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The influence of water motion on the growth rate of the kelp *Laminaria digitata*

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ABSTRACT

The shallow water kelp *Laminaria digitata*, abundant in coastal zones of the North Atlantic, is exposed to a range of hydrodynamic environments that makes it ideal for assessing the role of water motion on their growth rate. Here we quantify the growth of *L. digitata*, as a factor of blade and stipe elongation, at sites adjacent to Strangford Lough, Northern Ireland under different hydrodynamic conditions over a one year period. A modelling approach was used to numerically determine both the temporal and spatial variability of the hydrodynamic environment. Ambient seawater nutrient concentrations, temperature and irradiance were measured as well as the internal nutrient status of the *L. digitata* populations. Kelp populations growing in the greatest and lowest water motion showed the lowest growth rates. Differences observed in growth rate could not be attributed to seawater nutrient availability, temperature or light. The internal nutrient status also suggested no influence on the observed differences in growth rate. Therefore if there are minimal differences in light, temperature and nutrients between sites, then populations of *L. digitata* exposed to different water motions are likely to exhibit different growth rates. It is suggested that the growth rate differences observed were a function of water motion with the possibility that, in response to the hydrodynamic forces experienced by the algal cells, *L. digitata* kelps in the high energy environments were putting more energy into strengthening cell walls rather than blade elongation.

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1. Introduction

Kelp ecosystems are particularly associated with temperate inshore locations worldwide which are generally characterised by energetic hydrodynamic environments (Steneck et al., 2002). This has led to early broad conclusions being drawn between the requirements of temperate macroalgal communities for water movement to achieve optimal growth (Conover, 1968; Sundene, 1962). However, observational evidence for this remains equivocal and may indicate a degree of species-specificity in the response. Hepburn et al. (2007) were able to directly relate an increase in the growth rate of the kelp *Macrocystis pyrifera* in autumn to increased wave activity. However, for the macroalgae *Laminaria longicruris* and *Adamsiella chauvinii*, Gerard and Mann (1979) and Kregting et al. (2008) respectively were unable to establish a direct influence of water motion on macroalgal growth rate. In the case of *L. hyperborea*, Kregting et al. (2013) demonstrated no relationship between either frond growth rates or stipe elongation rates and water motion. In contrast, Sjøtun et al. (1998) reported that some unspecified factor connected with wave exposure enhanced the growth rate of only 4-year old *L. hyperborea* individuals.

Investigations of the effect of water motion on macroalgal growth rates is difficult because of the complex interactions between ancillary environmental variables, especially light, nutrients and water motion on macroalgal growth (Hurd, 2000). For example, wind-induced waves can influence turbidity through re-suspension of sediment (Lund-Hansen et al., 1997 and Schoellhamer, 1995) thereby reducing light levels. Water movement may counteract community shading or self-shading by directly moving blades allowing light flecks to penetrate into the sub-canopy or thalli optimizing photosynthetic tissue for carbon acquisition and photon capture (Dring and Lüning, 1994 and Kübler and Raven, 1996). In addition, during periods of low flow velocities (<0.06 m s^{-1}), conditions that can be found in habitats such as harbours and inlets or during slack water, macroalgae may for a time experience mass-transfer limitation, a process influencing the flux of molecules to and from the blade surface (Gerard, 1982; Hurd et al., 1996; Hurd, 2000 and Wheeler, 1980).

There are also difficulties in making comparative objective assessments of the form and magnitude of the water motion experienced by macroalgae at different locations. Previous approaches have relied on coarse scale predictors such as the biological-exposure scale (Lewis, 1964), wind-sector method (Sjøtun et al., 1998), and directional

wave-fetch approach (Burrows et al., 2008) which lack spatial and temporal resolution for detailed measurements of the effects of water motion on macroalgae. Oceanographic equipment such as wave systems and current metres have also been employed successfully (Gaylord et al., 2006, 2007 and Kregting et al., 2013); however there are limitations on the deployment of these instruments at several sites simultaneously owing to logistical reasons. These issues have led to the use of an engineering approach to model the hydrodynamic environment. While hydrodynamic models are not new for coastal engineering applications, the improved spatial and temporal resolution of models and their capability has led to increasing use in studies attempting to establish relationships between the physical environment and the biology of macroalgae (Brennan et al., 2014 and Wing et al., 2007).

L. digitata (Hudson) J.V. Lamouroux forms luxuriant canopies in the lower intertidal to shallow sub-tidal zone over large areas of North Atlantic coastlines. Within this zone kelp plants experience a wide range of waves and currents. The present investigation considers the effect of variations in the ambient hydrodynamic regime on the growth of *L. digitata* as recorded at several locations in a tidal sea lough in Northern Ireland (Fig. 1). Numerical modelling was used to describe the hydrodynamics at each sampling location. The possible confounding role of temperature and irradiance on the growth rates of *L. digitata* at the sampling sites was addressed using in situ measurements of both variables. Similarly the potential role of nutrients on kelp growth rates was considered using data obtained from ambient water samples and also from the internal nutrient status of the kelps.

2. Materials and methods

2.1. Strangford narrows hydrodynamic models

Prediction of the hydrodynamic conditions at the sampling sites (Fig. 1) for the duration of the study (May 2009 to June 2010) was derived from the combined output of the Strangford Lough flow and the Strangford Lough wave models based on MIKE 21 (DHI Water and Environment software package; www.dhisoftware.com). Full details of the development and calibration of the Strangford Lough flow model can be found in Kregting and Elsäßer (2014). The Strangford Lough wave model simulates the sink and source terms of a WAM (Wave Model) cycle 4 spectral wave model in quasi-steady mode. The wave model uses a fully spectral formulation based on the wave action conservation equation as described in Komen et al. (1994) and Young (1999), where the wave action spectrum is the dependent variable. The wave model used the same flexible mesh (FM) as the Strangford Lough flow model.

