



## SOIL & CROP SCIENCES | RESEARCH ARTICLE

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# An evaluation of *Sargassum* seaweed media compositions on the performance of hot pepper (*Capsicum chinense* Jacq.) seedling production

Marcus N.A. Ramdwar<sup>1\*</sup>, Valerie A. Stoute<sup>2</sup> and Brandon S. Abraham<sup>1</sup>

**Abstract:** There has been a massive influx of *Sargassum* in the Caribbean causing devastating consequences. The current study investigated the utilization of *Sargassum* (a mixture of the *Sargassum flutens* and *Sargassum natans* species) as a substrate formulated with imported promix. Experiments were conducted during April to May in 2016 to evaluate the performance and quality of hot pepper seedlings using *Sargassum*-promix formulations. The seedlings were visually evaluated on the 45th day after sowing by an independent panel to visually score seedling quality. At the same time, the sample mean and standard deviation were calculated for each of nine seedling physical characteristics. A one-way MANOVA analysis, with formulation ratio as the independent variable and these nine characteristics as correlated dependent variables, was carried out using SPSS V.22. Introducing *Sargassum* into the formulation caused statistically significant differences in the majority of the seedling characteristics. However, these actual differences are small and are unlikely to practically impact seedling production except with one characteristic, where the change is beneficial. The treatment formulations of *Sargassum* significantly improved the sturdiness of the seedlings. *Sargassum* can be successfully utilized to reduce the reliance on 100% imported commercial promix.

**Subjects:** Environment & Agriculture; Bioscience; Food Science & Technology

**Keywords:** *Sargassum*; seaweed; peat moss; promix; seedlings; sturdiness

### ABOUT THE AUTHORS

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### PUBLIC INTEREST STATEMENT

There has been an annual massive influx of *Sargassum* seaweed in the Caribbean within recent years impacting aquatic resources, fisheries, shorelines, waterways and tourism. *Sargassum* seaweed is an underexploited marine plant which can be explored for utilization in various spheres of agricultural productivity. The overharvesting of peat from wetlands for horticultural purposes is of an environmental concern globally. Crop production in Trinidad and Tobago and the wider Caribbean is dependent on the utilization of seedlings produced using imported promix composed of 90% sphagnum peat moss. The results presented demonstrate that *Sargassum* has the potential for reducing the reliance on 100% promix for seedling production thereby possibly reducing the environmental burden attributed to the demand for harvesting peat from wetlands.

## 1. Introduction

Internationally, seaweed is regarded as an underutilized bio-resource leading to many questions about its potential for utilization in agriculture and other areas of economic activity (Ashtalakshmi & Prabakaran, 2015). Several studies have already established a wide range of beneficial effects of seaweed extracts on plants, such as early seed germination, improved crop performance and yield and elevated resistance to biotic and abiotic stresses (Eryas et al., 2008; Jayaraman, Norrie, & Punja, 2011; Norrie & Keathley, 2006). Additionally, seaweeds can be used as a soil amendment (Gandhiyappan & Perumal, 2001; Han & Guoming, 2000), in pest control (Hong, Hien, & Son, 2007), in plant disease management (Jayaraj, Wan, Rahman, & Punja, 2008) and as a source of plant growth hormones (Han & Guoming, 2000). Seaweed extracts, as natural regulators, have induced increased crop yield and plant vigor to withstand adverse environmental effects (Anisimov & Chaikina, 2014). These extracts contain all major and minor plant nutrients, including those with bio-control properties. They also contain organic compounds such as auxins, gibberellins, and precursors of ethylene and betaine, which impact plant growth (Durand, Briand, & Meyer, 2003; Ördög, Stirk, Van Staden, Novák, & Strnad, 2004; Stirk, Novák, & Strnad, 2003). Crouch and van Staden (1993) reported that seaweed extracts are bioactive at low concentrations diluted to 1:1,000 or more. The presence of growth hormones, nutrients, and other important physiochemical compounds has been reported to have an ameliorating effect on germination behavior of seed under salt stress (Salma et al., 2014). Seaweed contains essential mineral elements which are not found in water or in crops grown in mineral-depleted soil (Thomas, Chauhan, Patel, & Panchal, 2013). The mineral fraction of some seaweed species can account for up to 40% of dry matter (Krishnaiah, Sarbatly, Prasad, & Bono, 2008).

