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Assessing Processing Waste from the Sea Urchin (*Centrostephanus rodgersii*) Fishery as an Organic Fertilizer

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Abstract: The longspined sea urchin, *Centrostephanus rodgersii*, is a climate-driven pest species in south-eastern Australia. The harvest of this species is highly encouraged and in Tasmania, the existing fishery is expanding resulting in a large amount of waste that needs disposal. Research into use of waste products as inputs for organic or biodynamic farming systems can help reduce costs of disposal and keep the industry profitable; by sustaining or incrementing sea urchin harvest the industry can assist in their control. In the current study, urchin waste was dried and finely ground to a powder and applied to tomato plants in a greenhouse to examine the effect on growth and productivity. Urchin waste powder (UWP) had a mineral composition of Ca (40 g 100 g^{-1}), Mg (1.7 g 100 g^{-1}), P (0.03 g 100 g^{-1}), Fe (19.34 mg kg^{-1}) and B (38 mg kg^{-1}), a pH 8.06 in water and an Electrical Conductivity (EC) value of 7.64 dSm^{-1}. Seven different treatment rates of UWP (0.3%; 0.5%; 0.8%; 1%; 2%; 3%; 5%), were added to 10 replicate pots containing 4 kg nutrient-poor potting mix planted with tomato (Variety K1) seedlings. Plant growth, yield, quality attributes and mineral content of tomato were measured under UWP treatments with comparison against a Hoagland solution control. UWP influenced tomato growth and productivity proportional to the quantity applied, however, the Hoagland solution control had a significantly greater yield. Potting mix pH increased from 6.8 to 7 and higher available P was detected in potting mix receiving higher rates of UWP. No phytotoxic effects were detected. The highest UWP treatment matched the Hoagland control in fruit quality and nutritional composition. Processing waste from the sea urchin fishery has potential as organic fertiliser or amendment providing plant-available Ca and some microelements such as Boron.

Keywords: seafood waste; sea urchin; *Centrostephanus rodgersii*; organic amendment; fertilizer; plant growth; tomato

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1. Introduction

Processing of seafood from industrial fisheries results in considerable waste, posing logistical disposal and environmental problems [1]. In Australia, the seafood industry produces more than 50,080 tons of fish waste at the manufacturing stage [2] and on average processors pay more than $200/t for removal to landfill [3]. The reuse and repurposing of waste is of growing importance as the world's population and the amount of waste produced continues to increase [4].

In this context, different sectors of industry and research have explored various ways to repurpose fish waste, for example, some studies have investigated the use of fishery by-products as sources of proteins and lipids in feed [5], extraction of bioactive compounds useful in pharmaceutical and cosmetic products [6] and as fertilizers in agricultural crops [7]. Products made from animal excreta, animal processing wastes or food processing waste can be used to improve the structure and stability of the soil [8–10] in addition to enhancing the yield and quality of the crop plants [11–13]. Investigation into the agronomic benefits of

processing waste as an alternative to synthetic fertilisers or others soils amendments could provide an opportunity to decrease costs while increasing environmental sustainability.

In particular, the waste proceeding from shellfish, mainly bivalves and from sea urchins was deemed potentially useful for the calciferous composition of the shell. A few studies demonstrated the potential use of calciferous waste generated by bivalve farming. Mussel shells (grounded and calcinated) and lime were compared as an amendment in soil with a low pH [14], resulting in comparable increases to soil pH, exchangeable Ca and decreased exchangeable alluminium. The amendment also led to an increase in dry matter yield and concentration of calcium (Ca) in the plants. Sea urchin (*Paracentrotus lividus*) waste was assessed as an amendment in acidic soil proving to significantly increase soil pH and electrical conductivity, available phosphorous (P), active carbonate as well as microbial abundance and activity [15].

Sea urchins represent an important part of invertebrate fisheries and are collected in coastal areas all around the world. Different studies have characterized the mineral composition of various species of sea urchins including *Strongylocentrotus intermedius*, *Mesocentrotus nudus*, *Scaphechinus mirabilis*, and *Echinocardium cordatum* from the Japan sea [16], the red (*Strongylocentrotus franciscanus*) and green (*Strongylocentrotus droebachiensis*) sea urchins from the West and East coasts of Canada, respectively [17] and *Paracentrotus lividus* from the coasts of Sardinia in the Mediterranean sea [15]. A high Ca and relatively high magnesium (Mg) content were found in all species with nitrogen (N), P, and potassium (K) in minor quantities, among the micronutrients identified were iron (Fe), zinc (Zn), manganese (Mn) and copper (Cu). Heavy metals such as cadmium (Cd) and lead (Pd) were found in trace amounts.

In Tasmania, Australia the expanding sea urchin fishing industry target the longspined sea urchin, *Centrostephanus rodgersii*, a large native echinoid of south-eastern Australian coastal waters. The species has expanded its range and abundance over the last few decades due to climate change. So that it is overgrazing reef habitats and harming coastal ecosystems [18]. This species is harvested for its roe and over the last 3 years, approximately 1500 tons were harvested along the east coast of Tasmania. Roe ranges between 5% and 15% of total body weight whilst the remaining parts (guts, test, spines and jaws) are considered waste. Preliminary characterization of the waste parts revealed an interesting composition in macro and micronutrients that could be beneficial in supporting plant nutrition. This study aimed to test the use of waste produced by the longspined sea urchin *C. rodgersii* fishery as a potential mineral fertilizer. We selected tomato as our model species due to its salt tolerance [19] and the expectation that sea urchin waste powder (UWP) is likely to increase the electrical conductivity (EC) of the soil. Using a base potting mix medium with known properties, we investigated the productivity, yield, fruit quality and nutrition of tomato using UWP at increasing rates against a standard nutrient fertiliser regime.

2. Materials and Methods

Longspined sea urchins were harvested between March and May 2017, along the east coast of Tasmania to extract roe for sale for human consumption. The processing waste including tests (endoskeletons), spines and jaws were rinsed with tap water to eliminate salt residue and oven dried for 24 h at 105 °C. Dried material was finely ground using a grinding mill (A11 analytical mill, IKA, Staufen, Germany) and samples were sent to SWEP Laboratory (Victoria, Australia) for nutrient analysis to determine elemental composition and physico-chemical parameters of the UWP. Specifically, P, K, sulphur (S), Ca, Mg, sodium (Na), Fe, Mn, Zn, Cu, cobalt (Co), boron (B), and molybdenum (Mo) were determined with inductively coupled plasma atomic emission spectroscopy (ICP-AES) after acid digestion. Nitrogen was determined by the Dumas method [20]. The pH and EC of the powder were measured in water (ratio 1:5) with a pH and EC reader [21] and organic carbon (C) with LECO carbon analyser following Rayment and Lyons [22].

A potting mix was prepared comprising of 90% composted pine bark, 5% sand, 5% cocopeat, plus 3 kg dolomite 0.5 m^{-3} to produce a consistent growing medium with sufficient structure for plant growth. Seven treatment rates (0.3%; 0.5%; 0.8%; 1%; 2%; 3%;

5% by weight) of UWP were added to 4 kg of the potting mix at the commencement of the trial with ten replicate pots per treatment (Table 1). The UWP was added with one application only at trial start. The potting mix was very low in macronutrients N, P, K and Ca (Table 1). The initial EC and pH were 0.470 dSm^{-1} and 7.30, respectively (Table 1). An additional treatment with a standard Hoagland solution (Table S1) with ten pot replicates was used as the control of which 400 mL was applied twice a week for 12 weeks. One week after the preparation of the pot treatments, three tomato (*Solanum lycopersicum*) seedlings (variety K1) were added to each pot and after two weeks the strongest plant was retained, and the others were discarded (Figure 1). pH and EC measurements of the potting mix for the seven treatments were recorded three days post-planting and before the addition of Hoagland solution in the control treatment and at the conclusion of the trial. Experimental plants underwent a natural photoperiod, in an uncontrolled temperature environment and received automatic irrigation for two minutes, six times over 24 h (Figure 1).

Table 1. Nutrient composition of the potting mix with UWP applied at different rates and the total applied in the Hoagland control.