In order to accurately predict the wave conditions at the five site locations, both offshore swell and wind conditions are required as well as the local wind conditions. To derive the boundary offshore swell conditions, data were taken from an in-house large Irish Sea wave model which is driven by 6 hourly wind (speed and direction) and wind sea and swell parameters (significant wave height (H_{m0}), period (T), direction, spreading index of wind waves and swell). The wind data for the Irish Sea model were obtained from the ERA-Interim project supplied by the European Centre for Medium-Range Weather Forecasts (ECMWF) (Dee et al., 2011). From this Irish Sea wave model the wave conditions were extracted at 3 hourly intervals and applied to the open boundary conditions of the Strangford Lough wave model along a deep water boundary (Fig. 1). To simulate the locally generated wave field, which dominates the wave condition in the inner part of the Lough and the Narrows, wind data observed at Orlock Point, approximately 20 km north of the northern end of Strangford Lough, were used as the 10 m wind speed for local wind wave generation. To validate the accuracy of the wave model, data were obtained from a Nortek Aquadopp (2 MHz) ADCP instrument. The instrument was deployed outside Strangford Lough (N 54°18.52 W 005°30.87) at a depth of 10 m for a 2-week period. Wave characteristics were measured at 1 Hz over a period of 17 min every 3 h providing significant wave height (H_m0).

Since macroalgae experience the combined effects of wave and current motion (Kregting et al., 2013), the output components from the flow and wave models, i.e. current speed (U) and wave-associated motion derived from the root-mean-square of the maximum horizontal particle velocity at 0.5 m above the bottom (Umax), were summed together to provide an estimate of the total velocity (m s^{-1}) experienced by the macroalgae. This approach allowed an objective comparison of dynamic conditions experienced at the sites to be made. In order to accommodate the fluctuations in velocity output from the model, velocities were expressed as the velocity that is exceeded 50% of the time
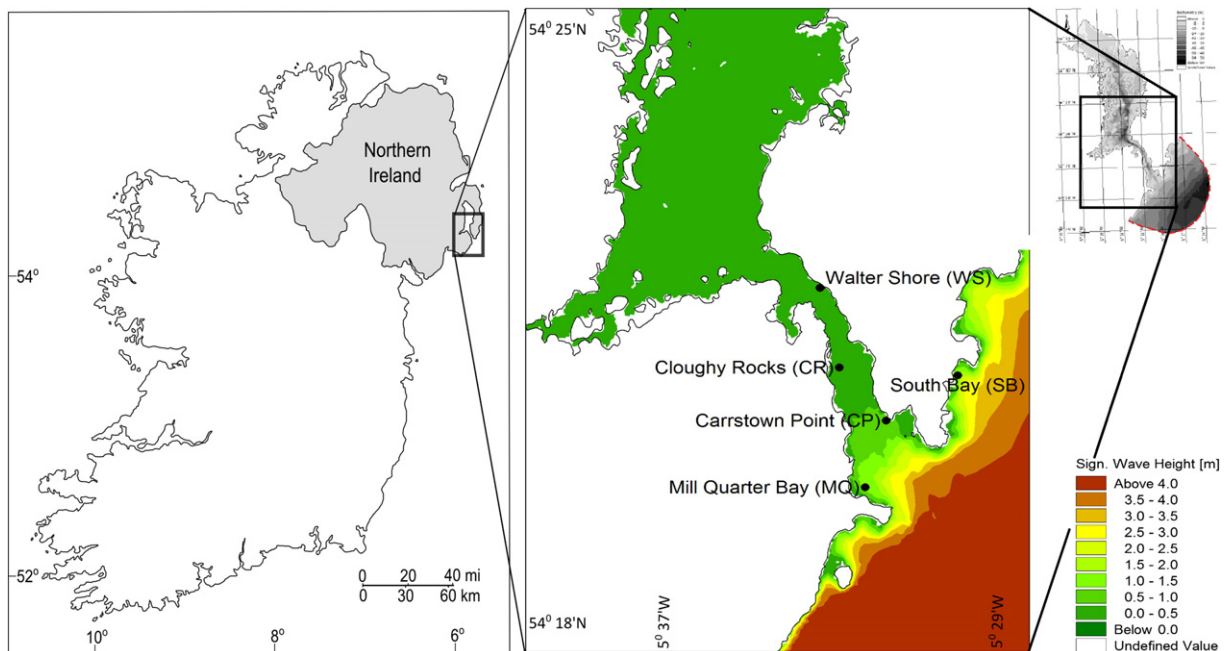

Fig. 1. Map of Northern Ireland and entrance to Strangford Lough showing the position of the five study sites and significant wave height (m) simulated for the 16 January 2010 from the wave model (insert showing the wave model domain with red dotted line showing the model boundary).

(P50) as calculated for each monthly period. P50 may thus be taken to represent the median flow velocity over the period that the growth measurements were made.

2.2. Environmental variables

Observations were carried out at five sites: South Bay on the coast close to the seaward entrance of the Narrows which experiences relatively high wave activity and low current flows, Mill Quarter Bay at the entrance of the Lough which experiences low wave activity and low current flows and Carrstown Point, Walter Shore and Cloghy Rocks in the Strangford Narrows which experience predominantly medium to high current flows but low wave activity (Figs. 1 & 2). The choice of sites selected for the study encompassed a range of water motion regimes located within a relatively small study area to ensure that other influencing environmental parameters on growth rate such as temperature and seawater nutrient concentrations were likely to be similar at all sampling sites.

In situ photon irradiance and temperature were measured using Odyssey integrating light (PAR, 400–700 nm) and temperature sensors (Dataflow Systems Pty Ltd., Christchurch, New Zealand) deployed simultaneously at four sites, South Bay, Mill Quarter Bay, Carrstown

Fig. 2. Photos of the *Laminaria digitata* populations at each site including a representative of the morphology of an individual at the site. The South Bay population is located outside the Strangford Lough and exposed to wave heights >4 m and low current flows (<0.2 m s^{-1}). Mill Quarter Bay populations experience low waves (<1 m) and low current flows (<0.6 m s^{-1}). Carrstown Point populations experience wind wave chop and medium current flow (max 1.5 m s^{-1}). Walter Shore populations experience wind wave chop and medium current flow (max 2 m s^{-1}) and Cloghy Rocks experiences wind wave chop and high current flow (max 3 m s^{-1}). Scale bar represents 0.3 m.

Point, and Walter Shore and from May 2009 to June 2010. Loggers were not placed at the fifth location, Cloghy Rocks, owing to difficulties in securing the instrumentation to the rock at this site. Data-loggers were attached to concrete blocks at each deployment site and located in small cleared areas (1 m^2) of kelp. The light and temperature sensors recorded integrated irradiance and temperature continuously over consecutive 10 min scan periods. Prior to deployment the light sensors were calibrated using a PAR Quantum sensor (Skye Instruments). To prevent error associated with fouling of the light sensors, sensors were changed on each sampling occasion (approximately 30 d intervals).