A serious environmental and economic issue, which has emerged in the last few years, particularly in 2015, is the massive influx of *Sargassum* seaweed along the coastlines of Trinidad and Tobago and other islands within the Caribbean. *Sargassum* seaweed is macroscopic brown floating invasive algae found in the Atlantic Ocean. The main species of *Sargassum*, which have been emerging in the Caribbean, are *Sargassum natans* and the *Sargassum fluitans*, both of which consolidate into large floating mats. Globally, both species of *Sargassum* have broad distributions offshore in the North Atlantic (Schneider & Searles, 1991) and, outside of the Sargasso Sea, there are more *Sargassum* mats floating in the Gulf of Mexico than anywhere else in the world (Gower & King, 2008). The invasion of *Sargassum* is negatively impacting the shorelines, fisheries, and tourism in many of the Caribbean islands. In Tobago, the *Sargassum* influx has been declared a disaster and as much as US 454,000 has been allocated for clean-up efforts across Tobago (George, 2015).

Many crops grown in Trinidad and Tobago are established from transplants grown in celled-trays using peat moss as the growing medium. Peat moss is imported into Trinidad and Tobago and is the common growing medium for the production of quality seedlings. Peat is a natural and nonrenewable resource obtained from wetlands and its large-scale utilization is of great environmental concern (Abad, Noguera, & Burés, 2001). As a consequence of overharvesting many countries have begun to limit the extent of peat moss mining, leading to price increases (Jung & Yang, 2014). Although both *Sargassum* and peat are natural resources, the former is a renewable marine resource.

Currently, *Sargassum* stockpiles from beach clean-up activities are not being utilized, making it imperative that suitable uses for this marine biomass be explored. Besides the value gained by reducing the environmental problem, the utilization of any local or regional substrate for crop production would have an economic benefit in food production. The current study investigates the performance of hot pepper (*Capsicum chinense* Jacq.) grown on *Sargassum* media to determine its suitability for the production of high quality seedlings.

## 2. Materials and methods

### 2.1. Study location

The study was conducted in a semi-enclosed type of shade house at the Eastern Caribbean Institute of Agriculture and Forestry (ECIAF), Centeno, Trinidad. ECIAF is located at 10°35'20.65" North 61°19'25.28" West, and is approximately 17 m above mean sea level.

### 2.2. Substrate preparation and treatments

*Sargassum* seaweed was collected from Manzanilla beach on the South East coast of Trinidad and washed to remove excess salt. Although other authors (Lum Lee, 2015) have identified these two species as those washing ashore on Caribbean beaches, actual verification of the species collected was done using the summary of morphological characteristics contained in a publication by Széchy, Guedes, Baeta-Neves, and Oliveira (2012). The seaweed was left exposed for aeration and allowed to decompose for a period of 6 months. It was frequently watered and turned to ensure uniform decomposition. The final compost was oven dried at a temperature of 60°C for 72 h until constant weight, following which it was manually crushed into a granular-like structure. The commercial substrate used for this study was sphagnum peat moss (certified to be a minimum of 90%). The treatments used for the study are summarized in Table 1.

