



Seasonal ammonium uptake kinetics of four brown macroalgae: Implications for use in integrated multi-trophic aquaculture

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Received: 21 November 2021 / Revised and accepted: 6 April 2022
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Abstract

The combined culture of fed species (bivalves, fish) and macroalgae, known as integrated multi-trophic aquaculture (IMTA), has been suggested as a method of mitigating localised nitrogen (N) increase from aquaculture, whilst simultaneously culturing macroalgae for commercial applications. The development of IMTA requires an understanding of the N ecophysiology of candidate macroalga species. We examined seasonal variations in ammonium (NH_4^+) uptake kinetics, carbon to nitrogen (C:N) ratio, pigment content and soluble tissue N of four macroalgae of the phylum Ochrophyta, *Ecklonia radiata*, *Macrocystis pyrifera*, *Lessonia corrugata*, and *Phyllospora comosa*, from Tasmania, Australia. This study aimed to determine, (1) if the N physiology of the four macroalgal species was suitable for IMTA applications and (2) whether the species had seasonal variations in N ecophysiology which would influence their suitability for IMTA. *Macrocystis pyrifera*, *L. corrugata*, and *E. radiata* exhibited saturable NH_4^+ uptake kinetics, with a maximum uptake rate (V_{\max}) during spring, summer and autumn of 200, 45.8 and 45 $\mu\text{mol gDW}^{-1} \text{h}^{-1}$ and half-saturation constants (K_s) of 361.3, 104.2 and 121 μM , respectively. *Phyllospora comosa* exhibited biphasic uptake patterns for three out of four months sampled. There were no noticeable seasonal patterns in pigment content or soluble tissue N for any species. C:N ratios increased from spring (October) to autumn (March) in both *E. radiata* (28.34 – 47.83) and *P. comosa* (24.99 – 51.62), indicating progressive N limitation though summer and into autumn. Results suggest that *M. pyrifera* and *P. comosa* are most suitable for IMTA due to their high NH_4^+ uptake potential.

Keywords ammonium · integrated-multi-tropic aquaculture · macroalgae · nitrogen physiology · Ochrophyta

Introduction

Anthropogenic N inputs into coastal waters have been increasing over the last century (Vitousek 1997; Seitzinger et al. 2002; Seitzinger et al. 2008), in part caused by intensive finfish aquaculture, as fish excrete additional NH_4^+ into the ecosystem (Handy and Poxton 1993; Wild-Allen et al. 2010). Such inputs can have broader scale ecological impacts outside of the farm footprint (Oh et al. 2015). Nutrient loading can cause eutrophication and anoxic sediments leading to broader scale changes in marine community

structure such as alteration of benthic fauna and native fish abundance, increased macroalgal growth and epiphyte loading (Black 2001; Read and Fernandes 2003; Soto and Norambuena 2004; Buschmann et al. 2006; Cubitt et al. 2008).

Integrated multi-trophic aquaculture (IMTA) is being tested as a mitigation solution for the effects of increased dissolved inorganic nitrogen (DIN) loading associated with intensive mono-specific aquaculture operations (Wu et al. 2015; Biswas et al. 2020; Knowler et al. 2020; Rugiu et al. 2021). In these systems, fed aquaculture species are farmed in conjunction with extractive species such as macroalgae which take up a portion of the excreted nutrients and can reduce the overall nutrient input into the environment (Chopin et al. 2001; Chopin 2006). Other organisms in the water column also take up DIN surrounding aquaculture facilities including phytoplankton, but IMTA operations utilise species which can then be harvested for commercial gain (Knowler et al. 2020). They can also provide key ecosystem services such as oxygenation and mitigate against coastal

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ocean acidification (Hasselström et al. 2018; Fernández et al. 2018). IMTA principles have been applied to aquaculture operations globally including Korea (Park et al. 2018), China (Wu et al. 2017), Chile (Buschmann et al. 1994, 2008, Vásquez 2008), Europe (Haglund and Pedersen 1993; Sanderson et al. 2008), Israel (Ashkenazi et al. 2019), and North America (Chopin et al. 1999; Carmona et al. 2006). Similarly, there is a strong interest to implement IMTA methods into Australian aquaculture facilities using local macroalga species (Kelly 2020).

Nitrogen is an essential nutrient for macroalgae, found in chlorophylls *a* and *b*, amino acids and cellular enzymes, thus is a key factor limiting macroalgal growth in the marine environment (Hurd et al. 2014). The two main sources of N used by macroalgae are ammonium (NH_4^+) and nitrate (NO_3^-). Ambient concentrations of inorganic N are generally low in seawater, ranging between <5–20 μM depending on location, however concentrations of NH_4^+ can exceed 150 μM immediately surrounding finfish aquaculture facilities (Neori and Shpigel 1999; Carmona et al. 2006). In temperate systems the availability of DIN varies seasonally being generally higher in winter and lower in summer (Hurd et al. 2014). However, spatial variations in inorganic N supply due to anthropogenic inputs can cause localised increases in N concentration regardless of the season (Herbert 1999; Howarth and Marino 2006; Van Alstyne 2018).

One key aspect to assess macroalgal species suitability for IMTA is understanding NH_4^+ uptake kinetics (Roleda and Hurd 2019) because it is the primary waste product of operations incorporating marine finfish, excreted through the gills (Randall and Wright 1987; Wilkie 1997). Also localised increases in NH_4^+ concentrations are observed surrounding finfish cages (Sanderson et al. 2008). NH_4^+ is readily available to macroalgae, where it is taken up through cell membranes via one or more of three mechanisms - passive diffusion, facilitated diffusion and active uptake (Hurd et al. 2014; Roleda and Hurd 2019). Passive transport involves uptake via simple diffusion, whereas facilitated diffusion and active transport utilise proteins to move ions into the cell.

Uptake kinetics can be used to gain an understanding of the mechanisms of inorganic N uptake (Harrison and Druehl 1982; Rosenberg and Ramus 1984; Phillips and Hurd 2004; Roleda and Hurd 2019). Active uptake is indicated by a plot of uptake rate vs. concentration exhibiting saturating kinetics and can be described by the Michaelis-Menten equation (Hurd et al. 2014):

$$V = V_{\max} \frac{S}{K_m + S}$$

From this relationship, the parameters maximum uptake rate (V_{\max}) and half-saturation constant (K_s) are determined. Desirable V_{\max} and K_s values for IMTA are dependent on

the objective of the IMTA operation (Chopin et al. 2001). Species with a high V_{\max} and K_s values can better take up NH_4^+ at high concentrations, which is desirable for IMTA applications and lower values of K_s indicate a greater ability to procure nutrients at a low concentration. Passive uptake is indicated by a linear relationship between concentration and uptake rate. A combination of linear and saturating components indicates that both active and passive uptake mechanisms are present, known as biphasic or multi-phasic uptake (Roleda and Hurd 2019, See Fig. 6.2 d in Hurd et al. 2014). Biphasic uptake mechanisms also prove desirable for IMTA applications, as the species can take up NH_4^+ at high external NH_4^+ concentrations.

In addition to NH_4^+ uptake kinetics, determining which macroalgal species are most suited to IMTA requires a comprehensive understanding of their underlying N ecophysiology (Neori et al. 2004). Of particular importance are the C:N ratios, photosynthetic pigment content, and soluble tissue nitrogen pools which are used to assess the nitrogen status of the macroalgal tissue (Roleda and Hurd 2019; Rugiu et al. 2021). Comparatively high C:N ratios, low pigment content and low soluble tissue N pools can indicate that macroalgal tissues are depleted in N (Roleda and Hurd 2019; Chopin et al. 1995; Vergara et al. 1993). N depleted species are able to uptake excess DIN in the environment, such as that released from aquaculture operations and are therefore useful in IMTA operations (Pedersen and Borum 1997; Hadley et al. 2018). Soluble tissue N is the amount of N that is stored within the macroalgae cells and provides an indication of nutrient storage capacity, and whether the macroalgae are N depleted at the time of sampling (Roleda and Hurd 2019). Seasonal variation in macroalgal N physiology has been observed for many temperate regions worldwide due to changing light, water temperature, wave motion and nutrient supply (Kain 1989; Lüning 1993). Macroalgae adapt to these changes by seasonally altering pigment content (Flukes et al. 2015), nutrient uptake and storage (Asare and Harlin 1983, Hurd and Dring 1990, Phillips and Hurd 2004), as well as biochemical composition (Wheeler and Björnsater 1992).

Macroalgae of the orders Laminariales and Fucales are key components of temperate reefs worldwide and are candidate species for IMTA in southern Australia. In Tasmania, there are a range of animal aquaculture operations including salmon, mussels, abalone and pacific oysters (DPIPWE 2020), with growing interest to develop IMTA to help mitigate DIN inputs and provide a commercial product. The proportion of DIN up taken by macroalgae in an IMTA setting varies depending on location, stocking densities and hydrodynamics of the area. Modelling studies in Tasmania indicate that *M. pyrifera* can remove up to 11% of DIN input from salmon aquaculture over a nine-month period (Hadley et al. 2018). Four macroalgal species, *Ecklonia radiata* (C. Agardh) J. Agardh, *Macrocystis pyrifera* (Linnaeus) C.

Agardh, *Lessonia corrugata* A.H.S. Lucas, and *Phyllospora comosa* (Labillardière) C. Agardh, have been identified as potential species for IMTA operations in Tasmania due to economic value and potential for high biomass production (Sanderson and Di Benedetto 1988; Kelly 2020). Members of the Laminariales are being trialled in other regions due their comparatively fast growth rate (Barrington et al. 2009), and although the growth rates of the Tasmanian kelps are not well studied, *Macrocystis* and *Ecklonia* are known to have growth rates similar to those of other Laminariales (Miller et al. 2011; Schiel and Foster 2015). Here we determined (1) if the NH_4^+ physiology of the four Tasmanian species studied were suitable for IMTA applications and (2) if the species exhibited seasonal patterns in their NH_4^+ physiology that affect their suitability for IMTA.

Materials and Methods

Sample Collection

Macroalgal samples were collected sub-tidally at 3–5 m depth from two sites in southern Tasmania. Collection occurred four times during Spring – Autumn 2018–2019 at Flowerpot Point, Blackmans Bay ($-43^\circ 0'27''\text{S}$, $147^\circ 19'44''\text{E}$) and the Tessellated Pavements, Eagle Hawk Neck ($43^\circ 0'30''\text{S}$, $147^\circ 56'7''\text{E}$) (Table 1). Mature blades were collected for *M. pyrifera* and *L. corrugata*. Mature, lateral blades were collected for *E. radiata* and *P. comosa*. Five individuals for each species ($n = 5$) were collected at each sampling event. Samples were individually wrapped in damp tissue paper, stored in a dark cool-box and transported back to the laboratory, with 30 min for Flowerpot Point and 60 min for Tessellated Pavements. At the laboratory, samples were immediately wiped with tissue to remove any epibionts and rinsed in filtered, UV sterilised seawater (filtered to $1 \mu\text{M}$ and UV-sterilised with an Emperor Aquatics Smart HO UV steriliser, 025050-2, 50 W lamp) before being divided into experimental sections (see below). At the time of each collection, replicate 10 mL water samples ($n = 3$) were taken for analysis of ambient NH_4^+ and NO_3^- in seawater. Samples were filtered through a $0.7 \mu\text{m}$ filter (Whatman GF/F) on-site

before being transported back to the laboratory and frozen at -20°C until analysis.