Element	Unit	Potting Mix	Total Hoagland mg/Pot	Urchin Waste Powder	Urchin Waste Powder Application Rates (g/Pot)						
					0.30%	0.50%	0.80%	1%	2%	3%	5%
					12	20	32	40	80	120	200
N	g 100 g^{-1}	0.0007 ± 0.0001	1950	0.500 ± 0.070	0.060	0.100	0.160	0.200	0.400	0.600	1.00
P	g 100 g^{-1}	0.0029 ± 0.0002	223	0.030 ± 0.003	0.004	0.006	0.010	0.012	0.024	0.036	0.060
K	g 100 g^{-1}	0.164 ± 0.005	1685	0.260 ± 0.030	0.031	0.052	0.083	0.104	0.208	0.312	0.520
Ca	g 100 g^{-1}	0.383 ± 0.010	2076	40.4 ± 0.670	4.85	8.08	12.9	16.1	32.3	48.5	80.8
Mg	g 100 g^{-1}	0.057 ± 0.001	353	1.77 ± 0.020	0.210	0.350	0.570	0.710	1.42	2.12	3.54
Na	g 100 g^{-1}	0.012 ± 0.001	0.0408	1.35 ± 0.160	0.160	0.270	0.430	0.540	1.08	1.62	2.70
S	g 100 g^{-1}	0.0057 ± 0.0013	461	0.470 ± 0.080	0.056	0.094	0.150	0.188	0.376	0.564	0.940
Cu	mg kg^{-1}	0.870 ± 0.100	0.240	0.600 ± 0.120	0.072	0.120	0.190	0.240	0.480	0.720	1.20
Zn	mg kg^{-1}	18.1 ± 1.21	0.600	6.36 ± 2.24	0.760	1.27	2.04	2.54	5.09	7.63	12.7
Fe	mg kg^{-1}	63.3 ± 7.34	3.60	19.3 ± 5.60	2.32	3.87	6.19	7.74	15.4	23.2	38.7
Mn	mg kg^{-1}	38.3 ± 2.63	5.64	1.87 ± 0.960	0.220	0.370	0.600	0.750	1.50	2.24	3.74
Mo	mg kg^{-1}	n/d	0.864	0.114 ± 0.027	0.014	0.023	0.036	0.046	0.091	0.140	0.230
B	mg kg^{-1}	0.680 ± 0.006	3.72	38.1 ± 1.86	4.57	7.62	12.2	15.2	30.5	45.7	76.2
EC	dSm^{-1}	0.470 ± 0.06	1.70	7.64 ± 0.974	0.490	0.510	0.690	0.690	0.860	0.850	1.15
pH Level	1:5 Water	7.30 ± 0.10	5.80	8.06 ± 0.100	7.40	7.50	7.50	7.60	7.60	7.60	7.70

Values represent average ± standard error. Number of replicates (n = 3). Values of EC and pH of UWP treatments represent measurements of the potting mix post treatments application at the start of the trial. The values represent a single measurement per treatment of pooled sub-samples from the ten pot replicates three days post planting of tomato seedlings and cannot be presented with standard errors.

The dynamic of plant growth was recorded with weekly measurements of a range of plant growth and reproductive characteristics. Individual plant height and width were obtained using a ruler, and stem cross-section area (CSA) using Vernier callipers. The number of fully grown branches, flowers and fruits was recorded for each plant weekly. At the end of the trial, the vegetative and reproductive weights of all tomato plants were calculated for each of the eight treatments. Each plant was cut at the base (potting mix surface) and the fresh weight was recorded, then plants were oven dried for 48 h at 60 °C and dry weight and moisture content were calculated. Five branches per plant from each treatment were cut and sent for nutrient analysis. Three replicates of standard potting mix before the addition of UWP and three replicates of potting mix from each treatment (10 g per sample) were collected at the end of the trial, sieved through a 2 mm mesh and air dried for approximately two weeks in aluminium foil trays. Dried samples from each treatment were pooled to make a composite sample and sent for nutrient analysis.

Figure 1. Experimental set up of tomato plants in greenhouse pot trial with UWP addition treatments and Hoagland control. Clockwise the five photos shows five stages of plant growth, from seedlings transplant to tomato fruit maturation.

After 12 weeks, fruits from each plant were counted and weighed, and the total yield per plant was calculated. To assess fruit quality attributes, fruits of similar ripe stages (maturity) were selected for comparison. Colour intensity was recorded with a colour meter (Chroma Meter CR-400, Konica Minolta, Tokyo, Japan) in three spots around the pericarp and values were averaged. The system used to record the colour was the international standard CIE L* a* b* that expresses colour as three values: L* for the lightness from black (0) to white (100), a* from green (−) to red (+), and b* from blue (−) to yellow (+). Hue angle and Chroma were then calculated from each measurement using the following formulas:

$$Hue = \arctan\left(\frac{b}{a}\right)$$

$$Chroma = \left(a^2 + b^2\right)^{0.5}$$

Values of *a** are negative in green tomato and become positive when red colour starts to develop. Negative values represent unripe fruit while a higher hue angle shows fruit in different ripening stages. Red is better represented by the hue angle which explains the colour change associated with the enzymatic degradation of chlorophylls and the appearance of lycopene [23]. A minimum positive hue angle represents fully ripe fruits and shows an intense red colour.

Fruit firmness was measured with a compression meter (Güss fruit texture analyser, Strand, South Africa) which expresses deformation of the pericarp in millimetres in response to the applied load of 50 g for 0.4 s on the surface of the fruit using a 2 mm cylindrical probe at 4 mm depth. Each fruit was also dissected transversely to count the number of locules and to measure the pericarp thickness in mm at two locations on each fruit with a Vernier calliper and values were averaged. A random sub-sample of the fruit was sliced, weighed and placed in an aluminium tray then oven-dried at 60 °C for four days. Samples

were weighed, and dry matter content and moisture were calculated. Dried fruit samples from the same replicate were pooled together and sent to CSBP Laboratories for nutrient composition analysis. Remaining fruits were pureed through a thin mesh, centrifuged and the extracted juice was used to estimate soluble solid content (SSC), pH and titratable acidity (TA). Soluble solid content was determined with a hand refractometer (Atago 3810 pal-1, Fukaya, Saitama, Japan). The refractometer was washed with distilled water after each assessment use and dried with blotting paper. Fruit pH and TA was determined using a titrator (HI84532 Hanna Instruments, Melbourne, Australia).

One-way ANOVA was performed to compare the treatment effects on tomato plant growth, yield, and fruit quality parameters. Homogeneity of variances was verified with Levene's test. Two-way ANOVA with repeated measures was used on stem height, branch number, stem CSA, flower number and fruit number to analyse the interaction between fertilizer treatments and weekly measurements. Differences at the 5% significance level were compared using Tukey's Honestly Significant Difference (HSD) test. Permanova tests were performed on Euclidean distance matrix for leaf and tomato fruit nutrient content and fruit characteristics between each treatment and control to indicate significance of tested factors. The nMDS ordination plots were performed on Euclidean distance matrix for leaf and tomato fruit nutrient content to visualise grouping patterns between treatments. The nMDS plots were overlayed with the results of a cluster analysis by group average (dendrogram) to display group clustering based on resemblance distance. Bubble plots were used to visualise the trend of increasing nutrient concentration with increasing UWP addition across treatments. Statistical analysis of One-Way ANOVA and Two-Way ANOVA with repeated measures were performed with SPSS (IBM SPSS Statistics for Windows, version 26.0. Armonk, NY, USA: IBM Corp.). Permanova test and nMDS plots were performed using PRIMER 7 (Plymouth Routines In Multivariate Ecological Research) [24].

3. Results

3.1. Effect of Sea Urchin Waste Powder Supplement on Weekly Plant Growth

All plant growth parameters increased with greater UWP application, with plant performance in some variables equivalent to that observed in the Hoagland solution control treatment (Table 2, Figure 1). Weekly measurements of height, branches number and stem CSA showed overall statistically significant differences in group means for the interaction between treatments and sampling time (Figure 2, Table S2). At the end of the experiment, shoot length and stem CSA of tomato plants receiving the highest UWP (Treatment 7) were not significantly different to the Hoagland's solution control (Table 2, $\alpha > 0.05$). Shoot length, stem CSA, branches number and plant width had a moderate increase in the lowest three UWP treatments with no significant difference in these plant parameters (Figure 2, Table 2, $\alpha > 0.05$). Mean plant width (canopy area) for T7 (5% UWP) was significantly higher than all other treatments including the Hoagland's control (Figure 2, $\alpha < 0.05$). Branches number of tomato plants receiving the highest two UWP treatments (T6, T7) was not significantly different from each other but had significantly fewer branches than the Hoagland's control (T8) (Figure 2), (Table 2, $\alpha < 0.05$). Treatment T7 showed significant statistical differences from T6 for shoot length, stem CSA and plant dry weight. Tomato plants receiving the Hoagland's control had the greatest total dry matter mass which significantly decreased with each UWP rate increase from T7 to T4. Dry matter content (%) was greatest for the Hoagland's control but not significantly different to the highest UWP treatments (Table 2 $\alpha > 0.05$).

3.2. Plant Nutrient Levels as a Measure of Potting Mix Nutrient Uptake

Total nutrient (N, P, K, Ca and Mg) concentrations in the vegetative (combined shoots and leaves) parts of the plant showed an increasing response to higher UWP treatments (Table 3). All macronutrients increased significantly between T6, T7 and T8 (Table 3, $\alpha < 0.05$). A similar trend of increasing micronutrient levels with increasing UWP was also observed (Table 3). Plants grown in the Hoagland's solution (T8) contained higher

nutrient concentrations than plants in the highest UWP treatment (T7) for all nutrients including Ca, Mg and micronutrients such as B, Zn, Fe and Mn (Table 3).

Table 2. Vegetative growth parameters of tomato plants in greenhouse pot trial with UWP treatments at the end of the trial.