Three replicate 60-ml seawater samples were collected from the five sites on each sampling trip at low tide by wading out to 0.5 m water depth. The samples were filtered through Whatman GF/C glass-fibre filters and frozen at −20 °C for later analysis of nutrient concentrations in the laboratory. The samples were analysed for nitrate (NO_3^-), ammonium (NH_4^+) and phosphate (PO_4^{3-}) concentrations using a Bran + Luebbe AutoAnalyzer 3.

2.3. Growth rate measurements

Elongation rates of stipes and growth rates of blades of *L. digitata* populations at the five sampling sites were determined over intervals of approximately 30 days from May 2009 to June 2010. At each site 20 mature *L. digitata* kelps with blade length > 1 m were haphazardly selected and tagged with a cable tie and cattle tag tied to the base of the stipe on the initial survey. If tagged seaweeds disappeared between surveys, new individuals were selected and tagged to maintain a population size of 20 individuals at each site. Stipe elongation was determined from the increase in stipe length, as measured to the nearest 0.5 cm, over each inter-survey period. Blade growth was determined using the hole punch method (Parke, 1948) where a circular hole of 0.7 cm diameter was initially punched ~4 cm from the stipe/laminar transition zone; the distance between the transition zone and the hole was measured on each sampling occasion with callipers to the nearest 0.5 mm. New holes were punched ~4 cm from the stipe/laminar transition zone each month if blade growth had occurred. Daily relative growth rates (RGR, d^{-1}) of the blades and daily elongation rates (RER, d^{-1}) of the stipes were determined after Evans (1972).

2.4. Tissue nutrient analyses

Tissue samples were collected from 5 haphazardly selected adult seaweeds with blades >1 m in length from each site at monthly intervals and analysed for soluble tissue nitrate, ammonium and phosphate concentrations using standard colorimetric procedures. From each blade two discs (2 cm diameter; ~0.15 g ww) were punched side by side ~3 cm distance from the stipe/laminar junction, cleaned of any sediment and epiphytic material, then wiped and blotted dry for weight determination. One disc was trimmed with a scalpel equal to the weight of the other and dried at 80 °C for 48 h until a constant dry weight was attained. The fresh tissue disc was tested for soluble nutrient pools using the boiling-water extraction method of Hurd et al. (1996). Preliminary experiments confirmed that two extractions were sufficient to remove all soluble nutrients from the tissue samples. Extracted samples were stored in 60 mL polyethylene bottles and frozen (−20 °C) for later analysis with the Bran + Luebbe AutoAnalyzer 3.

Total tissue carbon and nitrogen contents were determined for five kelp blades collected in summer, autumn, winter and spring (July 2009, September 2009, January 2010 and April 2010 respectively) from each site. Sub-samples of the tissue collected for the analysis of soluble tissue nutrients were dried and ground before being combusted in a Delta V Advantage Elemental Analyser (Thermo Scientific) from which the percentage tissue carbon and nitrogen contents were expressed on an atomic weight basis.

2.5. Statistical analysis

A bootstrap resampling method (1000 resamples) (MATLAB and Statistics Toolbox Release 2012b The MathWorks, Inc., Natick, Massachusetts, United States) was used to calculate the mean and confidence interval for the P50 velocity values derived from the numerical models.

Regression analysis was used to examine the relationship between the response variable blade relative growth rate (blade) and the explanatory variables; water velocity (P50), seawater nitrate levels (Nitrate) and seawater phosphate levels (Phosphate). Temperature and irradiance were not included in the analyses owing to data not being available at all sites for the duration of the study. Data exploration revealed the relationship was non-linear and therefore a Generalised Additive Model (GAM) with a Gaussian (normal) distribution was constructed. Model assumptions were tested by extracting the residuals from the GAM and examining the plots (Zuur, 2013). All calculations were conducted using R (R Development Core Team, 2014) and the GAMs were constructed using the mgcv package, which uses the default method of a 'thin-plate regression spline' for smoothing and automatic selection of smoothing parameters by cross-validation. The GAM model used was represented by:

$$y \sim s(w) + s(xn) + s(xp) \tag{1}$$

where y is blade relative growth rate and s is the non-parametrically smoothed functions of water velocity (w), seawater nitrate (xn) and phosphate concentrations (xp). The relationship between stipe elongation of *L. digitata* with the explanatory variables was also extensively investigated; however all model assumptions were violated.

The comparison between sites and month of the rates of elongation of stipes and the growth rates of blades of *L. digitata* as well as the environmental variables (current speed, irradiance, seawater nutrient concentrations and temperature), soluble tissue pools (nitrate, ammonium and phosphate), tissue nitrogen and carbon status (% nitrogen and % carbon, C:N ratio) was carried out using a two-way analysis of variance (ANOVA general linear model with or without replication) (Dytham, 2009). The factors were site (up to five levels: South Bay, Mill Quarter Bay, Carrstown Point, Walter Shore, Cloghy Rocks and month (up to 13 levels). Post-hoc Tukey HSD tests were used to determine which months or sites were significantly different. All data were tested for normality and a Levene's test was used to test for homogeneity of variances, as well as examination of the residuals plot. IBM SPSS Statistics 19 was used for computations. The level of significance was $P \leq 0.05$ for all statistical tests.

3. Results

3.1. Environmental variables

A greater than ten-fold difference in the P50 velocity values was apparent between sites; P50 values of around 0.1 m s^{-1} were observed at South Bay with equivalent values averaging ~0.5 m s^{-1} at Mill Quarter Bay (Fig. 3). The P50 velocities at Carrstown Point and Walter Shore averaged ~1.1 and 1.2 m s^{-1} respectively while at Cloghy Rocks the equivalent value was approximately 1.5 m s^{-1}, but with considerable fluctuations (Fig. 3). Average P50 values based on the year's data showed a statistically significant difference between all sites (South Bay = 0.21 CI 0.21 and 0.22 m s^{-1}; Mill Quarter Bay = 0.49 CI 0.48 and 0.49 m s^{-1}; Carrstown Point = 1.06 CI 1.05 and 1.07 m s^{-1}; Walter Shore = 1.18 CI 1.17 and 1.19 m s^{-1}; Cloghy Rocks = 1.54 CI 1.53 and 1.56 m s^{-1}).