NH_4^+ Uptake Kinetics

To determine the NH_4^+ uptake kinetics of each species, individual blades from each species ($n = 5$) were divided into seven discs using a 3 cm diameter cork borer for *M. pyrifera* ($\sim 0.5 \text{ g}$), *E. radiata* ($\sim 0.5 \text{ g}$) and *L. corrugata* ($\sim 1.0 \text{ g}$), or seven 5 cm individual apical blade sections for *P. comosa* ($\sim 0.25 \text{ g}$). Samples from each individual were placed into separate beakers with filtered seawater and placed on shaker tables set to 100 rpm and a photoperiod of 12:12, which was kept constant across each experiment (at $150 \mu\text{mol photons}^{-1} \text{ m}^{-2} \text{ s}^{-1}$) for 24 h to allow wound healing (McDowell et al. 2015).

To determine the maximum uptake rate (V_{max}) and half-saturation coefficient (K_s) of each macroalga species, a multiple flask, constant incubation time, experiment was conducted for (Philips and Hurd 2003) at each sampling event, within 48 h of sample collection. A total of four experiments were conducted for each species. For each species, $37 \times 250 \text{ mL}$ conical flasks were filled with 200 mL of filtered seawater and enriched with NH_4^+ from a stock solution of NH_4Cl (0.2 M) to give a concentration series of approximately 2, 10, 20, 40, 80, 160 and 240 μM , with five replicates ($n = 5$) for each species. Two additional conical flasks with no macroalgae were used as controls, with one containing filtered seawater and one with filtered seawater enriched to 240 μM .

Before addition of the macroalgae, an initial seawater sample was taken from each flask with a 12 mL syringe filtered through a $0.7 \mu\text{m}$ filter (Whatman GF/F). Samples were stored in 12 mL polyethylene tubes at -20°C . One piece of alga was then placed into each flask and set on a shaker table at 100 rpm under $150 \mu\text{mol photons m}^{-2} \text{ s}^{-1}$. All flasks were left for two h as no lag or surge phases were detected for any species in the preliminary time-course experiment (data not shown). After two hours, a final water sample was taken, and macroalgae were removed from each flask. Macroalgae were blotted dry, weighed for wet weight (WW) and photographed for surface area. The surface area was calculated using Adobe Photoshop CC 2018 (Adobe Software

Table 1 Location and date of each macroalgal collection event.

Species	Location	Collection Date			
		October 2018 (Spring)	November 2018 (Spring)	January 2019 (Summer)	March 2019 (Autumn)
<i>L. corrugata</i>	Flowerpot Point	4/10/2018	26/11/2018	13/01/2019	08/03/2019
<i>M. pyrifera</i>	Flowerpot Point	4/10/2018	27/11/2018	13/01/2019	08/03/2019
<i>E. radiata</i>	Tessellated Pavements	1/10/2018	29/11/2018	18/01/2019	11/03/2019
<i>P. comosa</i>	Tessellated Pavements	1/10/2018	29/11/2018	18/01/2019	11/03/2019

Inc). Algal pieces were then dried at 60°C for 48 h for determination of dry weight.

Seawater nutrient analysis

NH_4^+ concentrations from the uptake experiments and NO_3^- and NH_4^+ concentrations extracted from tissue (soluble pools) was determined using a QuickChem 8000 Automated Ion Analyser (LaChat Instruments) using the methods outlined in ‘Determination of nitrate/nitrite in brackish or seawater by flow injection analysis’ (Diamond 2008)

and ‘Determination of ammonia in brackish or seawater by flow injection analysis’ (Liao 2008).

Calculation of NH_4^+ uptake rates

Uptake rates of NH_4^+ in individual flasks were calculated using the following equation:

$$V = \frac{(S_i - S_f) \times Vol}{t \times DW}$$

where V = uptake rate ($\mu\text{mol g}^{-1} \text{DW h}^{-1}$), S_i = initial concentration of seawater NH_4^+ (μM), S_f = final concentration after time interval (μM), t = time interval (2 h) and DW = dry weight of macroalgal samples (g) (Harrison and Druehl 1982).

Soluble tissue NO_3^- and NH_4^+ pools

Soluble tissue NO_3^- and NH_4^+ pools were determined in November 2018, January and March 2019 by boiling water extraction (Hurd et al. 1996). One additional tissue sample was taken from each replicate ($n = 5$) of each of the four species prior to the NH_4^+ uptake experiment for analysis of soluble tissue N. These tissue samples were blotted dry and cut into sections of $0.25 \text{ g} \pm 0.01 \text{ g}$. Each piece was placed into a 50 mL boiling tube with 20 mL of deionised water and samples were refrigerated overnight (4°C). Test tubes were then placed in a boiling water bath for 20 min. The samples were left to cool, and the liquid was decanted and filtered through $0.7 \mu\text{m}$ glass filter paper (Whatman GF/F). The extract was stored at -20°C before analysis using an Automated Ion Analyser described above. This process was repeated three times to ensure all soluble tissue N was extracted. NH_4^+ and NO_3^- contents were calculated using the following equation:

$$N_T = \frac{(N_1 + N_2 + N_3) \times V}{WW}$$

where N_T is the total concentration of NH_4^+ or NO_3^- extracted, N_1 , N_2 , and N_3 are the concentrations of NH_4^+ or NO_3^- is the solution after subsequent boiling extractions

(μM), V is the volume of water in the boiling tube (L) and WW is the wet weight (g) of the seaweed sample.

Photosynthetic Pigment Content

Photosynthetic pigment content (chlorophyll *a*, chlorophyll *c* and fucoxanthin) was determined using the methods outlined in Seely et al. (1972). Samples of 0.1–0.15 g wet weight were taken from the five collected replicates of each species, frozen in liquid nitrogen and stored at -80°C until extraction. Samples were then defrosted and placed into test tubes. Although approximately the same weight (0.1–0.15 g) as other species, the blades of *L. corrugata* were thicker than other species, and so were cut into smaller pieces to facilitate extraction. 4 mL of dimethyl sulfoxide (DMSO) was added to each test tube and samples were left to extract for 10 min. The liquid was then decanted and collected in a test tube. Immediately afterwards 6 mL of 90% acetone v/v was added to the macroalgal tissue and left to extract for 30 min or until tissue was void of pigments, and subsequently decanted into a separate test tube. Test tubes were kept over ice and regularly agitated. Absorbance of the extracts were measured with a S-22 UV/Vis Spectrophotometer (Halo RB-10, Dynamica Scientific Ltd). The absorbance of the DMSO extract was measured at 665, 631, 582, and 480 nm and the acetone extract measured at 664, 631, 581 and 470 nm. Pigment contents were calculated using the equations given by Seely et al. (1972).

C, N and C: N ratio

Tissue carbon, tissue nitrogen and C:N ratio of three replicates per species were determined in October and November 2018 and January and March 2019 using the methods described in Cornwall et al. (2015). An additional 2 cm diameter disk was taken from each blade of the collected species and dried at 60°C for 48 h. A Carlo-Erba NA1500 elemental analyser coupled to a Thermo Scientific Delta V Plus via a ConFlo IV was used in analysis with combustion and reduction of samples was achieved at 1020°C and 650°C, respectively. Values were normalised to the Vienna Pee Dee Belemnite (VPDB) scale with a 3-point calibration and both precision and accuracy were $\pm 0.1 \%$ (1 SD).

Curve fitting and data analysis

The Michaelis-Menten function (i.e., a rectangular hyperbola) was fitted to each replicate of *M. pyrifera*, *L. corrugata*, and *E. radiata* using SigmaPlot (Systat Software Inc), and V_{max} and K_s obtained for each replicate. The mean values for V_{max} and $K_s \pm \text{SE}$ were then obtained for each species in each sampling month.

For *P. comosa* the Michaelis-Menten function could only be fitted for data collected in January. In October, December, and March, the pattern of uptake vs. concentration was biphasic with uptake of NH_4^+ appearing to saturate for concentrations $<160 \mu\text{M}$, however further increased linearly between $160 - 240 \mu\text{M}$. Michaelis-Menten curves were therefore fitted to uptake rates at NH_4^+ concentrations $<160 \mu\text{M}$, and a linear regression was applied to uptake rates at $\text{NH}_4^+ >160 \mu\text{M}$. Negative uptake values were excluded from curve fitting and manuscript figures but are presented in Appendix 1.

The means \pm SE were calculated for soluble tissue NH_4^+ and NO_3^- , pigment content, C:N ratio, C and N. Data were analysed using the statistical software R (R core development team 2017). Data were tested for conformity of assumptions of homogeneity of variances and normality of residuals by plotting residuals and fitted values. Transformations to meet these assumptions for V_{max} , K_s , soluble tissue N and pigment determined using the Box-Cox method, and log transformations were applied to the data before analysis. A two-way ANOVA with the factors 'Species', 'Season' and 'Species*Season' was applied where $p < 0.05$. This test compared differences between species, between season and interaction effects and was applied to V_{max} , K_s , soluble tissue N content, pigment content, and C:N ratio. For significant results, a-posteriori multiple comparisons were then conducted using a Tukey's HSD test to elucidate specific differences between species across seasons. Additionally, Pearson's correlation test ($p < 0.05$) was used to determine the relationship between V_{max} and K_s across the sampling months.

Results

Background seawater nutrient concentration

Ambient, *in situ* seawater nutrients did not exceed $3.32 \mu\text{M}$, with the highest concentrations recorded at the Tessellated Pavements (Table 2). The range of NO_3^- concentrations across seasons were similar at both Flowerpot Point and the Tessellated Pavements.

Table 2 Range of seawater NH_4^+ and NO_3^- concentrations recorded at each location during the collection events

Location	NH_4^+ concentration (μM)	NO_3^- concentration (μM)
Flowerpot Point	1.16 – 1.55	0.29 – 0.87
Tessellated Pavements	0.56 – 3.32	0.20 – 0.69

NH_4^+ uptake kinetics

Controls showed minimal change in NH_4^+ concentration ($0 - 10 \%$) over the two-hour experimental period and the depletion of NH_4^+ from the seawater with macroalgae was thus attributed to macroalgal uptake. *L. corrugata*, *E. radiata*, and *M. pyrifera* all exhibited saturable uptake (Fig. 1). In contrast, *P. comosa* uptake was biphasic for three of four sampling events (Fig. 2) and V_{max} and K_s values could not be determined.

V_{max} were significantly different between species ($p < 0.05$), and an interaction effect between species and season ($p < 0.05$), indicating that seasonal patterns in V_{max} were not consistent across species (Table 3, Fig 3). An *a-posteriori* Tukey's HSD test revealed no significant differences in *E. radiata* and *L. corrugata* V_{max} over the sampling months ($p > 0.05$). In contrast, *M. pyrifera* V_{max} values varied seasonally and were highest in October 2018 ($159 \mu\text{moles g}^{-1} \text{DW h}^{-1} \pm 23 \text{ SE}$, Tukey's test, $p < 0.01$) and January 2019 ($200 \mu\text{moles g}^{-1} \text{DW h}^{-1} \pm 23 \text{ SE}$, Tukey's Test, $p < 0.01$) (Table 3). The highest V_{max} values for *M. pyrifera* were approximately 4.5 times greater than those for *E. radiata* and *L. corrugata*, which ranged between 17 and $45 \mu\text{mol g}^{-1} \text{DW h}^{-1}$.