### 2.3. Sowing and seedling management

The polystyrene commercial germination trays (32 cm W, 64 cm L and 6 cm H) used had 128 conical cavities, each with a capacity of 25 ml. Before use, the germination trays were disinfected by submerging them for 20 min in chlorinated water (100 ppm). Moruga Red hot pepper (*Capsicum chinense* Jacq.) seeds were procured from the Caribbean Agricultural Research Development Institute (CARDI). The germination percentage for the pepper seeds, according to the International Seed Testing Association rules (ISTA, 1985), was calculated prior to experimentation to be 100%. The respective treatment substrates were watered up to field capacity. One seed was placed in each of the 128 cells in the germination tray on April 01, 2016. Each cell was filled to seven-eighths of its capacity with the specific treatment substrate. The seed was placed at 0.5 cm depth into each cell and covered with the substrate. The trays were watered liberally throughout the study such that water flowed from the holes at the bottom of the tray. The watering removed the residual salt from the respective treatments. The treatments were monitored daily and the emergence date was recorded for each treatment (Seeds were recorded as emerged when the radicals appeared above the surface of the substrate medium). The Speed of Germination Index (SGI) was calculated using the formula in Equation (1) for counts made at several intervals, "i", from the first count to the final count "n" after sowing.

$$\text{Speed of germination index (SGI)} = \frac{\sum_{i=1}^n (\text{No. of germinated seeds})_i}{\sum_{i=1}^n (\text{Days before } i\text{th count})} \quad (1)$$

The final count for germination and the treatment in tray germination percentage were recorded 10 day after sowing (DAS). The treatment germination percentage at day 10 was calculated using the formula in Equation (2):

**Table 1. Treatment compositions**

Treatment	Media composition	Ratio (w/w)
A	100% commercial substrate (90% sphagnum peat moss)	1:0
B	100% <i>Sargassum</i> compost	0:1
C	50% commercial substrate and 50% <i>Sargassum</i>	1:1
D	25% commercial substrate and 75% <i>Sargassum</i>	1:3

$$\text{Germination percentage} = \frac{\text{No. of germinated seeds 10 DAS}}{\text{No. of cells in the seedling tray}} \times 100 \quad (2)$$

The trays were initially kept in an enclosed area but once germination began the trays were moved to a greenhouse to prevent seedling etiolation. All samples were treated weekly with a complete foliar fertilizer (20:20:20) after the emergence of the cotyledon leaves. Given the semi-enclosed characteristics of the greenhouse used in this study, external weather conditions could have influenced internal greenhouse conditions. These external conditions were mean daily temperature of 29°C, humidity average of 75.7%, and mean daily rainfall of 2.5 mm

#### **2.4. Qualitative evaluation of seedlings**

The seedlings in all treatments at 45 DAS were visually evaluated for quality using a ranking from “1” to “5”, according to Justus and Kubota (2010). The panel for evaluating the seedlings was made up of five individuals comprising two hot pepper farmers, one agricultural student, one agronomist and one random individual, with no agricultural background. The panel was asked to select the tray which they thought would be the best option to transplant in the field.

#### **2.5. Quantitative evaluation of seedlings**

Twenty-five seedlings were randomly selected 45 DAS from each tray and washed with water to remove the substrate adhered to the roots. A total of 100 seedlings from all four treatments were evaluated. The measured variables for evaluation were seedling height (cm), stem diameter (mm), height to stem diameter ratio, leaf area, fresh weight of shoot (g), dry weight of shoot (g), fresh weight of root (g) and dry weight of root (g).

#### **2.6. Seedling height (cm)**

The seedling height was determined using a centimeter measuring tape. The height was determined by placing the tape on the substrate and the measurement was taken from the apical meristem.

#### **2.7. Stem diameter (mm) and shoot height to diameter ratio (sturdiness quotient)**

The diameter of each seedling was determined using digital vernier calipers with an accuracy of 0.01 mm. The stem diameter was measured on the main stem of the plant, 1 cm above the substrate. The stem diameter is important for providing robustness to seedlings and is the most practical characteristic for the classification and identification of high quality seedlings (Mello, Trevisan, & Steiner, 2016). The sturdiness quotient (SQ) [shoot length (cm)/collar diameter (mm)] was determined for each seedling. High SQ values, which are indicated of slender seedlings, are likely to result in an unfavourable response when transplanted.

