2012; Martynenko *et al.* 2016; Santaniello *et al.* 2017; Goni *et al.* 2018; Jithesh *et al.* 2019). Transcriptomics studies have found the application of seaweed extract can activate plant-defence-system responses (Goni *et al.* 2016; Islam *et al.* 2020; Omidbakhshfard *et al.* 2020). When applied to soil, seaweed extracts intrinsically provide a range of nutrients for soil microbes to assimilate and can alter their community structure to the benefit of crop growth (Renaut *et al.* 2019; Hussain *et al.* 2021).

Seaweed extracts have proven to be practical to use in agriculture. They are easy to apply through soil irrigation, foliar sprays, drenching, soaking or with synthetic fertilisers (Arioli *et al.* 2015; Shukla *et al.* 2019). Seaweed extracts are complementary to crop-protection solutions, and integral to regenerative agriculture and the transition to more sustainable farming practices. The use of seaweed extracts is appealing because they are inherently safe to use in farming, they are biodegradable, and their efficacy is resilient to challenging climate anomalies. Their use is economically viable when incorporated into conventional farming such as Australian strawberry and wine-grape production (Mattner *et al.* 2018; Arioli *et al.* 2021).

A previous study showed that the use of a seaweed extract increased sugarcane yield by 10% and reduced the need for inorganic fertilisers and estimated emissions of greenhouse gases (Singh *et al.* 2018). Despite the potential, there are few scientific publications about the effect of seaweed extracts on sugarcane production and economics in Australia (Farnsworth and Arioli 2018; Arioli *et al.* 2020). Furthermore, the biostimulant industry is advocating for the testing of seaweed extracts in real-world growing conditions and on a commercial scale (Ricci *et al.* 2019).

The aim of our study was to determine the effects of applications of seaweed extract on sugarcane yield and production economics in Australia. Our hypothesis was that sugarcane treated with recurring applications of a seaweed extract would increase yield across growing seasons and locations and improve revenue for growers.

METHODS

Commercial-scale sugarcane field trials

Trials were conducted across five growing seasons (2014–19) on two commercial sugarcane farms near Mareeba in northern Queensland, Australia (Table 1). Each field trial consisted of one randomly selected treatment plot and one adjacent control plot. The trial sites had sub-surface drip irrigation (SSDI) with driplines buried at a depth of around 30 cm, delivering water and nutrients straight to the root zone. Experimental variation was minimised by selecting field trial locations that had a uniform managed history.

The seaweed extract (Seasol, Seasol International, Australia) was made from two large cold-water species, *Durvillaea potatorum* (native to the Southern Hemisphere) and *Ascophyllum nodosum* (native to the Northern Hemisphere), using an alkaline extraction process. Detailed information about the seaweed extract such as composition, mineral content and manufacture has been published previously (Arioli *et al.* 2015; Wite *et al.* 2015). The seaweed extract was applied at 10 L/ha per month (1:400 concentration in irrigation water) in the trials and compared with untreated controls of the equivalent amount of irrigation water. In cases where it was too wet to irrigate due to excessive tropical rainfall, the application dose was doubled in the next application. Equal amounts of total irrigation water, fertiliser and pesticides were applied to the treatment and control plots at each field trial site, based on the growers' standard practices. The treatment application rate and concentration were chosen based on other efficacy studies conducted in Australia (Mattner *et al.* 2013, 2018; Farnsworth and Arioli 2018; Arioli *et al.* 2020).

Field trial 1 was started in 2014 using plant cane. The trial plots in this trial received N 145, P 50, K 71, S 23 (kg/ha) in each crop cycle. The soil type at this site was categorised as a pale, coarse, sandy gravel, clay loam duplex. Field trial 2 was started in 2014 using a first-ration crop. The trial plots in this field trial received N 180, P 36, K 90 (kg/ha) in each crop cycle. The soil type at this site was categorised as a dark, silty loam duplex. The sugarcane field trial sites were monitored at harvest using mechanical harvesters and the trial GPS coordinates. Cane yield and sugar content data for each harvest were tracked using harvest bin numbers and yield and quality results provided by the sugar mill to the grower at the time of harvest. The revenue for each year was based on QSL sugar prices at the time of harvest (\$400/t in 2015, \$660/t in 2016, \$420/t in 2017, \$430/t in 2018, \$399/t in 2019).

