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Nitrogen ecophysiology of intertidal seaweeds from New Zealand: N uptake, storage and utilisation in relation to shore position and season

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ABSTRACT: The nitrogen ecophysiology of 4 intertidal seaweeds (*Stictosiphonia arbuscula*, *Apophlaea lyallii*, *Scytothamnus australis*, *Xiphophora gladiata*) from southeastern New Zealand is described in terms of N status, N uptake rates and N utilisation. The species growing in the highest shore position had large internal NO_3^- and NH_4^+ pools. For all species, tissue NH_4^+ pools were greater than tissue NO_3^- pools. Total tissue N was directly related to shore position with high intertidal species having highest tissue N, while the opposite trend was observed for C:N ratios. The ability to take up inorganic (NO_3^-, NH_4^+) and organic (urea) N when one or all N forms were present in the culture medium was measured using time-course uptake experiments at initial concentrations of 5 and 30 µM. Nitrate uptake did not vary over time for any of the species. *S. arbuscula* and *S. australis* exhibited a surge phase of NH_4^+ uptake at both concentrations. Urea uptake at 5 µM was generally low and consistent over time; uptake at 30 µM was highly variable. All species were capable of simultaneous uptake of all N forms. The relative importance of each N form to overall N nutrition indicated that NH_4^+ was an important N source in winter for all species. Urea was an important N source in summer, contributing 27 to 33% to the total N acquisition for most species. A relative preference index indicated that in winter N sources were utilised in the order $NH_4^+ > NO_3^- >$ urea, while in summer the order was $NH_4^+ = NO_3^- >$ urea. Estimates of the amount of N that each species could acquire during a tidal cycle indicated that the high intertidal *S. arbuscula* had the greatest capacity for N acquisition, regardless of season.

KEY WORDS: Macroalgae · Intertidal seaweed · Nitrogen uptake · Nitrate · Ammonium · Urea · Season · Zonation · New Zealand

INTRODUCTION

The zonation of intertidal seaweeds on rocky shores is related in part to their ability to withstand disruptive and limitation stresses (Davison & Pearson 1996). It is well known that the abilities of seaweeds to recover physiological function following disruptive stresses are related to their position on the shore (e.g. Dring & Brown 1982, Dudgeon et al. 1989, 1990, Kübler & Davison 1993, Burritt et al. 2002). Less is known about the abilities of intertidal seaweeds to withstand limited supplies of essential inorganic nutrients such as nitrogen during tidal emersion (Davison & Pearson 1996). Since intertidal seaweeds are removed from their source of N when they are emersed, N-limitation may be one of the most important limitation stresses structuring intertidal algal communities (Thomas et al. 1987a). Several studies (Schonbeck & Norton 1979, Thomas et al. 1987a, b, Hurd & Dring 1990) have provided evidence that the growth of high intertidal seaweeds is nutrient-limited, although there has been no conclusive study demonstrating N-limitation in high shore species. Furthermore, it has not yet been established whether high-shore species tolerate limited

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periods of N availability by more rapid N uptake when submersed, lower N requirements for growth, greater N storage capacity, or through the ability to readily utilise different N forms.

The energetics of uptake and assimilation of different N forms will affect N utilisation. Following uptake, NO_3^- assimilation has a high energy demand to reduce it to NH_4^+ in a 2-step process (Lobban & Harrison 1994). Ammonium is a reduced N form that can be readily incorporated into amino acids (Nasr et al. 1968, Lobban & Harrison 1994). Much of the current knowledge on urea assimilation by microalgae indicates that urease releases NH_4^+ from urea, and the free NH_4^+ is then assimilated (Antia et al. 1991, Tamminen & Irmisch 1996). Ammonium and urea may therefore be energetically favourable to seaweeds, particularly in winter when reduced light levels may impose energy-limitation.

Uptake of nitrogenous ions is frequently variable over time, and may be influenced by the physiological status of the seaweed. For example, an initial phase of surge or enhanced uptake can occur as a response to depleted internal N storage pools in N-starved algae (Fujita et al. 1988, McGlathery et al. 1996). As internal N pools fill, feedback inhibition of membrane transport processes may result in a temporary reduction in subsequent uptake rates (Fujita et al. 1988, Pedersen 1994, McGlathery et al. 1996). This period of internally-controlled uptake represents the maximum uptake rate of N-replete cells where the rate-limiting step is the assimilation of N into organic compounds (Fujita et al. 1988, McGlathery et al. 1996). A delay in the activation of the uptake mechanism for a particular N form results in an initial lag in uptake rates. This lag phase has been reported for NO_3^- and is thought to represent an induction period of the uptake mechanism following exposure to high concentrations (Harrison et al. 1986). Induction of NO_3^- uptake was evident for *Macrocystis integrifolia* (Wheeler & Srivastava 1984), *Porphyra perforata* (Thomas & Harrison 1985) and *Laminaria groenlandica* (Harrison et al. 1986). Patterns of non-linear uptake, such as surge or lag phases, can be detected from a time-course incubation (Caperon & Meyer 1972), which allows a full description of the temporal response of an alga to the addition of nitrogen (Harrison et al. 1989).

When more than 1 N source is available for uptake by seaweeds, the ability to take up the different N forms may be affected. The presence of NH_4^+ inhibits NO_3^- uptake in certain green and red algal species, for example *Codium fragile* (Hanisak & Harlin 1978), *Hypnea musciformis* (Haines & Wheeler 1978), *Gracilaria foliifera* and *Neoagardhiella baileyi* (D'Elia & DeBoer 1978), although inhibition may only be partial or temporary. For many brown algae species, such as *Macrocystis pyrifera* (Haines & Wheeler 1978), *Macrocystis integrifolia* (Wheeler & Srivastava 1984), *Fucus spiralis* (Topinka 1978) and *Laminaria longicruris* (Harlin & Craigie 1978), the presence of NH_4^+ does not affect rates of NO_3^- uptake by mature plants. However, Harrison et al. (1986) found that NO_3^- uptake by first-year *Laminaria groenlandica* plants was completely suppressed for 30 min in the presence of NH_4^+. Although urea uptake in macroalgae is not well documented, the presence of inorganic N (e.g. NO_3^-, NH_4^+) in the culture medium can inhibit urea uptake rates in phytoplankton (Horrigan & McCarthy 1982, Antia et al. 1991).

This paper examines the ability of 4 intertidal seaweeds that grow at different vertical positions on the shore to take up inorganic and organic N when all or 1 N form is present in the culture medium. Time-course N uptake experiments at a low (5 µM) and high (30 µM) initial concentration were used to investigate temporal variation in uptake for evidence of lag or surge phases, the ability to sustain uptake over prolonged periods, preference for any particular N form, and evidence of inhibition of uptake. The nutrient status of each species and its likely effect on uptake rates were examined, and the relative importance of each N source to overall N nutrition and the capacity for each species to take up N during a tidal cycle are also reported.

MATERIALS AND METHODS

Species and collection site. We collected 4 species of macroalgae from 2 divisions, *Stictosiphonia arbuscula* (Harvey) King et Puttock, *Apophlaea lyallii* Hook. f. et Harvey, *Scytothamnus australis* (J.Agardh) Hook. f. et Harvey and *Xiphophora gladiata* (Labillardière) Montagne ex Harvey from the intertidal zone of a wave-exposed, rocky shore at Brighton Beach (45° 57′ S, 170° 20′ E), south-eastern New Zealand, which has a maximum tidal range of 2.1 m. Table 1 gives the taxonomic affinity of each species as well as details on their shore position and submersion times. *S. arbuscula* shows morphological variation across its vertical distribution and was therefore collected from the upper ('high shore') and lower ('low shore') vertical extremes.

Ambient seawater N concentrations. Seawater samples (n = 5) were collected monthly from Brighton Beach between January 1999 and March 2000, filtered (Whatman GF/C) and analysed for NO_3^- (nitrate + nitrite) and NH_4^+ concentrations using a QuikChem® 8000 Automated Ion Analyzer (Lachat Instruments). The minimum amount of N that could be detected in seawater samples was 0.03 and 0.07 µM of NO_3^- and NH_4^+, respectively (Lachat Instruments).

Table 1. *Stictosiphonia arbuscula*, *Apophlaea lyallii*, *Scytothamnus australis* and *Xiphophora gladiata*. Taxonomic affinity, shore position and submersion time for the 4 intertidal species used in this study. LAT: shore position (m above lowest astronomical tide). Submersion times (per tidal cycle) estimated using median shore position of each species and assuming maximum tidal fluctuation of 2.1 m at Brighton Beach, New Zealand

Species	Division	Order	LAT	Approx. submersion time (h)
Stictosiphonia arbuscula (high shore)	Rhodophyta	Ceramiales	2.25 – 3	2
Apophlaea lyallii	Rhodophyta	Hildenbrandiales	1.5 – 2	2.75
Stictosiphonia arbuscula (low shore)	Rhodophyta	Ceramiales	1.5 – 2.25	3.5
Scytothamnus australis	Phaeophyta	Scytothamnales	1 – 1.5	5
Xiphophora gladiata	Phaeophyta	Fucales	0 – 0.4	10

Tissue C and N and soluble tissue nitrate and ammonium. Monthly, between January 1999 and March 2000, approximately 5 (high- and low-shore *Stictosiphonia arbuscula*), 10 (*Apophlaea lyallii* and *Scytothamnus australis*) or 20 (*Xiphophora gladiata*) g wet weight of non-reproductive tissue was removed from each of 3 replicates of each species. Clean algal material was dried at 80°C for 48 h and total C and total N were determined using a Carlo Erba CHNS-O Elemental Analyzer (Model EA1108). The percent tissue C and percent tissue N were standardised to algal dry weight (g) and C:N ratios were determined on an atomic weight basis.

