



# Effects of Location and Season on Seaweed Spectral Signatures

Sadhvi Selvaraj, Bradley S. Case and W. Lindsey White\*

School of Science, Faculty of Health and Environmental Sciences, Auckland University of Technology, Auckland, New Zealand

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### \*Correspondence:

W. Lindsey White  
lindsey.white@aut.ac.nz

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Remote sensing is an effective tool for applications such as discriminating plant species, detecting plant diseases or drought, and mapping aquatic vegetation such as seagrasses and seaweeds. Each plant species has a unique spectral reflectance which can be used with remote sensing to map them. However, variations in season, illumination, phenological stages, turbidity or location may affect the spectral reflectance. The aim of this study is to understand the spatial and seasonal effect on two commonly found New Zealand native seaweed species, *Ecklonia radiata* (C. Agardh) J. Agardh. and *Carpophyllum maschalocarpum* (Turner) Grev. We collected hyperspectral data (using ASD Handheld2 Field spectrometer with wavelength range 325–1,075 nm) of the seaweed species from four locations across four seasons and used mixed effects modelling to determine the model that best described the spectral data of each seaweed species. The results showed some seasonal pattern across the four locations. In general, summer has an effect on both the species in all four locations; likely due to the higher rates of photosynthesis. However, location did not effect the spectral signature of either species in winter. This study shows the potential for analysis of other micro- and macro-environment factors of different species and provides an understanding of the degree of natural spectral variation in seaweeds enabling further assessment of the impact of anthropogenic activities and changing environment on their spectral characteristics and health. It also identifies a general trend for best season to collect data for better classification accuracy across larger areas.

**Keywords:** seaweed, spectral signature, season, location, *Ecklonia*, *Carpophyllum*

## INTRODUCTION

Remote sensing surveys use multispectral and hyperspectral sensors to map vegetation based on the unique spectral signature of the individual plant species. Typically for healthy plants, there is low spectral reflectance in the visible region and high reflectance in near infrared region of the spectrum attributed to its biochemical and biophysical properties (Clark et al., 2005). Effects of physiological stress due to disease, insects or drought change the plant's spectral signature and such characteristic spectral features can be exploited (using remote sensing) to address mass scale outbreaks by early detection (Kanemasu, 1974; Silva et al., 2008; Abdullah et al., 2018). However, in the terrestrial environment, there is also spectral variability within single healthy plant due to natural variations such as season, substrate features, illumination or phenological stages (Silva et al., 2008; Bue et al., 2015) which if accounted for, can improve mapping accuracy (Somers and Asner, 2014).

Anthropogenic activities such as urbanisation, deforestation and farming directly impacts the coastal environment globally (Seers and Shears, 2015), especially benthic seaweed communities (Benedetti-Cecchi et al., 2001), by reducing the amount of light that these communities receive for photosynthesis. These effects are of growing concern to fast urbanising New Zealand, particularly in the Hauraki Gulf located on the northeast of North Island (Blain and Shears, 2019). The Hauraki Gulf is home to riverine estuaries, sheltered harbours, coastal beaches and open-coast shores with recreational, cultural and economic importance (Seers and Shears, 2015) and high impacts of sedimentation on coastal environment makes it essential to monitor the health of its benthic habitats (Kelly et al., 2014). In addition, seaweeds are affected by other natural factors such as season, varying growth cycle in different seasons, nutrient flow or environment conditions (Fung et al., 2013; Kotta et al., 2014). While spectral variability due to natural variations are being explored in terrestrial environment and seaweeds are being mapped using satellite/aerial data around the world (Uhrin and Townsend, 2016), very little research has investigated the variability of spectral characteristics of seaweeds.