Trial site	Location	Cultivar	Timeframe	Number of seasons	Treatment	Number of applications per season	Plot size (ha)
Trial 1	Mareeba,	Q2280	2014–2019	5	Seaweed Extract 10 L/ha/month	5-10	4.2
I liai I	QLD	QZZO	2014-2019 5		Control	5-10	4.2
Trial 2	Mareeba,	Q2280	2014–2019	F	Seaweed Extract 10 L/ha/month	7-9	7.5
i nai Z	QLD	QZZ0 ⁽¹⁾	2014-2019	5	Control	7-9	3.5
20 measu	rements (bas	sed on 2 sit	tes x 2 plots =	4 plots; 4 pl	ots x 5 seasons = 20 measurement	s)	

Table 1. Locations, cultivar, timeframes, treatment, plot size information of the field trials.

Statistical analysis

The response variables analysed were cane yield (t/ha), commercial cane sugar content (CCS), sugar yield (t/ha) and revenue (\$/ha).

An analysis of variance (ANOVA) on each response variable divided the data from the field trials into three levels of variation – between sites, between plots within sites, and between seasons within plots. The variation between plots (experimental units) was then used to compare treatments (seaweed extract vs control), and the variation within plots was used to compare seasons and to evaluate the treatment by season interaction. If the variation between sites was estimated to be minimal, the effect of site was not incorporated in the analysis, and just two levels of variation were used in the statistical model.

Confidence intervals (95%) were calculated for the difference in means between the seaweed extract and control treatments (95% CI for difference). It is appropriate to consider this overall difference in making conclusions because the treatment by season interaction was not significant for any of the response variables. The analysis combining all trials across years used Genstat 18th edition (VSNI, UK).

RESULTS

Commercial cane yield

Treatment with seaweed extract significantly (P = 0.011) increased cane yield by an average of 17% (cane t/ha) (Table 2). The seaweed extract improved cane yield compared with the control in each year of the trials. The results show a general decline in cane yield over time, but this is consistent with the aging of the crop across the seasons (Garside *et al.* 2005).

Treatment			Sea	son		
Treatment	2014/15	2015/16	2016/17	2017/18	2018/19	Mean
Control	116	118	97	102	85	104
Seaweed extract	137	142	116	109	101	121
Difference	21	24	19	7	16	17
95% CI for difference						(9, 25)
P-value (Control vs Sea	aweed extrac	t)				0.011

Table 2. Cane yield (t/ha) in the commercial-scale field trials.

Commercial sugar content

Treatment with seaweed extract did not affect sugarcane sugar content (CCS) (Table 3). In each year of the trials, sugar content at harvest differed little between the seaweed extract treatment and the control.

Table 3. Commercial sugarcane sugar content (CCS) in the commercial-scale field trials.

Treatment	Season						
rreatment	2014/15	2015/16	2016/17	2017/18	2018/19	Mean	
Control	15.0	13.8	13.9	14.2	15.1	14.4	
Seaweed extract	14.6	13.6	14.0	14.7	15.4	14.5	
Difference	-0.4	-0.2	0.1	0.5	0.3	0.1	
95% CI for difference						(–1.3, 1.5)	
P-value (Control vs Sea	aweed extract	t)				0.563	

Commercial sugar yield

Treatment with seaweed extract significantly (P = 0.012) increased sugar yield by 17% (sugar t/ha) (Table 4). The seaweed extract improved sugar yield compared with the control in each year of the trials. The results show a decline in sugar yield over time, which is consistent with the aging of the crop across the seasons.

Treatment			Sea	ason					
Treatment	2014/15	2015/16	2016/17	2017/18	2018/19	Mean			
Control	17.4	16.3	13.4	14.5	12.5	14.8			
Seaweed extract	20.0	19.2	16.2	16.0	15.4	17.4			
Difference	2.6	2.9	2.8	1.5	2.9	2.6			
95% CI for difference									
P-value (Control vs Sea	aweed extract	t)	P-value (Control vs Seaweed extract)						

Table 4. Sugar yield (t/ha) in the commercial-scale field trials.