Soluble tissue NO_3^- and NH_4^+ were determined using a boiling-water extraction after Hurd et al. (1996). Non-reproductive apical segments (0.10 ± 0.01 g, n = 5) were chopped into small pieces and placed in boiling tubes containing 40 ml of high-purity water. The tubes were placed in a boiling water bath for 20 min, cooled, and the supernatant decanted, filtered (Whatman GF/C) and analysed for NO_3^- and NH_4^+. Preliminary experiments revealed that 3 extractions from the same algal material were sufficient to extract all soluble tissue NO_3^- and NH_4^+.

Collection and pre-treatment of algae for N uptake experiments. Algal material was collected during low tide on the day of each uptake experiment, between 07:00 and 08:30 h. For *Stictosiphonia arbuscula*, *Apophlaea lyallii* and *Scytothamnus australis* whole-plant segments were collected, while apical segments were collected for the larger *Xiphophora gladiata*.

Algal material was cleaned of all epiphytes and sand, trimmed to the required size where necessary, and placed into 1 l glass containers containing ~0.7 l filtered (Whatman GF/C) seawater from the collection site. Seaweeds were pre-treated for 2 h in a constant-temperature growth cabinet at 12°C (Conviron Model E15, Controlled Environments), with cool white fluorescent tubes (Sylvania F72T12-CW-VHO) providing an overhead irradiance of 170 to 200 µmol photons m^{-2} s^{-1} and orbital shakers providing water motion (Model SS70, Chiltern Scientific) at 110 rpm. Preliminary uptake trials revealed that the 2 h pre-treatment period was sufficient to allow all species to recover from any wounding or emersion effects.

Multiple-N-addition uptake experiments. Time-course uptake experiments involving the addition of 3 N forms to the seawater medium were conducted in winter (July) 1998, summer (January) 1999 and winter (July) 1999, under the same conditions as those used for pre-treatment. To reduce the possibility of diel variation in uptake, all experiments commenced between 12:00 and 15:00 h. An initial wet weight to volume ratio of 0.3 to 0.5 g l^{-1} was used for all species.

Uptake experiments were conducted in 1 l acid-washed (10% HCl) glass jars containing 500 ml of filtered seawater. Nitrate, ammonium and urea (as KNO_3, NH_4Cl and urea, respectively) were all added to each jar, at either 5 or 30 µM (in addition to background N in the seawater media). Jars were placed on orbital shakers in the growth cabinet and a 10 ml water sample was taken from each jar before algal material was added to determine initial N concentrations. Further water samples were taken every 15 min for the first hour thereafter, every 30 min for the second hour and a final sample was taken after 3 h: 3 replicates at each concentration were performed on 2 consecutive days, giving a total of 6 replicates per species; 2 controls (no algal material added) at each concentration were also performed each day. Algal material was removed and dried at 80°C for 48 h for dry weight determination.

Single-N-addition uptake experiments. In July 1999, time-course uptake experiments involving the addition of a single N source to the seawater medium were conducted. Methods were as for the multiple-N addition experiments described above, except that only 1 N source (either NO_3^-, NH_4^+ or urea) was added to each jar at a concentration of 5 µM.

For multiple and single-addition experiments, uptake rates for each N source during each time interval were calculated using the equation:

$$V = \frac{(S_i - S_f) \times \mathrm{vol}}{t \times \mathrm{dw}} \quad (1)$$

where V = uptake rate (µmol [g dry wt]$^{-1}$ h^{-1}), S_i = substrate concentration at start of time interval (µM), S_f = substrate concentration at end of time interval (µM), vol = volume at start of time interval (l), t = time (h), and dw = dry weight (g).

Relative importance of NO_3^-, NH_4^+ and urea. The relative importance of NO_3^-, NH_4^+ and urea was determined from the contribution of each to the total amount of N taken up, and by using the relative preference index (McCarthy et al. 1977, Varela & Harrison 1999), both of which were calculated from N uptake rates.

The contribution of each N form to the total N taken up (in µg) was determined using V_{0-90} from 5 µM multiple-N-addition uptake experiments. For each species, the uptake rate of each N form during replicate time-courses was multiplied by the atomic weight of N and the contribution to total N (expressed as %) calculated. The relative preference index (RPI) for each N source for each species was determined by comparing N uptake rates relative to N availability, after McCarthy et al. (1977) and Varela & Harrison (1999). The equation for calculating RPI was modified to include time-dependency of uptake, and to remove the disproportionate effect of negative uptake rates. The RPI for NO_3^- was calculated as follows:

$$\mathrm{RPI}_{\mathrm{nitrate}} = \frac{V(NO_3^-)_{0-15} \,/\, \left(V(NO_3^-)_{0-15} + V(NH_4^+)_{0-15} + V(\mathrm{urea})_{0-15}\right)}{[NO_3^-]_{0-15} \,/\, \left([NO_3^-]_{0-15} + [NH_4^+]_{0-15} + [\mathrm{urea}]_{0-15}\right)} \quad (2)$$

where V = uptake rate (µmol [g dry wt]$^{-1}$ h^{-1}); if negative, treated as zero; $_{0-15}$ = time interval of the time-course (min); and [ ] = mean concentration (µM) during time interval indicated.

The RPI for NO_3^- was calculated separately for each time interval of the first 90 min of the time-courses, and plotted against the total N concentration (i.e. any background N in the seawater plus N additions) during each interval. Where an RPI was zero (i.e. resulting from a negative uptake rate or release) it was excluded from the plots. This procedure was repeated for NH_4^+ and urea. Uptake rates proportional to availability are indicated by an RPI value of 1 (i.e. unity); values above or below indicate faster or slower uptake rates, respectively, relative to availability (McCarthy et al. 1977). RPI values do not indicate physiological preference; rather they allow inference as to which N form is taken up at a faster rate relative to its ambient concentration (Varela & Harrison 1999).

Total N acquisition over tidal cycle. The total amount of N that each species might take up over a 12 h tidal cycle was estimated from approximate submersion times during a single tide and from uptake rates of each N form during the 5 µM multiple-N-addition time-course experiments (after Hurd & Dring 1990).

The uptake rate of each N form during each time interval of the 3 h time-courses was used to determine the amount of N taken up (in µg [g dry wt]$^{-1}$) in that period. (Uptake rates over 3 h were chosen as it was considered that this would most accurately incorporate interspecific differences in the reduction in uptake rates due to internal pool-filling and/or N assimilation rates.) Where the uptake rate during a time interval was negative, the value was converted to zero (i.e. no contribution to total amount of N taken up in that period). Values were converted to µg N to allow accurate assessment of the contribution of the dinitrogen urea, and amounts were summed over the submersion period. Using the maximum tidal fluctuation of 2.1 m and the median shore position of each species, submersion times were estimated as 2 h for high-shore *Stictosiphonia arbuscula*, 2.75 h for *Apophlaea lyallii*, 3.5 h for low-shore *S. arbuscula*, 5 h for *Scytothamnus australis* and 10 h for *Xiphophora gladiata*. For the latter 3 species, uptake rates of each N form were assumed to remain constant for the remaining 0.5, 2 and 7 h of submersion, respectively.

Data analyses. We used 2-way analyses of variance (ANOVA) or Student's *t*-tests to examine for differences in uptake rates between species and time intervals, at $p < 0.05$. Transformations failed to remove heterogeneity of variances, so ANOVAs or *t*-tests proceeded using untransformed data as these analyses are relatively robust to violations of assumptions (Underwood 1981, 1997, Zar 1996). We also used 2-way ANOVAs ($p < 0.05$) to compare the percentage contribution of each N form between species. Where ANOVAs indicated significant differences between factors, post-hoc multiple comparisons were performed using multiple-comparison Tukey tests. All analyses were performed using SigmaStat (v2.03, SPSS, USA).

There were no consistent trends between season (ie. winter vs summer), and because our goal was to determine patterns of uptake during the time-course experiments, data from the 3 seasons were pooled (n = 18). Uptake rates during the first time interval (0 to 15 min, hereafter denoted V_{0-15}) and 'sustained' uptake rates, determined from the difference in N concentration at 15 min and after 90 min (denoted V_{15-90}), were compared within and between species. The period 15 to 90 min was chosen so that the N concentration in the seawater media was not reduced to below 50% of the initial concentration. For each data set, values that appeared erroneous (outliers) were excluded from analyses. From the remaining data, 7 replicates were ran-

domly assigned to each of V_{0-15} and V_{15-90} to achieve independence of data. V_{0-15} and V_{15-90} of each N source at either low (5 µM) or high (30 µM) initial concentrations were then compared separately by 2-way ANOVA, the factors being species and time, followed by Tukey multiple comparison tests where required. Surge uptake was considered to have taken place if the mean uptake rate during the first 15 min (V_{0-15}) was significantly higher than the mean rate of uptake during the subsequent period (V_{15-90}). Alternatively, if V_{0-15} was significantly lower than V_{15-90} this was considered to represent an initial lag phase in N uptake.