#### **2.8. Fresh and dry weights (g)**

All weights were obtained using a digital top loading balance. Individual seedlings from each treatment were separated into shoots and roots and the respective fresh weight (g) for each section was recorded. The shoots and roots for each seedling were placed into a small brown paper bag and dried in a forced air circulation oven for three days at 65°C. After the samples were allowed to cool in a desiccator, the dry weights (g) were recorded for the shoots and for the roots for each seedling.

#### **2.9. Number of leaves and leaf area**

Ten seedlings from each treatment were randomly selected and the true leaves were counted and removed. For each seedling, the total leaf area (cm<sup>2</sup>), for the leaves from each treatment, was determined using a CI-202 portable leaf area meter.

#### **2.10. Statistical analysis**

Sample means and standard deviations were calculated for the physical characteristics of the seedling samples in each treatment group. One-way MANOVA analyses, using SPSS V.22. General Linear Model (GLM) Multivariate routine, were carried out for “Treatment” as the independent variable and seedling height, stem diameter, height to diameter ratio, fresh and dry weights of shoots and roots

as the dependent variables in one analysis and number of leaves and leaf area (cm<sup>2</sup>) per plant as the dependent variables in another. Two separate MANOVA analyses had to be done because the two dependent variables in the second analysis have only 10 measurements for each treatment, whereas 25 measurements were made for each of the dependent variables in the first analysis.

### 3. Results

#### 3.1. Qualitative seedling quality assessment

All treatments received a visual score of “5” indicating that the seedlings were green and completely healthy with no visual decrease in quality. Although, the seedlings in all treatments were healthy, the pepper farmers and the agronomist independently selected treatment C (50% commercial substrate + 50% *Sargassum*) as the optimal treatment for field transplanting.

#### 3.2. Germination parameters

The germination % and SGI values are given in Table 2 for the four treatments. The commercial substrate used is promix. There was a decline in the germination index in relation to increasing levels of *Sargassum* for treatments C (50% *Sargassum*), D (75% *Sargassum*) and B (100% *Sargassum*). Treatment C had the SGI closest to the commercial substrate, Treatment A. The other *Sargassum* formulations had SGI values between 18 and 21% slower than treatments A and C, although their germination % values were not that far off—Table 2. Note that the SGI reflects how quickly the process of germination takes place but it is the equilibrium germination % achieved in a given practical period which is important. Although the 100% *Sargassum* germinated at the slowest speed, its final germination percentage after 10 days was nearly equal to that of the commercial product. This comparison cannot be tested statistically because each of these values is not a mean of individual replicate seedling values but a single parameter calculated for each treatment.

#### 3.3. Quantitative comparisons of treatment effects

The sample means and standard deviations are given in Table 3 for each of the nine physical characteristics of the seedlings in each of the four sample groups. The sample means of the seedling heights and the height to diameter ratios are largest for the 100% commercial substrate (promix) and the least for the 100% *Sargassum* treatment, but there appears to be little difference in the stem diameters produced in the seedlings exposed to the four different treatments. These differences in the means of the physical characteristics of the samples from the different treatment groups were tested statistically since the standard errors for the measurements, which measure the variability among replicates, must be taken into consideration in assessing whether a difference in two means is truly statistically significant or not.

The results of the four omnibus MANOVA tests show that, when the seven dependent variables of the first analysis are treated together as one linear combination (one variate), all four treatments give significantly different results (the centroids are significantly distinct since  $p = 0.000$  for all inference tests). The same is true for the omnibus tests for the 2nd analysis using number of leaves

**Table 2. Treatment effect on germination percentage and SGI**

Treatment	Germination(%)	Speed of germination index (SGI)
100% promix (A)	96	11.8
100% <i>Sargassum</i> (B)	93	9.3
50%/50% <i>Sargassum</i> /promix (C)	92	11.1
75%/25% <i>Sargassum</i> /promix (D)	85	9.7

