

Figure 33: Mean yield for Basil (Fresh) (**A**) and Basil (Dry) (**B**) for peat and peat-free substrates.Peat-free substrates demonstrated significantly lower yields comapred against the peat substrates. This is more apparent in Coriander than Basil. The addition of PGPR's appears to be detrimental to dry yield in Coriander, with the majority of yields significantly diminished against the Control values for both peat and peat-free substrates. However, improvements in yields for Peat based substrates from all species, 25 increase 0.44g

Table 12: Mean Yield, Basil

Dry	Sub	Crop	А	All	В	K	Control	0
Fresh Yield	Peat	Basil	35.00(7.63)	29.00(4.43)	29.33(4.62)	31.67(7.10)	30.71(5.23)	32.50(2.15)
	Peat-Free		25.33(2.67)	25.31(3.17)	23.67(2.46)	26.17(2.04)	23.86(8.87)	24.09(2.59)
Dry	Peat	Basil	3.56(0.87)	3.31(0.62)	2.99(0.49)	3.60(1.04)	3.29(0.79)	3.34(0.18)
	Peat-Free		2.89(0.32)	2.97(0.35)	2.55(0.29)	2.92(0.36)	2.62(1.48)	2.70(0.20)



Figure 34: Coriander growth at close to harvest. In figure (b), lower leaves can be seen suffering from necrosis. This is a symptom of light deprivation from increasing green leaf area above the lower leaves.

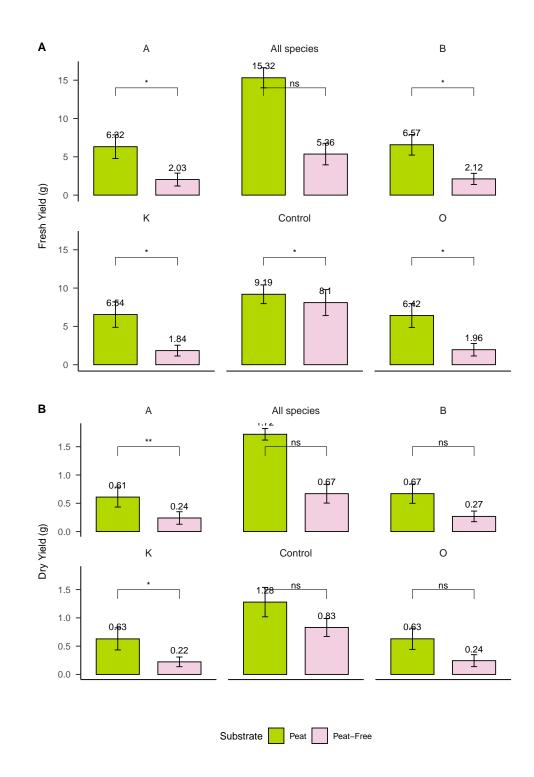


Figure 35: Mean fresh yield for Coriander (Fresh)  $(\mathbf{A})$  and Coriander (dry)  $(\mathbf{B})$  for peat and peat-free substrates. Fresh yield demonstrates significantly improved results regarding treatment All species, however only evident in peat-based substrates.

Freshyield	Sub	Crop	А	All	В	Κ	Control	0
Fresh yield	Peat	Coriander	$6.32(5.36)^*$	15.32(2.27)	6.57(4.67)	3.55(2.23)	9.19(7.03)	3.68(2.38)
	Peat-Free		2.03(2.91)	5.36(2.44)	2.12(2.41)	1.84(2.44)	10.00(10.50)	1.96(2.70)
Dry Yield	Peat	Coriander	0.61 (0.61)	1.72(0.18)	0.67(0.59)	0.28(0.27)	1.28(1.49)	0.29(0.24)
	Peat-Free		0.24(0.38)	0.67(0.29)	0.27(0.31)	0.22(0.30)	1.15(1.18)	0.24(0.35)

Table 13: Mean Yield, Coriander, standard deviation (\*)

## 5.3.3 Leaf Diameter

The highest mean leaf diameter for both crop types was Coriander (Figure 38), with a diameter of 2.62cm in Peat substrates. The most effective substrate for promoting increased leaf diameter was Peat for both crop types with a control mean of 2.33cm for Coriander and 1.82cm for Basil (see Figure 37). The most effective microbial amendment for increased leaf diameter was treatment "A" at 2.62cm, a 0.29cm increase on the control for the same substrate (Peat) and crop (Coriander), see Tables 15 and 14. Basil leaf diameter was significantly (p < 0.005) lower for the majority of treatments when compared to Coriander. Two randomly selected leaves for Coriander and Basil can be seen in Figure 36.



Figure 36: Random selection of Basil and Coriander leaf with cm ruler for scale.