To determine if inhibition of N uptake occurred when all 3 N sources were presented together, uptake rates of each N source at 5 µM were compared between multiple- and single-N addition time-course experiments conducted in July 1999. V_{0-15} and V_{15-90} for each species and each N source were compared by a *t*-test.

RESULTS

Seawater NO_3^- and NH_4^+ concentrations

NO_3^- concentrations in surface seawater at Brighton Beach in winter were approximately 10-fold higher than in summer, with a maximum of 4.5 µM recorded in June 1999 (Fig. 1). No seasonal pattern for NH_4^+ concentrations was evident, with a maximum concentration of 2.2 µM in March 1999 and a minimum of 0.6 µM in March 2000.

Tissue C and N

Tissue C content was lowest in low-shore *Stictosiphonia arbuscula* in all months apart from November 1999 and ranged from 27 to 33% of dry weight (Fig. 2a). *Apophlaea lyallii* had the consistently highest % C of all algae studied, with values between 35 and 41%. The 2 brown algae *Scytothamnus australis* and *Xiphophora gladiata* had intermediate levels of tissue C ranging from 32 to 37% and 30 to 36% C, respectively. No seasonal trends in % tissue C were evident for any of the 4 species examined.

For tissue N, high-intertidal species had the highest N content (as % dry weight), while lowest values were recorded for species occupying the lower shore positions (Fig. 2b). For all species a seasonal trend of greater tissue N in late winter/early spring following a steady increase after a summer minimum was evident, although this was most pronounced in the low- and high-shore *Stictosiphonia arbuscula.*

The brown algae *Scytothamnus australis* and *Xiphophora gladiata* had the highest C:N ratios; these were always above 18.5 and reached a maximum of ~30 (Fig. 2c). C:N ratios for *Stictosiphonia arbuscula* were lowest, and always below 15. For all species there was a general trend for higher C:N ratios during the summer months.

Soluble tissue N

For all species, NH_4^+ content g dry wt^{-1} was higher than NO_3^- content during the study period (Fig. 3). Soluble tissue NO_3^- content in high- and low- shore populations of *Stictosiphonia arbuscula* appeared to respond to the 10-fold increase in ambient seawater NO_3^- during winter (Fig. 1), reaching peaks of 28 and 42 µmol g dry wt^{-1}, respectively. Soluble tissue NO_3^- content among the remaining species was similar, and generally remained below 7 µmol g dry wt^{-1}.

Ammonium content varied among species, and species growing at the tidal extremes of the intertidal zone (i.e. *Stictosiphonia arbuscula* and *Xiphophora gladiata*) had similar levels of soluble tissue NH_4^+ (Fig. 3). Maximum soluble tissue NH_4^+-content in high- and low-shore *S. arbuscula* and *X. gladiata* was 58, 49 and 84 µmol g dry wt^{-1}, respectively. Soluble tissue NH_4^+ levels in *Apophlaea lyallii* and *Scytothamnus australis* were generally low throughout the study period, with levels between 4 and 15 µmol g dry wt^{-1} at most times.

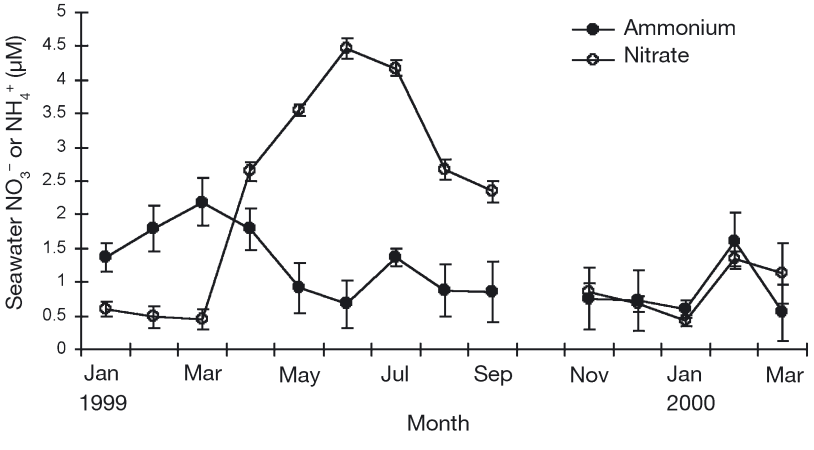

Fig. 1. Mean (± SE, n = 5) nitrate and ammonium concentrations in seawater at Brighton Beach, New Zealand, January 1999 to March 2000

Temporal variation in N uptake

The change in N concentration (NO_3^-, NH_4^+ and urea) in control solutions was minimal (<4%), and the disappearance of N from the seawater medium was therefore attributed to uptake by the macroalgae.

Nitrate uptake did not vary with time for any of the species examined, although uptake rates varied among species (Fig. 4a,b). High- and low-shore *Stictosiphonia arbuscula* had similar, high rates of NO_3^- uptake at both 5 and 30 µM initial concentrations (Tukey's, $p > 0.05$ in both comparisons). At high initial concentrations (30 µM), all species had higher uptake rates for V_{0-15} compared with V_{15-90}, although differences were not significant ($p = 0.06$) (Fig. 4b).

Ammonium uptake varied with time and among species, and there were significant interactions between these factors during both 5 and 30 µM experiments ($p < 0.01$) (Fig. 4c,d). *Stictosiphonia arbuscula* (high- and low-shore) and *Scytothamnus australis* exhibited surge uptake of NH_4^+ (V_{0-15}) that was significantly higher than V_{15-90}, regardless of the initial concentration (Tukey's, $p < 0.05$ in all cases). Uptake rates for *Apophlaea lyallii* and *Xiphophora gladiata* were similar (Tukey's, $p > 0.99$) and significantly lower than for the other species (Tukey's, $p < 0.01$) at high and low concentrations.

At low initial urea concentrations, high-shore *Stictosiphonia arbuscula* showed an initial release of urea (i.e. negative V_{0-15}), while uptake over time did not vary for the remaining species (Tukey's, $p > 0.1$) (Fig. 4e). Urea uptake at high urea concentrations was

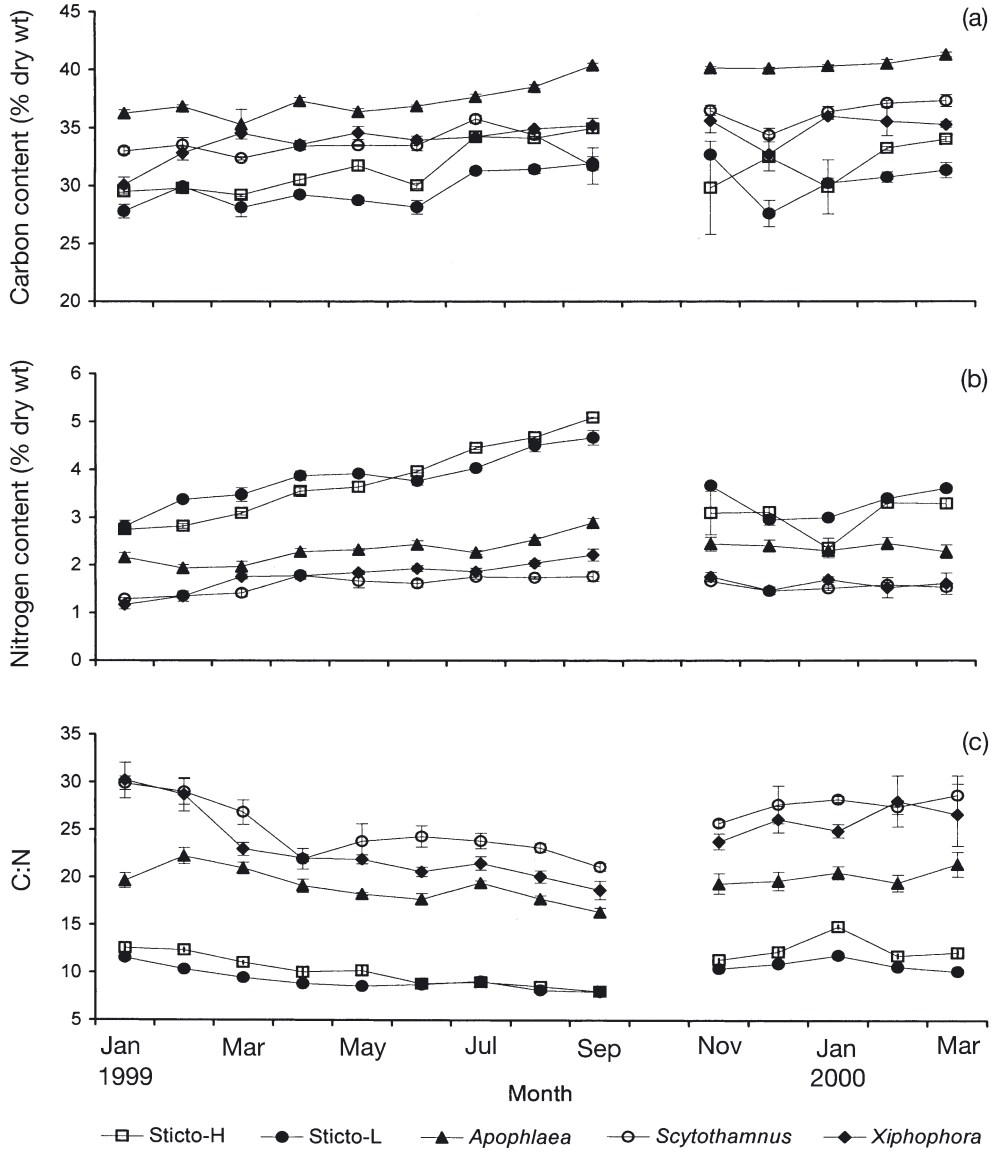

Fig. 2. *Stictosiphonia arbuscula*, *Apophlaea lyallii*, *Scytothamnus australis* and *Xiphophora gladiata*. Mean (± SE, n = 3) tissue C and N content between January 1999 and March 2000. (a) Tissue C content (% dry wt); (b) tissue N content (% dry wt); (c) tissue C:N ratio (at. wt). Sticto-H, Sticto-L: high-shore and low-shore *Stictosiphonia arbuscula*, respectively

highly variable and the large error associated with uptake rates meant there were no significant differences within or between species (Fig. 4f).

