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PHYSIOLOGICAL RESPONSES OF *ECKLONIA RADIATA* (LAMINARIALES) TO A LATITUDINAL GRADIENT IN OCEAN TEMPERATURE[1]

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We tested the ability of sporophytes of a small kelp, *Ecklonia radiata* (C. Agardh) J. Agardh, to adjust their photosynthesis, respiration, and cellular processes to increasingly warm ocean climates along a latitudinal gradient in ocean temperature (~4°C). Tissue concentrations of pigment and nutrients decreased with increasing ocean temperature. Concurrently, a number of gradual changes in the metabolic balance of *E. radiata* took place along the latitudinal gradient. Warm-acclimatized kelps had 50% lower photosynthetic rates and 90% lower respiration rates at the optimum temperature than did cool-acclimatized kelps. A reduction in temperature sensitivity was also observed as a reduction in Q_{10}-values from cool- to warm-acclimatized kelps for gross photosynthesis (Q_{10}: 3.35 to 1.45) and respiration (Q_{10}: 3.82 to 1.65). Respiration rates were more sensitive to increasing experimental temperatures (10% higher Q_{10}-values) than photosynthesis and had a higher optimum temperature, irrespective of sampling location. To maintain a positive carbon balance, *E. radiata* increased the critical light demand (E_c) exponentially with increasing experimental temperature. The temperature dependency of E_c was, however, weakened with increasing ocean temperature, such that the critical light demand was relaxed in kelp acclimated to higher ocean temperatures. Nevertheless, calculations of critical depth limits suggested that direct effects of future temperature increases are unlikely to be as strong as effects of reduced water clarity, another globally increasing problem in coastal areas.

Key index words: **acclimatization; depth limit; *Ecklonia radiata*; nutrients; photosynthesis; pigments; respiration; temperature**

***Abbreviations:* E, E_c, E_k: irradiance, light compensation point, onset of light saturation; GP, GP_{max}: gross photosynthesis, maximum gross photosynthesis; NP, NP_{max}: net photosynthesis, maximum net photosynthesis; R_d, dark respiration; T_{opt}, optimum temperature**

Temperature is a major determinant of the broad-scale distribution of organisms on land and in aquatic habitats (McQuaid and Branch 1984, Wang et al. 2003, Haidekker and Hering 2007). Tolerances to high temperatures (summer maxima) define the biogeographical boundaries of macroalgae, the main foundation species on temperate reefs (van den Hoek 1982, Lüning 1984). The temperature gradients that cause geographic changes in abundance and composition of macroalgal communities are often associated with continental-scale features in oceanography (Leliaert et al. 2000, Wernberg et al. 2003a). Many environmental gradients covary at such broad spatial scales, and it is therefore difficult to attribute causality to temperature (Kain 1989). However, the critical role of temperature in determining algal species distributions is also evident on much smaller spatial scales, such as where the cooling outlets of power plants have altered local temperature conditions (Schiel et al. 2004). Nevertheless, most species of macroalgae have distribution ranges that span temperature gradients of several degrees Celsius, and they are capable of growing and reproducing over a wide range of temperatures (Bolton and Lüning 1982, Egan et al. 1989). This broad tolerance suggests that many macroalgae have an ability to adjust and optimize carbon fixation (photosynthesis) and biosynthesis (respiration) to the prevailing temperature conditions.

Algae generally employ a range of mechanisms to optimize the photosynthesis-irradiance relationship at different temperatures. These mechanisms include alterations in pigment content and the ratio of photosynthetic pigments relative to photoprotective pigments (Falkowski and Laroche 1991), cellular carboxylation activity (Davison 1991), and membrane fluidity and electron chain transfer (Raison et al. 1980). In microalgae, cellular acclimation to long-term increases in growth temperature significantly changes the short-term temperature response of metabolic rates (Staehr and Birkeland 2006) through: (1) lower rates of respiration and photosynthesis at low incubation temperatures, (2)

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weaker response of respiration and photosynthesis to increasing experimental temperature, (3) higher optimum temperatures, and (4) better performance of respiration and photosynthesis at high temperatures. Similar responses seem likely for macroalgae. Furthermore, as optimum rates of photosynthesis are generally measured at lower temperatures than those for respiration, this implies a faster rise in demands than in new production of ATP and carbohydrates at high temperatures (Raven and Geider 1988). As a result, the irradiance needed for gross photosynthesis to compensate the oxygen demands for respiration (E_c) increases with increasing temperature (Maegawa et al. 1987, 1988). Consequently, unless a significant acclimatization takes place, such a relationship essentially implies a reduction in the potential depth limit of the macroalgae with increasing temperature.

This study examined differences in temperature dependency of metabolic rates of the sporophyte stage of *Ecklonia radiata*, a widely dominant kelp (order Laminariales) in Australasia, along a latitudinal gradient in ocean temperature. The aim was to investigate the importance of temperature in regulating metabolism in mature kelp sporophytes, and the degree of physiological adjustment to chronic exposure to warmer waters. Investigations were undertaken within a nutrient poor, clear water, coastal environment covering a latitudinal gradient in annual temperatures of 2°C–4°C (Pearce 1991, Fig. 1). This unique environment allowed us to test the hypothesis that growth at different temperatures causes a significant thermal acclimatization in the photosynthetic performance of *E. radiata*, and to identify the underlying mechanisms involved in thermal acclimatization of *E. radiata*. Moreover, empirical models of light compensation levels as a function of temperature were used to evaluate the potential effects of increasing ocean temperature on the depth distribution of this important kelp species.

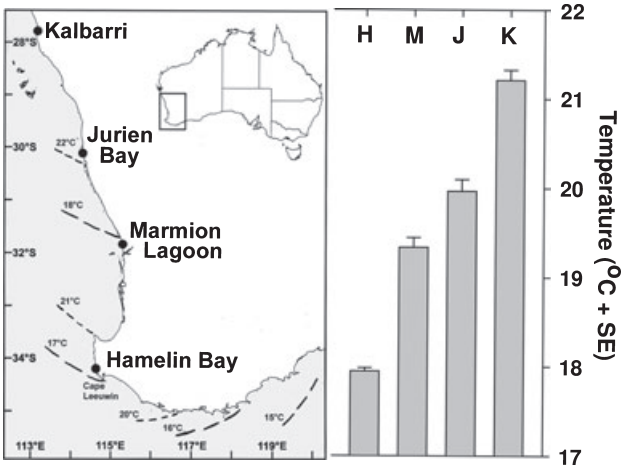

FIG. 1. Left panel: locations of study along the southwest coast of Australia with summer (short dash) and winter (long dash) isotherms (after Pearce 1991). Right panel: temperature at the reef surface, at the time of sampling kelp tissues for this study (mean + SE, $n = 4$; all samples were collected within 2 weeks in December 2005). H: Hamelin Bay, M: Marmion Lagoon, J: Jurien Bay, K: Kalbarri.