	Height (cm)	Branches (n°)	CSA (mm)	Width (cm)	Dry Weight (gr)	DMC (%)
T1	32.9 ± 2.08 a	15.0 ± 2.71 a	8.47 ± 0.730 a	39.7 ± 4.16 a	5.61 ± 1.29 a	17.7 ± 1.54 abc
T2	32.0 ± 12.3 a	15.4 ± 3.40 ab	8.66 ± 0.800 ab	43.1 ± 5.53 a	6.96 ± 2.19 ab	16.6 ± 1.29 a
T3	37.0 ± 3.94 a	18.1 ± 3.31 bc	8.93 ± 0.660 abc	46.2 ± 6.30 ab	8.39 ± 2.74 ab	16.9 ± 1.21 ab
T4	40.0 ± 3.40 ab	21.0 ± 3.59 bc	9.40 ± 0.720 abc	48.1 ± 4.38 ab	9.58 ± 2.84 b	17.5 ± 2.14 abc
T5	46.3 ± 5.93 bc	24.3 ± 4.00 c	9.83 ± 1.01 bcd	55.8 ± 7.30 bc	15.4 ± 4.96 c	17.8 ± 0.680 bc
T6	46.7 ± 7.57 bc	27.9 ± 5.02 d	10.7 ± 0.760 cd	59.9 ± 6.72 cd	21.5 ± 7.16 d	18.0 ± 0.860 cd
T7	53.5 ± 3.10 d	29.6 ± 4.55 d	11.2 ± 0.900 e	68.4 ± 5.02 e	32.1 ± 4.73 e	18.2 ± 0.580 d
T8	55.0 ± 4.19 d	33.9 ± 7.23 e	11.2 ± 0.550 e	62.7 ± 4.60 cd	38.6 ± 4.34 f	19.0 ± 0.470 d
p-Value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

For each parameter measured (column) different letters show significant differences ($p < 0.05$) among means by post hoc Tukey HSD Test. Treatments represent increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). Measurement values represent the average of ten replicates ($n = 10$) per treatment ± standard error.

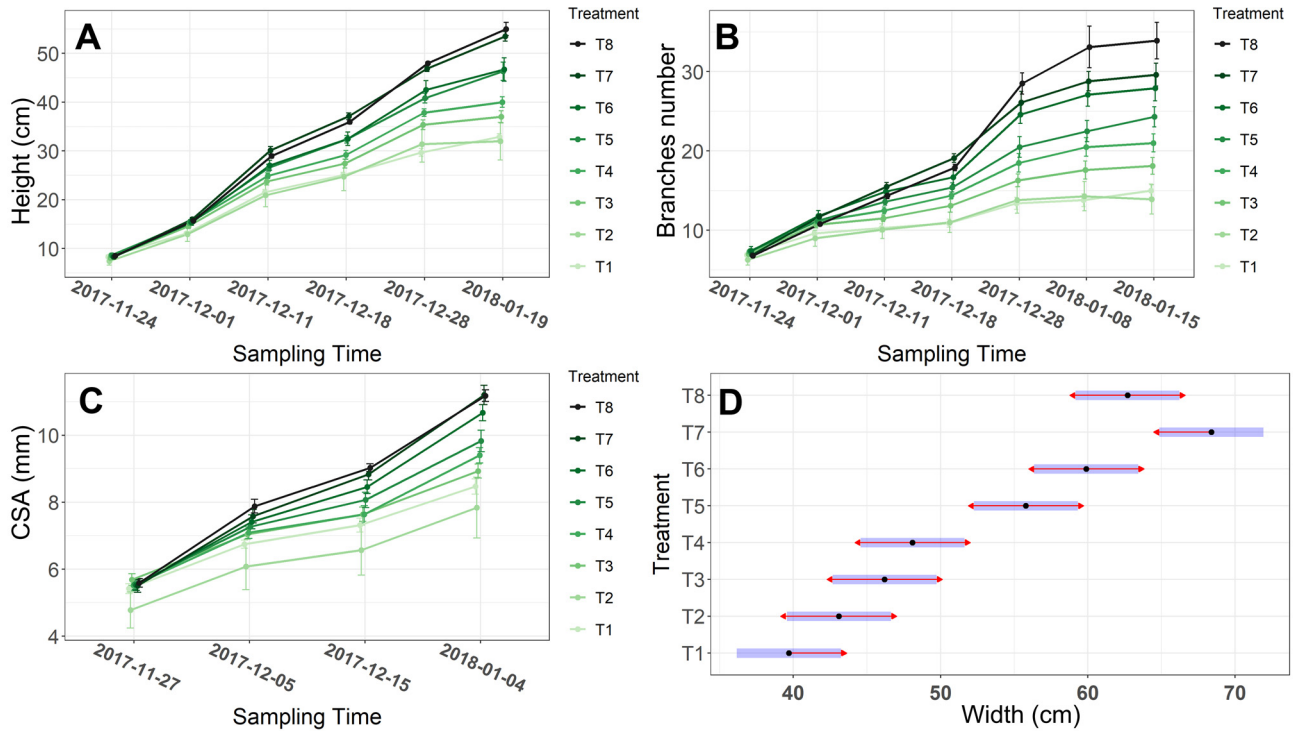

Figure 2. Line graphs of plant growth measurements including mean values of stem height in cm (**A**), number of branches (**B**), stem CSA in mm (**C**) and leaf width at the completion of the trial in cm (**D**) for increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). *x*-axis represents sampling event in weeks after planting. Error bars denote standard error (**A**–**C**). Plant width (**D**) represents a single sampling event. When arrows overlap among treatments, then the treatments are not significantly different.

Multivariate analysis on the proportion of leaf nutrient content showed overall statistically significant differences among treatment groups (Table S3). The nMDS plot shows four groups, however, the overlayed cluster analysis based on the resemblance distance reveals three clusters (Figure 3) of plant nutrient proportions. The four lowest UWP addition rates (T1–T4) form a tight grouping and clustered together with T5 and part of T6, while two separate clusters are evident for part of T6 with T7 and T8. The grouping within

treatments was evident but the increase in the proportion of nutrients in the first five to six treatments did not justify separate clustering. A shift occurred in T6 (3% rate of UWP addition) which splits between the lower cluster, (grouping with T5) and T7, the highest rate of UWP addition (Figure 3). The low stress (0.01) in the two dimensions indicates a good degree of agreement between the data distance and the ordination (Euclidean) distance matrices. Potassium increase is displayed by bubble size across treatments and control since its deficiency represents a limiting factor for plant growth and was provided in a small amount with UWP.

Table 3. Nutrient concentration in vegetative parts (combined shoot and leaf) of tomato plant in greenhouse pot trial with UWP treatments.

A	**N (g 100 g^{-1})**	**P (g 100 g^{-1})**	**K (g 100 g^{-1})**	**Ca (g 100 g^{-1})**	**Mg (g 100 g^{-1})**	**Na (g 100 g^{-1})**
T1	0.080 ± 0.001 a	0.018 ± 0.001 a	0.170 ± 0.005 a	0.160 ± 0.009 a	0.031 ± 0.002 a	0.002 ± 0.0004 a
T2	0.090 ± 0.006 a	0.018 ± 0.002 a	0.210 ± 0.021 a	0.260 ± 0.026 a	0.042 ± 0.002 a	0.003 ± 0.0001 a
T3	0.110 ± 0.003 a	0.018 ± 0.001 a	0.230 ± 0.001 a	0.310 ± 0.025 ab	0.050 ± 0.004 ab	0.003 ± 0.0003 a
T4	0.130 ± 0.008 a	0.018 ± 0.001 a	0.260 ± 0.012 ab	0.310 ± 0.011 ab	0.053 ± 0.002 ab	0.003 ± 0.0004 a
T5	0.220 ± 0.010 b	0.027 ± 0.001 a	0.360 ± 0.015 bc	0.500 ± 0.020 bc	0.076 ± 0.006 bc	0.004 ± 0.0006 a
T6	0.310 ± 0.011 c	0.029 ± 0.003 ab	0.440 ± 0.025 c	0.680 ± 0.073 cd	0.098 ± 0.012 c	0.008 ± 0.0008 b
T7	0.450 ± 0.014 d	0.040 ± 0.003 b	0.630 ± 0.027 d	0.800 ± 0.026 de	0.135 ± 0.002 d	0.014 ± 0.0001 c
T8	0.760 ± 0.019 e	0.076 ± 0.005 c	1.02 ± 0.033 e	0.940 ± 0.068 e	0.182 ± 0.013 e	0.010 ± 0.0014 b
p-Value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
B	**S (g 100 g^{-1})**	**Cu (mg kg^{-1})**	**Fe (mg kg^{-1})**	**Mn (mg kg^{-1})**	**Zn (mg kg^{-1})**	**B (mg kg^{-1})**
T1	0.023 ± 0.001 a	0.170 ± 0.060 a	3.25 ± 0.300 a	1.67 ± 0.110 a	0.950 ± 0.06 a	2.16 ± 0.090 a
T2	0.032 ± 0.001 ab	0.160 ±0.050 a	3.80 ± 0.290 a	2.89 ± 0160 ab	1.25 ± 0.120 a	2.87 ± 0.140 ab
T3	0.036 ± 0.003 ab	0.120 ± 0.030 a	4.06 ± 0.330 a	3.34 ± 0.420 bc	1.34 ± 0.110 ab	3.17 ± 0.190 ab
T4	0.035 ± 0.002 ab	0.100 ± 0.001 a	3.81 ± 0.070 a	3.53 ± 0.230 bc	1.41 ± 0.180 ab	3.29 ± 0.090 ab
T5	0.053 ± 0.002 bc	0.240 ± 0.090 a	6.77 ± 0.550 ab	4.86 ± 0.270 cd	2.26 ± 0.270 bc	4.93 ± 0.110 bc
T6	0.066 ± 0.007 c	0.350 ± 0.040 a	8.55 ± 0.700 b	6.40 ± 0.490 d	2.78 ± 0.380 c	6.83 ± 0.740 c
T7	0.076 ± 0.003 c	0.300 ± 0.030 a	12.4 ± 0.940 c	9.86 ± 0.340 e	4.11 ± 0.200 d	9.73 ± 0.660 d
T8	0.155 ± 0.013 d	1.07 ± 0.170 b	17.6 ± 1.52 d	13.3 ± 0.460 f	5.13 ± 0.120 e	15.9 ± 0.850 e
p-Value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