Reflectance properties of a species of marine vegetation can vary spatially and seasonally (Fyfe, 2003). There is little information on reflectance properties of a single marine vegetation species at different seasons (Casal et al., 2013). Researchers who have undertaken studies on eelgrass mapping in shallow waters in Canada (O'Neill et al., 2011), discrimination of invasive seaweed *Caulerpa* species in the Mediterranean (Kišević et al., 2011) and seaweeds in Baltic Sea (Kotta et al., 2014) have all recommended more research on reflectance properties of individual species across different seasons and locations for wide scale mapping applicability, accurate characterising of the discriminating spectral features of the species in question or true representation of training dataset for spectral library classification approach.

Spectral signatures from hyperspectral data enable us to identify the subtle differences in plant species better than multispectral datasets despite the common chemical composition within species (Cochrane, 2000; George et al., 2014). Therefore, in this study, using hyperspectral data, we analyse the effects of location and season on spectral characteristics of two commonly found, dominant, habitat forming seaweed species, *Ecklonia radiata* (C. Agardh) J. Agardh. and *Carpophyllum maschalocarpum* (Turner) Grev. in the Hauraki Gulf.

## MATERIALS AND METHODS

### Seaweed Collection and Spectral Measurements

Seven individuals of each species, *Ecklonia radiata* and *C. maschalocarpum*, from each of four locations located approximately 10 km apart from Te Haruhi Bay (174°49'2''E and 36°37'5''S) north to Motuora Island (174°47'36.59''E and 36°30'30.31''S), Kawau Island (174°52'33.02''E and 36°27'3.93''S), and Takatu Point (174°51'59.89''E and

36°22'12.18''S) (**Supplementary Figure 1**) in Spring (6th October 2017), Autumn (6th April 2018), Winter (23rd June 2018) and Summer (11th December 2018) were collected. Seasons were chosen as December to February (summer), March to May (autumn), June to August (winter), September to November (spring). Spectral readings of seaweed individuals, taken out of water and placed on a black mat, were measured using ASD Handheld-2 field spectrometer with wavelength range of 325–1075 and 1 nm sampling interval. The spectrometer was held at 10 cm above the individuals looking at nadir and ten readings of each seaweed individual over blade and stipe were recorded. A number of calibration readings were taken depending on the cloud conditions using Spectralon® panel. Readings in each location were recorded within an hour of taking the samples out of water. Every effort was taken to collect the readings within 2 h of solar noon but some were collected beyond the range and the location description was recorded (**Supplementary Table 1**).

## Data Analysis

### Data Pre-processing

The spectral data set from seven individuals of each seaweed species from four seasons and four locations comprised: 1,120 readings for *C. maschalocarpum*, 1,119 readings for *E. radiata*, 350 numeric wavelengths of the reflectance values between 400 and 750 nm and the categorical variables “seaweed species”, “season” and “location”. There is a missing data point for *E. radiata* due to an erroneous reading. The wavelengths included those that lie within the water transmission window (450–550 nm; Kieleck et al., 2001) as well as the NIR region that show the greatest discrimination between species (Fyfe, 2003). The data was standardised using Standard Normal Variate (SNV) technique. SNV scaling was performed on the data across the columns for each row to remove multiplicative noise from each spectrum (Wehrens, 2011). It was performed by subtracting the mean of values at all wavelengths, for each spectral signature, from the reflectance values at each wavelength and dividing the difference by standard deviation of the spectrum (Kotta et al., 2014).

### Qualitative Separation of Seaweed Species

Principal component analysis (PCA) was performed to qualitatively assess the separation between *E. radiata* and *C. maschalocarpum*. In case of good separation of the two species, the seaweeds were assessed for spatial and seasonal effect, separately. R statistical software package was used for the analysis (R Core Team, 2019).