No significant differences in either temperature or irradiance were recorded between the sites (Table 1). Significant differences between months were observed with both parameters showing typical seasonal variations (Temperature: $F_{9,18} = 290.98$, $P < 0.001$; Irradiance: $F_{9,18} = 9.419$, $P < 0.001$). Temperature minima (~7 °C) and irradiance minima
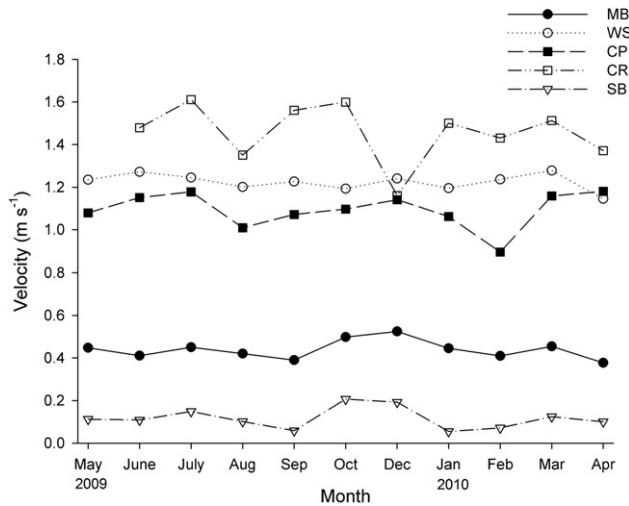
Fig. 3. Velocity value (P50 m s^{-1}) exceeded 50% of the time over each month of the observational period (May 2009 to June 2010) at the five sites (South Bay (SB), Mill Quarter Bay (MB), Carrstown Point (CP), Walter Shore (WS) and Cloghy Rocks (CR)).

(~1 mol m^{-2} day^{-1}) were recorded in winter (December 2009 to March 2010) and temperature maxima (~14 °C) and irradiance maxima (~12 mol m^{-2} day^{-1}) in September 2009 and May 2010 respectively (data not shown). Limited irradiance and temperature data were available for the South Bay site located outside the Lough due to severe weather events leading to equipment loss.

Significant differences in the mean seawater nitrate concentrations were observed between sites, but not for phosphate concentrations (Table 1). Seawater nitrate concentrations at Mill Quarter Bay and South Bay were significantly different but were similar to the values at the other sites (Walter Shore, Carrstown Point and Cloghy Rocks). Overall highest seawater nitrate concentrations were recorded at Mill Quarter Bay with particularly high concentrations being apparent in December 2009 and February 2010; high nitrate concentrations was also observed at Walter Shore in December 2009. Differences between months occurred for both nitrate and phosphate concentrations (nitrate: $F_{3,12} = 29.29$, $P < 0.001$; phosphate: $F_{3,12} = 13.941$, $P < 0.001$), with a trend for increased concentrations for both nutrients over the winter months (October 2009 to February 2010) (Fig. 4). Seawater ammonium concentrations were below the levels of detection throughout the study.

3.2. Relative blade growth rates and stipe elongation rates of Laminaria digitata

Significant differences in the relative growth rate (RGR) of the blades occurred between sites (Table 2). On average, minimal blade growth rates were observed at the South Bay site located outside the Lough

with RGR values up to 50% lower than at the other sites throughout the entire study period (Fig. 5). The second lowest average blade growth rates were recorded at Cloghy Rocks: the values at this location were again significantly different from those at other locations. Clear differences in the RGRs were particularly apparent during the summer to autumn period (July to December 2009). Kelps located at Carrstown Point grew at a lower rate than Mill Quarter Bay, similar to Walter Shore but higher than Cloghy Rocks (Tukey's test $P < 0.05$) (Fig. 5). Variations between months were observed in the RGR of the blades ($F_{12,46} = 5.241$, $P < 0.001$), and although all sites showed the same growth pattern, no strong seasonal effects were evident with only a weak indication of increased growth rate from late autumn to late spring (October 2009 to March 2010) but with the exception of a small but consistent decrease evident in February at all sites except Cloghy Rocks (Fig. 5).

The plot of residuals against linear predictors for the GAM model displayed some heterogeneity of variance and therefore the dependent variable was square root transformed. All three smoothed explanatory variables were significant in the model (water velocity $F_{4,32} = 35.89$, $P < 0.001$; nitrate $F_{2,24} = 5.42$, $P < 0.01$; phosphate $F_{5,95} = 3.81$, $P < 0.001$) and explained 32.6% of the deviance (Adj R^2 = 0.317) (Table 3; Fig. 6). Although water velocity explained most of the variation, deletion of seawater nitrate (xn) and phosphate concentrations (xp) from the model caused a highly significant increase in the residual deviance ($P < 0.001$) indicating they should remain in the model (Crawley, 2007), as also confirmed by the improved Akaike Information Criterion (AIC) values (Table 3).

The relative elongation rates (RER) of the stipes were overall an order of magnitude lower than blade growth rates (Fig. 5). However as for blade growth, significant differences in stipe elongation rates were observed between sites (Table 2). Tukey's test ($P < 0.05$) showed that a significantly lower stipe elongation rate occurred at South Bay than at either Mill Quarter Bay or Walter Shore but with the rates being similar to those at Carrstown Point and Cloghy Rocks. Both Mill Quarter Bay and Walter Shore had similar stipe RER, however Mill Quarter Bay was also similar to Carrstown Point and Cloghy Rocks. Significant differences occurred between months in stipe RER ($F_{12,46} = 5.516$, $P < 0.001$). All sites showed a similar elongation pattern with the lowest elongation occurring from July to October 2009 corresponding to the late summer and early autumn followed by a period of maximum elongation over the winter period (December 2009 to March 2010), although, as with blade growth, a minimum in stipe elongation was again apparent in February 2010 (Fig. 5).