Commercial production economics

Treatment with seaweed extract increased revenue (at the P < 0.1 level used by agronomists) from sugarcane by 18% (AU\$1,158) across 2014–18 (Table 5). The seaweed treatment increased crop revenue compared to the control in each year of the trials. The pattern was consistent across the growing seasons, despite the fluctuations in price of sugar across (AU\$399/t to AU\$660/t).

Treatment	Season						
Treatment	2014/15	2015/16	2016/17	2017/18	2018/19	Mean	
Control	6,960	9,876	5,628	5,422	4,994	6,576	
Seaweed extract	8,020	11,683	6,804	5,996	6,164	7,734	
Difference	1,060	1,807	1,176	574	1,170	1,158	
95% CI for difference						(–700, 3016)	
P-value (Control vs Sea	P-value (Control vs Seaweed extract)						

Table 5. Production economics (revenue AU\$/ha) in the commercial-scale field trials.

DISCUSSION

Our study showed for the first time that the application of a biostimulant seaweed extract to soil can increase sugarcane yield on a commercial scale over five seasons and two soil types in Australia. On average, our trials demonstrated that the seaweed extract increased sugarcane and sugar yield by 17%. Furthermore, the yield response to the seaweed extract was consistent across the five years of the trials (i.e. the season × treatment interaction was not significant). This suggests the effect of the seaweed extract was not cumulative over time and could be generated within a single cropping season. In economic terms, the average crop revenue (AU\$/ha) at the commercial sugarcane farms used in this study was increased by 18% or AU\$1,158 due to using the seaweed extract.

Our cane and sugar yield results are consistent with the few publications reporting the efficacy of seaweed extracts on sugarcane. Singh *et al.* (2018) conducted three seasons of small-plot field trials using plant cane and ratoon crops and found seaweed extract significantly increased cane yield by 12.5% and 8.0%. They also noted that the response was influenced by different application rates of the seaweed extract made from the red algae *Kappaphycus alvarezii*. Deshmukh and Phonde (2013) conducted a single-year small-plot field trial and reported increases in cane yield by 14.1% and sugar yield by 23.1% when the seaweed extract was applied as foliar or soil applications. Devi and Mani (2013) used seaweed extracts made from two different red algae, *K. alvarezii* and *Glacilaria* sp. In their single-year study, they found three foliar applications of high rates of the respective seaweed extracts (at the higher rates) significantly increased cane yield between 18.1 and 19.5%. Karthilkeyan and Shanmugam (2017) conducted a longer-term study using a potassium-rich (11.3% w/w) biostimulant made from *K. alvarezii* and found increases in cane yield between 20.5 and 28.8% across the four seasons of testing. Nori *et al.* (2019) reported that two different seaweed bioformulations increased cane yield by 19.1 and 23.5%. Collectively, the results demonstrate the efficacy of seaweed extracts on sugarcane using plant or ratoon cane, a range of production systems, applying seaweed extract by different methods (foliar and soil) and across different sugarcane growing geographies such as Australia and India.

The application of seaweed extract in the current trial did not change sugar content in sugarcane across the seasons tested. By comparison, the literature reports contrasting results with on sugar content, with some trials finding significant increases and others finding no significant effect when using seaweed extracts (Deshmukh and Phonde 2013; Devi and Mani 2013; Gomathi *et al.* 2017; Karthilkeyan and Shanmugam 2017; Singh *et al.* 2018; Nori *et al.* 2019). The reasons for these differences may be related to the cultivars, production practices and the intensity of sugarcane production, among others.

It is unlikely that the improvements in cane and sugar yields demonstrated in our trials were due to a fertiliser effect of the seaweed extract because the nutrient composition (Wite *et al.* 2015) is low in comparison to the nutrient program. Instead, *Arabidopsis* research using the same seaweed extract used in this study has uncovered a complex mode of action involving the upregulation of plant genes involved in different pathways related to plant growth, defence and stress tolerance, among others (Islam *et al.* 2020). The *Arabidopsis* gene expression responses are consistent with the idea that agricultural biostimulants initiate a series of beneficial plant responses rather than depending on their nutrient composition. Research using tomato and pepper plants has highlighted a link between plant and soil effects (Renaut *et al.* 2019; Hussain *et al.* 2021). In those studies, the application of seaweed extracts prompted concomitant effects: (i) increased growth phenotypes (such as flower and fruit numbers), and (ii) beneficial changes in soil microbiology. Although not integral to our yield analysis, we investigated the soil microbiology at one of our field trials using microbe diversity profiling approaches (using 16S and *nif*H DNA) and found the rhizosphere (soil-root interface) to be enriched in microbes linked to the nitrogen cycle (Appendix). This highlights an additional complexity when studying the effect of seaweed extracts on sugarcane yield.