Single versus multiple-N-addition experiments

Although V_{0-15} for NO_3^- uptake by all species was always higher when NO_3^- was presented as the sole N source than when all 3 N sources were available for uptake, this difference was only significant for *Apophlaea lyallii* ($p = 0.0008$; Fig. 5). V_{0-15} for all other species and N sources did not vary significantly when 1 or all N sources were available ($p > 0.06$ in all cases). Similarly, V_{15-90} for all N sources were similar in single- and multiple-N experiments for all species ($p > 0.05$).

Percentage contribution of each N form to total N acquisition

NO_3^- was a significantly more important N source in winter 1998 than during the other seasons ($p = 0.0001$ in both comparisons) and accounted for more than half the N taken up by high-shore *Stictosiphonia arbuscula* in this season (Fig. 6a). Among species, high-shore *S. arbuscula* took up significantly more NO_3^- than *Xiphophora gladiata* ($p = 0.02$).

Ammonium contributed a similar proportion of the total N acquired in 90 min in each season (2-way ANOVA, $p = 0.10$), but in summer 1999 the amount taken up by *Apophlaea lyallii* was greater than that by high-shore *Stictosiphonia arbuscula* ($p = 0.03$) (Fig. 6b). In winter 1999, NH_4^+ was the most important of all 3 N sources for all species, contributing up to 74% (Fig. 6c), which was significantly greater than during other seasons ($p < 0.005$). Within species, the percentage contribution of NH_4^+ varied between seasons for both high-shore *S. arbuscula* and *Scytothamnus australis* ($p < 0.05$) and was highest in winter 1999 for both species (Fig. 6c).

Urea contribution varied between seasons and species, and there was a significant interaction between these factors (2-way ANOVA, $p < 0.005$ in all cases) (Fig. 6). Urea contribution was low in both winters and did not vary between species ($p > 0.19$), but became a more important N source during summer with contribution increasing to 27–33% for all species except *Apophlaea lyallii* (10%) (Fig. 6).

Relative preference index (RPI)

Examination of the RPI among species revealed some general trends in the utilisation of each N

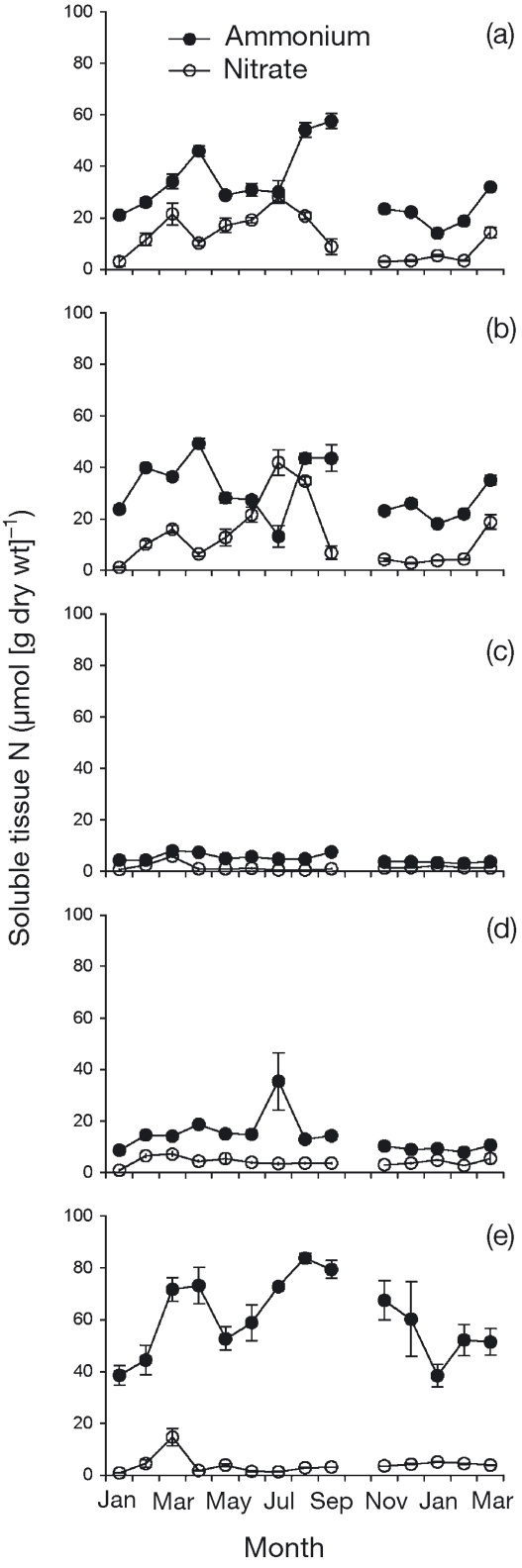

Fig. 3. (a,b) High- and low-shore *Stictosiphonia arbuscula* respectively; (c) *Apophlaea lyallii*; (d) *Scytothamnus australis*; (e) *Xiphophora gladiata*. Mean (±SE, n = 5) soluble tissue nitrate and ammonium in thalli, January 1999 to March 2000

source. Firstly, the ammonium RPI was usually above unity in all seasons and was typically higher than the RPI for NO_3^- or urea (Fig. 7), indicating that NH_4^+ uptake was faster than that of either NO_3^- or urea. Furthermore, the RPI_{NH4} was higher in winter than in summer for certain species such as high-shore *Stictosiphonia arbuscula* and *Scytothamnus australis* (Fig. 7).

All species had a higher RPI_{NO3} in summer than in winter, the former being close to RPI_{NH4} (Fig. 7), indicating that uptake of NO_3^- and NH_4^+ proceeded at similar rates. For low-shore *Stictosiphonia arbuscula* and