### Spatial and Seasonal Variability in Spectral Reflectance

Due to high dimensionality of the data, it was decomposed to few principal components (PCs) that encompassed the variation from influential wavelengths. This was implemented using PCA on the standardised data for the two seaweed species, separately. Principal component (PC) with most variance was chosen and the wavelengths that influenced this PC

were selected based on the loading values using “factoextra” package in R software (Kassambara and Mundt, 2017). For each seaweed species, to identify significant difference in spectral reflectance across various locations and seasons, a linear mixed effect model was built with each contributing PC as response variable, location and season as fixed effects, and seaweed individuals as the random effect. The difference between the individuals due to any in-plant variation in each location and season was modelled by the random effect. Linear mixed model separates the variance due to random sampling from fixed effect (Zuur et al., 2009). The explanatory variables location, season and seaweed individuals were factor variables with four, four and seven levels, respectively. Full model was built including both location, season and the interaction between the two using maximum likelihood (ML) method which was compared against a model with a term dropped (Table 1). The best model was selected using Akaike Information Criterion (AIC) and rerun using the method Restricted Maximum Likelihood. “nlme” package in R was used for this analysis (Pinheiro et al., 2018). The final model prediction plot with  $\pm 2$  Standard deviations at 95% confidence interval was plotted using “AICcmodavg” package in R software (Mazerolle, 2019). The goodness-of-fit of the model was assessed using marginal and conditional  $R^2$  value suited for mixed model (Nakagawa et al., 2013) and implemented using “MuMIn” package in R software (Barton, 2018). Marginal  $R^2$  value explains the variance due to fixed effects, conditional  $R^2$  value explains the total variance due to fixed and random effects.

## RESULTS

### Data Pre-processing and PCA

Principal component analysis (PCA) on SNV data of the two seaweed species combined showed clear grouping of *E. radiata* and *C. maschalocarpum* with the maximum variance of 86.5% on first PC. The two seaweed species were separated (Figure 1) for further analysis of effect of location and season on spectral reflectance using linear mixed effects model.

**TABLE 1** | Equations for the linear mixed effects model in R using nlme package used in this study.

Model	Formula
M1 <sub>seaweed</sub>	lme(PC1 ~ location * season, random =~ 1 replicate)
M2 <sub>seaweed</sub>	lme(PC1 ~ location + season, random =~ 1 replicate)
M3 <sub>seaweed</sub>	lme(PC1 ~ season, random =~ 1 replicate)
M4 <sub>seaweed</sub>	lme(PC1 ~ location, random =~ 1 replicate)

M1<sub>seaweed</sub> represents full model with all the variables included for a seaweed species, location, season and their interaction are fixed effects variables, random effects are represented by intercept varying over seaweed individuals; M2<sub>seaweed</sub> represents model with all the variables except interaction between location and season for a seaweed species; M3<sub>seaweed</sub> represents model with only season variable for a seaweed species; M4<sub>seaweed</sub> represents model with only location variable for a seaweed species.

### Spatial and Seasonal Variation on Spectral Reflectance of *C. maschalocarpum* Using Linear Mixed Modelling

Principal component analysis on standardised spectral data of *C. maschalocarpum* resulted in the maximum variance in first PC (93.4%).

#### Variables Contributing to PC1

Based on loading values, wavelengths that contributed to the variation in PC1 were 597–608, 693–718 and 733–750 nm. The spectral reflectance values of influential wavelengths between 733 and 750 nm were positively correlated to PC1 scores while that between 597–608 and 693–718 nm are negatively correlated and the latter has a higher magnitude.

#### Effect of Season and Location on PC1

The results of mixed effects modelling on PC1, using likelihood ratio test, showed that location, season and the interaction between them had significant effect on standardised spectral reflectance of *C. maschalocarpum*. Data is more likely under model M1<sub>C.maschalocarpum</sub> than it is under model M2<sub>C.maschalocarpum</sub> [ $L = 197.02$  ( $df = 9$ ,  $p < 0.001$ )] or model M3<sub>C.maschalocarpum</sub> [ $L = 1433.19$  ( $df = 12$ ,  $p < 0.001$ )] or model M4<sub>C.maschalocarpum</sub> [ $L = 423.77$  ( $df = 12$ ,  $p < 0.001$ )]. Model M1<sub>C.maschalocarpum</sub> performed well with lowest AIC and marginal and conditional  $R^2$  of 0.735 and 0.741, respectively (Table 2).