MATERIALS AND METHODS

E. radiata is a small kelp (~2 m, Wernberg et al. 2003b) that often dominates the macroalgal flora of temperate rocky reefs in the Southern Hemisphere (Wernberg et al. 2003a, Goodsell et al. 2004). In terms of size and canopy structure, *E. radiata* is similar to common kelps in the Northern Hemisphere (e.g., *Laminaria digitata* and *Eisenia arbora*), and it forms dense kelp beds of equally high productivity (Larkum 1986, Fairhead and Cheshire 2004b). *E. radiata* occurs in waters ranging in temperature from ~8°C to 24°C (Bolton and Anderson 1994). It has been characterized as a warm-temperate species, with a broad optimal temperature range for growth and reproduction of 12°C to 20°C, and decreasing performance outside this range (Novaczek 1984). The most northerly location of this study extends toward the warmer limit of the species' distribution.

Study sites and collection of algal material. The coast of central Western Australia is a transition zone between tropical coral reefs and algal-dominated temperate reefs. The ocean climate along this coast is determined by the southward flowing Leeuwin Current, which maintains a gradual transition in water temperatures (Pearce 1991, Lourey et al. 2006), and nutrient poor conditions throughout the entire year (nitrate below 0.5 μM, phosphate below 0.2 μM; Lourey et al. 2006). These conditions contrast with the western margins of other continents in the Southern Hemisphere (Pearce 1991), where changes in ocean temperature usually are abrupt and are associated with large changes in nutrient concentrations as a consequence of localized upwelling (Dayton et al. 1999). The unique oceanographic conditions of the Australian west coast, therefore, offer an opportunity to study climate-related effects of temperature unconfounded by effects of variable nutrient levels.

Algae and water were collected from subtidal rocky reefs (10–12 m depth) at four locations, each separated by ~2° latitude (~350 km coastal distance) (Fig. 1): Hamelin Bay (34°13.3 S, 115°01.7 E), Marmion Lagoon (31°49.4 S, 115°44.0 E), Jurien Bay (30°17.3 S, 115°02.5 E), and (Kalbarri, 27°42.5 S, 115°09.5 E). Depending on the season and local weather conditions, water temperatures range between 18°C and 25°C along this stretch of coast, but, at all times, there is a gradual temperature increase of 2°C–4°C from Hamelin Bay to Kalbarri (Pearce 1991, Lourey et al. 2006). Water temperature was measured ~5 cm above each reef at the same time as kelp sporophytes were collected for analysis (Onset tidbit logger logging every 5 min for a period of 30 min; WTA32-5 + 37, OneTemp, Adelaide, Australia). Four replicate kelps were collected from each location, one mature sporophyte from each of four reefs separated by >1 km. The kelps were kept in the dark, in aerated seawater, at ambient seawater temperature until measurements of oxygen exchange. Water was sampled for analysis of ammonia, nitrate, and phosphate. These samples were collected in clean syringes by drawing water from immediately above the kelp canopy. Water was filtered through a 0.45 μm filter and stored on ice and frozen before analysis according to Johnson (1982, 1983) and Switala (1993). All collections and measurements were done within 2 weeks in December 2005 (early summer) to ensure minimal influence of any temporal variation on the latitudinal pattern. Moreover, locations were not sampled in sequence (order: Hamelin Bay, Jurien Bay, Kalbarri, Marmion), and the weather remained consistent throughout the coastline over the entire 2-week sampling period.

Measurement of photosynthesis and respiration. Photosynthesis and respiration were determined from oxygen exchange of epiphyte-free sections (185 ± 58 mg dry weight [dwt], mean ± SD) of laterals from the lower third of the thallus. Measurements were made within a few hours of collection using a temperature-controlled incubation unit, which allowed simultaneous measurements on algal material exposed to six different light intensities (0, 13, 25, 50, 250, and 530 μmol photons $\cdot$ $m^{-2} \cdot s^{-1}$) covering light-limited to light-saturated photosynthesis (cf. Binzer et al. 2006). All algal material was cooled to 10°C and allowed a short acclimation period of ~30 min prior to measurements of photosynthesis and respiration successively at 10, 20, 25, and 30°C, corresponding to the likely range of contemporary and future (+5°C) temperatures encountered by kelps in the region. Changes in oxygen concentrations were measured using a Oxyguard Handy Polaris (OxyGuard International, Birkerød, Denmark) oxygen probe, at the beginning and at the end of each incubation period, which had a duration of ~90 min at 10°C and 30 min for each of the remaining temperatures. The electrode was calibrated regularly.

Tissue chl and nutrients. Chl and nutrient content were measured on tissue samples from each kelp. Algal material was stored at −20°C until freeze drying. Chl *a* and *c* were determined spectrophotometrically according to Jeffrey and Humphrey (1975). Organic carbon (C) and nitrogen (N) was measured using a Carlo-Erba EA-1108 CHN analyzer (Carlo Erba Instruments, Cambridge, UK). Organic phosphorus (P) was determined spectrophotometrically after wet oxidation with boiling H_2SO_4 (Strickland and Parsons 1968).

Parameter calculations. The photosynthetic efficiency at low irradiance (α: mg $O_2 \cdot g^{-1}$ dwt $\cdot$ mol^{-1} photons $\cdot$ m^{-2}) was determined as the initial linear slope between photosynthesis and irradiance at light limitation. The photosynthetic efficiency was used to calculate the light compensation point ($E_c = R_d/\alpha$), at which net oxygen exchange is zero. The maximum rate of gross photosynthesis (GP_{max}) was determined by nonlinear regression of net photosynthesis (*NP*) normalized to dry weight (dwt) on irradiance (*E*) according to a saturating exponential model (Webb et al. 1974) as

$$NP = GP_{max}[1 - \exp(-\alpha E/GP_{max})] - R_d \quad (1)$$

where GP_{max} is the calculated gross light-saturated photosynthetic rate and R_d is the dark respiration rate at $E = 0$ (positive value). Additional parameters calculated were the net maximum rate of photosynthesis, NP_{max} defined as $GP_{max} - R_d$, and the onset of light saturation, E_k, estimated as GP_{max}/α.

The temperature-responses of NP_{max}, GP_{max}, and R_d were fitted to the equations of Johnson et al. (1974) slightly modified by Staehr and Birkeland (2006). For each specimen and growth temperature, optimum temperature (T_{opt}) was determined for NP_{max}, R_d, and GP_{max} by a Gaussian curve fit to each temperature-response curve. Temperature-response curves were furthermore used to calculate the highest rate of NP_{max}, R_d, and GP_{max} at the optimum temperature. Curve-fitting was done by nonlinear regression using the Gauss iterative method in SAS/STAT software package (SAS Institute Inc. 1994).