**Table 3. Means and standard deviations for physical characteristics of treated seedlings**

Treatment		Seedling height (cm)	Stem diameter (mm)	Height/diameter ratio	Fresh weight shoot	Dry weight shoot	Fresh weight root	Dry weight root	No. of leaves	Leaf area (cm <sup>2</sup> )
100% promix	Mean	10.18	2.20	4.64	1.46	0.19	0.69	0.06	7.10	55.32
	N	25	25	25	25	25	25	25	10	10
	SD	0.85	0.09	0.51	0.20	0.04	0.21	0.01	0.88	8.65
100% <i>Sargassum</i>	Mean	5.42	2.04	2.67	0.82	0.13	0.24	0.04	7.10	38.854
	N	25	25	25	25	25	25	25	10	10
	SD	0.62	0.20	0.29	0.14	0.03	0.07	0.01	0.74	6.23
50/50 <i>Sargassum</i> /promix	Mean	8.71	2.33	3.75	1.59	0.25	0.68	0.07	7.70	58.33
	N	25	25	25	25	25	25	25	10	10
	SD	0.78	0.17	0.39	0.23	0.04	0.22	0.01	0.48	7.72
75/25 <i>Sargassum</i> /promix	Mean	6.54	2.19	2.98	1.25	0.21	0.60	0.10	8.00	49.87
	N	25	25	25	25	25	25	25	10	10
	SD	0.88	0.14	0.36	0.21	0.04	0.16	0.15	0.67	6.02
Total	Mean	7.71	2.19	3.51	1.28	0.19	0.55	0.07	7.48	50.59
	N	100	100	100	100	100	100	100	40	40
	SD	2.02	0.19	0.86	0.35	0.06	0.25	0.08	0.78	10.24

**Table 4. Tests of between subjects effects—analysis 1**

Source	Dependent variable	Type III sum of squares	df	Mean square	F	Sig.
Corrected model	Seedling height (cm)	342.289 <sup>a</sup>	3	114.096	182.908	0.000
	Stem diameter (mm)	1.070 <sup>b</sup>	3	0.357	14.357	0.000
	Height to diameter ratio	58.186 <sup>c</sup>	3	19.395	125.589	0.000
	Fresh weight shoot	8.603 <sup>d</sup>	3	2.868	73.434	0.000
	Dry weight shoot	0.197 <sup>e</sup>	3	0.066	44.068	0.000
	Fresh weight root	3.324 <sup>f</sup>	3	1.108	36.363	0.000
	Dry weight root	0.053 <sup>g</sup>	3	0.018	2.966	0.036
Intercept	Seedling height (cm)	5,949.037	1	5,949.037	9,536.897	0.000
	Stem diameter (mm)	480.486	1	480.486	19,348.446	0.000
	Height to diameter ratio	1,231.950	1	1,231.950	7,977.130	0.000
	Fresh weight shoot	164.019	1	164.019	4,200.022	0.000
	Dry weight shoot	3.814	1	3.814	2,563.887	0.000
	Fresh weight root	30.780	1	30.780	1,010.107	0.000
	Dry weight root	0.496	1	0.496	83.131	0.000
Treatment	Seedling height (cm)	342.289	3	114.096	182.908	0.000
	Stem diameter (mm)	1.070	3	0.357	14.357	0.000
	Height to diameter ratio	58.186	3	19.395	125.589	0.000
	Fresh weight shoot	8.603	3	2.868	73.434	0.000
	Dry weight shoot	0.197	3	0.066	44.068	0.000
	Fresh weight root	3.324	3	1.108	36.363	0.000
	Dry weight root	0.053	3	0.018	2.966	0.036

(Continued)