3.3. Nutrient status of Laminaria digitata

Significant differences were observed in the soluble tissue nitrate content of the *L. digitata* seaweeds between sites although comparable differences were not observed for ammonium or phosphate pools (Table 4). Overall, soluble tissue nitrate pools were highest at the Cloghy Rocks site and lowest at Carrstown Point although both values were similar to those at Mill Quarter Bay, Walter Shore and South Bay. All sites showed a strong seasonal trend in their nitrate pools ($F_{12,46} = 19.495$, $P < 0.001$) with maximum levels (30–80 μmol g dw^{-1}) occurring in winter to early spring (January to April 2010) and minimum levels (<5 μmol g dw^{-1}) in the summer to late autumn months (July to October 2009) (Fig. 7a). Seasonal changes in soluble nutrient pools were also observed for ammonium and phosphate (ammonium: $F_{12,46} = 10.106$, $P < 0.001$; phosphate: $F_{12,46} = 15.371$, $P < 0.001$) with a similar seasonal trend as for the nitrate pools with maximum levels during the winter months (December 2009 to February 2010) and minimum during the summer to late autumn months (June to October 2009) (Fig. 7b and c). Ammonium tissue concentrations were low with values <5 μmol g dw^{-1} during the summer reaching a high of approximately 10 μmol g dw^{-1} during the winter. In contrast, soluble tissue phosphate pools were consistently high with values above

Table 1
Mean values of irradiance (mol m^{-2} day^{-1}), temperature (°C), seawater nitrate and phosphate concentrations (μM) at each site (South Bay (SB), Mill Quarter Bay (MB), Carrstown Point (CP), Walter Shore (WS) and Cloghy Rocks (CR)) during the growth monitoring period (May 2009–June 2010). Significance levels for the factor site only from the two-way ANOVA (without replication) are shown.

Site	Irradiance	Temperature	Nitrate	Phosphate
SB			1.2[a]	0.5[a]
MB	5.2[a]	10.3[a]	10.4[b]	0.5[a]
CP	3.3[a]	10.3[a]	2.9[ab]	0.5[a]
WS	4.5[a]	10.4[a]	8.4[ab]	0.6[a]
CR			2.9[ab]	0.6[a]
	$F_{2,18} = 2.838$	$F_{2,18} = 0.080$	$F_{4,46} = 3.608$	$F_{4,46} = 1.314$
	$P = 0.085$	$P = 0.923$	$P = 0.012$	$P = 0.279$

Superscript a & b represent differences between sites (Tukey's test $P < 0.05$).
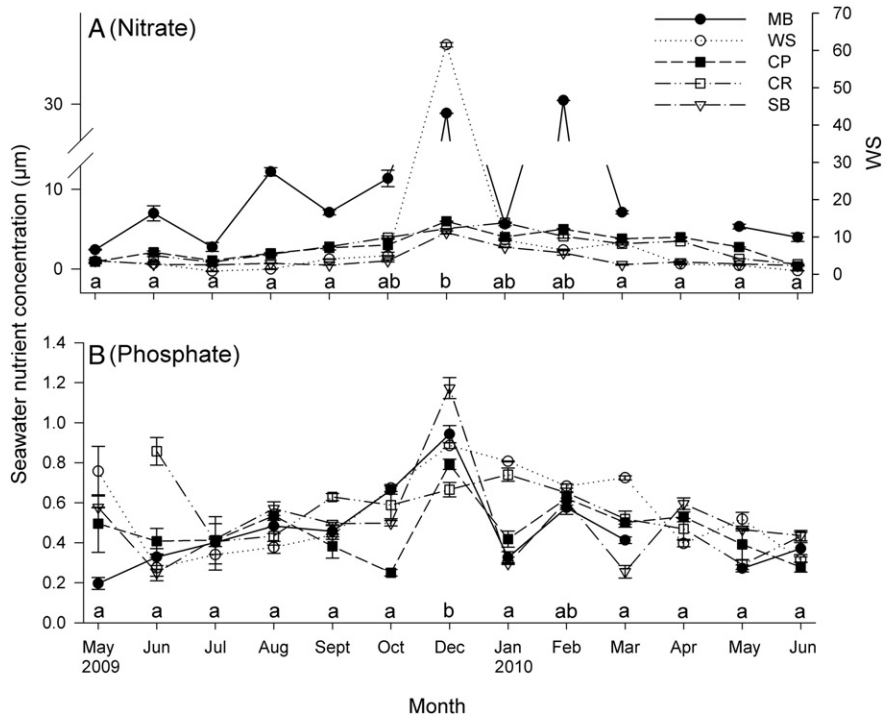
Fig. 4. Seawater nitrate (A) and phosphate (B) concentrations at the five sites (South Bay (SB), Mill Quarter Bay (MB), Carrstown Point (CP), Walter Shore (WS) and Cloghy Rocks (CR)) during the study period. Points represent means ± SE (n = 2–3). NB the two axes for seawater nitrate concentrations due to high values that were recorded for Walter Shore in December 2009. Superscript a & b represent differences between months (Tukey's test $P < 0.05$).

30 µmol g dw^{-1} in the summer and between 50 and 80 µmol g dw^{-1} in winter.

Significant differences were observed between sites for tissue nitrogen and carbon content of *L. digitata* (Table 4). Although results showed that kelps at Mill Quarter Bay had the greatest tissue nitrogen content while those at Carrstown Point had the lowest, the difference in mean values between sites was only 0.14%. Significant differences in %C were also observed between the kelps at Walter Shore and Mill Quarter Bay which had similar tissue nitrogen and carbon content to those at South Bay, Carrstown Point and Cloghy Rocks, although it may be argued that these differences were too small to be biologically relevant. This suggestion is supported by the data on the C:N ratios of the kelp tissue where no differences were observed between sites (Table 4).

Strong seasonal differences were observed between months for %N, %C and C:N ratios (%N; $F_{3,80} = 265.14$, $P < 0.001$; %C: $F_{3,80} = 33.58$, $P < 0.001$; C:N: $F_{3,12} = 29.29$, $P < 0.001$). Total nitrogen content (%N) was lowest in summer (1.5 in July 2009) which showed an increasing trend over autumn and winter (1.5 in September 2009 and 2.2 in

January 2010) to reach maximum values of 2.5 in spring (April 2010). Total carbon content showed a different pattern with the lowest carbon content observed in winter (~28 in January 2010) and highest in the summer/autumn period (~31 in July and September). The changes in %N and %C observed between seasons was reflected in the C:N ratio with maximum values observed during the summer/autumn period (~20 in July and September 2009) and minimum in the winter/spring period (~11 in January and April 2010).

4. Discussion

The combined components from the flow and wave models (current speed (*U*) and horizontal particle velocity (*Umax*) expressed as the median of the flow velocity (P50) during the growth period, described the greatest proportion of the variation in blade RGR of *L. digitata*. While seawater nitrate and phosphate should remain in the GAM model, their influence on the observed blade growth rate variation between sites was minimal. An unexpected and interesting result was that blade growth rate was clearly reduced at both the lowest and highest water velocities: to the best of our knowledge this result has not been observed before (Fig. 6). These results suggest that if there are minimal differences in light, temperature and nutrients between sites, parameters known to influence growth rates of macroalgae (Hurd, 2000), then populations of *L. digitata* exposed to different water motions are likely to exhibit different growth rates.