For K_s , the two-way ANOVA indicated significant differences in species and season (Table 3). As with V_{max} , an interaction effect of species and season was also present for K_s values, indicating that seasonal effects were not consistent across species. Graphs of mean $K_s \pm \text{SE}$ are shown in Fig. 3. The K_s of *L. corrugata* did not change significantly between sampling months (Tukey's test, $p > 0.05$), ranging between 104 and $134 \mu\text{M}$. In contrast, the K_s of *E. radiata* varied over the sampling months (Tukey's test, $p < 0.05$) and was approximately nine times higher in March 2019 ($204.33 \mu\text{M} \pm 52.35 \text{ SE}$) than October 2018 ($23.50 \mu\text{M} \pm 5.05 \text{ SE}$). The K_s of *M. pyrifera* also varied across sampling months, with November K_s values being significantly lower than the other seasons (Tukey's test, $p < 0.05$). Higher K_s values were positively correlated to higher V_{max} values (Pearson correlation, $R = 0.808$, $t = 10.45$, $\text{df} = 58$, $p < 0.01$). The high V_{max} values for *M. pyrifera* in October 2018 and January 2019 occurred with the highest observed K_s values ($325.85 \mu\text{M} \pm 95.83 \text{ SE}$ and $361.26 \pm 80.26 \text{ SE}$ respectively).

Soluble tissue NO_3^- and NH_4^+ pools

Soluble tissue NO_3^- and NH_4^+ content ($\mu\text{mol g}^{-1} \text{WW}$) were variable both across seasons and between species (Table 3). Additionally, an interaction effect of species and season was present with no clear seasonal pattern in soluble tissue N content. For all species, NH_4^+ content was higher than NO_3^- content for all seasons (Fig. 4). As such, variation in total N content reflected changes in tissue NH_4^+ . Average NH_4^+

Fig. 1 NH_4^+ uptake rates as a function of substrate concentration for *E. radiata*, *L. corrugata*, *M. pyrifera* and *P. comosa* in October and November 2018 and January and March 2019

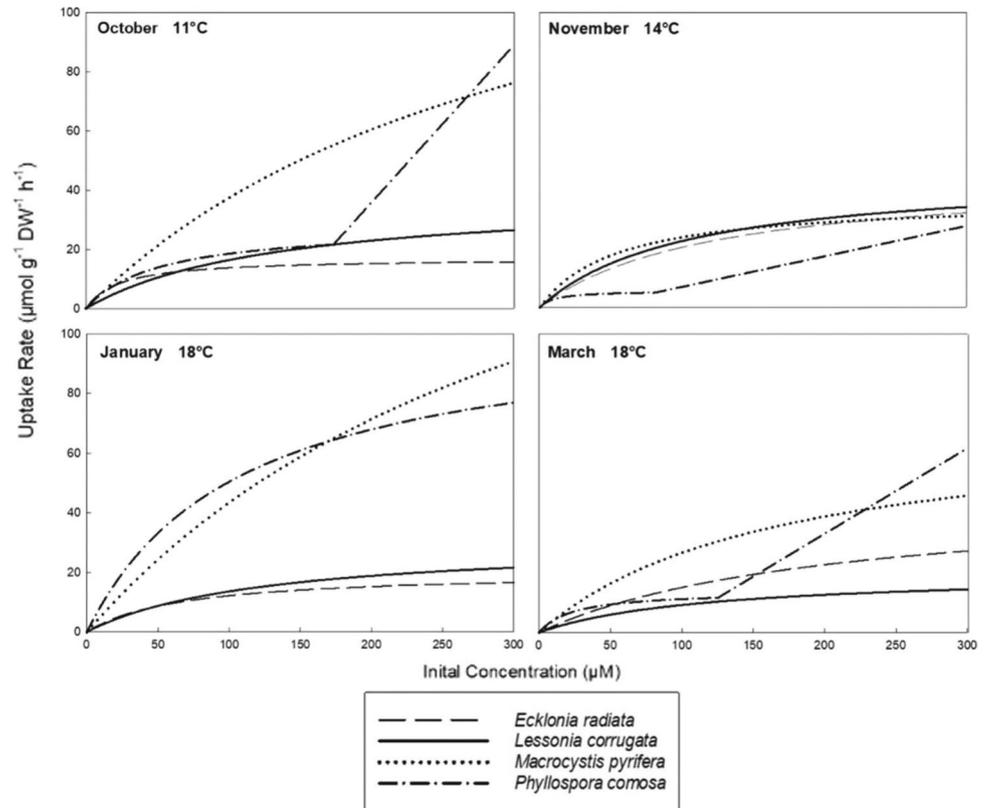
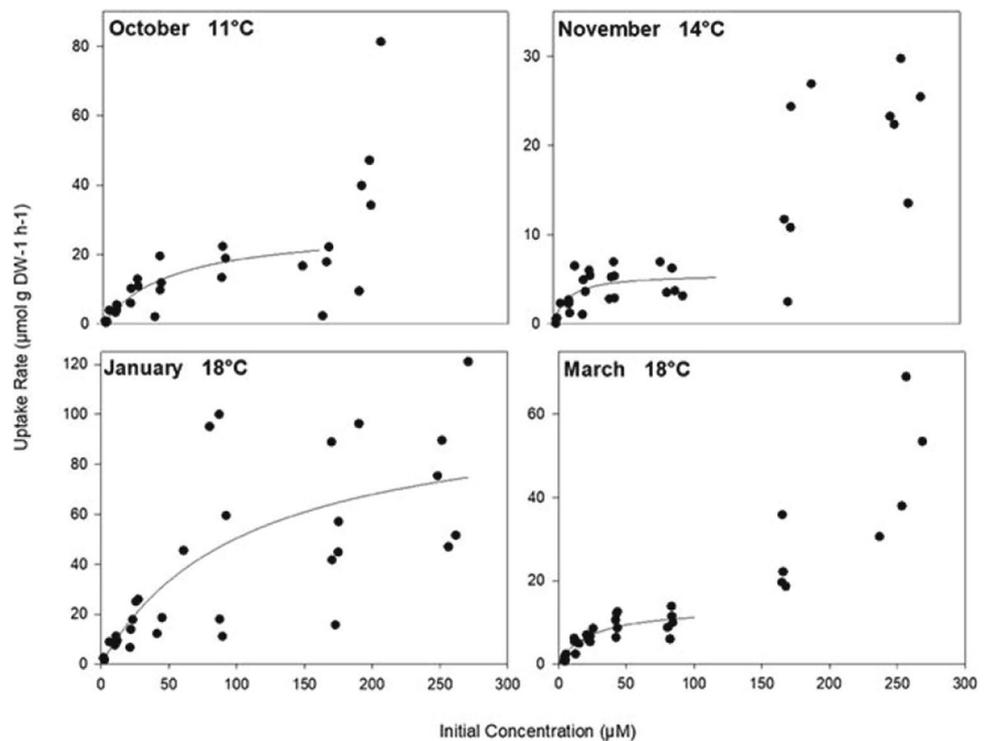


Fig. 2 Rates of NH_4^+ uptake as a function of NH_4^+ concentration for *P. comosa*. Note the y-axis scale range. Each point is a replicate seaweed ($n = 5$)



tissue content as a percentage of total N was 87.66% for *L. corrugata*, 95.49% for *M. pyrifera*, 85.82% for *E. radiata* and 92.87% for *P. comosa*.

For *L. corrugata*, total soluble N content was lowest in summer (January) ($0.928 \mu\text{mol g}^{-1} \text{WW} \pm 0.140 \text{ SE}$, Tukey's test $p < 0.01$) when compared to November and March (~ 4

Table 3 Summary of two-way ANOVA results for each parameter

Parameter	Degrees of Freedom	Sum of Squares	F Value	p-value
V_{max}				
Species	2	19.616	35.973	< 0.001
Season	3	0.602	0.736	0.536
Species*Season	6	13.5	8.252	< 0.001
K_s				
Species	2	11.31	7.928	< 0.01
Season	3	5.06	2.363	< 0.05
Species*Season	6	17.28	4.038	< 0.01
Soluble TissueN				
Species	2	6.153	30.459	< 0.001
Season	3	2.563	8.458	< 0.001
Species*Season	6	18.72	30.889	< 0.001
Pigment Content				
Species	3	9.480	13.026	< 0.001
Season	3	4.924	6.767	< 0.001
Species*Season	9	5.419	2.482	< 0.01
C: N Ratio				
Species	3	1.218	528.9	< 0.001
Season	3	1.766	766.8	< 0.001
Species*Season	9	1.102	159.5	< 0.001
TissueN				
Species	3	0.188	252.4	< 0.001
Season	3	0.191	256.0	< 0.001
Species*Season	9	0.274	22.4	< 0.001
Tissue C				
Species	3	0.416	28.23	< 0.001
Season	3	2.281	154.52	< 0.001
Species*Season	9	0.632	14.27	< 0.001

$\mu\text{mol g}^{-1} \text{ WW}$). A similar pattern was seen in *M. pyrifera*, where total soluble N content was approximately three times higher in March ($5.435 \mu\text{mol g}^{-1} \text{ WW} \pm 1.090 \text{ SE}$, Tukey's Test $p < 0.01$) than January ($0.581 \mu\text{mol g}^{-1} \text{ WW} \pm 0.114 \text{ SE}$). In contrast, total soluble N content for *E. radiata* in January was approximately double the values recorded for November and March ($4.519 \mu\text{mol g}^{-1} \text{ WW} \pm 0.693 \text{ SE}$, Tukey's test $p < 0.01$). *P. comosa* soluble tissue N content was highest in November ($2.42 \mu\text{mol g}^{-1} \text{ WW} \pm 0.292 \text{ SE}$, Tukey's test, $p < 0.01$).

Photosynthetic pigment content

Pigment content was significantly different between species and between seasons (Table 3). An interaction effect of species and season was also present ($p < 0.05$). Lowest mean pigment contents were recorded for *L. corrugata* samples in January 2019 ($0.0591 \text{ mg g}^{-1} \pm 0.0139 \text{ SE}$) and were highest in *P. comosa* in November 2018 ($0.326 \text{ mg g}^{-1} \pm 0.138 \text{ SE}$) (Fig. 5). There were no significant differences in pigment content across season for *L. corrugata*, *M. pyrifera* and *P. comosa* (Tukey's test, $p > 0.05$ for all pairwise comparisons). Mean total pigment content of *E. radiata* was approximately three times higher in October ($0.309 \text{ mg g}^{-1} \pm 0.0212 \text{ SE}$) than January and March ($p < 0.01$). November pigment content was lower than October ($0.149 \text{ mg g}^{-1} \pm 0.0167 \text{ SE}$, $p > 0.01$) and the lowest mean total pigment content for this species was recorded in March ($0.0898 \text{ mg g}^{-1} \pm 0.0119 \text{ SE}$).