**Table 4. (Continued)**

Source	Dependent variable	Type III sum of squares	df	Mean square	F	Sig.
Error	Seedling height (cm)	59.884	96	0.624		
	Stem diameter (mm)	2.384	96	0.025		
	Height to diameter ratio	14.826	96	0.154		
	Fresh weight shoot	3.749	96	0.039		
	Dry weight shoot	0.143	96	0.001		
	Fresh weight root	2.925	96	0.030		
	Dry weight root	0.572	96	0.006		
Total	Seedling height (cm)	6,351.210	100			
	Stem diameter (mm)	483.940	100			
	Height to diameter ratio	1,304.961	100			
	Fresh weight shoot	176.372	100			
	Dry weight shoot	4.154	100			
	Fresh weight root	37.030	100			
	Dry weight root	1.121	100			
Corrected total	Seedling height (cm)	402.173	99			
	Stem diameter (mm)	3.454	99			
	Height to diameter ratio	73.012	99			
	Fresh weight shoot	12.352	99			
	Dry weight shoot	0.339	99			
	Fresh weight root	6.249	99			
	Dry weight root	0.625	99			

<sup>a</sup> $R^2 = 0.851$  (adjusted  $R^2 = 0.846$ ) – seedling height.

<sup>b</sup> $R^2 = 0.310$  (adjusted  $R^2 = 0.288$ ) – stem diameter.

<sup>c</sup> $R^2 = 0.797$  (adjusted  $R^2 = 0.791$ ) – height to diameter ratio.

<sup>d</sup> $R^2 = 0.696$  (adjusted  $R^2 = 0.687$ ) – fresh weight shoot.

<sup>e</sup> $R^2 = 0.579$  (adjusted  $R^2 = 0.566$ ) – dry weight shoot.

<sup>f</sup> $R^2 = 0.532$  (adjusted  $R^2 = 0.517$ ) – fresh weight root.

<sup>g</sup> $R^2 = 0.085$  (adjusted  $R^2 = 0.056$ ) – dry weight root.

and leaf area as the dependent variables ( $p = 0.000$  for all 4 omnibus tests). In Tables 4 and 5, on the other hand, results are detailed for when each outcome is tested independently, instead of as a group. Also given, in rows at the bottom of the table, are the  $R^2$  values for the goodness of fit of each linear model for a single outcome. In the first analysis, the four treatments are significantly different for each single one of the seven outcome characteristics ( $p = 0.000$  for all except the dry weight root, for which  $p = 0.036$ ).  $R^2$  values vary between 0.517 and 0.851, indicating good model fits except for stem diameter ( $R^2 = 0.31$ ) and dry weight root ( $R^2 = 0.085$ ) as outcomes. Further,  $R^2$  and  $R^2_{adj}$  values are close to each other in all cases so there is little variance inflation in the  $R^2$  values. For analysis 2, all four treatments produce significantly different results both in the number of leaves found and in the leaf area, with the latter having a good model fit ( $R^2 = 0.539$ ). The Linear Model for each outcome then has the generic form given in Equation (3):

$$\text{Seedling characteristic} = \text{Intercept (base value)} + \text{Treatment impact on the characteristic} + \text{Error} \quad (3)$$

**Table 5. Tests of between subjects effects—analysis 2**

Source	Dependent variable	Type III sum of squares	df	Mean square	F	Sig.
Corrected model	Number of leaves	6.075 <sup>a</sup>	3	2.025	4.073	0.014
	Leaf area (cm <sup>2</sup> )	2,204.799 <sup>b</sup>	3	734.933	14.046	0.000
Intercept	Number of leaves	2,235.025	1	2,235.025	4,495.022	0.000
	Leaf area (cm <sup>2</sup> )	102,381.007	1	102,381.007	1,956.723	0.000
Treatment	Number of leaves	6.075	3	2.025	4.073	0.014
	Leaf area (cm <sup>2</sup> )	2,204.799	3	734.933	14.046	0.000
Error	Number of leaves	17.900	36	0.497		
	Leaf area (cm <sup>2</sup> )	1,883.617	36	52.323		
Total	Number of leaves	2,259.000	40			
	Leaf area (cm <sup>2</sup> )	106,469.422	40			
Corrected total	Number of leaves	23.975	39			
	Leaf area (cm <sup>2</sup> )	4,088.416	39			

<sup>a</sup>R<sup>2</sup> = 0.253 (adjusted R<sup>2</sup> = 0.191) – number of leaves.