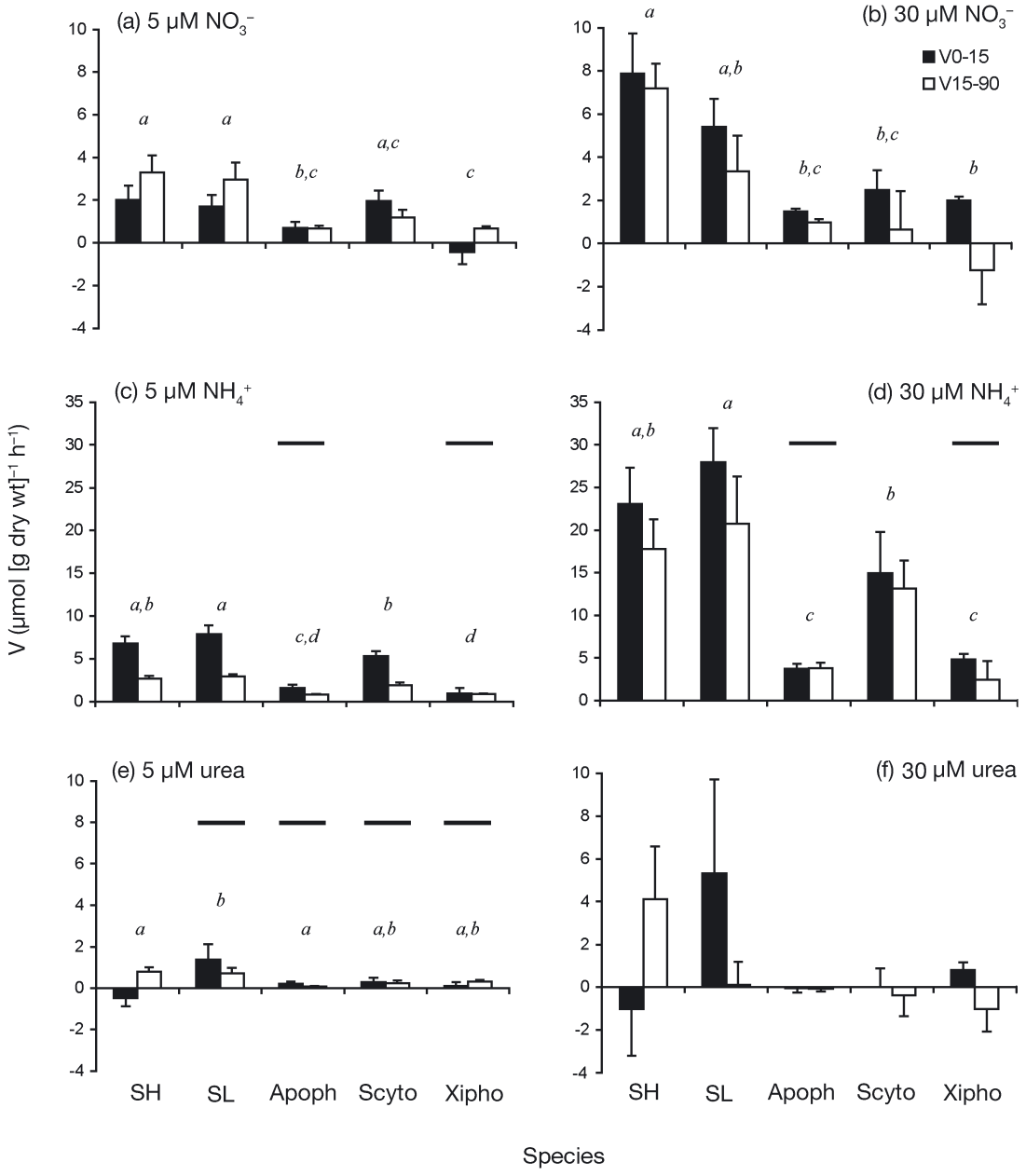

Fig. 4. *Stictosiphonia arbuscula* (SH: high-shore; SL: low-shore), *Apophlaea lyallii* (Apoph), *Scytothamnus australis* (Scyto) and *Xiphophora gladiata* (Xipho). Temporal variation in uptake rates (V, mean ± SE, n = 7) of nitrate, ammonium and urea during multiple-N-addition (5 or 30 µM) time-course experiments. Uptake rates were determined during the first 15 min (V_{0-15}) and subsequent 15 to 90 min (V_{15-90}) of 3 h time-course experiments. Different letters above bars indicate significant difference in uptake rates ($p < 0.05$; data pooled from both time intervals) between species; shared horizontal lines above bars indicate no significant difference ($p > 0.05$; Tukey's test). Note different scales on *y*-axes

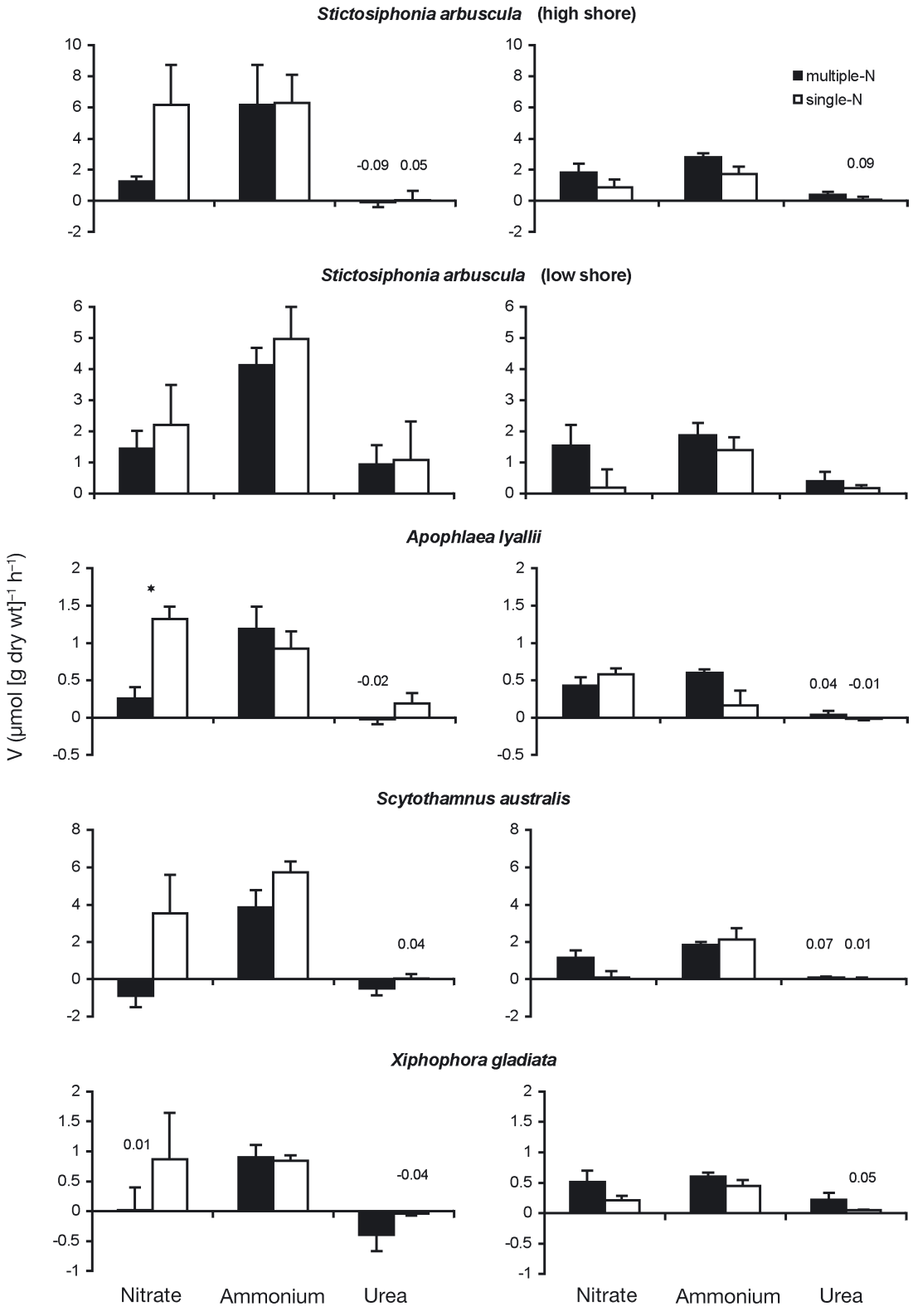

Fig. 5. *Stictosiphonia arbuscula*, *Apophlaea lyallii*, *Scytothamnus australis* and *Xiphophora gladiata.* Comparison of N uptake rates (V, mean ± SE, n = 6) during multiple- and single-N addition (5 µM) time-course experiments. Uptake rates were determined during the first 15 min (V_{0-15}) and subsequent 15 to 90 min (V_{15-90}) of 3 h time-course experiments. *: Significant differences ($p < 0.05$; Student's t-test) in uptake rates. Note different scales on y-axes

Xiphophora gladiata, this trend was evident at all N concentrations in summer experiments (Fig. 7).

It is interesting when that total N concentrations fell below 5 to 6 µM in summer, the RPI_{urea} for *Stictosiphonia arbuscula* and *Scytothamnus australis* was close to or approaching unity (Fig. 7). This indicates that urea uptake at low concentrations was proportional to availability for these species. In contrast, when total N was abundant in either summer or winters, the RPI_{urea} was typically below 1 for all species. A notable exception to this was for low-shore *S. arbuscula* in winter 1999, when the RPI_{urea} was at or above unity at most N concentrations (Fig. 7).

When results for all species across the range of total N concentrations are summarised, seasonal trends in the utilisation of the 3 N sources are evident. In winter, the RPI indicated that N sources were utilised in the