C, N and C:N ratio

The two-way ANOVA revealed differences in tissue N, tissue C and C:N ratio between seasons and species, with and additional interaction effect (Table 3). For all species,

Fig. 3 Maximum uptake rates (V_{max} in $\mu\text{mol g}^{-1} \text{ DW h}^{-1}$) and half saturation constants (K_s in μM) for *E. radiata*, *L. corrugata* and *M. pyrifera* (Mean \pm SE, $n = 5$). Means with different letters are significantly different (Tukey's test, $p < 0.05$). Note that V_{max} and K_s values could not be determined for *P. comosa* due to the biphasic uptake pattern identified in October, November and March

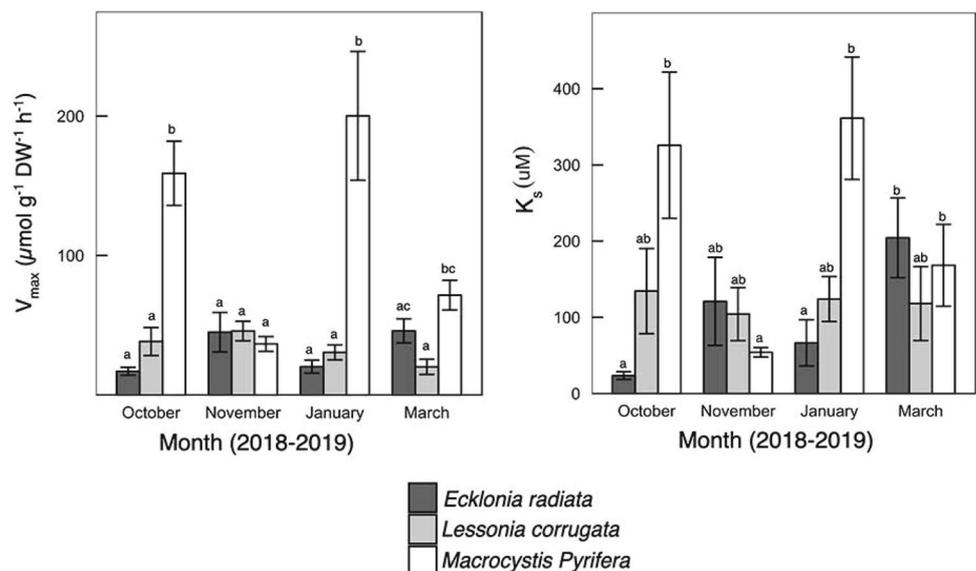


Fig. 4 Soluble tissue NO_3^- and NH_4^+ mean concentrations expressed as $\mu\text{mol g}^{-1}$ WW from (A) *L. corrugata*, (B) *M. pyriferera*, (C) *E. radiata* and (D) *P. comosa*. Bars represent mean \pm SE, $n = 5$. * denotes total tissue N mean significantly different to other months within a species (Tukey's test, $p < 0.05$)

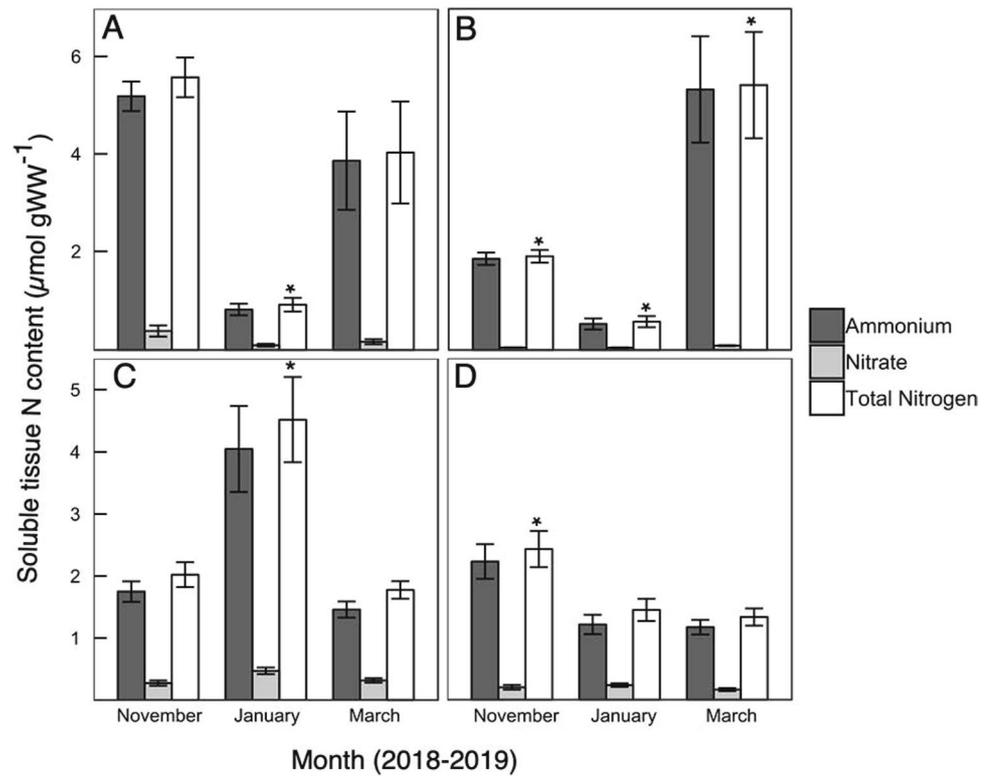
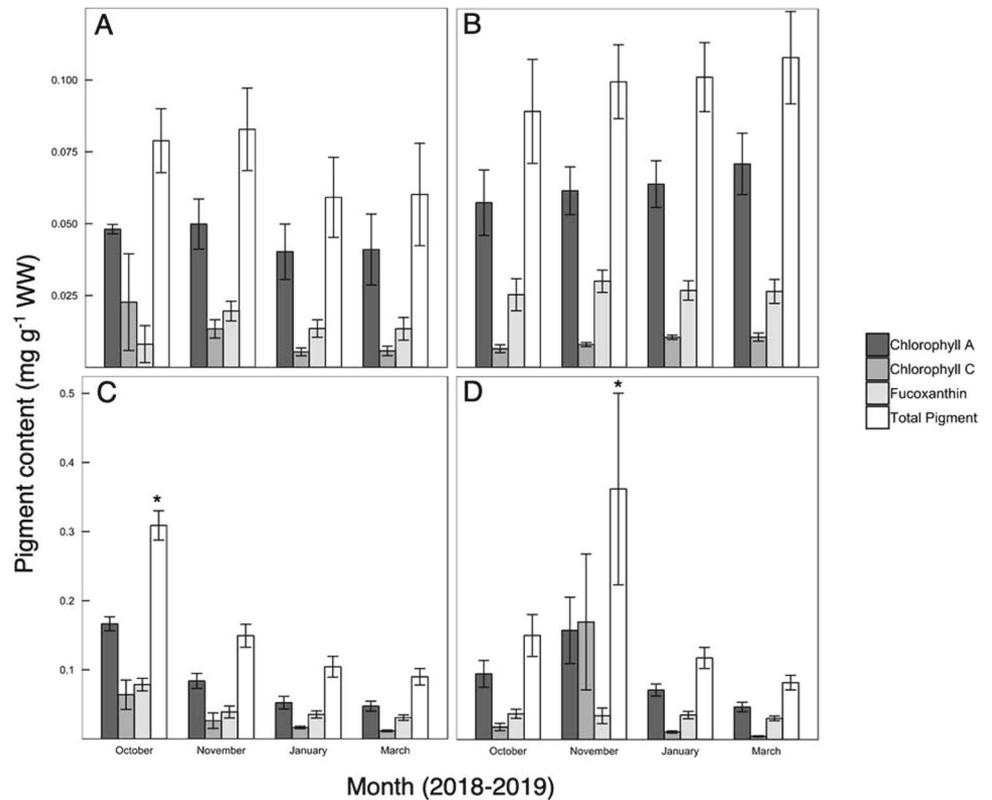


Fig. 5 Pigment content of chlorophyll *a*, chlorophyll *c*, fucoxanthin and total pigment content expressed as mg g^{-1} for (A) *L. corrugata*, (B) *M. pyriferera*, (C) *E. radiata* and (D) *P. comosa*. Bars represent mean \pm SE, $n = 5$. * denotes significant difference in total pigment content from other months within a species (Tukey's test, $p < 0.05$)



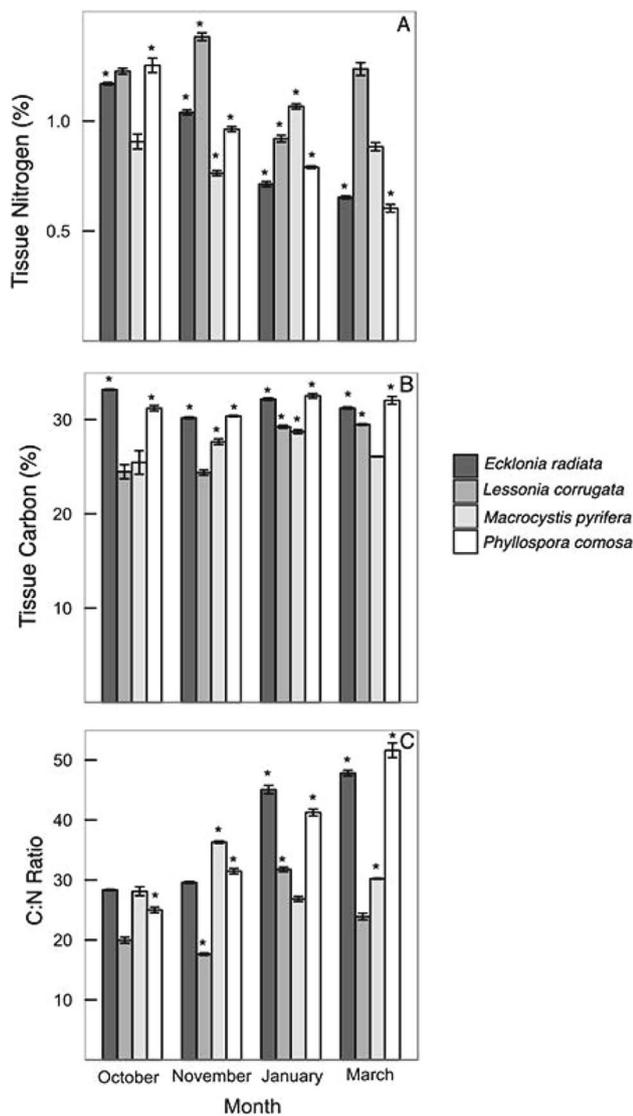


Fig. 6 Tissue C (A) and tissue N (B) expressed as % of total DW and C:N ratio (based on %) across sampling months. Bars represent mean \pm SE, $n=3$. * denotes significant difference (Tukey's test, $p < 0.05$) from other sampling months of the same species

tissue nitrogen did not exceed 1.4%. For *E. radiata* and *P. comosa*, tissue nitrogen decreased by approximately half from October to March ($p < 0.01$) (Fig. 6). Highest N values were recorded in November for *L. corrugata* ($1.38\% \pm 0.017$ SE) and January for *M. pyrifera* ($1.06\% \pm 0.012$ SE). The highest tissue C values were in *E. radiata* and *P. comosa* samples, where C content was always above 30%. For *L. corrugata*, tissue C increased between October 2018 and March 2019 (Tukey's test, $p < 0.01$). For *M. pyrifera*, tissue C was highest in November and January, at 27.6% and 28.72% respectively. The C:N ratio for *E. radiata* increased between October 2018 and March 2019 (Tukey's test, $p < 0.05$ for all pairwise comparisons). The same trend was seen

in *P. comosa*. *L. corrugata* and *M. pyrifera* showed no clear seasonal pattern in C:N ratio across the sampling months.