<sup>b</sup>R<sup>2</sup> = 0.539 (adjusted R<sup>2</sup> = 0.501) – leaf area.

The results of Hypothesis tests, given in Tables 6 and 7, show how each treatment compares to the reference treatment which is 100% commercial substrate (promix). For each seedling characteristic in the columns of these two tables, the performance of the seedlings, under each treatment formulation, shown in the rows, is compared to that for the reference. Information is detailed for the point estimate for the mean difference (contrast estimate) as well as for the lower and upper bounds for the 95% confidence interval for the difference in the mean value of the characteristic. When the contrast estimate is negative, this is because the value for the reference is bigger than that for the particular treatment in that row. The rows of “sig” values (*p*-values) indicate whether a mean value for a particular treatment is significantly different from that of the reference (*p* < 0.05).

**Table 6. Contrasts of physical characteristic values for different treatment groups**

Treatment simple contrast <sup>a</sup>		Dependent variable							
		Seedling height (cm)	Stem diameter (mm)	HD ratio	Fresh weight (g) shoot	Dry weight (g) shoot	Fresh weight (g) root	Dry weight (g) root	
100% <i>Sargassum</i> vs. reference	Contrast estimate	-4.76	-0.16	-1.973	-0.64	-0.07	-0.45	-0.02	
	Sig.	0.00	0.00	0.00	0.00	0.00	0.00	0.27	
	95% CI for difference	LB	-5.19	-0.25	-2.19	-0.75	-0.09	-0.54	-0.07
		UB	-4.31	-0.07	-1.75	-0.53	-0.05	-0.35	0.02
50% <i>Sargassum</i> vs. reference	Contrast estimate	-1.47	0.13	-0.89	0.13	0.05	-0.01	0.01	
	Sig.	0.00	0.00	0.00	0.02	0.00	0.86	0.70	
	95% CI for difference	LB	-1.92	0.04	-1.11	0.02	0.03	-0.11	-0.04
		UB	-1.03	0.22	-0.67	0.24	0.07	0.09	0.05
75% <i>Sargassum</i> vs. reference	Contrast estimate	-3.64	-0.00	-1.66	-0.20	0.01	-0.09	0.04	
	Sig.	0.00	0.93	0.00	0.00	0.23	0.07	0.07	
	95% CI for difference	LB	-4.08	-0.09	-1.88	-0.32	-0.08	-0.19	-0.00
		UB	-3.19	0.08	-1.44	-0.09	0.04	0.01	0.08

<sup>a</sup>Reference category = 1 = 100% promix.

**Table 7. Contrasts of physical characteristic values for different treatment groups**

Treatment simple contrast <sup>a</sup>		Dependent variable		
		Number of leaves	Leaf area (cm <sup>2</sup> )	
100% <i>Sargassum</i> vs. reference	Contrast estimate		0.00	-16.47
	Sig.		1.00	0.00
	95% confidence interval for difference	Lower bound	-0.64	-23.03
		Upper bound	0.64	-9.91
50% <i>Sargassum</i> vs. reference	Contrast estimate		0.60	3.00
	Sig.		0.07	0.36
	95% confidence interval for difference	Lower bound	-0.04	-3.56
		Upper bound	1.24	9.56
75% <i>Sargassum</i> vs. reference	Contrast estimate		0.90	-5.46
	Sig.		0.01	0.10
	95% confidence interval for difference	Lower bound	0.26	-12.02
		Upper bound	1.54	1.11

<sup>a</sup>Reference category = 1 = 100% promix.