More broadly our sugarcane productivity improvements are consistent with other Australian crop studies (broccoli, strawberry, tomato, wine grape) using the same type of seaweed extract (Mattner *et al.* 2013, 2018; Arioli *et al.* 2015, 2021; Hussain *et al.* 2021). In terms of application rates, the sugarcane yield improvements parallel strawberry and wine-grape field trials (that used monthly application of 10 L/ha). An important feature in all these field trials is the recurring applications of seaweed extract to the crops during the growing season. In addition, the efficacy of seaweed extract on sugarcane productivity was resilient across an extended number of growing seasons. This finding parallels research showing the same positive effect of seaweed extract on wine-grape yield spanning 6 years (Arioli *et al.* 2020).

The economics of using seaweed extract in sugarcane production is important for growers. We found that the use of seaweed extract improved grower net returns, and this feature was consistent across all the trials and seasons. Some costs may increase for growers from the adoption of the seaweed extract, but these are small relative to the revenue gains found in the current trials. For example, the increased yields from the seaweed extract may attract an additional harvest cost, and the average cost (per season) of the seaweed extracts in the trials was AU\$400 (based on 100 L/ha). Furthermore, SSDI is not universally adopted through the industry, and may require an additional outlay for growers if they wished to apply the seaweed extract using this method. Similarly, economics and production analyses of multi-season field trials in wine grapes and strawberries in Australia and sugarcane in India reported that the use of seaweed extract improved grower returns and resulted in a favourable benefit-to-cost ratio (Gomathi *et al.* 2017; Mattner *et al.* 2018; Singh *et al.* 2018; Nori *et al.* 2019; Arioli *et al.* 2020, 2021). This is an important attribute because the progression from conventional to more sustainable farming practices can be financially challenging for growers. Based on our results, the use of seaweed extract can be integrated into conventional sugarcane operations in an economical way. Overall, our results support the use of seaweed extracts to increase sugarcane productivity and farm revenue.

CONCLUSION

We found that the regular use of a liquid seaweed extract (made from both *Durvillaea potatorum* and *Ascophyllum nodosum*) improved cane and sugar yield, and grower return in Australia. The commercial-scale field-trial experiments found the application of seaweed extract significantly increased cane yield on average by 17% and sugar yield by 17%. Application of seaweed extract also increased grower return by an average of 18%. This is the first publication to demonstrate the efficacy of seaweed extracts applied by sub-surface drip irrigation in commercial-scale field trials across five cropping seasons.

This research is relevant to Australian sugarcane growers because the results were obtained in Australian production conditions, across five cropping seasons. It expands the limited number of Australian sugarcane

research publications related to the use of seaweed extracts. Future sugarcane research will investigate the effect of seaweed extract on soil microbiology (in sugarcane field trials) and the duration between sugarcane replanting.

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APPENDIX - Diversity profiling of soil microbes using roots collected from field trial Site 2

Operational Taxonomic Units (OTUs) of 16S rRNA sequences - Treatment with seaweed extract increased and decreased the representation of certain bacterial members in sugarcane root tissue. A total of 991 OTUs were identified.

Table A. 16S profiling showing the top 5 OTUs taxonomies (at order level) *increased* in sugarcane root tissue (top 5 entries with a \log_2 Fold Change >= 2, P value(adj) < 0.05).

OTU	log ₂ fold change	P value (adj)	Relevance	Taxonomy (Order)
OTU_95	3.3	2.71E-04	Nitrogen cycle	Rhizobiales
OTU_178	2.4	6.48E-03	Unclear	Fibrobacterales
OTU_12	2.3	5.78E-03	Nitrogen cycle	Rhizobiales
OTU_99	2.3	8.12E-03	Unclear	Chitinophagales
OTU_64	2.3	2.16E-03	Nitrogen cycle	Betaproteobacteriales

Table B. 16S profiling showing the top 5 OTUs taxonomies (at order level) *decreased* in sugarcane root tissue (top 5 entries with a \log_2 Fold Change >= -2, P value(adj) < 0.05).