Discussion

For IMTA applications with a primary goal of reducing NH_4^+ output into the environment, macroalgal species with a high potential for DIN uptake (high V_{\max}) are required. The maximum V_{\max} was recorded for *M. pyrifera* and was ~ 4.5 times greater than for *L. corrugata* and *E. radiata*. The higher nutrient uptake rates seen for *M. pyrifera* are consistent with the fast growth rate of this species (Gerard 1982) and higher NH_4^+ uptake rates have been observed in species with high growth rates (Pedersen and Borum 1997). *M. pyrifera* exhibited the highest K_s value, indicating that this species is better able to take up NH_4^+ at higher concentrations, compared to species with a lower K_s (Roleda and Hurd 2019). Concentrations of NH_4^+ immediately surrounding fin fish aquaculture facilities can exceed $150 \mu\text{M}$ (Neori and Shpigel 1999; Carmona et al. 2006), compared to background concentrations of DIN of $< 20 \mu\text{M}$, which as suggested by our results would be rapidly taken up by *M. pyrifera*. Indeed, it has been utilised in IMTA in Chile (Buschmann et al. 2008) and has a high market value compared to other kelp species (Correa et al. 2016; Camus et al. 2019). Of the four species studied here, *M. pyrifera* appears the most suitable for IMTA operations in Tasmania.

M. pyrifera, *L. corrugata* and *E. radiata* all exhibited saturable NH_4^+ uptake kinetics. Saturable NH_4^+ uptake is common in macroalgae, and a summary of species exhibiting saturable NH_4^+ uptake is provided in Table 3. Active uptake mechanisms are particularly important in regions where concentrations are low and can be limiting, as algae must be able to actively pump DIN into their cells against a concentration gradient, for storage and growth. In contrast to the laminarians, the fuclean brown seaweed *P. comosa* exhibited biphasic uptake patterns for all months except summer (January). To our knowledge, this is only the third record of biphasic NH_4^+ uptake in the class Phaeophyceae, and biphasic uptake has been observed only for members of the Fucales: *Fucus distichus* (Thomas et al. 1985) and *Fucus spiralis* (Topinka 1978).

The likely presence of both active and passive N uptake mechanisms, as indicated by bi-phasic uptake as seen in *P. comosa*, is considered an adaptation to areas with large variations in nutrient concentrations (Collos et al. 1997; Lomas and Glibert 1999). Biphasic uptake allows the macroalgae to operate active uptake when nutrient concentrations are low, and passive uptake when nutrient concentrations are high (Buchanan et al. 2000). As such, *P. comosa* may have uses in IMTA operations as its biphasic uptake mechanisms

Table 4 Summary of literature values of V_{\max} and K_s values of NH_4^+ for other seaweed species

Macroalgae	Temp (°C)	V_{\max} ($\mu\text{moles g}^{-1} \text{DW h}^{-1}$)	K_s	Location	Reference
Ochrophyta:					
<i>Chorda filum</i>	9	23.6	3.44	Sweden, Baltic	Wallentinus 1984
<i>Chordaria flagelliformis</i>	11	61.9	4.35	Nova Scotia, Canada	Probyn and Chapman 1982
<i>Dictyosiphon foeniculaceus</i>	9	54.43	3.6	Sweden, Baltic	Wallentinus 1984
<i>Ecklonia cava</i>	16	29.2	76.8	S. Korea	Kang et al. 2013
<i>Ecklonia radiata</i>	18	Passive uptake	Passive uptake	W. Australia	Paling 1991
<i>Ecklonia radiata*</i>	18	45	121 ± 57.8	Tasmania	This Study
<i>Ecklonia radiata*</i>	11	17	23.5 ± 5.05	Tasmania	This Study
<i>Ectocarpus siliculosus</i>	9	39.8	3.46	Sweden, Baltic	Wallentinus 1984
<i>Elachista fucicola</i>	9	133.9	20.93	Sweden, Baltic	Wallentinus 1984
<i>Eudesme virescens</i>	10	38.1	4.78	Sweden, Baltic	Wallentinus 1984
<i>Fucus distichus</i>	15	60	3 – 5	British Columbia	Thomas et al. 1985
<i>Fucus distichus</i>		3.6	13.90	Nova Scotia	Rosenberg and Ramus 1984
<i>Fucus spiralis^c</i>	15	0.29	5.8	Massachusetts	Topinka 1978
<i>Fucus spiralis^d</i>	15	0.35	9.6	Massachusetts	Topinka 1978
<i>Fucus spiralis^c</i>	10	0.26	6.4	Massachusetts	Topinka 1978
<i>Fucus spiralis^d</i>	5	0.18	6.4	Massachusetts	Topinka 1978
<i>Fucus vesiculosus</i>	15	41	21	Denmark	Pedersen and Borum 1997
<i>Hincksia sordida</i>	15	24.3	12.5	S. Australia	Campbell 1999
<i>Laminaria abyssalis</i>	18	2	4.6	Brazil	Dā Costa Braga and Yoneshigue-Valentin 1996
<i>Laminaria groenlandica</i>	18	Passive uptake	Passive uptake	British Columbia	Harrison et al., 1986a, b
<i>Lessonia corrugata*</i>	14	45.8	104	Tasmania	This Study
<i>Macrocystis pyrifera</i>	16	23.8	5.3	S. California	Haines and Wheeler 1978
<i>Macrocystis pyrifera*</i>	14	200	361	Tasmania	This Study
<i>Macrocystis pyrifera*</i>	14	36.6	54.1	Tasmania	This Study
<i>Macrocystis pyrifera</i>	6 – 9	23.6	50	S. California	Wheeler 1979
<i>Phyllospora comosa*</i>	18	Biphasic	Biphasic	Tasmania	This Study
<i>Pilayella littoralis</i>	8	35.9	3.57	Sweden, Baltic	Wallentinus 1984
<i>Sargassum baccharia</i>		13	4.8	N. Australia	Schaffelke and Klumpp 1998
<i>Scytosiphon lomentaria</i>	6	69	3.9	Sweden, Baltic	Wallentinus 1984
<i>Scytothamnus australis</i>	15	76	42.8	New Zealand	Phillips 2001b
<i>Undaria pinnatifida</i>	16	10.7	90.9	S. Korea	Kang et al. 2013
<i>Undaria pinnatifida^a</i>	15	56.7	9.2	S. Australia	Campbell 1999
<i>Undaria pinnatifida^b</i>	15	32.8	12.4	S. Australia	Campbell 1999
<i>Undaria pinnatifida</i>	10	350	172	Japan	Sato et al. 2016
<i>Undaria pinnatifida</i>	4	Passive uptake	Passive Uptake	New Zealand	Dean and Hurd 2007
<i>Xiphophora chondrophyll</i>	17.5	Passive uptake	Passive uptake	New Zealand	Taylor et al. 1998
<i>Xiphophora gladiata</i>	15	8.7	36.9	New Zealand	Phillips 2001b
Chlorophyta:					
<i>Acrosiphonia centralis</i>	2	115.2	19.07	Baltic	Wallentinus 1984
<i>Caulerpa cupressoides</i>		8.7	48.00	Virgin Islands	Williams and Fisher 1985
<i>Chaetomorpha linum</i>	15	132 ± 29	13 ± 12	Denmark	Pedersen and Borum 1997
<i>Cladophora glomerata</i>	12	356.4	13.2	Sweden, Baltic	Wallentinus 1984
<i>Cladophora serica</i>	15	81	25	Denmark	Pedersen and Borum 1997
<i>Cladophora serica</i>	15	122	13	Denmark	Pedersen and Borum 1997
<i>Cladophora sp.</i>	23	130	20.7	W. Australia	Gordon et al. 1981
<i>Codium decorticatam</i>		13.4	12	N. Carolina	Rosenberg and Paerl 1981
<i>Codium fragile</i>	15	240 ± 61	21 ± 16	Denmark	Pedersen and Borum 1997
<i>Codium fragile</i>	6	13	1.5 ± 0.2	Rhode Island	Hanisak and Harlin 1978

Table 4 (continued)

Macroalgae	Temp (°C)	V _{max} (μmoles g ⁻¹ DW h ⁻¹)	K _s	Location	Reference
<i>Codium fragile</i>	24	28	1.4 ± 0.2	Rhoda Island	Hanisak and Harlin 1978
<i>Enteromorpha ahlnieriana</i>	13	409.4	16.6	Sweden, Baltic	Wallentinus 1984
<i>Enteromorpha compressa</i>	14	36.8	24	Sweden, Baltic	Kautsky 1982
<i>Enteromorpha prolifera</i>	12 - 14	39 – 188	9.3 – 13.4	Oregon	O'Brien and Wheeler 1987
<i>Enteromorpha</i> sp.	17.5	Passive uptake	Passive uptake	New Zealand	Taylor et al. 1998
<i>Enteromorpha</i> sp.	20	Biphasic	Biphasic	Massachusetts	Fujita 1985
<i>Ulva compressa</i>	16	140.3	268.1	S. Korea	Kang et al. 2013
<i>Ulva lactuca</i>	20	138 ± 78	40.7 ± 8.5	Massachusetts	Fujita 1985
<i>Ulva lactuca</i>	15	190 ± 23	17 ± 6	Denmark	Pedersen 1994
<i>Ulva lactuca</i>		50	5.2	Israel	Cohen and Neori 1991
<i>Ulva</i> sp.	17.5	Passive uptake	Passive uptake	New Zealand	Taylor et al. 1998
<i>Ulva</i> sp.	15	0.9	8.2	S. Australia	Campbell 1999
<i>Ulva rigidia</i>		Passive uptake	Passive uptake	W. Australia	Lavery and McComb 1991
Rhodophyta:					
<i>Agardhiella subulata</i>	20	15.9	3.9	Massachusetts	D'Elia and DeBoer 1978
<i>Apophlaea lyallii</i>	15	11.6	42.08	New Zealand	Phillips 2001b
<i>Arthrocardia</i> sp.	12	2.07		New Zealand	Nguyen et al. 2020
<i>Ceramium rubrum</i>	2	271	29	Denmark	Pedersen and Borum 1997
<i>Ceramium rubrum</i>	15	25.2	3.6	Massachusetts	DeBoer et al. 1983
<i>Ceramium tenuicornea</i>	15	192.1	9	Sweden, Baltic	Wallentinus 1984
<i>Chondrus crispus</i>	15-16	62	35.5	France	Amat and Braud 1990
Crustose coralline sp.	12	0.58		New Zealand	Nguyen et al. 2020
<i>Furcellaria lumbricalis</i>	9	4.88	6.53	Sweden, Baltic	Wallentinus 1984
<i>Gracilaria foliifer</i>	20	23.8	1.6	Massachusetts	D'Elia and DeBoer 1978
<i>Gracilaria gracilis</i>	20	206.7	76.4	South Africa	Smit 2002
<i>Gracilaria incurvata</i>	16	50.6	151.7	S. Korea	Kang et al. 2013
<i>Gracilaria pacifica</i>	16	Passive uptake	Passive uptake	Oregon	Naldi and Wheeler 1999
<i>Gracilaria pacifica</i>	15	30	10	British Columbia	Thomas et al. 1987
<i>Gracilaria tikvahiae</i>	16	Passive uptake	Passive uptake	Florida	Friedlander and Dawes 1985
<i>Gracilaria tikvahiae</i>	20	2.67	24.8	Massachusetts	Fujita 1985
<i>Gracilaria vermiculophylla</i>	20	N/A	N/A	Portugal	Abreu et al. 2009
<i>Gracilariopsis lemaneiformis</i>	27	68	40	N. Carolina	Vergara et al. 1995
<i>Hemineura frondosa</i> [#]	12 ^e	0.12	152.93	Tasmania	Paine et al. 2020
<i>Hemineura frondosa</i> [#]	12 ^f	0.01	16.17	Tasmania	Paine et al. 2020
<i>Hypnea musciformis</i>	26	115	16.6	Virgin Islands	Haines and Wheeler 1978
<i>Phyllophora truncata</i>	14	9.71	7.03	Sweden, Baltic	Wallentinus 1984
<i>Polysiphonia decipiens</i>	15	4.5	5.7	S. Australia	Campbell 1999
<i>Porphyra</i> sp.	20	Passive uptake	Passive uptake	New Zealand	Taylor et al. 1998
<i>Porphyra yezoensis</i>	16	111.5	248.6	S. Korea	Kang et al. 2013
<i>Pterocladia capillacea</i>	16	65	45	New Zealand	Taylor et al. 1998
<i>Palmaria Palmata</i>	16	12.5 – 19.4	9.28 – 19.81	Spain	Martinez and Rico 2004
<i>Rhodomela confervoides</i>	4	38.1	2318	Sweden, Baltic	Wallentinus 1984
<i>Stictosiphonia arbuscula</i>	15	Passive uptake	Passive uptake	New Zealand	Phillips 2001b