Assessment of possible differences in the temperature responses of GP_{max}, NP_{max}, and R_d at suboptimal temperatures (below T_{opt}) was based on Q_{10}-values calculated as

$$Q_{10} = (\text{Rate}_2/\text{Rate}_1)^{[10/(T2-T1)]} \quad (2)$$

where, Rate 1 and 2 are metabolic rates measured at temperatures *T*2 (high temperature) and *T*1 (low temperature), respectively. Since the choice of *T*1 and *T*2 had a systematic effect on the calculated Q_{10}-values, calculations were made for the temperature intervals 10°C to 20°C and 20°C to 25°C. The reported Q_{10}-values were eventually calculated as a mean of these two values. To assess whether growth temperature had a significant effect on the supraoptimal (above T_{opt}) responses of GP_{max}, NP_{max}, and R_d, we calculated the decrease in rate from T_{opt} to 30°C.

Effects of long-term acclimatization (growth) temperatures on physiological parameters and estimates of the fitted temperature-response curves were compared with a linear model one-way analysis of variance (ANOVA). In the linear model, in situ water temperature measured during sampling was used as a quantitative parameter to represent the gradual increase in water temperatures from Hamelin Bay to Kalbarri. This comparison allowed us to evaluate the extent to which the temperature gradient explains differences in physiological and metabolic response variables. All data had homogeneous variances (Levene's test, $P > 0.05$) and satisfied the assumptions of normality (Kolmogorov–Smirnov *D*-test, $P < 0.05$). All data analysis was performed using SAS/STAT (SAS Institute Inc. 1994).

Temperature effects on the critical depth. To evaluate the importance of the observed acclimatization for the performance of *E. radiata* under different temperature conditions, we calculated the critical depths for plants acclimatized to the coolest and warmest locations in this study (~4°C difference). Assuming the critical depth to be the depth at which average daily light intensity equals the light compensation level (E_c), we calculated the critical depth at water temperatures between 20°C and 30°C. This interval covers current summer temperatures at Hamelin Bay (~20°C) and Kalbarri (~25°C), and an increase in temperature of ~5°C, corresponding to the projection by 2090 according to the A2 scenario of the United Nation's climate panel (IPCC 2007), at both locations. Light intensities at depth were calculated as a function of surface PAR irradiance, depth, and light attenuation as:

$$E_{z\_daily} = E_{0\_daily} e^{-K_D z} \quad (3)$$

where, E_{z_daily} is the daily average intensity (μmol photons $\cdot$ $m^{-2} \cdot s^{-1}$) at depth *z* (m), K_D (m^{-1}) is the light attenuation coefficient, and E_{0_daily} is an estimate of the daily average PAR surface irradiance (μmol photons $\cdot$ $m^{-2} \cdot s^{-1}$) during midsummer, calculated as:

$$E_{0\_daily} = \int_{Sunrise}^{Sunset} E_{0\_max} \times \text{Sin}\left(\frac{\text{hours} \times \pi}{\text{daylength (h)}}\right) \quad (4)$$

where, daylength is 14 h, E_{0_max} is the daily maximum radiation assumed to be 2,000 μmol photons $\cdot$ $m^{-2} \cdot s^{-1}$ at noon, and hours is the time since sunrise. To compare the importance of increasing temperatures to decreasing water clarity, another pressure on coastal ecosystems presumed to increase in the future, critical depth limits were calculated for four different light attenuation conditions (K_D = 0.05, 0.1, 0.2, and 0.4 m^{-1}) representing the range of conditions recorded near Perth, the largest city in Western Australia (Thompson and Waite 2003).

RESULTS

Pigment and nutrient content. Significant changes occurred in the cellular content of pigments and nutrients from the southern cooler waters at Hamelin Bay to the northern warmer waters at Kalbarri (Tables 1 and 3). A general decrease was observed in cellular concentrations of chl *a*, but not in chl *c*, causing a significant reduction in the ratio

TABLE 1. Tissue characteristics of *Ecklonia radiata* sampled at four locations. Chl *a* and chl *c* ($\mu g \cdot g^{-1}$ dwt). Carbon (C), nitrogen (N), and phosphorus (P) ($mg \cdot g^{-1}$ dwt). Ratios of C, N, and P were calculated on a molar basis. Data are mean ± SE, $n = 4$.

	Hamelin Bay	Marmion Lagoon	Jurien Bay	Kalbarri
Chl *a*	2.61 ± 0.15	2.08 ± 0.31	1.81 ± 0.18	1.71 ± 0.19
Chl *c*	1.60 ± 0.05	1.46 ± 0.04	1.51 ± 0.05	1.42 ± 0.11
Chl *a*:chl *c*	1.64 ± 0.10	1.42 ± 0.20	1.20 ± 0.13	1.20 ± 0.10
C	282 ± 5	308 ± 8	303 ± 1	301 ± 10
N	12.9 ± 0.8	10.8 ± 0.1	8.1 ± 0.6	8.7 ± 0.4
P	2.43 ± 0.09	2.25 ± 0.25	1.76 ± 0.14	1.83 ± 0.20
C:N	25.7 ± 1.1	33.4 ± 1.2	44.1 ± 3.3	40.7 ± 2.0
C:P	302 ± 11	370 ± 49	453 ± 39	447 ± 72
N:P	11.9 ± 0.9	11.0 ± 1.1	10.6 ± 1.6	10.9 ± 1.5

dwt, dry weight.

of chl *a* to chl *c* from Hamelin Bay to Kalbarri. The latitudinal increase in ocean temperature had no effect on cellular carbon content, whereas a gradual decrease in nitrogen and phosphorus was observed (Tables 1 and 3). As a result, the ratios of carbon to nitrogen and phosphorus increased from south to north, indicating a gradual decrease in the nutritious value of the thallus with increasing growth temperature. Covariant nutrient availability could, however, bias this interpretation. However, our measurements of inorganic nutrients in water sampled just above the kelp canopy supported previous investigations (Lourey et al. 2006) with consistently low concentrations and insignificant differences in nutrients at the four locations: ammonia (0.67 ± 0.62 μM; mean ± SD; $P = 0.83$, one-way ANOVA), nitrate (0.4 ± 0.30 μM; $P = 0.14$), and phosphate (0.33 ± 0.15 μM; $P = 0.23$).

Temperature effects on the light-photosynthesis relationship. Photoinhibition was not observed in any of the experiments (Fig. 2), and the nonlinear P–E model fitted the data with an overall level of determination (r^2) of 0.97 ± 0.03 (mean ± SD, $n = 64$) for all of the measured P–E curves. Metabolic rates followed the same dependency of increasing experimental temperatures for all locations (Fig. 3). Higher temperatures caused a gradual increase in net photosynthesis (Fig. 3A) up to an optimum ~25°C, after which a decrease in photosynthesis occurred. Dark respiration increased gradually toward 30°C with no clear temperature optimum (Fig. 3B).