For each element (column) different letters show significant differences ($p < 0.05$) among means by post hoc Tukey HSD Test. Treatments represent increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). Nutrient values represent the average of three replicates ($n = 3$) per treatment ± standard error.

3.3. Effect of UWP on Flowering and Fruit Production

Overall, statistically significant differences were observed in the weekly measurements between group means of flowers and fruit productivity for the interaction between treatments and sampling time (Table S4). Plant flowering and fruiting success measured at the end of the trial were variable across the treatments. A trend for increasing number of flowers and fruit was observed with increasing application of UWP (Figure 4), however, there were no statistical differences between the lowest four UWP treatments (Table 4, $\alpha > 0.05$). A main effect of UWP fertiliser was evident in T5, T6, and T7 for these factors. Flower number receiving the highest UWP (T7) was not significantly different (Table 4, $\alpha > 0.05$) from the standard Hoagland's solution. Average fruit number ranged from one fruit per plant in the lowest UWP treatment to eight fruit per plant in the Hoagland's control (Table 4) with significant differences in the mean values observed between the three highest rate treatments (T5 to T7) and the control (T8), ($F_{(7,72)} = 52.57$, $p = 0.0001$). Average fruit size (diameter) and overall fruit weight (total yield per plant) increased with increasing UWP (Table 4, $F_{(7,176)} = 2.844$, $p = 0.008$). Plants receiving the highest UWP (T7) yielded 238 g of fresh fruit which was almost half of the fresh fruit produced by plants in the Hoagland's treatment (448 g), (Table 4, $\alpha < 0.05$). Average fruit fresh weight again increased with increasing UWP, being very low in T1 (28.9 g) and showing a mild increase from T3 to T5, no clear statistical differences were observed in the first six treatments. Tomato fruits from

the highest rate of UWP addition T7 and the control weighed both 150 g, were statistically different from T6 but not significantly different from each other (Table 4).

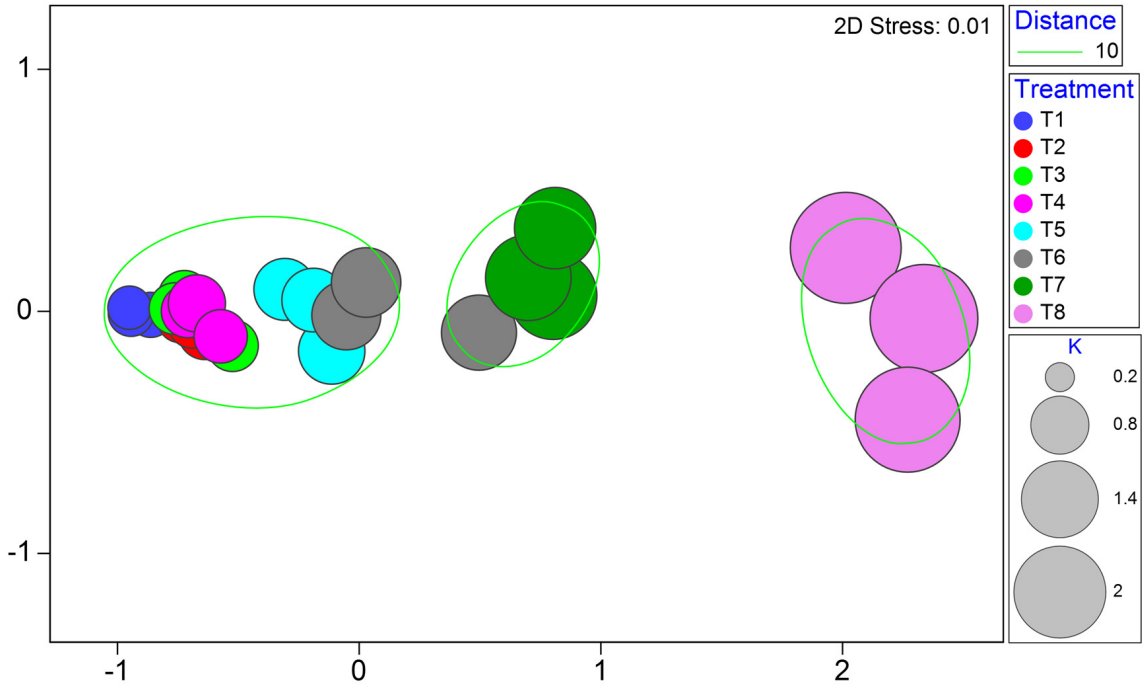

Figure 3. Non-metric multidimensional scaling (nMDS) bubble plot of Euclidian distances between the proportion of nutrients in tomato plant vegetative parts in treatments receiving increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). Samples treatments grouping are the results of an overlayed cluster analysis by group average of resemblance distance. Bubble size indicates K content (g 100 g^{-1}).

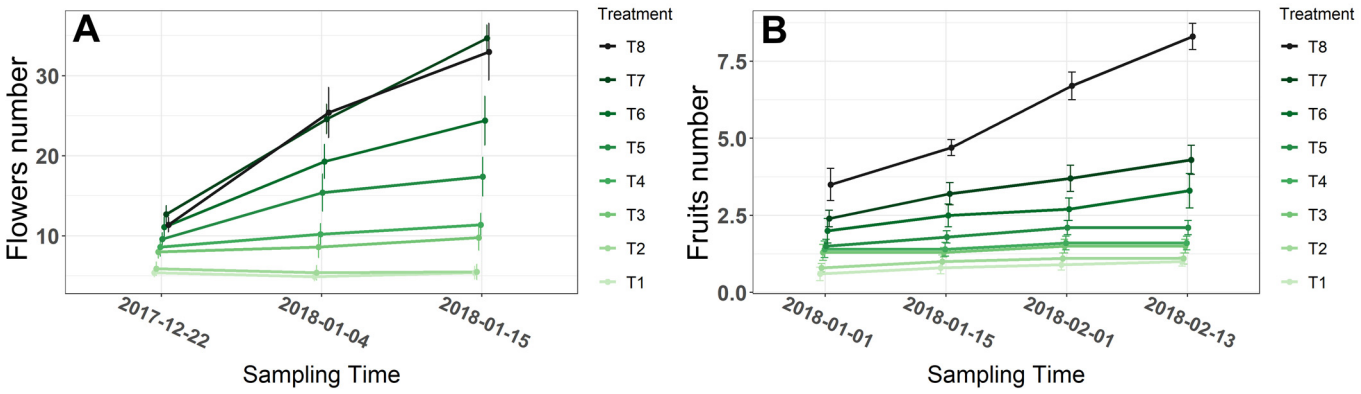

Figure 4. Line graphs including mean values of flower (**A**) and fruit (**B**) production for eight treatments representing increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). *x*-axis represents sampling events in weeks after planting. Error bars denote standard errors.

While there was a trend of decreasing fruit dry matter (%) from T1 (7.3%)–T7 (6.7%) and T8 (6.5%), there were no significant differences in mean fruit dry matter per plant ($F_{(6,64)}$ = 0.915, p = 0.501). Locule number was similar across all treatments varying from (6.00) to (8.00), ($F_{(7,72)}$ = 0.489, p = 0.840). Pericarp thickness ranged from a minimum of 3.60 (T1) mm and a maximum of 5.75 (T8) mm and significantly increased with higher UWP rates (Table S5), ($F_{(7,72)}$ = 2.951, p = 0.009).

Table 4. Average flower and fruit number, fruit size, yield per plant and fruit fresh weight of tomato plants in greenhouse pot trial under UWP treatments (T1–T7) and Hoagland's control (T8).