OTU	log ₂ fold change	P value (adj)	Relevance	Taxonomy (Order)
OTU_46	-2.7	8.88E-04	Unclear	Streptosporangiales
OTU_117	-2.5	1.75E-03	Unclear	Catenulisporales
OTU_295	-2.1	1.44E-02	Nitrogen cycle	Rhizobiales
OTU_657	-2.1	1.63E-02	Unclear	Chloroflexales
OTU_111	-2.0	1.94E-02	Unclear	Dongiales

Operational Taxonomy Units of *nifH* gene sequences - Treatment with seaweed extract increased and decreased the representation of certain bacterial members in sugarcane root tissue. A total of 1970 OTUs were identified.

Table C. nifH profiling showing the OTUs, and corresponding bacteria, with *increased* representation in sugarcane roots due to the seaweed extract treatment (top 15 entries with a log₂ Fold Change >2, P value(adj) <0.05).

OTU	log ₂ fold change	P value (adj)	Relevance	Taxonomy (Genus)
OTU_53	7.1	3.92E-18	Bioremediation	Desulfovibrio
OTU_149	6.4	4.32E-17	Nitrogen cycle	Uncultured & nitrogen-fixing bacteria
OTU_39	6.1	2.00E-16	Nitrogen cycle	Burkholderia
OTU_135	5.3	2.86E-15	Nitrogen cycle	Burkholderia
OTU_125	5.8	4.91E-15	Nitrogen cycle	Bradyrhizobium
OTU_35	6.1	9.36E-14	Bioremediation	Geobacter
OTU_134	5.7	3.28E-13	Unclear	Opitutaceae
OTU_16070	5.2	4.77E-13	Bioremediation	Geobacter
OTU_11842	5.0	5.28E-13	Nitrogen cycle	Bradyrhizobium
OTU_27131	5.2	7.47E-13	Nitrogen cycle	Bradyrhizobium
OTU_370	5.0	3.68E-12	Nitrogen cycle	Uncultured & nitrogen-fixing bacteria
OTU_353	4.9	4.58E-12	Bioremediation	Geobacter
OTU_474	4.8	6.82E-12	Nitrogen cycle	Uncultured & nitrogen-fixing bacteria
OTU_612	4.7	1.03E-11	Nitrogen cycle	Bradyrhizobium
OTU_207	5.2	1.22E-11	Oxidation	Methylococcus

OTU	log ₂ fold change	P value (adj)	Relevance	Taxonomy (Genus)
OTU_67	-7.7	5.30E-28	Nitrogen cycle	Bradyrhizobium
OTU_8	-7.5	1.24E-23	Unclear	Pantoea
OTU_179	-6.4	2.42E-22	Nitrogen cycle	Azospirillum
OTU_24392	-6.4	1.53E-19	Unclear	Pelomonones
OTU_69	-6.5	6.82E-18	Nitrogen cycle	Uncultured & nitrogen-fixing bacteria
OTU_14412	-5.0	3.40E-16	Nitrogen cycle	Burkholderia
OTU_247	-5.5	1.23E-13	Nitrogen cycle	Sinorhizobium
OTU_295	-5.4	1.67E-13	Nitrogen cycle	Uncultured & nitrogen-fixing bacteria
OTU_58	-5.5	4.16E-12	Disease	Serratia
OTU_709	-4.9	5.10E-12	Nitrogen cycle	Bradyrhizobium
OTU_429	-4.6	2.80E-11	Nitrogen cycle	Uncultured & nitrogen-fixing bacteria
OTU_743	-4.0	8.61E-10	Nitrogen cycle	Uncultured & nitrogen-fixing bacteria
OTU_930	-3.7	1.67E-09	Nitrogen cycle	Bradyrhizobium
OTU_286	-3.8	2.15E-09	Nitrogen cycle	Uncultured & nitrogen-fixing bacteria
OTU_970	-3.9	2.30E-09	Oxidation	Desulfobulbus

Table D. nifH profiling showing the OTUs, and corresponding bacteria, with *reduced* representation in sugarcane roots due to the seaweed extract treatment (top 15 entries with a log_2 Fold Change > -2, P value(adj) < 0.05).