Net photosynthesis at suboptimal incubation temperatures (10°C and 20°C) was slightly higher for algae collected from the southern cooler waters (Fig. 3A). Optimum temperatures (T_{opt}) for maximum rates of net photosynthesis showed a small and nonsignificant increase ($P = 0.09$, one-way linear ANOVA) with increasing ocean temperatures. No differences in T_{opt} were determined for GP_{max} between locations (Tables 2 and 3), and 75% of the respiration samples were found to have a T_{opt} of 30°C, corresponding to the highest incubation temperature applied. T_{opt} was accordingly much higher for respiration (>30°C), compared to GP_{max} (26.1 ± 2.0°C) and NP_{max} (24.4 ± 1.6°C). Metabolic rates at T_{opt} decreased linearly with increasing ocean temperature, the effect being most clear for respiration (rates taken at 30°C) (Tables 2 and 3).

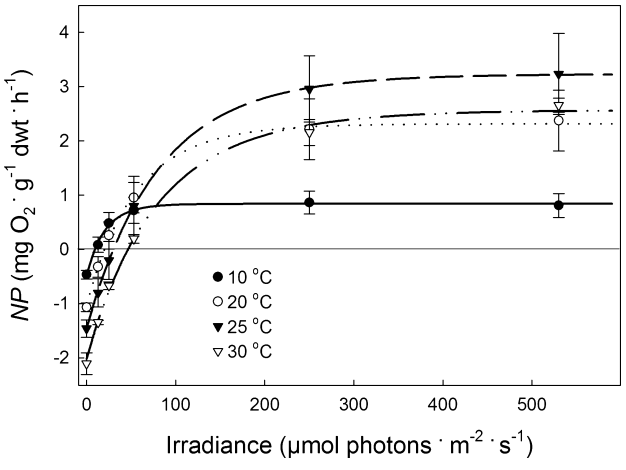

FIG. 2. Example of net photosynthesis (*NP*) versus irradiance relationships for kelp samples collected in Marmion Lagoon and exposed to different temperatures (mean ± SE, $n = 4$).

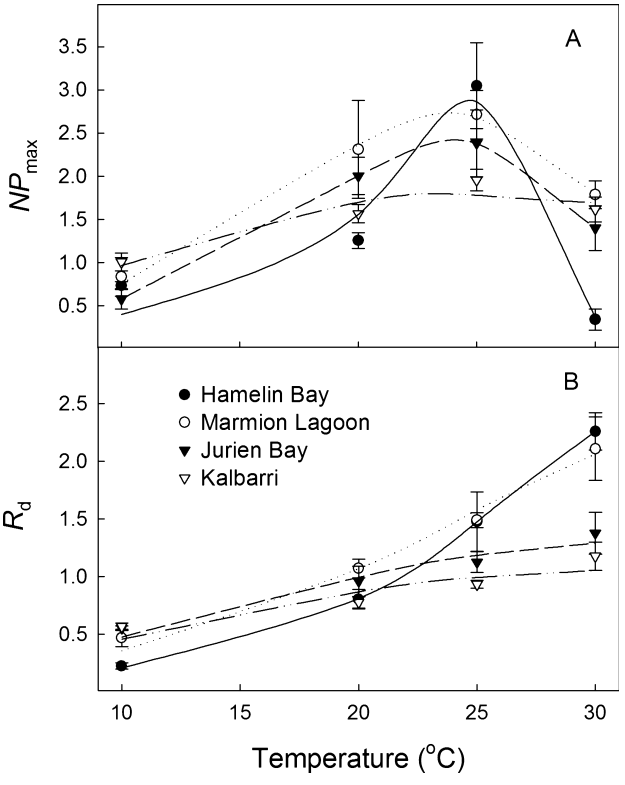

FIG. 3. Temperature dependency of (A) light saturated net photosynthesis, NP_{max}, and (B) dark respiration, R_d, in mg $O_2 \cdot g^{-1}$ dwt $\cdot h^{-1}$ (mean ± SE, $n = 4$).

TABLE 2. Parameter estimates from fitted temperature-response curves for gross and net photosynthesis and respiration of *Ecklonia radiata* sampled at four locations along a latitudinal temperature gradient. T_{opt} (°C) is the temperature at which maximum rates were found. Rates at T_{opt} were determined for the rate of net and gross light-saturated photosynthesis (NP_{max} and GP_{max}; mg $O_2 \cdot g^{-1}$ dwt $\cdot h^{-1}$). For dark respiration, R_d, the rate at T_{opt} is the rate measured at 30°C, as no optimum temperature was determined (nd = not determined). Reduction above T_{opt} is the change in rates (mg $O_2 \cdot g^{-1}$ dwt $\cdot h^{-1}$) from T_{opt} to rates at 30°C. No reduction occurred for dark respiration. Q_{10} is the relative increase in metabolism with a 10°C increase in experimental temperature. Data are mean ± SE, $n = 4$.

Parameter	Location	T_{opt}	Rate at T_{opt}	Reduction above T_{opt}	Q_{10}
NP_{max}	Hamelin Bay	23.6 ± 0.1	3.05 ± 0.50	2.71 ± 0.29	3.40 ± 0.23
	Marmion Lagoon	24.3 ± 0.2	2.71 ± 0.28	0.93 ± 0.06	2.48 ± 0.35
	Jurien Bay	24.1 ± 0.6	2.38 ± 0.39	0.98 ± 0.12	2.48 ± 0.26
	Kalbarri	25.5 ± 1.4	1.96 ± 0.13	0.34 ± 0.04	1.46 ± 0.06
R_d	Hamelin Bay	nd	2.26 ± 0.16	nd	3.82 ± 0.19
	Marmion Lagoon	nd	2.11 ± 0.28	nd	2.65 ± 0.14
	Jurien Bay	nd	1.38 ± 0.18	nd	1.91 ± 0.18
	Kalbarri	nd	1.18 ± 0.12	nd	1.65 ± 0.20
GP_{max}	Hamelin Bay	25.1 ± 0.2	4.43 ± 0.31	1.84 ± 0.17	3.35 ± 0.12
	Marmion Lagoon	26.9 ± 1.0	4.25 ± 0.26	0.37 ± 0.12	2.35 ± 0.29
	Jurien Bay	25.2 ± 1.1	3.46 ± 0.53	0.71 ± 0.23	2.11 ± 0.23
	Kalbarri	27.1 ± 1.3	3.04 ± 0.15	0.15 ± 0.11	1.45 ± 0.04

TABLE 3. Linear model one-way analysis of variance (ANOVA) on the effect of temperature on physiological conditions and on parameters describing the temperature dependence of NP_{max}, R_d, GP_{max}, and E_c (the light compensation level). In the linear model, in situ water temperatures at the time of sampling (see Fig. 1) were used to represent the gradual increase in annual water temperatures. Dependent variables are described in Table 2. Degrees of freedom = 3. The slope coefficient indicates the direction of the relationships, and r^2 represents the proportion of variability accounted for by increasing temperature. Significant effects ($P < 0.05$) are highlighted in bold.