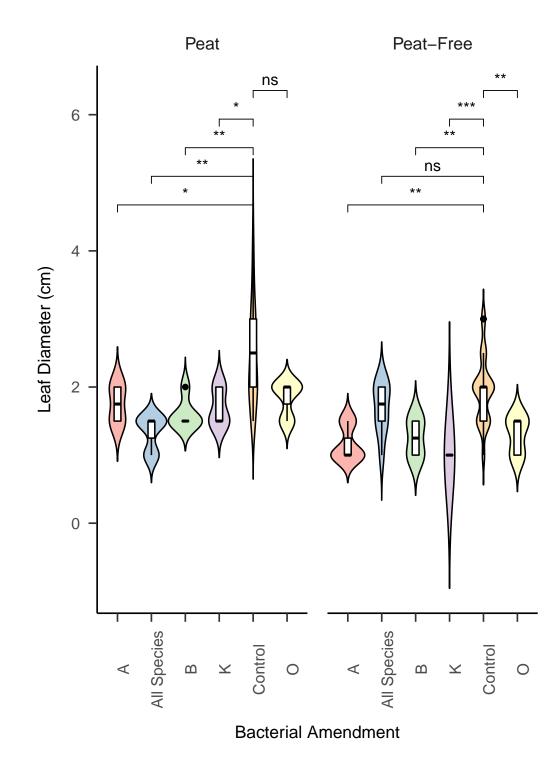


Figure 37: Leaf Diameter for Basil croppings in Peat and Peat-Free substrates shown in a violin graph. The distribution of the data is visible either side of the box-plots. The peat substrates show significant differences for species A, All species, B and K compared to the control with a more consistent, although decreased leaf diameter. Peat-free substrates are less evenly distrubuted regarding leaf diameter compared to peat, which may be a reflection of decreased substrate homogenity rather than treatment effect.

PeatBasil $1.75 (0.29)^*$ $1.33 (0.29)$ $1.60 (0.22)$ $1.70 (0.27)$ $1.82 (0.24)$ $1.83 (0.29)$ $0.052$ Peat-Free- $1.17 (0.29)$ $1.69 (0.37)$ $1.25 (0.29)$ $1.00 (0.00)$ $1.76 (0.33)$ $1.30 (0.27)$ $<0.00$	Substrate	Crop	А	All	В	К	Control	0	*p*
Peat-Free - $1.17(0.29)$ $1.69(0.37)$ $1.25(0.29)$ $1.00(0.00)$ $1.76(0.33)$ $1.30(0.27)$ < $0.00$	Peat	Basil	$1.75 (0.29)^*$	1.33(0.29)	1.60(0.22)	1.70(0.27)	1.82(0.24)	1.83(0.29)	0.052
	Peat-Free	-	1.17(0.29)	1.69(0.37)	1.25(0.29)	1.00(0.00)	1.76(0.33)	1.30(0.27)	$<\!0.001$

Table 14: Mean Leaf Diameter Basil, standard deviation (\*)

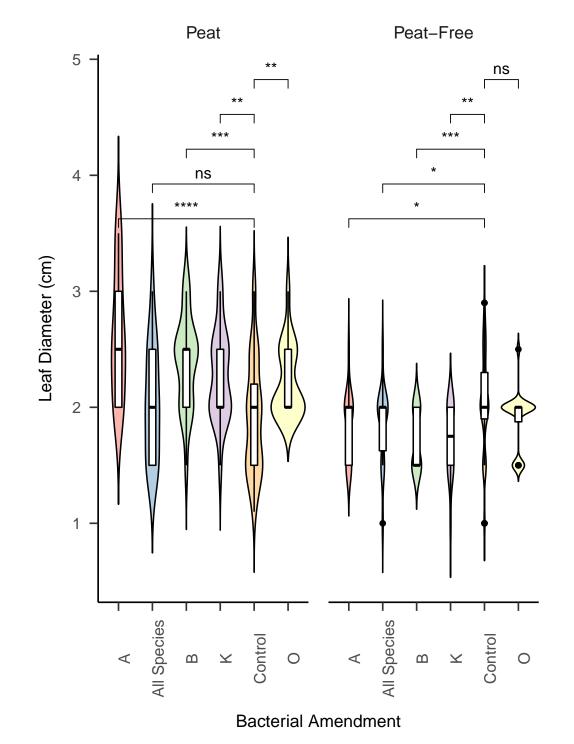


Figure 38: Leaf Diameter for Coriander in both Peat and Peat-Free substrates. Peat substrates out-performed peat-free in regards to increased leaf diameter, most notably with species A with a mean leaf diameter of 2.62cm vs the control of 2.33cm. Peat-free substrate achieved a lower mean than the control for peat with a mean control value of 1.8cm for peat-free control and no treatment surpassing a mean leaf diameter of 2cm. Less data was available for Peat-Free, therefore a reduction in the violins distribution is evident.