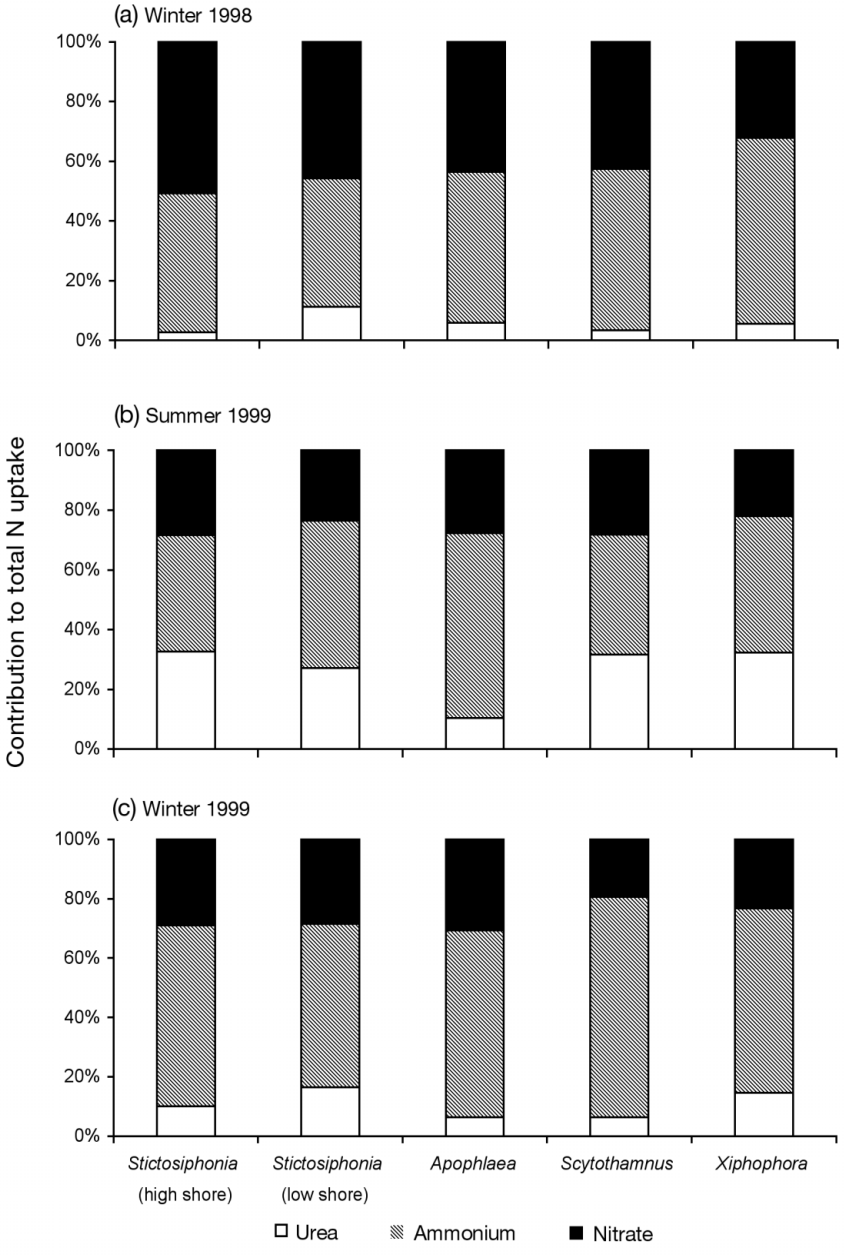

Fig. 6. *Stictosiphonia arbuscula*, *Apophlaea lyallii*, *Scytothamnus australis* and *Xiphophora gladiata*. Mean contribution of nitrate, ammonium and urea to total amount of N (µg) taken up over 90 min. Data derived from uptake rates during multiple-N-addition experiments at low (5 µM) initial concentrations

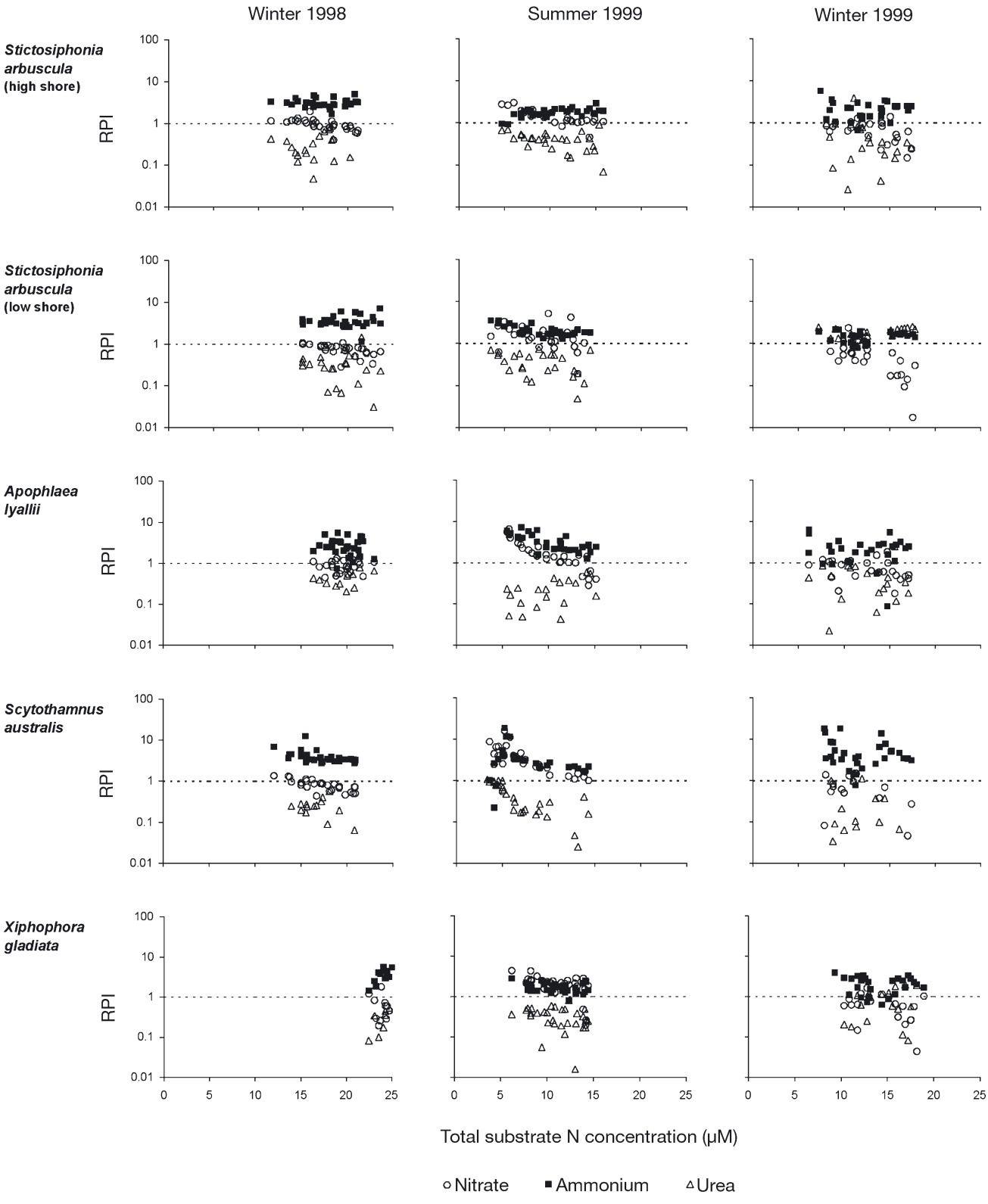

Fig. 7. *Stictosiphonia arbuscula*, *Apophlaea lyallii*, *Scytothamnus australis* and *Xiphophora gladiata*. Relative preference index (RPI) of nitrate, ammonium and urea on 3 sampling occasions. Data derived from low-concentration (5 µM) multiple-N-addition time-course experiments (negative values excluded). Total substrate N = NO_3^- + NH_4^+ + urea in seawater medium available for up take (including any background N present in seawater prior to N addition)

Table 2. *Stictosiphonia arbuscula*, *Apophlaea lyallii*, *Scytothamnus australis* and *Xiphophora gladiata*. Total amount of nitrate, ammonium and urea (in µg [g dry wt]$^{-1}$) taken up by 4 intertidal seaweeds over a 12 h tidal cycle. Data for each N source are mean values (n = 6) of 3 h multiple-N-addition time-course uptake experiments at low (5 µM) initial concentrations

Species	Approx. submersion time (h)	Winter (July) 1998				Summer (January) 1999				Winter (July) 1999			
		NO_3^-	NH_4^+	Urea	Total N	NO_3^-	NH_4^+	Urea	Total N	NO_3^-	NH_4^+	Urea	Total N
Stictosiphonia arbuscula (high shore)	2	127.5	104.7	43.9	276	49.1	60.6	64.3	174	51.8	76.9	26.0	155
Apophlaea lyallii	2.75	36.6	31.2	15.0	83	14.7	20.2	10.9	46	15.0	17.6	8.4	41
Stictosiphonia arbuscula (low shore)	3.5	234.2	111.9	76.1	422	45.3	93.6	105.7	245	72.9	72.0	66.0	211
Scytothamnus australis	5	110.6	88.9	24.8	224	19.7	28.9	65.2	114	36.7	48.4	17.4	102
Xiphophora gladiata	10	94.1	120.8	26.8	242	15.9	58.4	112.9	187	19.5	49.1	16.8	85

order of $NH_4^+ > NO_3^- >$ urea, while in summer the order was $NH_4^+ = NO_3^- >$ urea.

Total N acquisition over tidal cycle

The total amount of each N form that each species could take up over a 12 h tidal cycle during each season is shown in Table 2. Amounts of each N form, in µg g dry wt^{-1}, have been summed to give an estimate of total N acquisition possible in a single tide, assuming uptake to be equal in light and dark.

During winter 1998, low-shore *Stictosiphonia arbuscula* had the potential to take up the greatest amount of N of all species, followed by high-shore populations of the same species (Table 2). The brown algae *Scytothamnus australis* and *Xiphophora gladiata* took up similar amounts of N, while estimates for *Apophlaea lyallii*, which grows in the *S. arbuscula* zone of the high intertidal, were 2.5 to 5 times less N than all other species.

In summer 1999, the higher uptake rates of dinitrogen urea indicated that this contributed the greatest amount of total N during a single tide for all species except *Apophlaea lyallii*. Furthermore, in contrast to the previous winter, NO_3^- accounted for the smallest fraction of total N acquired by most species. When the amounts of all N forms were combined, low-shore *Stictosiphonia arbuscula* again seemed able to take up considerably more N than all other species in a 12 h tidal cycle. *Xiphophora gladiata* and high-shore *S. arbuscula* took up similar intermediate amounts of N, while *A. lyallii* was estimated to take up 2.5 to 5 times less N than the other species. In winter 1999, the pattern among species in regard to their estimated ability to acquire N over a tidal cycle was similar to the trend in the previous winter. However, total N was lower for all species and was closer to estimates during summer 1999.