Winter and spring affect spectral reflectance at wavelengths between 733 and 750 nm but there was no significant difference between the two seasons at each location (Figure 2). However, in summer, spectral reflectance between 693 and 718 nm described *C. maschalocarpum* in Motuora Island and was significantly different compared to rest of the locations. The spectral reflectance between 597 and 608 nm described the seaweed in Autumn across all locations.

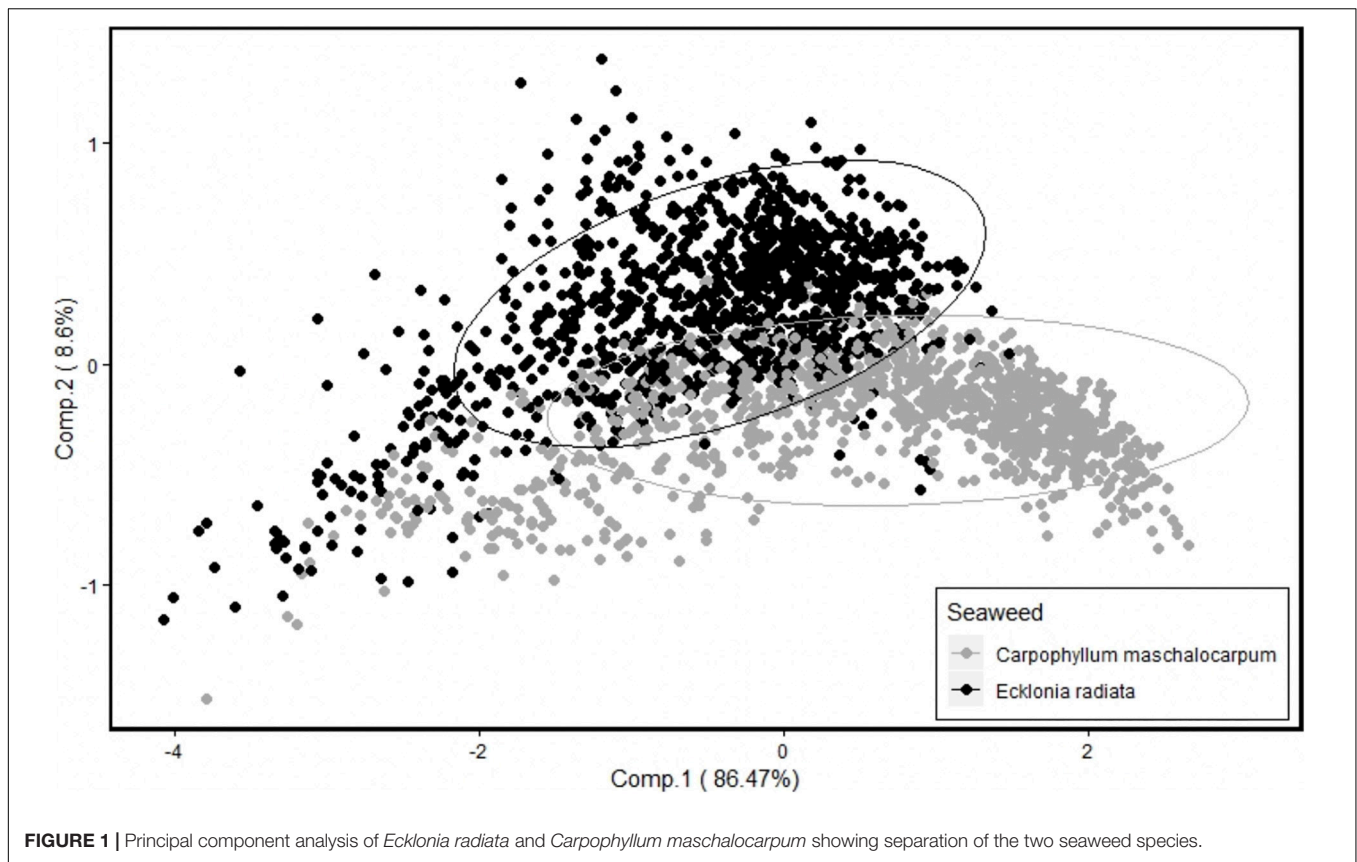
### Spatial and Seasonal Variation on Spectral Reflectance of *E. radiata* Using Linear Mixed Modelling