Variable	Type II SS	F	Slope	P	r^2
Chl *a*	1.77	9.10	−0.29	**0.0093**	0.39
Chl *c*	0.055	2.92	−0.05	0.11	0.17
Chl *a*:chl *c*	0.45	5.83	−0.14	**0.03**	0.29
C	623	2.42	5.39	0.14	0.15
N	46.10	20.38	−1.47	**0.0005**	0.59
P	0.97	6.90	−0.21	**0.02**	0.33
C:N	582	18.72	5.21	**0.0007**	0.57
C:P	51,982	5.99	49.21	**0.0282**	0.30
N:P	2.42	0.38	−0.34	0.55	0.03
T_{opt} for NP_{max}	7.04	3.28	0.57	0.09	0.19
T_{opt} for GP_{max}	5.19	1.31	0.49	0.27	0.09
Rate at T_{opt} for NP_{max}	2.59	6.07	−0.35	**0.0273**	0.30
Rate at T_{opt} (30°C) for R_d	2.94	18.06	−0.37	**0.0008**	0.56
Rate at T_{opt} for GP_{max}	4.68	10.71	−0.47	**0.0056**	0.43
Q_{10} for NP_{max}	7.21	30.39	−0.58	**<0.0001**	0.68
Q_{10} for R_d	10.52	61.96	−0.70	**<0.0001**	0.82
Q_{10} for GP_{max}	7.36	52.53	−0.59	**<0.0001**	0.79
Reduction of NP_{max}	10.76	22.45	−0.71	**0.0003**	0.62
Reduction of GP_{max}	5.02	10.21	−0.48	**0.0065**	0.42
E_c slope	0.004	6.56	−0.015	**0.0237**	0.34
E_c intercept	113	4.10	2.45	0.06	0.24

The biomass-specific rates of metabolism were accordingly the highest for plants with a high content of N, P, and chl *a*, acclimatized to cooler temperatures.

Q_{10}-values for NP_{max}, respiration, and GP_{max} were the highest at Hamelin Bay and decreased significantly toward the warmer locations in the north (Tables 2 and 3). Metabolic rates in the cool-acclimatized kelps were more strongly stimulated by the short-term temperature experiments than plants acclimatized to a warmer ocean climate. Acclimatization to warmer conditions was furthermore seen in a less pronounced reduction in metabolic rates at supraoptimal temperatures (Tables 2 and 3), suggesting that warm-acclimatized kelps in the northern waters perform better at high (>25°C) temperatures.

The light level needed for *E. radiata* to maintain a positive net metabolism (E_c) increased exponentially with increasing incubation temperature at all locations (Fig. 4A). However, the temperature dependency of E_c, seen as the slope of the exponential curves, decreased significantly

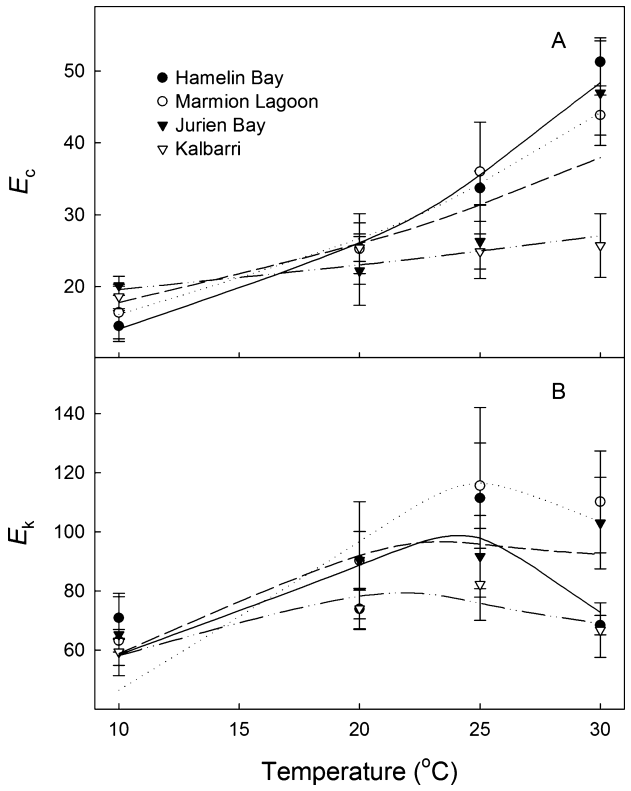

FIG. 4. (A) Light compensation level (μmol photons · m^{-2} · s^{-1}) and (B) light saturation level (μmol photons · m^{-2} · m^{-1}) as a function of experimental temperature (mean ± SE, n = 4).

from Hamelin Bay toward Kalbarri (Table 3). This finding implies that *E. radiata* has a considerable ability to optimize cellular metabolism to warmer conditions. Regardless of the growth temperature, the amount of light needed to saturate photosyntesis (E_k) increased with increasing incubation temperature (Fig. 4B). The increase was slightly lower for the warm-acclimatized kelp, signifying that acclimatization to growth temperature also compensated for the short-term effect of temperature on E_k.

Temperature effects on the critical depth. Calculations of critical depths for waters of different temperature (20°C–30°C) and attenuation (0.05–0.40 m^{-1}) clearly suggested that changes in water clarity are likely to impose more severe limitations on kelps than changes in temperature (Fig. 5). Regardless of water clarity, a temperature increase of 5°C caused a 10% decrease in the depth limit for cool-acclimatized (Hamelin Bay) kelps and a 2% decrease for warm-acclimatized (Kalbarri) kelps. In contrast, a change in light attenuation from clear waters with K_D = 0.1 m^{-1} to slightly more turbid waters with K_D = 0.2 m^{-1}, caused a decrease in depth limits by 50%. Adding a temperature rise from 25°C to 30°C would only cause a further reduction in depth limits of 5% for cool-acclimatized and 1% for warm-acclimatized kelp.