DISCUSSION

Ambient and experimental N concentrations

Nitrate concentrations in surface waters at Brighton Beach were characteristically high in winter, but overall were low compared to some northern hemisphere regions of the Pacific Ocean where levels may reach up to 30 µM (Wheeler & Srivastava 1984, Germann et al. 1987, Whitney et al. 1998). Although NO_3^- levels at Brighton Beach are fairly low, concentrations were never below detection limits, as has been observed during spring/summer in northern hemisphere regions such as the NW Pacific (Germann et al. 1987, Thomas & Harrison 1987, Thomas et al. 1987a) and southern California (Wheeler & North 1981, Zimmerman & Kremer 1986). In New Zealand, seawater NO_3^- concentrations and seasonal fluctuations at Brighton Beach were similar to those reported for 2 nearby locations — Kuri Bush, 12 km SW of Brighton (Brown et al. 1990) and Aquarium Point in Otago Harbour, 28 km NE (Brown et al. 1997) — and for Mokohinau Islands in NE New Zealand (Taylor et al. 1998), indicating that Brighton Beach is not locally or regionally unique in its low seawater NO_3^-. Seawater NH_4^+ concentrations at Brighton Beach were generally low throughout the year and showed no seasonal pattern; this is consistent with reports from coastal waters of New Zealand (Brown et al. 1990, 1997, Gillanders & Brown 1994a, Taylor et al. 1998) and temperate regions elsewhere (Chapman & Craigie 1977, Asare & Harlin 1983, Probyn & Chapman 1983).

Our experimental N concentrations were somewhat higher than ambient seawater concentrations, particularly for NH_4^+. Although our experimental low NO_3^- concentration (5 µM) was similar to naturally occurring winter levels at Brighton Beach, it is important to note that the uptake rates determined in this study do not represent uptake potential under natural conditions. Nitrogen uptake rates are usually enhanced at higher

N concentrations (eg. Thomas & Harrison 1985), and therefore our results are not intended to represent uptake rates per se for each species; rather, our intention is to demonstrate the effects of zonation on N dynamics and their consistency or otherwise at different N concentrations.

N status of intertidal algae

Ammonium is considered toxic at high internal concentrations and feedback inhibition of uptake may prevent the accumulation of sizeable internal pools (McGlathery et al. 1996). Ammonium reserves in the macroalgae *Ulva rigida* and *Chaetomorpha linum* are small and considered of little importance as storage pools (Fujita et al. 1988, McGlathery et al. 1996). In the present study, however, all seaweeds accumulated internal NH_4^+ pools that were generally somewhat larger than NO_3^- pools. The considerable internal NH_4^+ in the Brighton seaweeds is also unusual, since NH_4^+ pools are generally considered to be transient due to fast turnover rates — of the order of hours during N-starvation of *C. linum* (McGlathery et al. 1996)— although large NH_4^+ pools have also been reported for *Ulva fenestrata* (~25 µmol N g dry wt^{-1}) and *Gracilaria pacifica* (~75 µmol N g dry wt^{-1}) (Naldi & Wheeler 1999). The accumulation of NH_4^+ pools in the seaweeds examined in the current study could have resulted from an imbalance between uptake and assimilation into other forms of soluble N (Fujita et al. 1988) or the assimilation of NO_3^- into NH_4^+. Alternatively, as it is energetically 'cheaper' to use NH_4^+ than NO_3^- (Syrett 1962), the presence of larger NH_4^+ pools may have resulted from the preferential uptake of this N form by the species in this study.

Intracellular NO_3^- pools vary on a seasonal basis (Chapman & Craigie 1977, Asare & Harlin 1983) in response to ambient NO_3^- availability, and accumulate during periods of low N demand for growth, typically in winter. However, in this study, only *Stictosiphonia arbuscula* populations showed increased internal NO_3^- storage during winter, when seawater NO_3^- levels were maximal, although a lag between peak seawater NO_3^- and tissue NO_3^- was evident. Asare & Harlin (1983) found a similar response to seawater NO_3^- for the intertidal browns *Fucus vesiculosus*, *Ascophyllum nodosum* and *Laminaria saccharina*, and the red alga *Chondrus crispus*, although maximum tissue NO_3^- values for these species were considerably higher (10 to 80 µmol g dry wt^{-1}) than those reported here. A possible reason for the relatively low NO_3^- pools in this study may be that nitrate reductase (NR) remains active in the seaweeds throughout the year, resulting in NO_3^- utilisation rather than storage. This is supported by the similar NO_3^- uptake rates over time for all species and suggests that NR activity remains fairly constant so as to prevent the accumulation of NO_3^- pools from inhibiting further uptake. High NR activity may be the seaweeds' response to lack of an N supply during tidal emersion, i.e. an optimisation of N sources during submersion (Thomas et al. 1987a). Alternatively, N storage may occur as N compounds such as amino acids, proteins, chlorophyll and accessory pigments (Gagné et al. 1982, Ekman et al. 1989, Fredriksen & Rueness 1989, Naldi & Wheeler 1999) rather than as soluble vacuolar/cytoplasmic NO_3^-.

The ability of macroalgae to store N as a variety of compounds suggests that total N may be a better indicator of N status. Hanisak (1979b, 1983) and O'Brien & Wheeler (1987) proposed that a tissue N of ca. 2% of dry weight is the critical N concentration for seaweeds, below which growth would be N-limited. Lapointe & Duke (1984), however, argued that a seaweed could not be assigned a single critical N concentration since the N requirement for growth and photosynthesis are affected by light. The brown algae *Scytothamnus australis* and *Xiphophora gladiata* rarely had tissue N above 2%, which indicates either N-limitation or lower N requirements. In contrast, the red algae *Stictosiphonia arbuscula* and *Apophlaea lyallii* usually had tissue N content above 2%, with a maximum of 5% in the former species. This high tissue N content may reflect a high N demand or critical N content required to sustain growth (Fujita et al. 1988, Pedersen & Borum 1996). Although variations in tissue N also reflect phylogenetic differences in the ability to store excess N, accumulation of tissue N in all species in this study generally began in mid-winter (July) and continued through to September, despite seawater N having declined steadily following the period of highest concentrations from April to July. In other studies, a lag between peak seawater N and peak tissue N was either not evident or not as pronounced as that described here (Asare & Harlin 1983, Wheeler & Björnsäter 1992, Pedersen & Borum 1996). The asynchrony between peak seawater N and peak tissue N observed in this study suggests that storage is uncoupled from supply, and that species are well-adapted to maximising available N during periods of lower N demand.

The nutrient status of macroalgae can also be inferred from their C:N ratio, with values above the proposed critical limit of 10 (D'Elia & DeBoer 1978, Harrison & Druehl 1982) indicating N-limitation and values below suggesting N storage. Among all species investigated in this study, there was an inverse relationship between shore position and C:N ratio, although this may be partly attributable to phylogenetic differences in N storage and utilisation, as brown algae tend to have higher C:N ratios than red algae (Niell 1976, La-

pointe 1989). Nevertheless, this inverse relationship is interesting; Thomas et al. (1987b) reported an opposite trend between C:N ratios and shore position of 5 intertidal seaweeds from 3 algal divisions in the Pacific northwest. Furthermore, the low C:N ratios (8 to 15) of *Stictosiphonia arbuscula* populations are comparable to those for *Gracilaria pacifica* growing at different intertidal locations (Thomas et al. 1987a) and the 9 intertidal red algae included in Neill's (1976) survey. This suggests that *S. arbuscula*, along with other intertidal seaweeds, may not always become N-limited despite long periods of tidal emersion.

Nitrogen uptake is strongly influenced by the N status of seaweeds (e.g. D'Elia & DeBoer 1978, Hanisak 1983, O'Brien & Wheeler 1987), with higher uptake rates usually observed in more N-deficient algae. Based on C:N ratios and tissue N content, all species in this study showed a greater degree of N-limitation in summer, yet surprisingly this did not result in enhancement of uptake rates. Conversely, uptake rates were highest during the first winter experiments when algae were more N-replete. Furthermore, *Stictosiphonia arbuscula* populations had a higher N content and storage pools as well as the highest N uptake rates, indicating that feedback inhibition of uptake did not occur. This suggests that the control that internal N content exerts over uptake rates may not be as generic as previously thought, and that the benefits of maintaining high N uptake capability during periods of low N availability (i.e. summer) may outweigh any costs (Cochlan & Harrison 1991). This may be particularly relevant to intertidal seaweeds on the east coast of New Zealand's South Island, where seawater N levels are low but never fall to undetectable levels.

Temporal variation in uptake rate

In contrast to other studies, a lag phase or induction period in NO_3^- uptake was not evident in seaweeds examined in this study. It is possible that a lag phase in NO_3^- uptake existed, however, but had ended before the first sampling event (15 min after exposure to NO_3^-), or that the NO_3^- uptake mechanism is maintained in a fully active state, despite the metabolic cost of doing so (Thomas & Harrison 1985, Cochlan & Harrison 1991). A lag phase in NO_3^- uptake reportedly lasted for up to 20 min in low intertidal *Gracilaria pacifica* (Thomas & Harrison 1987) and up to 1 h in the subtidal *Laminaria groenlandica* (Harrison et al. 1986) and *Chaetomorpha linum* (McGlathery et al. 1996). For the Brighton Beach seaweeds, the absence of an induction period for NO_3^- uptake and the ability to maintain constant uptake rates over 90 min may represent an adaptation to maximising NO_3^- procurement in an environment where N supply is low and periodic, and where the supply of the other main inorganic N source, NH_4^+, is variable.