Principal component analysis on standardised spectral data of *E. radiata* resulted in the maximum variance of 83.4% in first PC and variance of 9.3% in second PC.

**TABLE 2** | Linear mixed model result.

Model	<i>Carpophyllum maschalocarpum</i>		<i>Ecklonia radiata</i>	
	AIC	$R^2_m$ , $R^2_c$	AIC	$R^2_m$ , $R^2_c$
M1	2255.64	0.738, 0.743	2522.54	0.457, 0.466
M2	2434.66	0.689, 0.694	2984.36	0.176, 0.181
M3	2655.41	0.62, 0.625	3068.75	0.107, 0.112
M4	3664.84	0.069, 0.07	3114.89	0.069, 0.073

$R^2_m$ , marginal  $R^2$  value;  $R^2_c$ , conditional  $R^2$  value.



### Important Variables Contributing to Components PC1 and PC2

Based on loading values, wavelengths that contributed to the variation in PC1 were 572–613, 693–715 and 730–750 nm. The spectral reflectance values in wavelengths between 730 and 750 nm are positively correlated to PC1 scores while that between 572–613 and 693–715 nm are negatively correlated and the latter has a higher magnitude.

### Effect of Season and Location on PC1

The results of mixed effects modelling on PC1, using likelihood ratio test, showed that location, season and the interaction between them had a significant effect on wavelengths that contributed to PC1. The test revealed that data is more likely under model M1<sub>*E.radiata*</sub> than it is under model M2<sub>*E.radiata*</sub> [ $L = 479.82$  ( $df = 9$ ,  $p < 0.001$ )] or model M3<sub>*E.radiata*</sub> [ $L = 616.35$  ( $df = 12$ ,  $p < 0.001$ )] or model M4<sub>*E.radiata*</sub> [ $L = 570.21$  ( $df = 12$ ,  $p < 0.001$ )]. Model M1<sub>*E.radiata*</sub> performed well with marginal and conditional  $R^2$  of 0.457 and 0.465, respectively (Table 2).

Autumn and spring had a similar effect pattern on the spectral reflectance values in wavelengths 572–613 nm in all locations (Supplementary Figure 4A). There was no seasonal effect on spectral reflectance values in wavelengths 730–750 nm that described the *E. radiata* in Kawau Island (Supplementary Figure 4B).