Table 15: Mean Leaf Diameter Coriander

Substrate	Crop	А	All	В	Κ	Control	0	Pval
Peat	Coriander	2.62(0.55)	2.03(0.48)	2.38(0.39)	2.26(0.39)	2.33(0.37)	2.22(0.31)	0.004
Peat-Free	-	1.84(0.29)	1.86(0.33)	1.73(0.26)	1.72(0.31)	1.80(0.34)	1.90(0.26)	0.359

## 5.3.4 Root length

Root length was measured at select harvest intervals of 3, 5 and 14days.

The root length of each harvested plant was measured against a ruler from the point of surface emergence on the main stem to end of root, indicated by a sudden lack of cholorphyll on the stem (see Figure 39 (a) and (b)). Measurements of root length only took place on Coriander and only included two treatments, a control and the all species mix. Differences between substrate type (Peat vs. Peat-Free) were insignificant, as was the effect of bacterial treatments on increasing root length (see Figure 40 and Table 16).



(a) Cleaned root

(b) Root in-situ

Figure 39: Root growth for Coriander. (a) demonstrating root length at GS11 (First true leaf emerging). (b) shows roots in situ at a much later growth stage. Both pictures show the difficulty in extracting whole roots from the substrate. Retaining intact specimens proved challenging and may not be a true relection of actual root length.

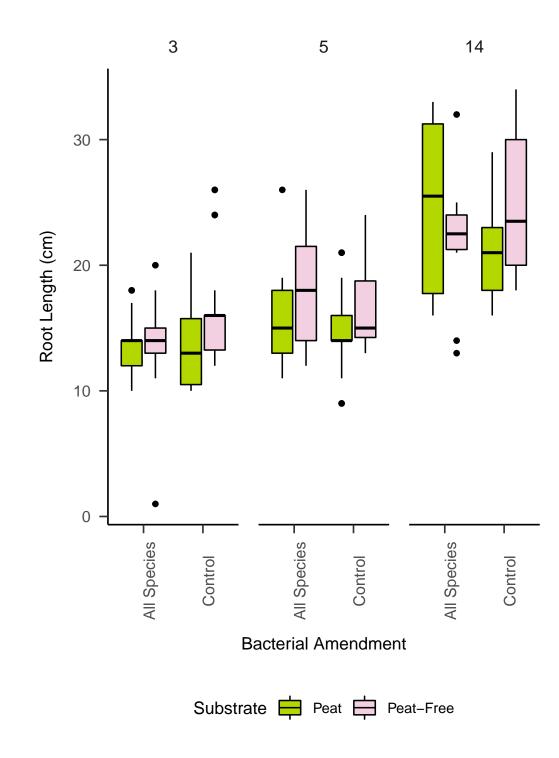


Figure 40: Root length, Coriander. Root length can be a clear indicator of crop health and substrate suitability. This graph shows increased mean length in roots for peat substrate at day 14 for the all species treatment. This is not repeated in any other assessment with the control out-performing the treatment and no comparisons of means were statistically significant.

Substrate	Day	Treatment	Root Length(cm)
Peat	3	allrb	13.87(2.29)
Peat	5	allrb	$15.71 \ (3.79)$
Peat	14	allrb	24.75(7.40)
Peat	3	none	$13.60 \ (3.06)$
Peat	5	none	14.68(2.86)
Peat	14	none	$21.30 \ (3.56)$
Peat-Free	3	allrb	$13.67 \ (4.17)$
Peat-Free	5	allrb	17.93(4.73)
Peat-Free	14	allrb	22.00(5.42)
Peat-Free	3	none	15.93(4.04)
Peat-Free	5	none	$16.53\ (2.97)$
Peat-Free	14	none	25.05(5.92)

Table 16: Root Length for Coriander

## 5.3.5 RGB

Basil was assessed further at harvest for Red-Blue-Green colour values, see Figure 41. This assessment was implemented in order to gain additional insight into treatment effects on crop quality, increased values of either Red, Green or Blue light can be directly correlated to increasing concentrations of various phytochemical compounds (Özreçberoğlu and Kahra-manoğlu 2020). Basil was selected due to limited Phytometric changes on croppings apparent from biological amendments. The sensor recorded data at x4 randomly selected leaves at crop cover height per pot. There was no significant changes in RGB values for either substrate type or treatment (see Table 17).