Although light and temperature were not included in the GAM model because observations were not recorded during the entire study period or at every site, it is not believed that these variables would have any major influence on the variation observed in growth rates between sites. In the absence of previous data records, it was assumed when designing the study that there would be only minimal differences in these parameters at the five sampling locations owing to their proximity and perceived minimal anthropogenic influence. Seawater temperature at the three sites, where measurements were

Table 2
Mean values of relative growth rate (RGR) for blades and relative elongation rates (RER) for stipes of *L. digitata* at the five sites (South Bay (SB), Mill Quarter Bay (MB), Carrstown Point (CP), Walter Shore (WS) and Cloghy Rocks (CR)) during the growth monitoring period (May 2009 – June 2010). Significance levels for the factor site only from the two-way ANOVA (without replication) are shown.

Site	Blade	Stipe
SB	0.013[d]	0.001[c]
MB	0.027[a]	0.006[ab]
CP	0.024[b]	0.005[bc]
WS	0.026[ab]	0.010[a]
CR	0.021[c]	0.003[bc]
	$F_{4,46} = 75.644$	$F_{4,46} = 9.090$
	$P < 0.001$	$P < 0.001$

Superscript a, b, c & d represent differences between sites (Tukey's test $P < 0.05$).
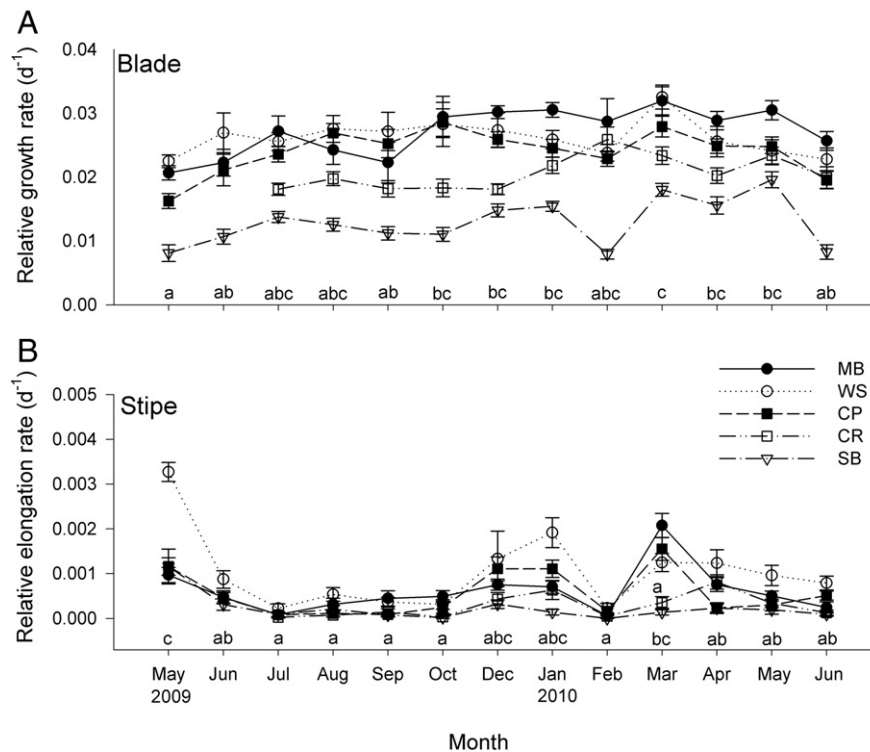
Fig. 5. Relative growth rate of *L. digitata* blades (A) and relative elongation rate of stipes (B) at South Bay (SB), Mill Quarter Bay (MB), Carrstown Point (CP), Walter Shore (WS) and Cloghy Rocks (CR) locations. Points represent means ± SE (n = 8–20). Superscript a, b & c represent differences between months (Tukey's test *P* < 0.05).

obtained, were indeed similar exhibiting standard seasonal cycles and it was extrapolated that there would be little difference between the five sites.

Similar conclusions were drawn for incident irradiance. As for temperature, irradiance levels were similar between the sites where it was measured and showed a seasonal variation typical of mid-latitude regions (Kain, 1989). However it is recognised that small local variations in light climate between the sites may have been present. The light climate in the intertidal is heterogenous and varies on a wide range of temporal scales. Factors such as tidal fluctuations, aspect and density of thalli (self-shading) may influence light availability for photosynthesis (Kübler and Raven, 1996; Dring and Lüning, 1994; Kirk, 2011). At those sites where data loggers were able to be positioned (Mill Quarter Bay, Carrstown Point and Walter Shore), minor differences in the light climate between sites were observed during the late spring and summer months but not from September to March (data not shown). However similar blade elongation rates were recorded at these sites over the observational period suggesting that light had no major influence on the growth patterns observed. When selecting the sampling sites for the study, there was no expectation that the ambient nutrient concentrations would be significantly different at the five locations. The observation of increased ambient nitrate concentrations at Mill Quarter Bay explained some of the deviation in the GAM model. Thus the results of

seawater nutrients in the GAM model emphases the value of routinely monitoring these core variables important for growth during comparable studies. However this increase in nitrate at Mill Quarter Bay was not reflected in the internal nutrient status of *L. digitata*. There was no clear trend in the data indicative of any one site having higher or lower tissue nitrate content than other sites. There was a clear trend of nitrate storage for all the populations from December 2009 to April 2010 when soluble tissue nitrate levels increased to 30–80 μmol g dw^{-1}, a trend also reflected in the soluble tissue ammonium and phosphate levels. This typical pattern of nutrient limitation in summer and luxury consumption in winter has been observed in several other macroalgal species (Chapman and Craigie, 1977; Conolly and Drew, 1985; Martínez and Rico, 2002 and Wheeler and North, 1981). The C:N ratios reflected the internal nutrient pools which were indicative of nutrient limitation during the summer and autumn months (values > 15) and nutrient replete in winter and spring (values ~10) (D'Elia and DeBoer, 1978 and Harrison and Druehl, 1982). Although throughout the summer the nitrogen status of the kelps was generally low, the *L. digitata* populations exhibited growth suggesting that ambient seawater nutrients were not low enough to affect growth rates.