	Flowers (n°)	**Fruits (n°)**	**Diameter (cm)**	**Yield (gr/Plant)**	**Fresh Weight (gr)**
T1	5.40 ± 2.50 a	1.10 ± 0.470 a	34.3 ± 14.3 a	23.6 ± 17.6 a	28.9 ± 4.53 a
T2	5.50 ± 3.10 a	1.20 ± 0.570 a	40.1 ± 15.7 ab	41.1 ± 20.1 ab	46.4 ± 4.21 ab
T3	9.80 ± 4.96 ab	1.50 ± 0.710 ab	47.3 ± 10.0 b	65.2 ± 22.1 bc	62.3 ± 7.08 abc
T4	11.4 ± 4.43 ab	1.60 ± 0.700 ab	47.1 ± 9.38 bc	69.2 ± 25.3 bc	63.1 ± 8.49 abc
T5	17.4 ± 7.69 bc	2.10 ± 0.740 b	44.1 ± 11.6 b	84.9 ± 27.9 c	68.9 ± 9.71 bc
T6	24.4 ± 9.59 cd	3.30 ± 1.77 c	54.9 ± 10.3 cd	144 ± 45.1 d	98.5 ± 8.01 c
T7	34.7 ± 5.19 e	4.30 ± 1.49 d	57.5 ± 11.9 d	238 ± 63.2 e	150 ± 15.0 d
T8	33.0 ± 11.2 de	8.30 ± 1.34 e	60.3 ± 9.38 d	448 ± 74.0 f	150 ± 6.08 d
p-Value	<0.001	<0.001	<0.001	<0.01	<0.001

For each parameter measured (column) different letters show significant differences ($p < 0.05$) among means by post hoc Tukey HSD Test. Treatments represent increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). Measurements values represent the average of ten replicates ($n = 10$) per treatment ± standard error. Fruits fresh weight mean values are represented by the eight ($n = 8$) biggest fruits at the same stage of maturity for each treatment and control.

3.4. Fruit Nutrient Levels as a Measure of Potting Mix Nutrient Uptake

Fruits nutrient concentrations increased in response to higher rates of UWP (Table 5). An increasing level of macronutrients (N, P, K and Ca and Mg) was observed with increasing rate of UWP applied. Both macro and micronutrients concentrations showed a mild increase in the first five treatments but were not significantly different (Table 5, $\alpha > 0.05$). In contrast, nutrient content significantly increased between T6, T7 and T8 (Table 5, $\alpha < 0.05$). Harvested fruits from tomato plants receiving the Hoagland's solution (T8) contained higher nutrient concentrations than fruits from plants receiving the highest UWP treatment (T7) for all the macro and micro-nutrients, with the exception for Na which was higher in T7 than T8 but not significantly different (Table 5).

Table 5. Nutrient concentration in fruit of tomato plants receiving UWP treatments (T1–T7) and the Hoagland's control (T8).

A	**N (g 100 g^{-1})**	**P (g 100 g^{-1})**	**K (g 100 g^{-1})**	**Ca (g 100 g^{-1})**	**Mg (g 100 g^{-1})**	**Na (g 100 g^{-1})**
T1	0.039 ± 0.003 a	0.017 ± 0.001 a	0.146 ± 0.004 a	0.006 ± 0.0003 a	0.005 ± 0.0003 a	0.001 ± 0.0001 a
T2	0.062 ± 0.004 ab	0.029 ± 0.002 a	0.272 ± 0.023 ab	0.013 ± 0.002 ab	0.009 ± 0.001 ab	0.002 ± 0.0002 a
T3	0.084 ± 0.008 ab	0.034 ± 0.002 a	0.359 ± 0.024 b	0.021 ± 0.001 ab	0.012 ± 0.001 ab	0.003 ± 0.0005 ab
T4	0.091 ± 0.005 ab	0.035 ± 0.003 a	0.383 ± 0.021 b	0.024 ± 0.001 ab	0.012 ± 0.001 ab	0.002 ± 0.0003 ab
T5	0.128 ± 0.017 b	0.040 ± 0.003 ab	0.444 ± 0.019 b	0.025 ± 0.001 ab	0.014 ± 0.001 b	0.003 ± 0.0004 ab
T6	0.264 ± 0.022 c	0.077 ± 0.008 bc	0.851 ± 0.046 c	0.038 ± 0.003 b	0.027 ± 0.003 c	0.008 ± 0.001 ab
T7	0.451 ± 0.026 d	0.110 ± 0.016 c	1.28 ± 0.080 d	0.064 ± 0.00 c	0.041 ± 0.002 d	0.021 ± 0.011 b
T8	0.873 ± 0.016 e	0.246 ± 0.012 d	2.55 ± 0.038 e	0.116 ± 0.014 d	0.087 ± 0.002 e	0.016 ± 0.002 ab
p-Value	<0.001	<0.001	<0.001	<0.001	<0.001	0.035
B	**S (g 100 g^{-1})**	**Cu (mg kg^{-1})**	**Fe (mg kg^{-1})**	**Mn (mg kg^{-1})**	**Zn (mg kg^{-1})**	**B (mg kg^{-1})**
T1	0.005 ± 0.0003 a	0.160 ± 0.034 a	1.72 ± 0.110 a	0.320 ± 0.020 a	0.700 ± 0.053 a	0.670 ± 0.028 a
T2	0.010 ± 0.0005 ab	0.210 ± 0.040 a	2.90 ± 0.020 a	0.570 ± 0.057 ab	1.26 ± 0.09 ab	1.26 ± 0.112 ab
T3	0.013 ± 0.0005 ab	0.240 ± 0.025 a	3.29 ± 0.240 a	0.720 ± 0.042 ab	1.52 ± 0.137 ab	1.77 ± 0.09 bc
T4	0.015 ± 0.0003 b	0.260 ± 0.005 a	3.86 ± 0.060 a	0.810 ± 0.081 ab	1.66 ± 0.048 ab	1.97 ± 0.052 bc
T5	0.018 ± 0.001 b	0.360 ± 0.050 a	5.08 ± 0.570 a	0.930 ± 0.076 b	2.26 ± 0.254 b	2.31± 0.028 c
T6	0.034 ± 0.003 c	0.780 ± 0.014 ab	9.85 ± 1.24 b	1.71 ± 0.142 c	4.66 ± 0.365 c	3.91 ± 0.209 d
T7	0.053 ± 0.003 d	1.14 ± 0.208 b	14.2 ± 1.34 c	2.86 ± 0.193 d	7.48 ± 0.57 d	6.35 ± 0.295 e
T8	0.100 ± 0.002 e	2.57 ± 0.336 c	23.6 ± 1.06 d	5.66 ± 0.136 e	10.1 ± 0.496 e	12.0 ± 0.382 f
p-Value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

For each element (column) different letters show significant differences ($p < 0.05$) among means by post hoc Tukey HSD Test. Treatments represent increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). Nutrient values represent the average of three replicates ($n = 3$) per treatment ± standard error.

The multivariate analysis on the proportion of tomato fruit nutrient content showed overall statistically significant differences among treatment groups (Table S6). The nMDS plot showed four clear groups where clustering was also confirmed by the overlayed cluster analysis of group average (Figure 5). A tight group is represented by the five lowest UWP rates (T1–T5), while a sharp distinction is evident for T6, T7 and T8 groups, reflecting the outcomes of fruit production number and yield in these three treatments. The low stress (0.01) in the two dimensions indicates a good degree of agreement between the data distance and the ordination (Euclidean) distance matrices. Nitrogen increase is displayed by bubble size across treatments and control since its deficiency represents a limiting factor for plant growth and was provided in a relatively small amount with UWP and an adequate amount in the control.