^aImmature thalli; ^bmature thalli; ^cdark-acclimated; ^dlight acclimated; ^esaturation light; ^flimiting light; * maximum and minimum values from species from this study; [#]V_{max} measured in μmol cm⁻¹ h⁻¹

allows a high NH_4^+ uptake capacity at high external NH_4^+ concentrations.

To compare the values of V_{\max} and K_s obtained in our study with those of other macroalgal species, a search of published literature was undertaken and the uptake kinetics of 62 species are reported in Table 4. There were 26 Rhodophyta, 24 Ochrophyta and 16 Chlorophyta, in addition to the four Ochrophyta species studied here. In general, the phyla Chlorophyta and Rhodophyta had higher V_{\max} and K_s values than the phylum Ochrophyta, Class Phaeophyceae. This is the first study to examine the NH_4^+ uptake mechanisms of *P. comosa* and *L. corrugata*, and the V_{\max} for these species fell within the range of studies on macroalgae of the order Laminariales. For *E. radiata*, Paling (1991) suggested a passive uptake mechanism for NH_4^+ where uptake rate increased proportionally to NH_4^+ concentration, but we found evidence of active uptake as saturating kinetics were observed.

For kelps, a C:N ratio of >15 - 20 is often considered to indicate N limitation (Hurd et al. 2014). For some macroalgae from mid and low latitudes, there are strong seasonal patterns of tissue nitrogen and C:N ratio. For example, C:N ratios of *Ulva olivascens* have been shown to change from 12.9 to 39.4 moving from spring to summer (Altamirano et al. 2000) due to higher levels of nitrogen in the water column in winter months. Such seasonal patterns in soluble tissue N content, N uptake, pigment content and macroalgal growth have been demonstrated by Topinka (1978), Küppers and Weidner (1980), Asare and Harlin (1983), Rosell and Srivastava (1985), Brown et al. (1997), Abreu et al. (2011) and Bearham et al. (2013). In this study, tissue N in *E. radiata* and *P. comosa* declined over summer and C:N ratio increased, indicating progressive N limitation from spring to summer. In contrast, *L. corrugata* and *M. pyrifera* showed no clear seasonal pattern in C:N ratio. The C:N ratio of *M. pyrifera*, *E. radiata*, and *P. comosa* was >20 for all months, indicating these species are N limited year-round (Hurd et al. 2014) and would therefore be able to uptake NH_4^+ all year in an IMTA scenario. *Lessonia corrugata* was the only species which exhibited a relatively low (<20) C:N ratio year-round. As seawater DIN at the collection locations remained relatively constant over the sampling months, it is possible that *E. radiata* and *P. comosa* had increased growth in the summer months due to higher irradiance and warmer waters, and therefore used any stored N. Additional DIN sources from IMTA operations may prove beneficial for the growth of the species during summer.

Of the species studied, *M. pyrifera* and *P. comosa* appear to be the most suitable for IMTA applications in Tasmanian waters due to their high NH_4^+ uptake potential. *Lessonia corrugata* and *M. pyrifera* did not show any distinct patterns in N ecophysiology that could be attributed to the changing seasons, however *P. comosa*

had a depletion in tissue N over the summer, indicating N limitation. Other studies have proposed the introduction of a multi-cultured approach at which different species are grown at different depths. Experiments conducted in Chile have demonstrated that *M. pyrifera* can be best cultivated at 3 m and can be farmed in conjunction with *Gracilaria chilensis* (Buschmann et al. 2008). This approach may be an option in Tasmania. *Ecklonia radiata* and *L. corrugata* have high commercial value as food products (Sanderson and Di Benedetto 1988) and as sources of extracts (Lorbeer et al. 2015). Farmed in conjunction with a species of high environmental value, such as *M. pyrifera*, a multi-cultured approach may be the best option environmentally and economically.

As the commercial markets for farmed seaweeds grow, the methods used in this study may be used to assess the suitability of other local species including Rhodophyta and Chlorophyta for use in IMTA operations. The work conducted in this study is the first to confirm that *M. pyrifera* in Tasmania has desirable NH_4^+ uptake kinetics for use in IMTA. Additionally in the exploration of NH_4^+ uptake kinetics of *P. comosa*, the works identify a potential species for IMTA that has, to the best of our knowledge, not previously been utilised in aquaculture operations.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s10811-022-02743-w>.

Acknowledgements We thank John Berges, Erica Young for their expert advice, and Charlotte Levi, Bailee Woolley, and Hugh Nichols for help with field collection.

Authors contributions J. Smart: experimental design, conducted experiments, drafting, and editing of manuscript; M. Schmid: experimental design, laboratory assistance, editing manuscript; E. Paine: experimental design, experiment assistance, laboratory assistance, editing manuscript; D. Britton experimental design, experiment assistance, laboratory assistance; A. Revill: analysis and interpretation of C: N data; C. Hurd: original concept, experimental design, editing manuscript

Funding Open Access funding enabled and organized by CAUL and its Member Institutions. This study was funded by supervisor funds from the Institute for Marine and Antarctic Studies to CLH

Data Availability Contact corresponding author

Declarations

Conflicts of Interest The authors declare they have no conflicts of interest

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References

- Abreu MH, Pereira R, Buschmann AH, Sousa-Pinto I, Yarish C (2011) Nitrogen uptake responses of *Gracilaria vermiculophylla* (Ohmi) Papenfuss under combined and single addition of nitrate and ammonium. *J Exp Mar Biol Ecol* 407:190–199
- Abreu MH, Varela DA, Henríquez LM, Villarroel A, Yarish C, Sousa-Pinto I, Buschmann AH (2009) Traditional vs. integrated multi-trophic aquaculture of *Gracilaria chilensis*. *Aquaculture* 293:211–220
- Altamirano M, Flores-Moya A, Conde F, Figueria FL (2000) Growth seasonality, photosynthetic pigment, and carbon and nitrogen content in relation to environmental factors: a field study of *Ulva olivascens* (Ulvales, Chlorophyta). *Phycologia* 39:50–58
- Amat M, Braud J (1990) Ammonium uptake by *Chondrus crispus* Stackhouse (Gigartinales, Rhodophyta) in culture. *Hydrobiologia* 204:467–471
- Asare S, Harlin M (1983) Seasonal fluctuations in tissue nitrogen for five species of perennial macroalgae in Rhode Island sound. *J Phycol* 19:254–257
- Ashkenazi D, Israel A, Abelson A (2019) A novel two-stage seaweed integrated multi-trophic aquaculture. *Rev Aquac* 11:246–262
- Barrington K, Chopin T, Robinson S (2009) Integrated multi-trophic aquaculture (IMTA) in marine temperate waters. FAO, Rome (Italy)
- Bearham D, Vanderklift M, Gunson J (2013) Temperature and light explain spatial variation in growth and productivity of the kelp *Ecklonia radiata*. *Mar Ecol Prog Ser* 476:59–70
- Biswas G, Kumar P, Ghoshal TK, Kailasam M, De D, Bera A, Mandal B, Sukumaran K, Vijayan KK (2020) Integrated multi-trophic aquaculture (IMTA) outperforms conventional polyculture with respect to environmental remediation, productivity and economic return in brackishwater ponds. *Aquaculture* 516:734626
- Black K (2001) Environmental impacts of aquaculture. Sheffield Academic, Florida, USA
- Bristow L, Mohr W, Ahmerkamp S, Kuypers M (2017) Nutrients that limit growth in the ocean. *Curr Biol* 27:431–510
- Brown N, Nyman M, Keogh J, Chin N (1997) Seasonal growth of the giant kelp *Macrocystis pyrifera* in New Zealand. *Mar Biol* 129:417–424
- Buchanan BB, Gruissem W, Russell LJ (2000) Biochemistry and Molecular Biology of Plants. American Society of Plant Physiologists, Rockville, MD
- Buschmann AH, Mora OA, Gómez P, Böttger M, Buitano S, Retamales C, Vergara PA, Gutierrez A (1994) *Gracilaria chilensis* outdoor tank cultivation in Chile: Use of land-based salmon culture effluents. *Aquac Eng* 13:283–300
- Buschmann AH, Riquelme VA, Hernández-González MC, Varela D, Jiménez JE, Henríquez LA, Vergara PA, Guíñez R, Filún L (2006) A review of the impacts of salmonid farming on marine coastal ecosystems in the southeast Pacific. *ICES J Mar Sci* 63:1338–1345
- Buschmann AH, Varela DA, Hernández-González MC, Huovinen P (2008) Opportunities and challenges for the development of an integrated seaweed-based aquaculture activity in Chile: determining the physiological capabilities of *Macrocystis* and *Gracilaria* as biofilters. *J Appl Phycol* 20:571–577
- Campbell S (1999) Uptake of ammonium by four species of macroalgae in Port Phillip Bay, Victoria, Australia. *Mar Freshw Res* 50:515
- Camus C, Infante J, Buschmann AH (2019) Revisiting the economic profitability of giant kelp *Macrocystis pyrifera* (Ochrophyta) cultivation in Chile. *Aquaculture* 502:80–86
- Carmona R, Kraemer G, Yarish C (2006) Exploring Northeast American and Asian species of *Porphyra* for use in an integrated fin-fish–algal aquaculture system. *Aquaculture* 252:54–65
- Chopin T (2006) Integrated Multi-Trophic Aquaculture. What it is and why you should care... and don't confuse it with polyculture. *North Aquac* 12:4
- Chopin T, Gallant T, Davison I (1995) Phosphorus and nitrogen nutrition in *Chondrus crispus* (Rhodophyta): effects on total phosphorus and nitrogen content, carrageenan production and photosynthetic pigments and metabolism. *J Phycol* 31:283–293
- Chopin T, Sharp G, Belyea E, Semple R, Jones D (1999) Open-water aquaculture of the red alga *Chondrus crispus* in Prince Edward Island, Canada. *Hydrobiologia* 398:417–425
- Chopin T, Buschmann AH, Halling C, Troell M, Kautsky N, Neori A, Kraemer GP, Zertuche-González JA, Yarish C, Neefus C (2001) Integrating seaweeds into marine aquaculture systems: a key towards sustainability. *J Phycol* 37:975–986
- Chopin T, Yarish C, Sharp G (2007) Beyond the monospecific approach to animal aquaculture—the light of integrated multi-trophic aquaculture. In: Bert TM (ed) *Ecological and Genetic Implications of Aquaculture Activities*. Springer, Dordrecht, pp 447–458
- Cohen I, Neori A (1991) *Ulva lactuca* biofilters for marine fishpond effluents. I. Ammonia uptake kinetics and nitrogen content. *Bot Mar* 34:475–482
- Collos Y, Siddiqi MY, Wang MY, Glass ADM, Harrison PJ (1992) Nitrate uptake kinetics by two marine diatoms using the radioactive tracer ^{15}N . *J Exp Mar Biol Ecol* 163:251–260
- Collos Y, Vaquer A, Bibent B, Slawyk G, Garcia N, Souchu P (1997) Variability in nitrate uptake kinetics of phytoplankton communities in a Mediterranean coastal lagoon. *Estuar Coast Shelf Sci* 44:369–375
- Cornwall CE, Revell AT, Hurd CL (2015) High prevalence of diffusive CO_2 uptake by macroalgae in a temperate subtidal ecosystem. *Photosynth Res* 124:181–190
- Correa T, Gutiérrez A, Florea R, Buschmann AH, Cornejo P, Bucarey C (2016) Production and economic assessment of giant kelp *Macrocystis pyrifera* cultivation for abalone feed in the south of Chile. *Aquac Res* 47:698–707
- Cubitt F, Butterworth K, McKinley RS (2008) A synopsis of environmental issues associated with salmon aquaculture in Canada. In: Culver K, Castle D (eds) *Aquaculture, innovation and social transformation*. Springer, Dordrecht, pp 123–162
- Cumming E, Matthews TG, Sanderson J, Ingram B, Bellgrove A (2020) Growth and survivorship of *Phyllospora comosa* (Phaeophyceae, Fucales) on different mariculture seeding twines in a hatchery setting. *Aquaculture* 523:735216
- Dã Costa Braga A, Yoneshigue-Valentin Y (1996) Nitrogen and phosphorus uptake by the Brazilian kelp *Laminaria abyssalis* (Phaeophyta) in culture. In: Lindstrom SC, Chapman DJ (eds) *Fifteenth International Seaweed Symposium*. Springer, Dordrecht, pp 445–450
- Dayton PK, Tegner MJ, Edwards PB, Riser KL (1999) Temporal and spatial scales of kelp demography: the role of oceanographic climate. *Ecol Monogr* 69:219–250
- Dean PR, Hurd CL (2007) Seasonal growth, erosion rates, and nitrogen and photosynthetic ecophysiology of *Undaria pinnatifida* (Heterokontophyta) in southern New Zealand. *J Phycol* 43:1138–1148