Note the 50% *Sargassum* treatment produces seedlings which have essentially the same dry and fresh root weights, number of leaves and leaf areas as those sown in 100% commercial substrate, the reference ( $p > 0.05$  for each characteristic)—Tables 6 and 7. The 75% *Sargassum* has values which are statistically the same as the reference in five of the nine characteristics in Tables 6 and 7, namely stem diameter, dry shoot weight, fresh and dry root weights, and leaf area. The 100% *Sargassum* has significantly smaller values than the 100% commercial substrate (promix) for every physical characteristic except the dry root weight (Table 6) and number of leaves (Table 7). Although the mean values for the stem diameter are significantly different from the reference for 100 and 50% *Sargassum* treatments, the actual differences are relatively small from a practical perspective (with the largest difference of -0.160 being just 8% of the smaller mean value). The same is essentially true for all of the other physical characteristics in both Tables 6 and 7, except seedling height and height/diameter. Except with these last two characteristics, differences from the reference, even when significant, are relatively small. This suggests that although statistically significant, the differences in most of the physical characteristics of seedlings, when *Sargassum* is introduced into the growth medium, may not matter in the practical sense for seedling production. This could be tested further. With height, though, the difference from the reference does matter. The *Sargassum* formulations lead to shorter seedlings, which have roughly the same stem diameters as those seedlings treated with the commercial substrate. This could lead to improved robustness in the *Sargassum* treatment samples. The statistical significance with small mean differences is important in that it indicates that intragroup variability (standard error) is also small, suggesting that the results in this study should be reproducible elsewhere, and at another time, under the same conditions specified here.

#### 4. Physical observations of all treated plants

The seedlings from all treatments with *Sargassum* were shorter compared to the commercial imported promix. Although the stem diameter was comparable for all treatments in the study, the internode length for seedlings grown in the promix appeared noticeably longer than the internode length for the seedlings grown in the various *Sargassum* incorporated with promix treatments. An additional evaluation beyond the main objectives of this study assessed the performance of plants exposed to each treatment, when transplanted into top-soil filled grow bags under field conditions, up to the initiation of flower bud production and development into flowers. The seedlings from the commercial promix treatment responded less favorably to transplanting when compared to the *Sargassum* treatments. The seedlings from the *Sargassum* treatments developed a fuller canopy with markedly more lateral branching, when compared to the seedlings grown using only the commercial promix media. It was observed that the seedlings grown using *Sargassum* took a shorter

time to initiate floral development and presented with a substantially greater number of flower buds and flowers, supported by the enhanced fuller canopy. Possibly, the favorability of *Sargassum* grown seedlings to withstand transplanting stresses as well as the suggested phytohormone properties within *Sargassum* contributed to the enhanced canopy development and the earlier and more prolific floral production observed (Durand et al., 2003; Ördög et al., 2004; Stirk et al., 2003).

## 5. Conclusion and future prospects

The use of *Sargassum* in the formulations with promix has proven to be effective for the commercial production of hot pepper seedlings based on the parameters investigated. Formulations replaced by as much as seventy-five percent *Sargassum* have proven to produce seedling quality comparable to that seen with conventional sphagnum based promix. For one specific characteristic, the introduction of *Sargassum* as a formulation component with promix was shown to yield even better quality. It improves the sturdiness of the seedlings, a recognized measure for seedling quality. Sturdy seedlings are less susceptible to damage from handling, wind, and drought and provide a better seedling for field transplanting purposes. The reduction in the use of promix through replacement in formulations with *Sargassum*, an underexploited renewable marine resource, results in benefits from reductions in the cost of promix purchases, in the presence of this weed on beaches, and in the overharvesting of sphagnum peat found in global wetlands. One future project involves exploration of the *Sargassum* and promix for other agronomic and horticultural uses. Another will focus on testing whether the reported ability of *Sargassum*, to act as a biosorbent for heavy metals is also seen in the species which washes up on Caribbean beaches (Davis, Volesky, & Vieira, 2000).

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The authors declare no competing interest.

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