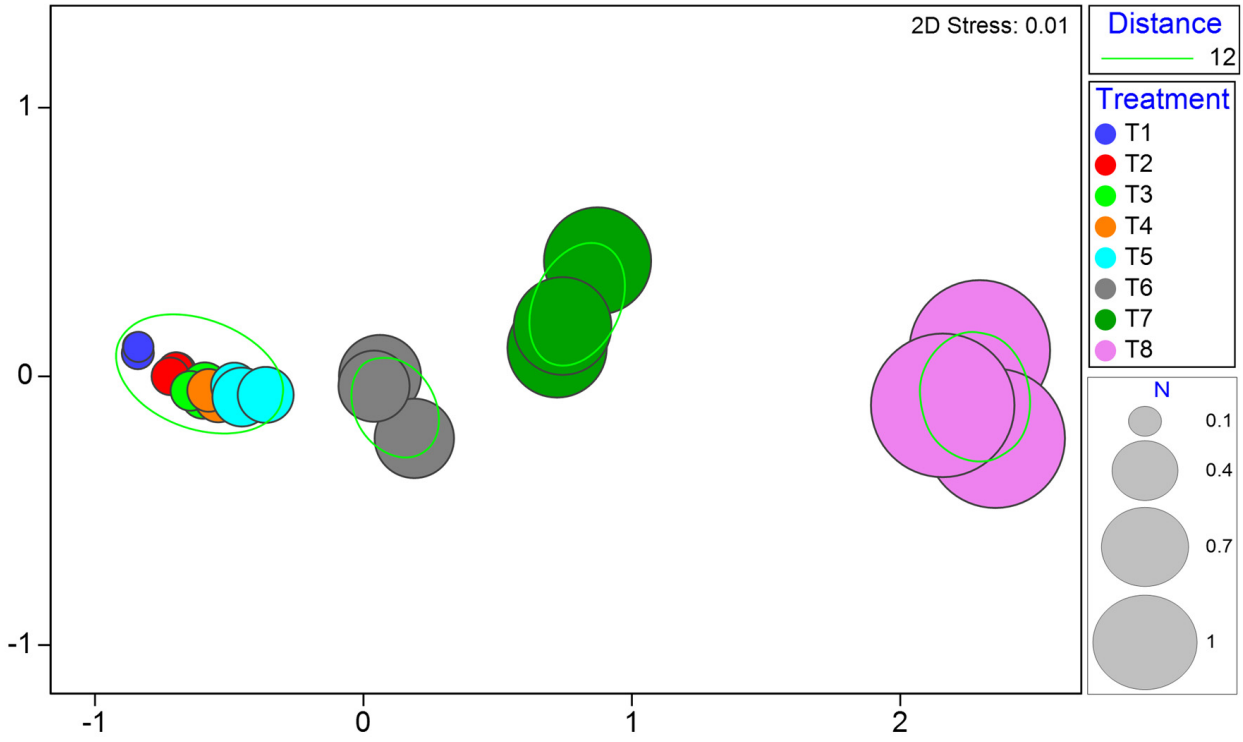

Figure 5. Non-metric multidimensional scaling (nMDS) bubble plot of Euclidian distances between the proportion of nutrients in tomato fruits in treatments receiving increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). Samples treatments grouping are the results of an overlayed cluster analysis by group average of resemblance distance. The bubble size indicates N content (g 100 g^{-1}).

3.5. Fruit Ripeness

UWP treatments had a significant influence on the maturation of tomato fruit as measured by fruit firmness and colour. As fruit ripen over time, firmness values (as determined by resistance to the probe during the pressure test) decrease as the flesh gets softer and the red colour of fruit increases. Fruit firmness results were statistically higher in T1 compared to all other treatments (Table 6, $\alpha < 0.05$) because fruit did not develop properly, remaining small and under-ripe. Fruit harvested from plants receiving the Hoagland's solution were on average firmer than fruit from T7, but not significantly different (Table 6, $\alpha > 0.05$). Flesh firmness was inversely correlated to colour values and dry matter content in fruit with the lowest rate of UWP.

Fruit colour analysis was consistent with other ripening variables and again followed a response curve to the treatments. L* values were higher in fruit treated with less UWP (46.6) in T1, reflecting a lower degree of ripeness—hence colour ranged from white green to pale red, while riper fruit showed an intense red colour that translate into lower L* values

(35.18 and 36.81) as observed in T7 and T8, respectively (Table 6). Overall, Permanova analysis on the tomato fruit colour coordinates and firmness did not show statistically significant difference among treatment groups (Table S7, $p = 0.108$).

Table 6. Colour and firmness parameters in tomato fruit receiving UWP (T1–T7) and Hoagland solution (T8) treatments.

	L	a	b	Hue Angle	Chroma	Firmness
T1	46.6 ± 2.98 a	18.2 ± 2.45 a	31.0 ± 2.98 a	1.02 ± 0.090 a	36.9 ± 2.15 a	0.670 ± 0.060 a
T2	37.4 ± 0.370 b	22.7 ± 1.08 abc	21.3 ± 0.630 bc	0.760 ± 0.010 b	31.2 ± 1.18 abc	0.490 ± 0.020 b
T3	40.1 ± 0.660 b	25.3 ± 1.25 bc	25.6 ± 0.890 ab	0.800 ± 0.030 b	36.2 ± 1.22 a	0.460 ± 0.010 b
T4	40.0 ± 0.790 b	27.0 ± 1.27 c	25.6 ± 1.29 ab	0.760 ± 0.020 b	37.3 ± 1.64 a	0.490 ± 0.020 b
T5	40.2 ± 1.79 b	22.6 ± 1.62 abc	24.3 ± 2.00 bc	0.810 ± 0.060 b	33.8 ± 1.55 ab	0.480 ± 0.030 b
T6	37.9 ± 0.970 b	22.5 ± 1.60 abc	21.8 ± 1.65 bc	0.770 ± 0.020 b	31.4 ± 2.20 abc	0.440 ± 0.020 b
T7	35.2 ± 0.320 b	19.6 ± 0.830 ab	17.7 ± 0.630 c	0.740 ± 0.010 b	26.4 ± 0.970 c	0.400 ± 0.020 b
T8	36.8 ± 1.19 b	18.7 ± 0.770 a	19.2 ± 1.30 bc	0.790 ± 0.040 b	27.0 ± 0.960 bc	0.490 ± 0.030 b
p-Value	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

For each parameter measured (column) different letters show significant differences ($p < 0.05$) among means by post hoc Tukey HSD Test. Treatments represent increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). Measurements values represent the average of ten replicates ($n = 10$) per treatment ± standard error.

There was no significant difference for pH ($F_{(7,37)} = 1.235$, $p = 0.309$), with treatment means ranging from 4.06 to 4.24 while significant differences were observed for TA ($F_{(7,37)} = 12.23$, $p = 0.0001$) with 5.77 mg/100 mL in T1, 3.51 mg/100 mL in T7 and 3.43 mg/100 mL in T8 (Figure 6A, $\alpha < 0.05$). Mean values of SSC ranged from 4.3 to 5.9 (Figure 6B, $\alpha < 0.05$) with the Hoagland's control and T7 having significantly lower SSC than all the other treatments ($F_{(7,37)} = 9.026$, $p = 0.0001$).

3.6. Potting Mix Nutrient Content

The application of UWP increased the initial pH and EC of the potting mix as the rate of UWP application increased (Table 1). Values of the potting mix post treatments application at the start of the trial are not shown here, since represent a single measurement per treatment from pooled sub-samples from the ten pot replicates three days post planting of tomato seedlings and cannot be presented with standard errors.

The pH of the potting mix at trial end shows an increasing trend in the three lower treatments from pH 6.7 to pH 6.9 and stabilizing slightly above pH 7 from T4 to T7 with significant differences between them (Figure 7A, $F_{(6,14)} = 15.14$, $p = 0.0001$). The EC instead varied between 0.428 dSm^{-1} in T1, 0.563 dSm^{-1} in T6 and 0.693 dSm^{-1} T7 with no clear differences among treatments (Figure 7B, $F_{(6,14)} = 1.618$, $p = 0.214$).

No statistical differences between the treatments were observed for N, P, Mg, Na, Cu, Zn and B. The value of K in T7 was significantly lower than the first five treatments (Table 7, $\alpha < 0.05$), indicating that K was actively taken up by the plants in this treatment. The content of other elements in the pot appears more irregular. Ca increased from T1 to T5 then declined again to T7 with statistical difference between T1 and T5 (Table 7, $\alpha < 0.05$). Calcium decreases in T6 and T7 potting mix could be related to the greater vegetative production in these two treatments that led to more Ca uptake. A trend of increasing Na concentration can be observed from T1 to T7, but there are not clear statistical differences, while S was significantly higher in T7 compared to the first five treatments (Table 7, $\alpha < 0.05$). Conversely, the remaining Fe and Mn were higher in the lower treatments with a decline towards T7.

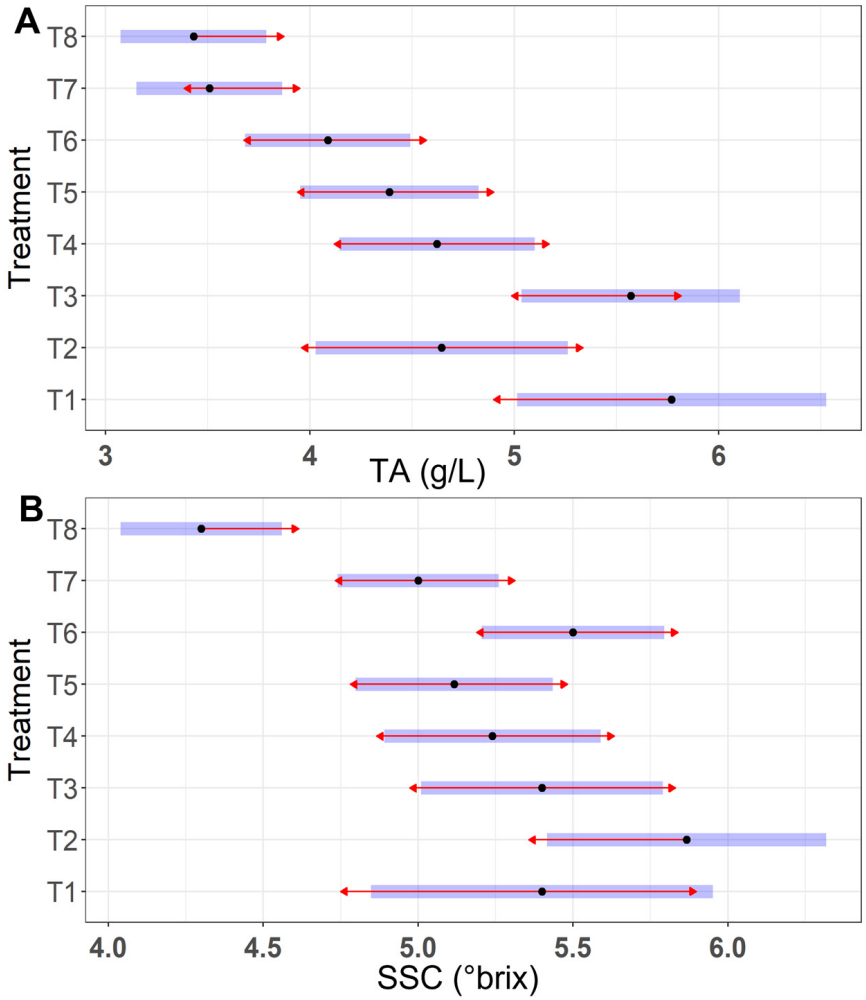

Figure 6. Average values of fruit TA (**A**) and SSC (**B**) across seven treatment rates of UWP (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%), and Hoagland's solution (T8). Dots indicate averages of fruit pooled together across the ten replicate pots in each treatment. Bars denote the confidence limit. When the red arrows overlap among treatments, then the treatments are not significantly different.