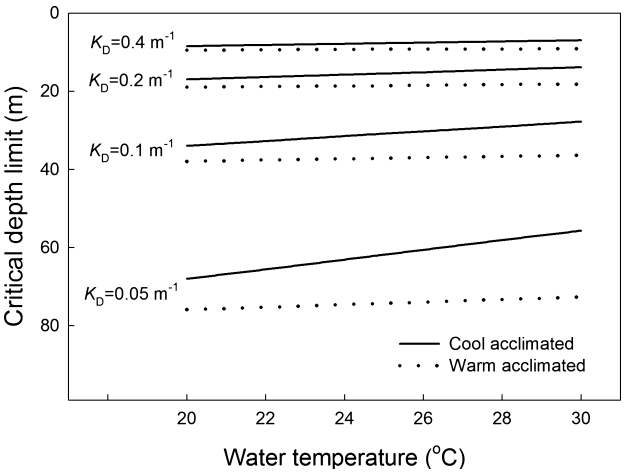

FIG. 5. Calculated critical depths in cool- and warm-acclimatized kelp for different temperatures and, under very clear (K_D = 0.05 m^{-1}) to turbid (K_D = 0.4 m^{-1}) attenuation conditions. The natural depth limit of *Ecklonia radiata* in southwestern Australia exceeds 50 m (T. Wernberg, personal observation).

DISCUSSION

This study showed that significant physiological differences exist in the sporophyte stage of a widely dominant perennial macroalga along a latitudinal gradient in ocean temperature. Recent investigations provide strong support for the idea that morphological and physiological differences in *E. radiata* across a wide range of environmental gradients represent phenotypic variability rather than genotypic divergence (Wernberg et al. 2003b, Fowler-Walker et al. 2006, Wing et al. 2007). Thus, assuming the gradual northward increase in chronic temperature exposure to be the main driver for the cellular and metabolic changes in *E. radiata*, increasing growth temperatures significantly decreased the cellular pigment and nutrient content of the kelp. At the same time, there was a significant decrease in the temperature sensitivity of metabolic rates (decreasing Q_{10}-values and better performance at supraoptimal temperatures) as well as a lowering of the light intensity needed for gross photosynthesis to compensate the oxygen demands for respiration (E_c). These results reflect an ability of *E. radiata* to optimize the metabolic balance to the prevailing temperature conditions by adjustment of the light capture and enzymatic processes to a lower level of activity.

Cellular changes. Cool-acclimatized kelps from the southern locations had higher concentration of chl *a* and organically bound nitrogen and phosphorus. The chl *a* to chl *c* ratio has previously been reported to vary seasonally in *E. radiata*, with the highest ratio during winter (Fairhead and Cheshire 2004a). These cellular changes are consistent with increasing enzyme (Raven and Geider 1988), lipid (Raison et al. 1980), and photosynthetic pigment (Falkowski

and Laroche 1991) concentrations to reduce constraints of low temperatures. Temperature has previously been associated with changes in tissue content of pigments, but these observations have typically been made in comparisons of seasonal changes, where concurrent changes in light availability also explained the changes in pigment concentrations (Campbell et al. 1999, Fairhead and Cheshire 2004b). Light is unlikely to be a major covariate of temperature in our study. The annual difference in global insolation between the latitudes of the northern and the southern locations is on the order of 10% more light at Kalbarri compared with Hamelin Bay (Kain 1989), with smaller differences (~8%) during summer (T. Wernberg unpublished data). Irradiance reaching the kelp bed furthermore depends on water transparency and depth. Measurements of water transparency during summer 2005 (T. Wernberg unpublished data) gave light extinction coefficients (0.16 ± 0.02; mean ± SD) similar to those reported by Thompson and Waite (2003) with no significant differences between the investigated locations ($P = 0.10$, one-way ANOVA). Kelp was sampled randomly at 10 to 12 m within locations. This variation alone causes a larger variability (25%) in the amount of light reaching the kelp beds than the variability caused by differences in latitude. Therefore, although the observed change in pigmentation with decreasing ocean temperatures is similar to the expected response of kelp acclimatized to light-limited conditions (Campbell et al. 1999), it seems most likely to be a result of long-term physiological acclimatization to different growth temperatures where slightly cooler conditions favor the synthesis of chl *a* in preference to antennae pigments and thus increase the chl *a* content.

Ambient concentration of nutrients, particularly inorganic nitrogen, could also have contributed to the observed patterns of pigment content and photosynthetic potential. Nitrogen limitation may reduce pigment synthesis and decrease carbon metabolism (Falkowski and Raven 1997). Productivity in the upper mixed surface waters (0–100 m depth) of the entire southwestern Australian shelf region is severely nitrogen limited all year round with a molar N:P ratio of typically <4 (Lourey et al. 2006). A low concentration of phytoplankton (chl *a* always below $1\ \mu g \cdot L^{-1}$), with no differences between Kalbarri and Hamlin Bay, is further evidence of low nutrient levels year-round (Lourey et al. 2006). Also, at the time of sampling, nutrients levels were low at all of the investigated locations. Considering the nutrient poor status of the region, increasing temperature-induced nutrient limitation would explain the negative relationship between the lower N and P content of *E. radiata* at increasingly warm locations.

Given the very similar light and nutrient conditions along the coast of southwestern Australia, it seems probable that the gradient of increasing ocean temperatures toward the north is responsible for the observed patterns in cellular nutrient and pigment composition. The most plausible explanation is a combination of two mechanisms where increasing temperatures relax the enzymatic requirements to meet the constraints of cool conditions and simultaneously raise the metabolic demand for nutrients and thereby the likelihood of nutrient limitation.

Temperature effects on metabolic performance. Cellular changes induced by differences in ocean temperature had profound effects on the short-term (1–2 h) temperature response of both photosynthetic and respiratory rates. Warm-acclimatized kelp with less nutritive tissue and lower pigment content generally had lower metabolic rates (most evident at T_{opt}) than did cool-acclimatized plants. Similar results have been reported for the kelp *Saccharina latissima* (Davison et al. 1991) and microalgae (Staehr and Birkeland 2006). Like microalgae (Staehr and Birkeland 2006), *E. radiata* acclimatized to warmer growth conditions by reducing the immediate response to increasing temperatures (low Q_{10}-values) and by exerting a higher tolerance to supraoptimal temperatures.

Optimum temperature for net photosynthesis (T_{opt}) for *E. radiata* was determined to be ~25°C. Sakanishi et al. (1989) observed similar values for *Ecklonia cava* in a seasonal study, where they also found slightly higher T_{opt}-values in summer than in winter. The lack of difference in optimum temperature between the locations in this study most likely reflects a search for relatively small changes in optimum temperatures using too few experimental temperatures. For all locations, however, we found lower optimum rates for photosynthesis than for respiration. Respiration was also more sensitive to increasing experimental temperatures (higher Q_{10}-values) than photosynthesis (Table 2), implying that increasing temperatures raise the respiratory energy demand faster than photosynthetic production of new ATP and carbohydrates. To maintain a positive carbon balance, *E. radiata* therefore increases the critical light demand (E_c) when temperatures increase. This phenomenon was previously suggested by Fairhead and Cheshire (2004a) to explain seasonal variation in E_c, but the exponential-type temperature dependency of E_c has not been documented before. More importantly, we determined that this temperature dependency was weakened with increasing growth temperature, such that the critical light demand is relaxed in kelp acclimatized to higher temperatures, and a similar response was observed for the light saturation level (E_k). Consequently, kelp from Hamelin Bay (cool) and Kalbarri (warm) were able to achieve similar rates of light-limited photosynthesis and similar E_c and E_k values at their respective growth temperature. Davison et al. (1991) also reported that acclimatization responses in *S. latissima* to growth temperature

compensated for the short-term effect on E_c and E_k in the laboratory.