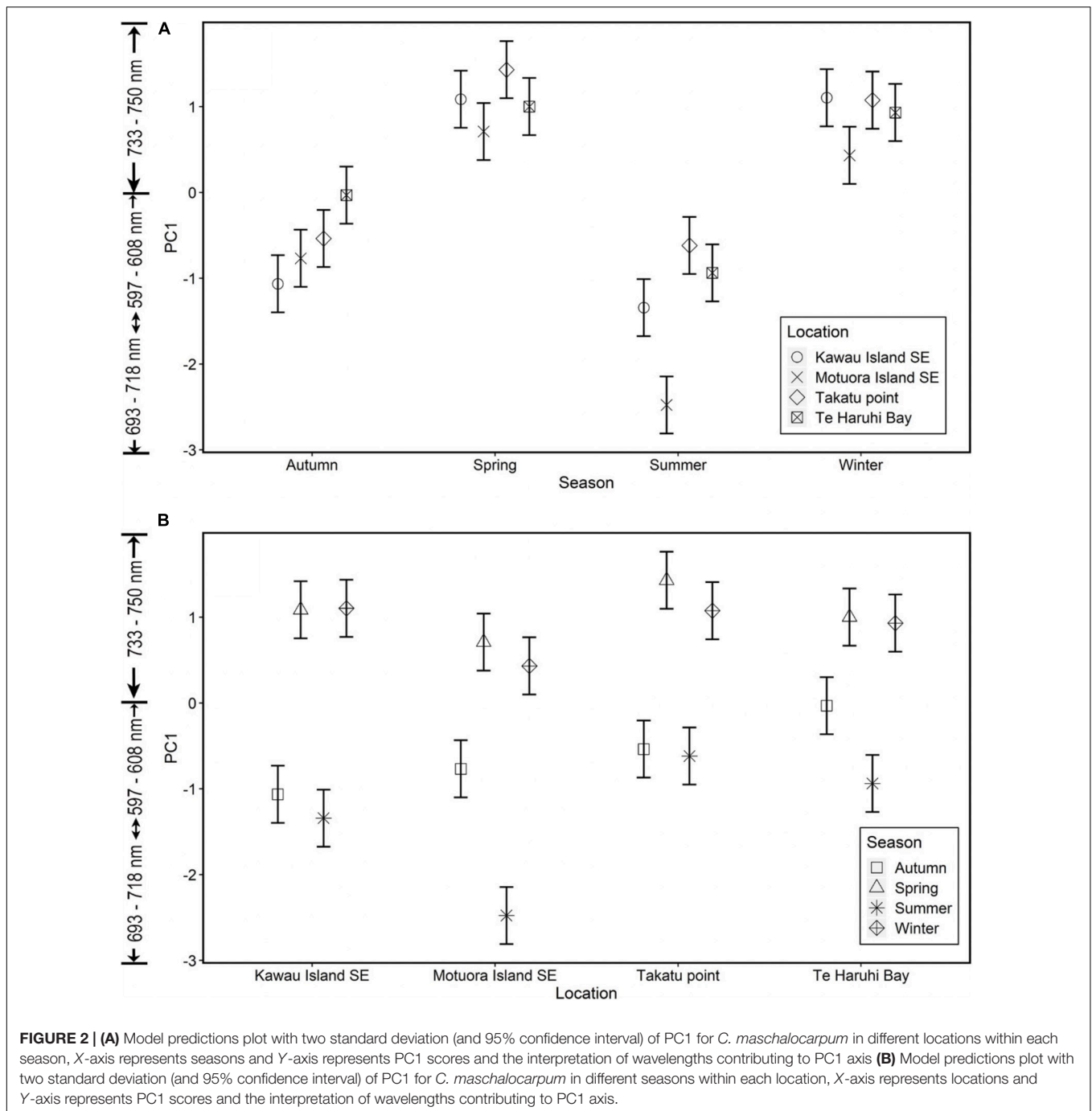
In summer, the wavelengths that described *E. radiata* in Motuora Island and Te Haruhi Bay were 693–715 nm

and, in Kawau Island and Takatu point were 730–750 nm (Supplementary Figure 4A). Location does not have a significant effect on spectral reflectance of *E. radiata* in winter and the seaweed was described by wavelengths between 730 and 750 nm.

## DISCUSSION

This study provides the first detailed investigation into the influence of season and location on the spectral signatures of *E. radiata* and *C. maschalocarpum* and addresses the shortcomings of similar past research that did not account for natural variations (Kišević et al., 2011; Kotta et al., 2014). Season and location were expected to affect the spectral reflectance of the two commonly found New Zealand native seaweed species. This study has found that wavelength regions describing *E. radiata* and *C. maschalocarpum* varies with season within each location which is indicated by significant location and season interaction in the mixed effects modelling. The result of this study is consistent with other studies that account for temporal and spatial variation in terrestrial (Somers and Asner, 2014) and marine environments (Fyfe, 2003).