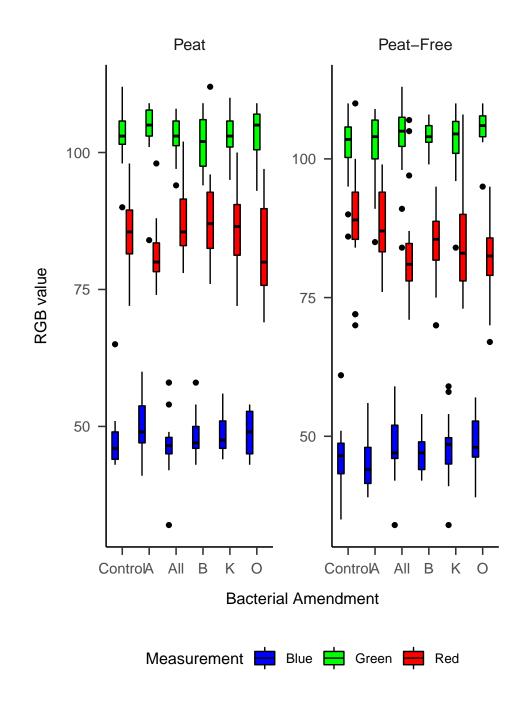


Figure 41: Red, Green, Blue values for crop quality using Rhziobacterial and AMF on potted Basil. The boxplots show the range each colour registered per treatment. Outliers shown here omitted for analysis.

Table 17: The meadian values combined RGB values throughout the trial period for each bacterial species (A, B, K, O) and AMF.

	A 5 4 1	D 541	TZ 741	0 5.11
	A, $n = 541$	B, $n = 541$	K, $n = 541$	O, $n = 541$
Peat	80(54, 103)	86 (48, 102)	87(50, 98)	86 (51, 101)
Peat-Free	85~(48,~100)	81 (52, 103)	86~(49,~103)	$83\ (50,\ 101)$
	Control, $n = 541$	All Species, $n = 541$		
Peat	86(49, 101)	80 (53, 100)		
Peat-Free	88 (49, 101)	82(53, 104)		

## 5.4 Discussion

Overall, individual PGPR had a varied and ill-defined effect on crop growth. All species mix of PGPR (A + B + K + O) showed significant effects on improving crop height for both Peat and Peat-Free media, but only in Coriander, Basil has however been previously demonstrated as significantly influenced by the introduction of PGPR previously (Ordookhani, Sharafzadeh, and Zare 2011). Leaf diameter did not follow this trend, with species "A" having the most significant impact on growth. Root length in Coriander was not affected by the addition of microbial amendments. Fresh Yield demonstrated significant increase in biomass in Peat growing media with the combined treatment of A + B + K + O, with a similar pattern repeating in Dry Yield.

Individual bacterial species performed relatively poorly. This result may be subject to the unique element of multiple PGPR species working at various levels to promote plant health (such as the adsorption of Phosphorus resources), rather than an independent introduction such as a single species.

This effect of co-inoculated species out-performing single species inoculation may have been influenced by several key factors. For instance; the lack of a gnotobiotic or axenic environment (Basic and Bleich 2018). Each substrate type was not sterile, as the cost of such a procedure would be commercially uneconomical. The presence of fungi and bacteria *in situ* prior to inoculation may have had an inhibition effect on a single species.

PGPR can be demonstrated as having a range of productivity in regards to increasing plant health or stress resistance, most notably by proximity and affinity for the rhizome of the target species (Gray and Smith 2005). The proximity of a single species may be not have been effective in retaining or gaining access to root sites due to increased competition from microbiological activity already present.

Ahemad and Kibret (2014) demonstrated the ability for multiplicities mixes of PGPR to perform well in potted crops, compared to single-inoculated species. However, this study also highlighted the ability for suppression of activity due to the synthesis of products such as antibiotics from various rhizobacterial species. This suppression may have been subdued in co-inoculated pots due to the different microbial processes from a variety of PGPR species, such as nitrogen fixation, phosphate absolution, habitation of rhizosphere etc.

Another argument is that lack of Arbuscular Mycorrhizal Fungi *in situ*, and resultant lack of mobile Phosphorus resources from symbiosis (Smith and Read 2008) inhibits PGPR, specifically antibacterial, to fix atmospheric nitrogen, demonstrated through the increased root modulation of PGPR under influence (Leij 1998; Jia, Gray, and Straker 2004).

In future studies, further study of microbial interactions through the isolation of metabolic products produced by both *in situ* and introduced microbiota may support this hypothesis of invasive co-inoculated species out-performing single species introductions.

The co-inoculation of PGPR appears to be effective in increasing crop yield and height for both Peat and Peat-Free growing media, with significantly less impact on Basil croppings compared to Coriander.