Importance of temperature acclimation. Dense kelp beds of *E. radiata* exist below 50 m depth in Western Australia (Bolton and Anderson 1994, T. Wernberg personal observation), suggesting that the calculated depth limits are reasonable. Calculations of critical depth under different scenarios of water temperature, water clarity, and temperature acclimatization highlighted some important points. Most importantly, given the ability of *E. radiata* to acclimatize its carbon metabolism, increasing global temperatures in themselves are not likely to cause severe reductions in the critical depth limit. In comparison, deteriorating light conditions are a much greater threat to the depth limits of this ecologically important kelp. This relationship is important because indirect effects of increasing global temperatures include altered weather patterns and sea level rise (IPCC 2007), which in addition to other human activities are likely to cause increased terrestrial runoff of sediments and nutrients and ultimately diminish water clarity (Vitousek et al. 1997, Airoldi and Beck 2007). It is therefore likely that predicted changes in temperature and turbidity will have a negative synergistic effect on the current depth distribution of sporophytes or their spatial distribution around populated areas prone to reduced water quality (Airoldi and Beck 2007). Moreover, while our results are restricted to the direct effects of ocean temperature on mature sporophytes, it is possible that these are more stress-tolerant than the microscopic gametophytes (Matson and Edwards 2007). However, this possibility seems questionable given that gametophytes of *E. radiata* become fertile over a wider range of temperatures than the local range of ambient sea temperatures where the species is found (Bolton and Anderson 1994). Also, a negative correlation between growth rate of mature plants and sea temperatures >18°C–20°C (Kirkman 1984) supports the idea that low tolerance of sporophytes to high temperatures may limit the distribution of the species (Hatcher et al. 1987) rather than growth and fertility of the haploid phase. We find that mature *E. radiata* displays pronounced physiological and metabolic adjustments to rising temperatures. The key to understanding how organisms will respond to future environmental changes is understanding the extent to which acclimatization is important (Edmunds and Gates 2008). In a climate change perspective, our study shows that for kelps, deteriorating light conditions associated with environmental degradation is likely to have a more negative effect on depth distributions of the sporophytes than increasing temperatures. However, more research aimed at highlighting the interactive effects of increasing temperatures and reduced water quality on physiological processes at different life stages is needed to understand the new constraints of future environments on kelp distribution.

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Airoldi, L. & Beck, M. W. 2007. Loss, status and trends for coastal marine habitats of Europe. Oceanogr. *Mar. Biol. Annu. Rev.* 35:345–405.

Binzer, T., Sand-Jensen, K. & Middelboe, A. L. 2006. Community photosynthesis of aquatic macrophytes. *Limnol. Oceanogr.* 51:2722–33.

Bolton, J. J. & Anderson, R. J. 1994. *Ecklonia. In* Akatsuka, I. [Ed.] *Biology of Economic Algae.* SPB Academic Publishing, The Hague, pp. 385–406.

Bolton, J. J. & Lüning, K. 1982. Optimal growth and maximal survival temperatures of Atlantic *Laminaria* species (Phaeophyta) in culture. *Mar. Biol.* 66:89–94.

Campbell, S. J., Bite, J. S. & Burridge, T. R. 1999. Seasonal patterns in the photosynthetic capacity, tissue pigment and nutrient content of different developmental stages of *Undaria pinnatifida* (Phaeophyta: Laminariales) in Port Phillip Bay, southeastern Australia. *Bot. Mar.* 42:231–41.

Davison, I. R. 1991. Environmental effects on algal photosynthesis, temperature. *J. Phycol.* 27:2–8.

Davison, I. R., Greene, R. M. & Podolak, E. J. 1991. Temperature acclimation of respiration and photosynthesis in the brown alga *Laminaria saccharina. Mar. Biol.* 110:449–54.

Dayton, P. K., Tegner, M. J., Edwards, P. B. & Riser, K. L. 1999. Temporal and spatial scales of kelp demography: the role of oceanographic climate. *Ecol. Monogr.* 69:219–50.

Edmunds, P. J. & Gates, R. D. 2008. Acclimatization in coral reefs. *Mar. Ecol. Prog. Ser.* 361:307–10.

Egan, B., Vlasto, A. & Yaris, C. 1989. Seasonal acclimation to temperature and light in *Laminaria longicruris* de la Pyl. (Phaeophyta). *J. Exp. Mar. Biol. Ecol.* 129:1–16.

Fairhead, V. A. & Cheshire, A. C. 2004a. Seasonal and depth related variation in the photosynthesis–irradiance response of *Ecklonia radiata* (Phaeophyta, Laminariales) at West Island, South Australia. *Mar. Biol.* 145:415–26.

Fairhead, V. A. & Cheshire, A. C. 2004b. Rates of primary productivity and growth in *Ecklonia radiata* measured at different depths, over an annual cycle, at West Island, South Australia. *Mar. Biol.* 145:41–50.

Falkowski, P. G. & Laroche, J. 1991. Acclimation to spectral irradiance in algae. *J. Phycol.* 27:8–14.

Falkowski, P. G. & Raven, J. A. 1997. *Aquatic Photosynthesis.* Blackwell, Oxford, UK, 375 pp.

Fowler-Walker, M. J., Wernberg, T. & Connell, S. D. 2006. Differences in kelp morphology between wave sheltered and exposed localities: morphologically plastic or fixed traits? *Mar. Biol.* 148:755–67.

Goodsell, P. J., Fowler-Walker, M. J., Gillanders, B. M. & Connell, S. D. 2004. Variations in the configuration of algae in subtidal forests: implications for invertebrate assemblages. *Austral Ecol.* 29:350–7.

Haidekker, A. & Hering, D. 2007. Relationship between benthic insects (Ephemeroptera, Plecoptera, Coleoptera, Trichoptera) and temperature in small and medium-sized streams in Germany: a multivariate study. *Aquat. Ecol.* DOI: 10.1007/s10452-007-9097-z.

Hatcher, B. G., Kirkman, H. & Wood, W. F. 1987. Growth of the kelp *Ecklonia radiata* near the northern limit of its range in Western Australia. *Mar. Biol.* 95:63–73.

van den Hoek, C. 1982. Phytogeographic distribution of groups of benthic marine algae in the North Atlantic Ocean. A review of

experimental evidence from life history studies. *Helgol. Meeresunters.* 35:153–214.