- D'Elia C, DeBoer J (1978) Nutritional studies of two red algae: Kinetics of ammonium and nitrate uptake. *J Phycol* 14:266–272
- Departments of Primary Industries, Parks, Water and the Environment (DPIPWE) (2020) *Annual Report 2020*. Tasmanian Government, Hobart
- Deyssher LE, Dean TA (1986) In situ recruitment of sporophytes of the giant kelp, *Macrocystis pyrifera* (L.) CA Agardh: effects of physical factors. *J Exp Mar Biol Ecol* 103:41–63
- Diamond D (2008) Determination of nitrate/nitrite in brackish or seawater by flow injection analysis. LaChat Instruments, Loveland, USA, p 19
- Fernández P, Leal P, Henríquez L (2018) Co-culture in marine farms: macroalgae can act as chemical refuge for shell-forming molluscs under an ocean acidification scenario. *Phycologia* 58:542–551
- Flukes E, Wright J, Johnson C (2015) Phenotypic plasticity and biogeographic variation in physiology of habitat-forming seaweed: response to temperature and nitrate. *J Phycol* 51:896–909
- Friedlander M, Dawes CJ (1985) In situ uptake kinetics of ammonium and phosphate and chemical composition of the red seaweed *Gracilaria tikvahiae*. *J Phycol* 21:448–453
- Fujita R (1985) The role of nitrogen status in regulating transient ammonium uptake and nitrogen storage by macroalgae. *J Exp Mar Biol Ecol* 92:283–301
- Gerard V (1982) Growth and utilization of internal nitrogen reserves by the giant kelp *Macrocystis pyrifera* in a low-nitrogen environment. *Mar Biol* 66:27–35
- Gordillo F, Dring M, Savidge G (2002) Nitrate and phosphate uptake characteristics of three species of brown algae cultured at low salinity. *Mar Ecol Prog Ser* 234:111–118
- Gordon D, Birch P, McComb A (1981) Effects of inorganic nitrogen on the growth of an estuarine *Cladophora* in culture. *Bot Mar* 24:93–106
- Hadley S, Wild-Allen K, Johnson C, Macleod C (2018) Investigation of broad scale implementation of integrated multitrophic aquaculture using a 3D model of an estuary. *Mar Pollut Bull* 133:448–459
- Haglund K, Pedersén M (1993) Outdoor pond cultivation of the subtropical marine alga *Gracilaria tenuistipitata* in brackish water in Sweden. Growth, nutrient uptake, co-cultivation with rainbow trout and epiphyte control. *J Appl Phycol* 5:271–284
- Haines K, Wheeler P (1978) Ammonium and nitrate uptake by the marine macrophytes *Hypnea musciformis* (Rhodophyta) And *Macrocystis pyrifera* (Phaeophyta). *J Phycol* 14:319–324
- Handy RD, Poxton MG (1993) Nitrogen pollution in mariculture: toxicity and excretion of nitrogenous compounds by marine fish. *Rev Fish Biol Fish* 3:205–241
- Hanisak M, Harlin M (1978) Uptake of inorganic nitrogen by *Codium fragile* spp. *tomentosoides* (Chlorophyta). *J Phycol* 14:450–454
- Harrison PJ, Druehl LD (1982) Nutrient uptake and growth in the Laminariales and other macrophytes: a consideration of methods. In: Srivastava LM (ed) *Synthetic and Degradative Processes in Marine Macrophytes*. Walter de Gruyter, Berlin, pp 99–120
- Harrison P, Hurd C (2001) Nutrient physiology of seaweeds: Application of concepts to aquaculture. *Cah Biol Mar* 42:71–82
- Harrison P, Druehl L, Lloyd K, Thompson P (1986a) Nitrogen uptake kinetics in three year-classes of *Laminaria groenlandica* (Laminariales: Phaeophyta). *Mar Biol* 93:29–35
- Harrison PJ, Parslow JS, Conway HL (1986b) Determination of nutrient uptake kinetic parameters: a comparison of methods. *Mar Ecol Prog Ser* 52:301–312
- Hasselström L, Wouter V, Gröndahl F, Nylund GM, Pavia H (2018) The impact of seaweed cultivation on ecosystem services – a case study from the west coast of Sweden. *Mar Pollut Bull* 133:53–64
- Herbert RA (1999) Nitrogen cycling in coastal marine ecosystems. *FEMS Microbiol Rev* 23:563–559
- Howarth RW, Marino R (2006) Nitrogen as the limiting nutrient for eutrophication in coastal marine ecosystems: Evolving views over three decades. *Limnol Oceanogr* 51:364–376
- Hurd C (2017) Shaken and stirred: the fundamental role of water motion in resource acquisition and seaweed productivity. *PiP* 4:73–81
- Hurd C, Dring MJ (1990) Phosphate uptake by intertidal algae in relation to zonation and season. *Mar Biol* 107:281–298
- Hurd C, Harrison P, Druehl L (1996) Effect of seawater velocity on inorganic nitrogen uptake by morphologically distinct forms of *Macrocystis integrifolia* from wave-sheltered and exposed sites. *Mar Biol* 126:205–214
- Hurd C, Harrison P, Bischof K, Lobban C (2014) *Seaweed ecology and physiology*, 2nd edn. Cambridge University Press, Cambridge
- Kain JM (1989) The seasons in the subtidal. *Br Phycol J* 24:203–215
- Kang Y, Hwang J, Chung I, Park S (2013) Development of a seaweed species-selection index for successful culture in a seaweed-based integrated aquaculture system. *J Ocean Univ China* 12:125–133
- Kautsky L (1982) Primary production and uptake kinetics of ammonium and phosphate by *Enteromorpha compressa* in an ammonium sulfate industry outlet area. *Aquat Bot* 12:23–40
- Kelly J (2020) Australian Seaweed Industry Blueprint. Publ 20-072, AgriFutures Australia, Wagga Wagga 44 pp
- Knowler D, Chopin T, Martinez-Espineira NA, Nobre A, Noce A, Reid G (2020) The economics of Integrated Multi-Trophic Aquaculture: where are we now and where do we need to go? *Rev Aquac* 12:1579–1594
- Küppers U, Weidner M (1980) Seasonal variation of enzyme activities in *Laminaria hyperborea*. *Planta* 148:222–230
- Lavery P, McComb A (1991) The nutritional ecophysiology of *Chaetomorpha linum* and *Ulva rigida* in Peel Inlet, Western Australia. *Bot Mar* 34:251–260
- Liao N (2008) Determination of ammonia in brackish or seawater by flow injection analysis. LaChat Instruments Loveland, USA
- Liu H, Wang F, Wang Q, Dong S, Tian X (2016) A comparative study of the nutrient uptake and growth capacities of seaweeds *Caulerpa lentillifera* and *Gracilaria lichenoides*. *J Appl Phycol* 28:3083–3089
- Lomas MW, Glibert PM (1999) Temperature regulation of nitrate uptake: a novel hypothesis about nitrate uptake and reduction in cool-water diatoms. *Limnol Oceanogr* 44:556–572
- Lorbeer AJ, Lahnstein J, Fincher GB, Su P, Zhang W (2015) Kinetics of conventional and microwave-assisted fucoidan extractions from the brown alga, *Ecklonia radiata*. *J Appl Phycol* 27:2079–2087
- Lüning K (1993) Environmental and internal control of seasonal growth in seaweeds. *Hydrobiologia* 260:1–14
- Martinez B, Rico J (2004) Inorganic nitrogen and phosphorus uptake kinetics in *Palmaria palmata* (Rhodophyta). *J Phycol* 40:642–650
- McDowell RE, Amsler MO, Li Q, Lancaster JR, Amsler CD (2015) The immediate wound-induced oxidative burst of *Saccharina latissima* depends on light via photosynthetic electron transport. *J Phycol* 51:431–441
- Miller SM, Hurd CL, Wing SR (2011) Variations in growth, erosion, productivity and morphology of *Ecklonia radiata* (Alariaceae; Laminariales) along a Fjord in southern New Zealand. *J Phycol* 47:505–516
- Naldi M, Wheeler P (1999) Changes in nitrogen pools in *Ulva fenestrata* (Chlorophyta) and *Gracilaria pacifica* (Rhodophyta) under nitrate and ammonium enrichment. *J Phycol* 35:70–77
- Neori A, Shpigel M (1999) Using algae to treat effluents and feed invertebrates in sustainable integrated mariculture. *WorldAquaculture* 30:46–49
- Neori A, Chopin T, Troell M, Buschmann AH, Kraemer GP, Halling C, Shpigel M, Yarish C (2004) Integrated aquaculture: rationale,