It is unclear as to what factors may be of importance in reducing growth rate at the sites with the highest and lowest flow rates. It was assumed when designing the study that the type of flow, whether it was current or wave dominated, would not have different effects on the growth rates of the kelps, as observed for the kelp *L. hyperborea* (Kregting et al., 2013). In the dynamic intertidal environment, kelp populations experience acceleration and drag forces arising from the oscillatory motion of the waves or a steady tidally driven current flow. Output from the modelling confirmed that the sites Carrstown Point, Walter Shore and Cloghy Rocks within the Narrows of Strangford Lough experience primarily current driven flow conditions. Mill Quarter Bay is sheltered by a headland and the kelp populations at this site are exposed to a mixture of low current flows and small (<1 m) waves. South Bay is located outside the Narrows where the kelp populations

Table 3
Model comparison for GAMs of blade relative growth rate; y is blade relative growth rate and s is the non-parametrically smoothed functions of water velocity (w), seawater nitrate levels (x$_n$) and seawater phosphate levels (x$_p$). Akaike information criterion (AIC), a measure of the relative quality of each statistical model.

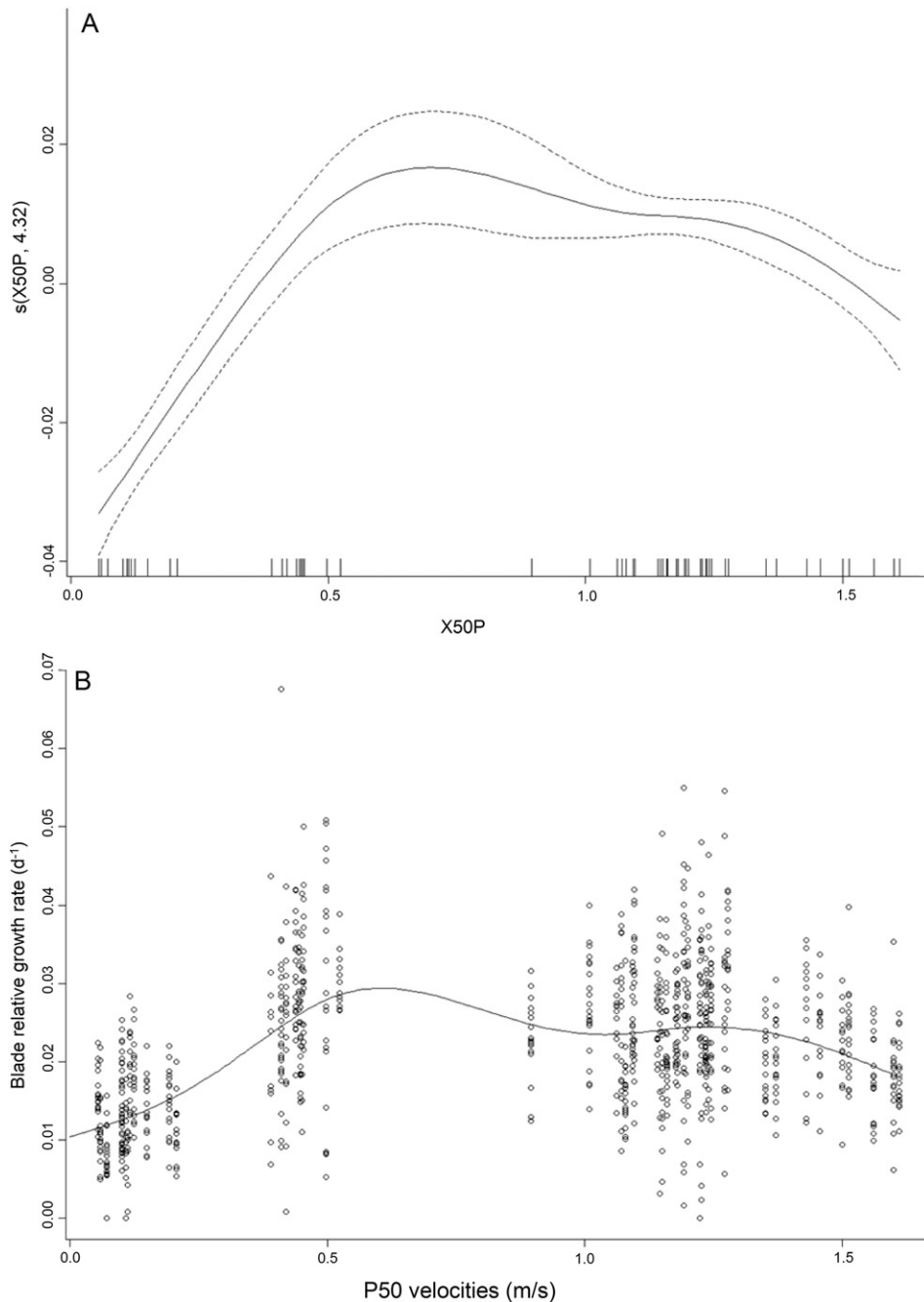| Model	Deviance explained	Adj R^2	AIC
y ~ s(w)	30.30%	0.299	−4060.06
y ~ s(w) + s(x$_n$) + s(x$_p$)	32.60%	0.317	−4077.96

Fig. 6. Smoother for water velocity (X50P) obtained by the GAM in Eq. (1) (A) and observed (open circles) and fitted values (line) for *L. digitata* blade relative growth rate in relation to water velocity (B).

are predominantly exposed to waves which can at times reach significant wave heights >4 m which are not reflected in the P50 values due to the sporadic and short duration (hours) of the large wave events that occur at this site (Kregting et al., 2013). Both types of water motion (current and waves) invoke well-documented biomechanical and morphological responses in macroalgae (e.g. Duggins et al., 2003; Harder et al., 2006; Johnson and Koehl, 1994 and Kitzes and Denny, 2005). Here we hypothesise that the growth rate differences observed were a function of water motion and may be related to trade-offs between an increase in tissue strength versus an increase in blade growth in the kelps.