Rapid or 'surge' NH_4^+ uptake was evident for high- and low-shore *Stictosiphonia arbuscula* and for *Scytothamnus australis*. For these species, the lack of feedback inhibition of NH_4^+ uptake despite the presence of intracellular NH_4^+ may have resulted from assimilation rates close to maximum uptake rates, allowing maximum utilisation of pulses of high NH_4^+ concentration. Surge uptake of NH_4^+ occurred when exposed to NH_4^+ levels 2 to 12 times higher than maximum ambient seawater concentrations, demonstrating the ability of *S. arbuscula* and *S. australis* to exploit any high NH_4^+ pulses resulting from events such as upwelling, runoff or sewage discharge (Rosenberg et al. 1984, Zimmerman & Kremer 1984) that may not be detected in monthly sampling of ambient seawater concentrations.

Surge uptake of urea was not observed for any species in this study. Surge uptake of urea has only been demonstrated for 1 macroalga, *Chordaria flagelliformis* (Probyn & Chapman 1982, 1983), although enhanced urea uptake by phytoplankton can result from N-starvation (Rees & Syrett 1979, Cochlan & Harrison 1991) and surge uptake may occur during the first 2 min of uptake (Antia et al. 1991). *C. flagelliformis* is a summer annual brown seaweed, with finely branched construction, that supports colonies of the amphipod *Gammarellus angulosus* and littorinid snails *Littorina vincta* (Probyn & Chapman 1983). The ability for high transient urea uptake appears to be an adaptive response to utilising urea excreted by its associated fauna during periods when seawater N availability in low (Probyn & Chapman 1983). Since surge uptake was not evident in the Brighton Beach seaweeds, it may be that surge uptake of urea is characteristic of opportunistic or ephemeral species only, although further investigation is required to support this.

Single versus multiple nutrient additions

Ammonium at least partially inhibits the uptake of NO_3^- in some macroalgae (D'Elia & DeBoer 1978, Haines & Wheeler 1978, Thomas et al. 1985, Thomas & Harrison 1985, Brenchley et al. 1997), but not in others (Harlin & Craigie 1978, Topinka 1978, Wheeler & Srivastava 1984). In this study, *Apophlaea lyallii* was the only species in which NO_3^- V_{0-15} was significantly reduced by the presence of NH_4^+, but this inhibition may have been concentration-dependent. Since NH_4^+ levels below 5 µM did not suppress NO_3^- uptake by *Gracilaria foliifera* (D'Elia & DeBoer

1978), it is possible that at lower naturally-occurring NH_4^+ concentrations inhibition of NO_3^- uptake may disappear in *A. lyallii*. There was no evidence of inhibition of urea uptake for any seaweed investigated in this study, even though the presence of inorganic N (NH_4^+, NO_3^-) in the culture medium is known to inhibit urea uptake by certain phytoplankton (Antia et al. 1991, Tamminen & Irmisch 1996, Peers et al. 2000).

It is worth noting that all species were capable of simultaneous uptake of all 3 N sources (NO_3^-, NH_4^+, urea) despite partial suppression of NO_3^- uptake by *Apophlaea lyallii*. Evidence to date suggests that many intertidal seaweeds are capable of simultaneous uptake of more than one N form (Topinka 1978, Thomas et al. 1985, Thomas & Harrison 1987, Thomas et al. 1987a, Brenchley et al. 1997), including the intertidal estuarine alga *Gracilaria chilensis* from New Zealand (Pillai 1992). Although the total number of seaweeds examined so far is relatively small, it appears that the ability for simultaneous uptake of multiple N forms may be a widespread ecological adaptation among intertidal seaweeds.

Relative importance of each N form

Preference for particular N sources can be inferred from uptake rates and/or the relative contribution of each source to the N nutrition of a seaweed. Among phytoplankton, the reduced N forms (i.e. NH_4^+, urea) are typically taken up in preference to NO_3^- (Butler et al. 1979, Tamminen & Irmisch 1996 and references therein). Among macroalgae, NH_4^+ is usually taken up in preference to other N forms (Lobban & Harrison 1994). In our study, the order of preference in terms of uptake rates as indicated by the RPI was $NH_4^+ > NO_3^-$ > urea. The apparent preference for NH_4^+ shown by the seaweeds in this study may be the result of diffusive uptake (Taylor et al. 1998). Alternatively, the energetics of uptake and assimilation of NH_4^+ may dictate the preference for this N form, since NH_4^+ is already in a reduced state and can be directly incorporated into amino acids (Lobban & Harrison 1994).

It should be noted, however, that the RPI does not infer physiological preference, but simply which N source is taken up at a faster rate relative to availability. If the contribution of each N form to total N acquisition is used instead to infer preference, NO_3^- and NH_4^+ were utilised equally in winter 1998 by all species except the low-intertidal *Xiphophora gladiata*, which showed a preference for NH_4^+. The following winter, NH_4^+ was the preferred source for all species. In summer, urea contribution exceeded that of NO_3^- for most species, despite low rates of uptake. These findings are in direct contrast to the assumption by Fujita et al. (1989) that dissolved organic N sources, such as urea, are not important N sources for rocky intertidal macroalgae.

The importance of urea as an N source for macroalgae has been largely overlooked, despite the well-documented evidence that many microalgal species readily utilise urea (e.g. Carpenter et al. 1972, Bekheet & Syrett 1979, Price & Harrison 1988). For macroalgae, the current knowledge of urea utilisation is restricted to the ability of urea to sustain the growth of a number of seaweeds (Nasr et al. 1968, Mohsen et al. 1974, Adamich et al. 1975, Hanisak 1979a, Probyn & Chapman 1983, Thomas et al. 1985, Mairh et al. 1999, Navarro-Angulo & Robledo 1999, Lotze & Schramm 2000) and the demonstrated uptake of urea by *Chordaria flagelliformis* (Probyn & Chapman 1982, 1983) but not by *Ecklonia maxima* (Probyn & McQuaid 1985). From the present study, it appears that urea is a seasonally important N source for 4 intertidal seaweeds in New Zealand, with the greatest contribution to overall N nutrition occurring in summer when ambient seawater N concentrations are minimal.

Influence of zonation on N uptake

In studies examining the nutrient uptake abilities of intertidal seaweeds in relation to zonation, no obvious correlation between N or P uptake rates and shore position has been noted (Thomas & Harrison 1987, Hurd & Dring 1990) in a range of seaweeds from different algal divisions or with different morphologies. However, Hurd & Dring (1990) found higher PO_4^{3-} uptake rates with higher shore position for 3 *Fucus* species with similar surface area:volume (SA:V) ratios and morphologies. In contrast, the seaweeds in this study generally showed higher rates of N uptake with decreasing submersion times, regardless of differences in taxonomy, morphology or N status. Furthermore, the amount of N that could be acquired in a tidal cycle was highest for the high-intertidal *Stictosiphonia arbuscula*, despite comparatively short submersion times. *Apophlaea lyallii* was an exception to this generalised trend, as it had the lowest N acquisition of all species examined. *A. lyallii* has been observed to grow slowly in the field; this, coupled with its low N uptake rates, suggests that this species has a low N requirement that allows it to survive in the upper intertidal. The other faster-growing species overcome potential N-limitation imposed by an intermittent N supply through a capacity for proportionally faster N uptake (Davison & Pearson 1996, Pedersen & Borum 1997). It is important to note, however, that uptake experiments were con-

ducted using fully-hydrated seaweeds, so the amounts of N acquired are likely to be overestimates; nevertheless, they are still useful for comparative purposes. The results of this study indicate that the degree of N limitation in intertidal seaweeds is not always correlated to shore position.

Conclusions

This is the first comprehensive study of the N ecophysiology of intertidal seaweeds across a whole shore system. We have demonstrated that the seaweeds examined employ a range of strategies to overcome potential N limitation, namely simultaneous uptake of all 3 N forms (NO_3^-, NH_4^+, urea), absence of a lag or induction phase prior to N uptake a predominant lack of inhibition of N uptake in the presence of other N forms, a lack of feedback inhibition of N uptake in the presence of internal N pools, and the ability to accumulate sizeable N stores. Furthermore, the degree of N limitation (as indicated by tissue N content and C:N ratios) and the estimated N acquisition on a single tide were both inversely correlated with shore position, except in the case of *Apophlaea lyallii*. Although some differences in N uptake and storage may be attributable to functional form, morphology or SA:*V* ratios, mass-specific NO_3^- uptake has been shown to scale to SA:*V* ratios for these seaweeds (T. A. V. Rees, J. C. Philips, B. C. Dobson, M. Bijl, B. Morelissen, C. L. Hurd unpubl.). The prevailing evidence of the current study is that intertidal seaweeds in southeastern New Zealand are not necessarily N-limited and are adapted to maximising N procurement from an intermittent N supply.

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