- evolution and state of the art emphasizing seaweed biofiltration in modern mariculture. *Aquaculture* 231:361–391
- Nguyen HTT, Pritchard DW, Hepburn CD (2020) Nitrogen and phosphorus ecophysiology of coralline algae. *J Appl Phycol* 32:2583–2597
- O'Brien M, Wheeler P (1987) Short term uptake of nutrients by *Enteromorpha prolifera* (Chlorophyceae). *J Phycol* 23:547–556
- Oh E, Edgar G, Kirkpatrick J, Stuart-Smith R, Barrett N (2015) Broad-scale impacts of salmon farms on temperate macroalgal assemblages on rocky reefs. *Mar Pollut Bull* 98:201–209
- Ozaki A, Mizuta H, Yamamoto H (2001) Physiological differences between the nutrient uptakes of *Kjellmaniella crassifolia* and *Laminaria japonica* (Phaeophyceae). *Fish Sci* 67:415–419
- Paine E, Schmid M, Revill AT, Hurd CL (2020) Light regulates inorganic nitrogen uptake and storage, but not nitrate assimilation, by the red macroalga *Hemineura frondosa* (Rhodophyta). *Eur J Phycol* 56:174–185
- Paling EI (1991) The relationship between nitrogen cycling and productivity in macroalgal stands and seagrass meadows. PhD thesis, University of Western Australia, Perth
- Park M, Shin SK, Do YH, Yarish C, Kim JK (2018) Application of open water integrated multi-trophic aquaculture to intensive monoculture: A review of the current status and challenges in Korea. *Aquaculture* 497:174–183
- Pedersen M (1994) Transient ammonium uptake in the macroalga *Ulva lactuca* (chlorophyta): nature, regulation, and the consequences for choice of measuring technique. *J Phycol* 30:980–986
- Pedersen M, Borum J (1997) Nutrient control of algal growth in estuarine waters. Nutrient limitation and the importance of nitrogen requirements and nitrogen storage among phytoplankton and species of macroalgae. *Mar Ecol Prog Ser* 142:261–272
- Phillips JC, Hurd CL (2003) Nitrogen ecophysiology of intertidal seaweeds from New Zealand: N uptake, storage and utilization in relation to shore position and season. *Mar Ecol Prog Ser* 264:31–40
- Phillips J (2001a) Marine macroalgal biodiversity hotspots: why is there high species richness and endemism in southern Australian marine benthic flora? *Biodivers Conserv* 10:1555–1577
- Phillips J (2001b) The nitrogen ecophysiology of intertidal seaweeds. PhD thesis, University of Otago, Dunedin
- Phillips J, Hurd CL (2004) Kinetics of nitrate, ammonium and urea uptake by four intertidal seaweeds from New Zealand. *J Phycol* 40:534–545
- Probyn TA, Chapman ARO (1982) Nitrogen uptake characteristics of *Chordaria flagelliformis* (Phaeophyta) in batch and continuous mode experiments. *Mar Biol* 71:129–133
- R Core Team (2021) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. <http://www.R-project.org/>
- Randall DJ, Wright PA (1987) Ammonia distribution and excretion in fish. *Fish Physiol Biochem* 3:107–120
- Read P, Fernandes T (2003) Management of environmental impacts of marine aquaculture in Europe. *Aquaculture* 226:139–163
- Roleda MY, Hurd CL (2019) Seaweed nutrient physiology: application of concepts to aquaculture and bioremediation. *Phycologia* 58:552–562
- Rosell K, Srivastava L (1985) Seasonal variations in total nitrogen, carbon and amino acids in *Macrocystis integrifolia* and *Nereocystis luetkeana* (Phaeophyta). *J Phycol* 21:304–309
- Rosenberg G, Paerl H (1981) Nitrogen fixation by blue-green algae associated with the siphonous green seaweed *Codium decortictum*: effects on ammonium uptake. *Mar Biol* 61:151–158
- Rosenberg G, Ramus J (1984) Uptake of inorganic nitrogen and seaweed surface area: volume ratios. *Aquat Bot* 19:65–72
- Rosenberg G, Probyn T, Mann K (1984) Nutrient uptake and growth kinetics in brown seaweeds: response to continuous and single additions of ammonium. *J Exp Mar Biol Ecol* 80:125–146
- Rugiu L, Hargrave MS, Enge S, Sterner M, Nylund GM, Pavia H (2021) Kelp in IMTAs: small variation in inorganic nitrogen concentrations drive different physiological responses of *Saccharina latissimi*. *J Appl Phycol* 33:1021–1034
- Ryther JH, Dunstan WM (1971) Nitrogen, phosphorus, and eutrophication in the coastal marine environment. *Science* 171:1008–1013
- Sanderson J, Bamler J (2012) A census of the marine Benthic Flora of Tasmania. Institute for Marine and Antarctic Studies, University of Tasmania, Hobart
- Sanderson J, Di Benedetto R (1988) Tasmanian Seaweeds for the Edible Market. Department of Sea Fisheries, Tasmanian Government, Hobart
- Sanderson J, Cromey C, Dring M, Kelly M (2008) Distribution of nutrients for seaweed cultivation around salmon cages at farm sites in north-west Scotland. *Aquaculture* 278:60–68
- Sato Y, Hirano T, Niwa K et al (2016) Phenotypic differentiation in the morphology and nutrient uptake kinetics among *Undaria pinnatifida* cultivated at six sites in Japan. *J Appl Phycol* 28:3447–3458
- Schaffelke B, Klumpp D (1998) Nutrient-limited growth of the coral reef macroalga *Sargassum baccularia* and experimental growth enhancement by nutrient addition in continuous flow culture. *Mar Ecol Prog Ser* 164:199–211
- Schiel DR, Foster MS (2015) The Biology and Ecology of Giant Kelp Forests. University of California Press, California, USA
- Seely G, Duncan M, Vidaver W (1972) Preparative and analytical extraction of pigments from brown algae with dimethyl sulfoxide. *Mar Biol* 12:184–188
- Seitzinger SP, Kroeze C, Bouwman A, Caraco N, Dentener F, Styles RV (2002) Global patterns of dissolved inorganic and particulate nitrogen inputs to coastal systems: recent conditions and future projections. *Estuaries* 25:640–655
- Seitzinger S, Sorensen L, Uematsu M et al (2008) Impacts of atmospheric anthropogenic nitrogen on the open ocean. *Science* 320:893–897
- Smit A (2002) Nitrogen uptake by *Gracilaria gracilis* (Rhodophyta): adaptations to a temporally variable nitrogen environment. *Bot Mar* 45:196–209
- Soto D, Norambuena F (2004) Evaluation of salmon farming effects on marine systems in the inner seas of southern Chile: a large-scale mensurative experiment. *J Appl Ichthyol* 20:493–501
- Taylor R, Peek J, Rees T (1998) Scaling of ammonium uptake by seaweeds to surface area: volume ratio: geo-graphical variation and the role of uptake by passive diffusion. *Mar Ecol Prog Ser* 169:143–148
- Thomas TE, Harrison PJ, Taylor EB (1985) Nitrogen uptake and growth of the germlings and mature thalli of *Fucus distichus*. *Mar Biol* 84:267–274
- Thomas TE, Harrison PJ, Turpin DH (1987) Adaptations of *Gracilaria pacifica* (Rhodophyta) to nitrogen procurement at different intertidal locations. *Mar Biol* 93:569–580
- Topinka J (1978) Nitrogen uptake by *Fucus spiralis* (Phaeophyceae). *J Phycol* 14:241–247
- Van Alstyne K (2018) Seawater nitrogen concentration and light independently alter performance, growth, and resource allocation in the bloom-forming seaweeds *Ulva lactuca* and *Ulvaria obscura* (Chlorophyta). *Harmful Algae* 78:35
- Vásquez J (2008) Production, use and fate of Chilean brown seaweeds: resources for a sustainable fishery. *J Appl Phycol* 20:457–467
- Vergara J, Niell F, Torres M (1993) Culture of *Gelidium sesquipedale* (Clem.) Born. et Thur. in a chemostat system. Biomass production and metabolic responses affected by N flow. *J Appl Phycol* 5:405–415

- Vergara J, Bird K, Niell F (1995) Nitrogen assimilation following NH_4^+ pulses in the red alga *Gracilariopsis lemaneiformis*: effect of C metabolism. *Mar Ecol Prog Ser* 122:253–263
- Vitousek P (1997) Human Domination of Earth's Ecosystems. *Science* 277:494–499
- Voss M, Bange HW, Dippner JW, Middelburg JJ, Montoya JP, Ward B (2013) The Marine nitrogen cycle: recent discoveries, uncertainties and the potential relevance of climate change. *Philos Trans R Soc Lond B* 368:20130121
- Wallentinus I (1984) Comparisons of nutrient uptake rates for Baltic macroalgae with different thallus morphologies. *Mar Biol* 80:215–225
- Wheeler PA (1979) Uptake of methylamine (an ammonium analogue) by *Macrocystis pyrifera* (Phaeophyta). *J Phycol* 15:12–17
- Wheeler W (1980) Pigment content and photosynthetic rate of the fronds of *Macrocystis pyrifera*. *Mar Biol* 56:97–102
- Wheeler PA, Björnsater BR (1992) Seasonal fluctuations in tissue nitrogen, phosphorus, and N:P for five macroalgal species common to the Pacific northwest coast. *J Phycol* 28:1–6
- Wild-Allen K, Herzfeld M, Thompson PA, Rosebrock U, Parslow J, Volkman JK (2010) Applied coastal biogeochemical modeling to quantify the environmental impact of fish farm nutrients and inform managers. *J Mar Syst* 81:134–147
- Wilkie M (1997) Mechanisms of ammonia excretion across fish gills. *Comp Biochem Physiol A* 118:39–50
- Williams S, Fisher T (1985) Kinetics of nitrogen-15 labelled ammonium uptake by *Caulerpa cupressoides* (Chlorophyta). *J Phycol* 21:287–296
- Wu H, Huo Y, Han F, Liu Y, He P (2015) Bioremediation using *Gracilaria chouae* co-cultured with *Sparus macrocephalus* to manage the nitrogen and phosphorus balance in an IMTA system in Xiangshan Bay, China. *Mar Pollut Bull* 91:272–279
- Wu H, Kim J, Huo Y, Zhang J, He P (2017) Nutrient removal ability of seaweeds on *Pyropia yezoensis* aquaculture rafts in China's radial sandbanks. *Aquat Bot* 137:72–79

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