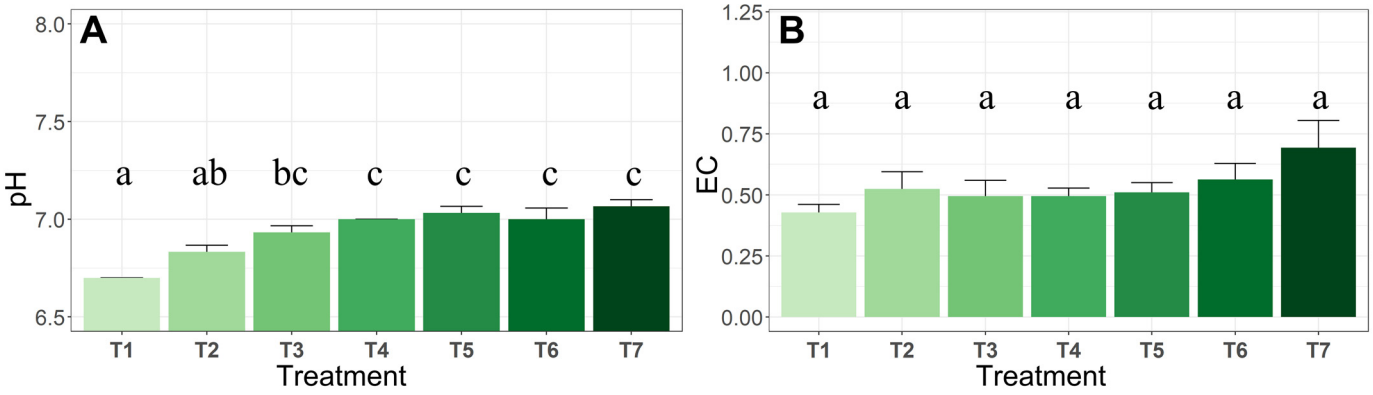

Figure 7. Bar chart of pH (**A**) and EC (**B**) of potting mix with addition of the seven UWP treatments rate (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%) at the end of the trial. Error bars denote averages of three replicates per each treatment. For each treatment (bar) different letters show significant differences ($p < 0.05$) among means by post hoc Tukey HSD Test.

Table 7. Nutrient concentration in potting mix with addition of UWP at seven rates at the end of the trial.

A	N (mg kg^{-1})	P (mg kg^{-1})	K (g 100 g^{-1})	Ca (g 100 g^{-1})	Mg (g 100 g^{-1})	Na (g 100 g^{-1})
T1	9.00 ± 2.00	12.0 ± 0.700	0.066 ± 0.004 a	0.496 ± 0.019 a	0.050 ± 0.002	0.010 ± 0.001 a
T2	10.0 ± 3.00	13.0 ± 0.300	0.058 ± 0.006 a	0.501 ± 0.012 a	0.044 ± 0.001	0.017 ± 0.004 ab
T3	7.00 ± 1.00	11.0 ± 0.300	0.047 ± 0.005 ab	0.523± 0.024 ab	0.041 ± 0.002	0.014 ± 0.001 ab
T4	9.00 ± 2.00	12.0 ± 0.300	0.058 ± 0.010 a	0.527 ± 0.014 ab	0.043 ± 0.002	0.017 ± 0.002 ab
T5	8.00 ± 3.00	14.0 ± 0.700	0.039 ± 0.004 abc	0.625 ± 0.040 b	0.048 ± 0.002	0.022 ± 0.004 ab
T6	11.0 ± 1.00	14.0 ± 2.30	0.021 ± 0.005 cd	0.589 ± 0.018 ab	0.045 ± 0.002	0.025 ± 0.003 ab
T7	18.0 ± 6.00	13.0 ± 1.90	0.015 ± 0.004 d	0.559 ± 0.018 ab	0.042 ± 0.003	0.032 ± 0.009 b
p-Value	ns	ns	<0.01	0.010	ns	ns
B	**S (g 100 g^{-1})**	**Cu (mg kg^{-1})**	**Fe (mg kg^{-1})**	**Mn (mg kg^{-1})**	**Zn (mg kg^{-1})**	**B (mg kg^{-1})**
T1	0.003 ± 0.001 a	12.1 ± 0.980	54.5 ± 1.86 a	47.7 ± 2.23 a	20.4 ± 1.66	0.520 ± 0.009
T2	0.004 ± 0.001 ab	7.70 ± 0.840	43.5 ± 4.89 ab	41.4 ± 2.71 ab	13.5 ± 0.620	0.520 ± 0.030
T3	0.004 ± 0.001 ab	7.23 ± 1.14	38.2 ± 1.16 bc	39.9 ± 0.340 ab	14.0 ± 0.720	0.540 ± 0.030
T4	0.004 ± 0.001 ab	9.78 ± 0.440	37.4 ± 1.49 bc	39.8 ± 1.19 ab	15.0 ± 0.230	0.530 ± 0.019
T5	0.004 ± 0.001 ab	12.5 ± 0.430	37.5 ± 2.00 bc	41.8 ± 0.390 ab	16.1 ± 0.920	0.550 ± 0.003
T6	0.008 ± 0.001 bc	12.1 ± 1.92	37.3 ± 1.75 bc	41.7 ± 2.38 ab	17.3 ± 1.86	0.520 ± 0.010
T7	0.009 ± 0.002 c	11.1 ± 1.61	30.3 ± 2.35 bc	35.1 ± 2.31 b	19.0 ± 3.20	0.507 ± 0.017
p-Value	0.005	0.026	<0.001	0.017	ns	ns

For each element (column) different letters show significant differences ($p < 0.05$) among means by post hoc Tukey HSD Test. Treatments represent increasing UWP supplement rates (T1 = 0.3%; T2 = 0.5%; T3 = 0.8%; T4 = 1%; T5 = 2%; T6 = 3%; T7 = 5%). Nutrient values represent the average of three replicates ($n = 3$) per treatment ± standard error.

4. Discussion

Powdered sea urchin waste improved the growth and productivity of tomato plants with increased performance at higher rates. For all parameters measured, significant improvements were often observed with each increasing rate. The standard Hoagland's fertiliser regime (control treatment) produced tomato plants with similar vegetative size characteristics to the highest UWP treatment (T7—5% *w*/*w*) yet were substantially bigger and healthier than plants receiving the lower UWP treatments. Consistent vegetative growth of the tomato seedlings was observed in the early stage of the trial with shoot growth of all treatments matching the control plants. Growth rate of plants in the low-rate UWP treatments (T1–T4) significantly slowed after four weeks suggesting a nutrient depletion under these treatments. Plants receiving treatments T6 and T7 were the best performing of all UWP treatments. However, tomato fruit yield in Hoagland's control was double the yield of the highest rate UWP (T7) even though plant size was similar, which reflects the higher and more readily absorbed soluble nutrient supply throughout the trial.

There was clear evidence of nutrient uptake by tomato plants receiving UWP, yet as expected plants receiving Hoagland's control solution were better performing. The Hoagland solution provided 1950 mg N in T8, twice the amount supplied to the plants receiving the highest UWP (T7). Almost four times the amount of P was provided in Hoagland's solution (223 mg) compared to 65 mg in T7 and three times the amount of K was provided. In contrast, some macro elements like Ca, Mg and S and microelements like B, Cu, Zn and Fe were supplied in higher proportions through the UWP in the higher rate treatments which may have supported comparable vegetative growth of tomato plants observed in T7 in the context of limited N supply. Boron, Zn and Fe were supplied in lesser amounts through the Hoagland's solution compared to the highest rates of UWP, however, these micronutrients were present in higher concentrations in the vegetative parts of plants in T8. The Hoagland solution facilitated better uptake of micronutrients in their soluble form to the plants as the total supply of nutrients was likely better balanced to meet plant requirements [25,26]. In contrast, plants receiving the UWP treatments including T7 towards the end of the trial showed signs of nutrient deficiency, especially (N, P, K) possibly impairing the uptake of other micronutrients.