Season plays a larger role in influencing spectral reflectance of *E. radiata* compared to location. The wavelengths that describe *E. radiata* varies in summer but remains the same in winter (730–750 nm) across all locations (Supplementary Figure 5A). This indicates that lowest irradiance level is in winter and the



subsequent increase in Chlorophyll-a pigment (Blain and Shears, 2019) levels remain the same across all locations. Photosynthetic pigments such as Chlorophyll-a and accessory pigments such as Chlorophyll-c and Fucoxanthin present in brown seaweeds such as *E. radiata* and *C. maschalocarpum* are responsible for the unique spectral signatures depending on the pigment concentration levels (Casal et al., 2012). Although it is reported that the wavelengths correlated to Chlorophyll-a are in green (550–560 nm) and red edge (680–750 nm) region, it is difficult to quantify the concentration levels of individual pigments remotely

(Huang et al., 2015). In summer, the wavelengths that describe *E. radiata* in Motuora Island and Te Haruhi Bay is different to that in Takatu point and Kawau Island. This is likely due to the high irradiance levels in summer (Blain and Shears, 2019) and varying wave exposure levels in different locations, for example, sites in Te Haruhi Bay and Motuora Island are sheltered while that in Kawau Island and Takatu point are very exposed. Variations in the degree of water movement around plants among other factors influence photosynthesis (Hillman et al., 1989) and in turn affects the spectral response of seaweed.

There is a very different seasonal pattern exhibited in spectral data of *C. maschalocarpum* across all locations compared to *E. radiata* despite both species being collected from the same locations. *C. maschalocarpum* individuals were spectrally similar across all locations in winter and spring and was described by wavelengths 733–750 nm (**Supplementary Figure 5B**). This is likely due to the lower irradiance levels during those seasons and no chlorophyll production. However, in summer, the wavelengths (597–608 and 693–718 nm) that described *C. maschalocarpum* varied significantly from those in spring and winter (**Supplementary Figure 6B**). It is likely that expectedly high photosynthetic activity in summer may cause some variation in wavelengths describing *C. maschalocarpum* in different locations (**Figure 2A**). The spectral reflectance values of both seaweed species in summer are significantly different in Motuora Island compared to other locations (**Figure 2B** and **Supplementary Figure 4B**). In Motuora Island, spectral signatures of both seaweed species are represented by wavelengths 692–715 nm in summer. For *E. radiata*, the spectral signatures are not significantly different in all other season. Whereas, for *C. maschalocarpum*, spectral reflectances are not significantly different in spring and winter while that in autumn is significantly different from summer, spring and winter. This could be due to the nature of the site (**Supplementary Table 1**) enabling higher photosynthetic activity.

Multispectral and hyperspectral sensors have the capability of capturing high resolution (spatially and spectrally) data that could resolve individual seaweeds. Due to the nature of the water current and movement of seaweed underwater, it is important to understand the within plant spectral variation across various parts of the seaweed individual such as blade and stipe. This study accounts for that variation and has found very low spectral variation within each sample indicated by the minimal difference

between marginal and conditional  $R^2$  values of mixed modelling for both species.

Detecting within species spectral variation due to factors such as season and location using remote sensing imaging sensors could be difficult. When combined with other factors such as water column properties, depth and turbidity, detecting these inherent within species differences could prove more difficult. Modelled spectral signatures have been used for distinguishing three seaweeds belonging to each broad taxon at different turbidity levels and depths (Vahtmäe et al., 2006), classifying seaweeds at a broad taxa level from hyperspectral images (Casal et al., 2013). However, spectral signatures from this study could be modelled for various water column properties using radiative transfer models or approached empirically for further research.

## DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

## AUTHOR CONTRIBUTIONS

All authors listed have made a substantial, direct and intellectual contribution to the work, and approved it for publication.

## SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fevo.2021.581852/full#supplementary-material>

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**Conflict of Interest:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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