Laboratory studies on juvenile macroalgae grown under a steady velocity of 1.2 m s^{-1} (Kraemer and Chapman, 1991a) indicated that thalli were twice as strong (breaking strain), twice as stiff (modulus of elasticity) and had a 1.5 times greater work fracture than macroalgae grown under low energy conditions (velocity < 1 cm s^{-1}); in addition there is evidence suggesting that the composition of the alginic acid and cellulose within the macroalgae were different although the results were inconsistent with treatments (Hackney et al., 1994 and Kraemer and Chapman, 1991a, 1991b,). Martone (2007) proposed that macroalgae growing under imposed wave forces have two growth strategies to reduce breakage: (1) an increase in cross sectional area and (2) an increase in tissue strength. Kelp species have been shown to follow the first strategy by increasing stipe girth with increased wave exposure (Stevens et al., 2002). *L. digitata* may however be exhibiting both strategies over a range of hydrodynamic gradients. The slower growth rate at

Table 4
Mean values for tissue nutrients (μmol gdw^{-1}) for *L. digitata* at the five sites (South Bay (SB), Mill Quarter Bay (MB), Carrstown Point (CP), Walter Shore (WS) and Cloghy Rocks (CR)) measured for the season's winter, spring, summer and autumn. Significance levels for the factor site only from the two-way ANOVA (with and without replication) are shown.

Site	Nitrate	Ammonium	Phosphate	%N	%C	C:N
SB	14.7[ab]	4.5[a]	56.1[a]	1.91[ab]	30.10[ab]	17.00[a]
MB	17.4[ab]	4.9[a]	47.0[a]	2.03[a]	30.91[b]	16.19[a]
CP	12.6[a]	4.9[a]	47.0[a]	1.89[b]	30.12[ab]	16.98[a]
WS	20.0[ab]	4.7[a]	50.6[a]	1.92[ab]	29.40[a]	17.31[a]
CR	25.6[b]	4.9[a]	50.9[a]	1.95[ab]	29.90[ab]	16.05[a]
	$F_{4,46} = 2.852$	$F_{4,46} = 0.573$	$F_{4,46} = 2.660$	$F_{4,80} = 2.574$	$F_{4,80} = 3.937$	$F_{4,12} = 0.259$
	$P = 0.034$	$P = 0.684$	$P = 0.054$	$P = 0.044$	$P = 0.006$	$P = 0.899$*

Superscript a, b & c represent differences between sites (Tukey's test $P < 0.05$).
* Two-way ANOVA without replication.

the sites imposing the greatest forces (wave or current) on the blades (Cloghy Rocks and South Bay) may potentially be a transition of metabolic cost associated with increased medullary thickness rather than blade growth (Demes et al., 2011), although this topic has yet to be explored in detail.

The findings in this study contrast with comparable observations for *L. hyperborea* where no direct influence of water motion could be established on the growth rate of the kelps (Kregting et al., 2013). The results also contrast with similar data obtained for the kelp *M. pyrifera*, where wave action enhanced growth rate although it was clearly demonstrated in this instance that the growth rate increase reflected enhanced nutrient availability (Hepburn et al., 2007; Stephens and Hepburn, 2014). It is well documented that macroalgae growing on wave swept shores are smaller and tougher (Dudgeon and Johnson, 1992; Gutierrez and Fernández, 1992 and Wing et al., 2007) and their smaller size may be a trade-off of investing their energy into increased structural tissue

strength. A similar effect may be operative in the case of *L. digitata* where reduced growth rates in both the highest current flow (Cloghy Rocks) and kelps exposed to waves (South Bay) may be offset by energy being changed into strengthening the components of the blades. This result requires further exploration, in particular separating the effects of waves and currents on growth rate of kelp populations.
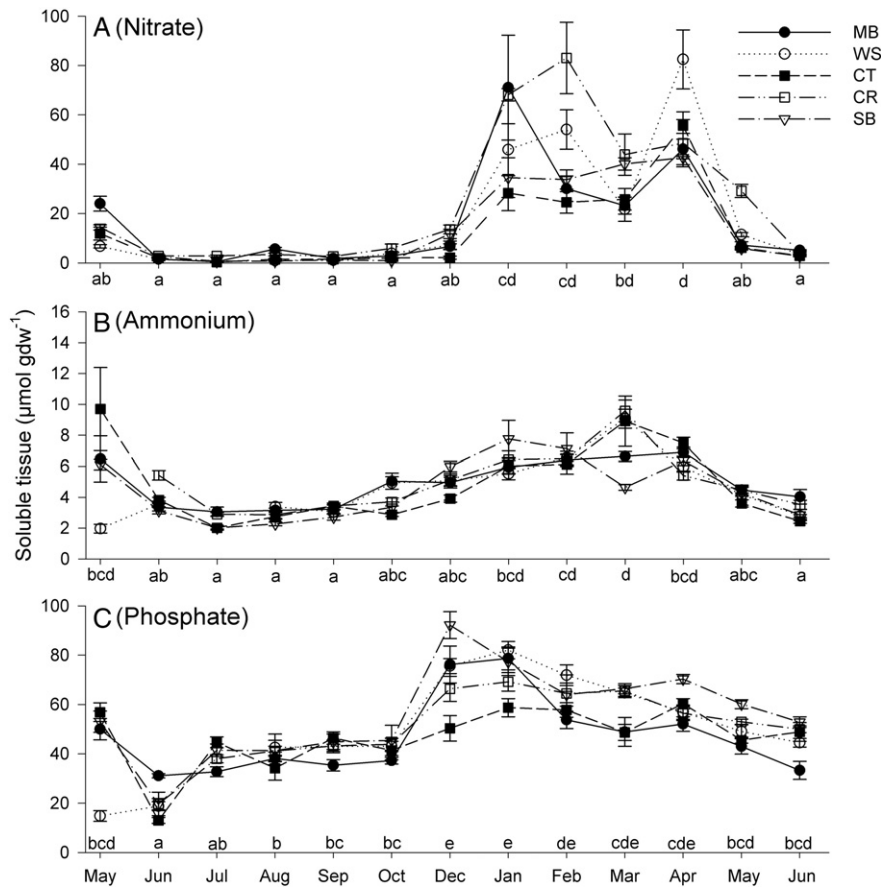
Acknowledgements

Fig. 7. Soluble tissue concentrations of nitrate (A), ammonium (B) and phosphate (C) of *L. digitata* blade tissue between May 2009 and June 2010 for the five sites (South Bay (SB), Mill Quarter Bay (MB), Carrstown Point (CP), Walter Shore (WS) and Cloghy Rocks (CR)). Points represent means ± SE (n = 5). Superscript a, b, c, d & e represent where soluble tissue concentrations are significantly different among months (Tukey's test $P < 0.05$).

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