Adequate N supply has been shown to significantly increase tomato vegetative growth [27], plant yield and fruit quality [28], whereas insufficient N content can lead to limited vegetative growth, reduced shoot length and leaf area [29], net photosynthetic rate decline [30] and blossom drop with subsequent low yields [31]. Symptoms of N deficiency were visible in plants receiving the lower UWP treatments where the four-week-old leaves became chlorotic, had completely yellowed and subsequently dehisced. Nitrogen in the Hoagland's solution was in the form of ammonia and nitrate which are both readily available forms for plant uptake. In contrast, N provided in UWP treatments was in the form of amino acids and bound peptides which require proteinaceous transporters to facilitate the transfer of N compounds across cellular membranes [32].

Inadequate K nutrition in tomatoes has been shown to negatively affect growth, fruit set, dry matter distribution, and fruit quality [33,34]. Physiological disorders such as blotchy ripening, greenback, yellow shoulder, decreased lycopene content, and irregularly shaped and hollow tomato fruit are associated with K deficiency [35,36]. Fruit appearance in UWP treatments was not affected negatively by the low K content but together with limited N may have contributed to the reduced fruit set in T6 and T7 as well as the poor plant performance in the lowest rate UWP treatments.

Phosphorous deficiency in tomato plants reduces CO_2 assimilation [37], leading to a decrease in biomass production [38]. Whilst biomass increased with each higher rate of UWP, the highest rate (T7) resulted in comparable plant biomass to the T8 control, even though the P content of that T7 was four times lower than in the Hoagland control suggesting the P derived from UWP was relatively plant available. Uchida [39] showed that the mobilization of P from old parts of the plant to new tissue causes the appearance of dark to blue-green (purpling) coloration on older leaves. Symptoms of leaf purpling (Figure S1) were most obvious in T1 to T5, however in T6 and T7 only the lower and oldest leaves were affected.

Limited magnesium can result in decreased biomass production and lower yield in greenhouse tomatoes [40], however, the relatively high content of Mg in T6 and T7 is likely to have promoted vegetative growth despite low levels of N and K.

Boron plays a key role in the growth of many fruit and vegetable plants and many studies have highlighted the importance of B in tomato fruit quality [41–44]. Davis, [45] demonstrated that foliar and root application of B increased tomato growth and promoted the uptake of N, Ca and K in plant tissue whilst improving fruit shelf life and firmness. Boron deficiency in tomatoes is associated with damaged fruit through concentric and radial cracking [45], while blossom-end rot in tomato is a physiological fruit disorder caused by insufficient Ca availability [46] and can reduce the marketability of the fruit [47]. In this study, both B and Ca were provided in the UWP at higher rates than the Hoagland's control and evidence of uptake of these micronutrients can be seen in the leaf and fruit nutrient dry matter analyses. As B is a very particular micronutrient, the range of normality and toxicity with the highest UWP addition (T7—5%) is close [48,49]. Due to the high B content in UWP, the consequent B level in the substrate is high. Sensitive crops could be negatively affected by the accumulation of B in the soil if UWP is applied at high rates.

Osmotic pressure in the root area is important for plant health. Whilst low levels of EC affect both plant growth and yield, high EC limits water absorption [50]. The EC limit for tomato is indicated at 2.5 dSm^{-1} [51]. Eltez, [52] reported a decrease in tomato yield when the EC of the treatment solution exceeded 2.0 dSm^{-1}. In this study, the EC never reached toxic levels even in the treatment with the highest application of UWP (T7—1.15 dSm^{-1}). The potting mix showed an immediate beneficial change soon after application, but the EC level dropped in all treatments at the end of the trial.

The normal range of soil pH for optimum tomato growth is from 5.5 to 7.0 [53] and Kang, [54] showed that soil pH at <4 and >8, led to limited growth of tomato seedlings, and that dry and fresh weight and shoot and root areas were particularly affected by pH 8. The potting mix used in our study had a base pH of 7.2 which increased after the application of UWP in each treatment rate to pH 7.7 in T7. At the end of the trial, the potting mix recorded

a decrease in pH in each treatment and plateaued around pH 7.0 from T4 onwards. We did not observe a negative influence on tomato productivity and nutrition of the higher pH in T7 suggesting that EC and pH were still in an optimal range to facilitate cation exchange in the root area.

The Hoagland's control solution produced significantly improved yield and fruit quality in most parameters tested. Higher fruit firmness was generally recorded in the early stages of fruit ripening, where the pericarp was less elastic and prone to perforation regardless of treatment, reducing as fruit become less firm as they matured. However, fruit harvested from plants receiving the Hoagland's solution were firmer than the T7, even though they were of similar maturity which may be a consequence of greater water content (bigger fruit) in these plants. This increases the tautness of the flesh and is further evidence for the superior quality fruit harvested from plants receiving this treatment. Fruit firmness is also related to total soluble solids content and can positively influence fruit flavour and shelf life [55].

Increased DMC is generally associated with greater vegetative growth and photosynthesis under improved nutrient conditions (specifically N) [56]. We observed this result for both fruit and plant total DMC where plants from T1 to T6 had significantly lower total DMC compared to plants receiving T7 and T8 UWP.

Fruit colour parameters pointed towards increased ripeness in fruit harvested from the highest UWP and Hoagland's solution treatments. Decreasing values of L* from T1 to T8 were observed. Decreasing L* values indicate the darkening of the red colour (from pink to full red) due to the synthesis of red colour pigments associated with fruit ripening. The a * component showed a clear increase between ripening stages from green (not ripe) to light red (ripening). The changes of a* from negative (green colour) to positive (red colour) values are attributed to chlorophyll degradation and lycopene synthesis. The b* values were higher at the pink-light red stage, the pale-yellow colour is due to the ζ-carotenes that reach their highest concentration before full ripening, where lycopene (red colour) and β-carotene (orange colour) are predominant [57,58]. However, the lower values of Chroma in T7 and T8 compared to the lesser rate UWP treatments may reflect the start of fruit senescence rather than a major accumulation of lycopene in those treatments.

5. Conclusions

The UWP used here as an organic fertiliser increased productivity of tomato plants with better performance at higher rates. Plant growth was directly related to the rate of UWP, with best plant performance for all parameters measured in T7 (5% rate addition) for all replicates. Although vegetative growth for the highest UWP treatment (T7—5%) compared well with the Hoagland's solution, this did not result in comparable yield to plants treated with the Hoagland fertiliser. These results suggest that while UWP can provide plant-available nutrients, supplementary addition of macronutrients to overcome deficiency in N and P is likely to be required as these were clearly exhausted during the vegetative growth of plants and flowers and were no longer available during fruiting.

Further research could investigate if multiple smaller additions of UWP on the topsoil during plant growth can provide a more balanced distribution of nutrients as opposed to one big application. Whilst the UWP used here comprised only the spines, jaws and tests of the urchin, the addition of urchin-derived liquid gut waste can be tested in future studies, since it may provide additional plant available nutrients to overcome some of the deficiencies identified. Alternatively, UWP could be combined with other liquid fish fertilisers that provide higher amounts of NPK. Given the high ratios of Ca, Mg and B in the UWP relative to N, P and K, there is a risk of oversupply of these nutrients, which may limit the amount of UWP that can be applied as a fertiliser to avoid nutrient toxicity. Multiple applications of UWP should be trialed to test for possible toxicity thresholds.

Gypsum and lime are often used as soil amendments, both containing high content of Ca plus SO_4^{2-} in gypsum and Mg in lime. However, they lack an array of other macro and micronutrients that are alternatively found in the UWP including K, P, Fe, Zn and B.

After Zn, B deficiency in plants is the most widespread micronutrient deficiency around the world and causes large losses in crop production and crop quality. Results from this trial suggest that the UWP could be used at lower rates as a soil amendment as an alternative to expensive soil supplements if it can be produced in sufficient quantity at a reasonable cost.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agronomy12122919/s1, Table S1: Composition of the Hoagland's stock solution used as control treatment in greenhouse pot trial with tomato plants. Table S2: Two-way Anova with repeated measure of weekly tomato plant growth parameters. Table S3: Permanova analysis of proportion of leaf nutrient content for UWP treatments (T1–T7) and the Hoagland's control. Table S4: Two-way Anova with repeated measure of weekly tomato plant measurement of flower and fruit production. Table S5: Average fruits dry matter content (%), fruits loculi number and fruit pericarp thickness of tomato plants in greenhouse pot trial under UWP treatments (T1–T7) and Hoagland's control (T8). Table S6: Permanova analysis of proportion of fruit nutrient content for UWP treatments (T1–T7) and Hoagland control (T8). Table S7: Permanova analysis of fruit ripeness parameters (colour coordinates and firmness) and quality attributes (pH, TA and SSC) for UWP and treatments (T1–T7) and Hoagland control (T8). Figure S1: On the left tomato plant treatments randomly arranged on benches inside the greenhouse (left to right T6, T8, T6, T2, T4). On the right close up of leaf purpling.

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