IPCC. 2007. *Climate Change 2007: The Physical Science Basis - Summary for Policy Makers.* Intergovernmental Panel on Climate Change, WMO/UNEP, Paris, 21 pp.

Jeffrey, S. W. & Humphrey, G. F. 1975. New spectrophotometric equations for determining chlorophylls a, b, c_1 and c_2 in higher plants, algae and natural phytoplankton. *Biochem. Physiol. Pflanzen.* 167:191–4.

Johnson, K. S. 1982. Determination of phosphate in seawater by flow injection analysis with injection of reagent. *Anal. Chem.* 54:1185–7.

Johnson, K. S. 1983. Determination of nitrate and nitrite in seawater by flow injection analysis with injection of reagent. *Limnol. Oceanogr.* 28:1260–6.

Johnson, F. H., Eyring, H. & Stover, B. J. 1974. *The Theory of Rate Processes in Biology and Medicine.* Wiley-Interscience, New York, pp. 175–272.

Kain, J. M. 1989. The seasons in the subtidal. *Br. Phycol. J.* 24:203–15.

Kirkman, H. 1984. Standing stock and production of *Ecklonia radiata* (C. Ag.) J. Agardh. *J. Exp. Mar. Biol. Ecol.* 76:119–30.

Larkum, A. W. D. 1986. A study of growth and primary production in *Ecklonia radiata* Laminariales at a sheltered site in Port Jackson New South Wales, Australia. *J. Exp. Mar. Biol. Ecol.* 96:177–90.

Leliaert, F., Anderson, R. J., Bolton, J. J. & Coppejans, E. 2000. Subtidal understorey algal community structure in kelp beds around the Cape Peninsula (Western Cape, South Africa). *Bot. Mar.* 43:359–66.

Lourey, M. J., Dunn, J. R. & Waring, J. 2006. A mixed-layer nutrient climatology of Leeuwin Current and Western Australian shelf waters: seasonal nutrient dynamics and biomass. *J. Mar. Syst.* 59:25–51.

Lüning, K. 1984. Temperature tolerance and biogeography of seaweeds: the marine algal flora of Helgoland (North Sea) as an example. *Helgol. Meeresunters.* 38:305–17.

Maegawa, M., Kida, W., Yokohama, Y. & Aruga, Y. 1988. Comparative studies on critical light conditions for young *Eisenia bicyclis* and *Ecklonia cava*. *Jpn. J. Phycol.* 36: 166–74.

Maegawa, M., Yokohama, Y. & Aruga, Y. 1987. Critical light conditions for young *Ecklonia cava* and *Eisenia bicyclis* with reference to photosynthesis. *Hydrobiologia* 151/152:447–55.

Matson, P. G. & Edwards, M. S. 2007. Effects of ocean temperature on the southern range limits of two understory kelps, *Pterygophora californica* and *Eisenia arborea*, at multiple life-stages. *Mar. Biol.* 151:1941–9.

McQuaid, C. D. & Branch, G. M. 1984. Influence of sea temperature, substratum and wave exposure on rocky intertidal communities: an analysis of faunal and floral biomass. *Mar. Ecol. Prog. Ser.* 19:145–51.

Novaczek, I. 1984. Response of gametophytes of *Ecklonia radiata* (Laminariales) to temperature in saturating light. *Mar. Biol.* 82:241–5.

Pearce, A. F. 1991. Eastern boundary currents of the Southern Hemisphere. *J. R. Soc. West. Aust.* 74:35–45.

Raison, J. K., Berry, J. A., Armond, P. A. & Pike, C. S. 1980. Membrane properties in relation to the adaptation of plants to temperature stress. *In* Turner, N. C. & Kramer, P. J. [Eds.] *Adaptation of Plants to Water and Temperature Stress.* John Wiley and Sons, New York, pp. 261–73.

Raven, J. A. & Geider, R. J. 1988. Temperature and algal growth. *New Phytol.* 110:441–61.

Sakanishi, Y., Yokohama, Y. & Aruga, Y. 1989. Seasonal changes of photosynthetic activity of a brown alga *Ecklonia cava* Kjellman. *Bot. Mag. Tokyo* 102:37–51.

SAS Institute Inc. 1994. *SAS/STAT Users Guide, Vol. 2, Version 6*, 4th ed. SAS Institute, Cary, North Carolina.

Schiel, D. R., Steinbeck, J. R. & Foster, M. S. 2004. Ten years of induced ocean warming causes comprehensive changes in marine benthic communities. *Ecology* 85:1833–9.

Staehr, P. A. & Birkeland, M. 2006. Temperature acclimation of two mesophilic microalgae. *Phycologia* 45:648–56.

Strickland, J. D. H. & Parsons, T. R. 1968. *A Practical Handbook of Seawater Analysis.* Fisheries Research Board of Canada, No. 167. Ottawa, Canada, 311 pp.

Switala, K. 1993. *Determination of Ammonia by Flow Injection Analysis Colorimetry (Dialysis).* Latchet Instruments, Milwaukee, Wisconsin.

Thompson, P. & Waite, A. 2003. Phytoplankton responses to wastewater discharges at two sites in Western Australia. *Mar. Freshw. Res.* 54:721–35.

Vitousek, P. M., Aber, J. D., Howarth, R. H., Likens, G. E., Matson, P. A., Schindler, D. W., Schlesinger, W. H. & Tilman, D. G. 1997. Human alteration of the global nitrogen cycle: source and consequences. *Ecol. Appl.* 7:737–50.

Wang, H. G., Owen, R. D., Sanchez-Hernandez, C. & Romero-Almandez, M. D. 2003. Ecological characterization of bat species distributions in Michoacan, Mexico, using a geographic information system. *Glob. Ecol. Biogeogr.* 12:65–85.

Webb, W. L., Newton, M. & Starr, D. 1974. Carbon dioxide exchange of *Alnus rubra*: a mathematical model. *Oecologia* 17:281–91.

Wernberg, T., Coleman, M., Fairhead, A., Miller, S. & Thomsen, M. 2003b. Morphology of *Ecklonia radiata* (Phaeophyta: Laminariales) along its geographic distribution in south-western Australia and Australasia. *Mar. Biol.* 143:47–55.

Wernberg, T., Kendrick, G. A. & Phillips, J. C. 2003a. Regional differences in kelp-associated algal assemblages on temperate limestone reefs in south-western Australia. *Divers. Distrib.* 9:427–41.

Wing, S. R., Leichter, J. J., Perrin, C., Rutger, S. M., Bowman, M. H. & Cornelisen, C. D. 2007. Topographic shading and wave exposure influence morphology and ecophysiology of *Ecklonia radiata* (C. Agardh 1817) in Fiordland, New Zealand. *Limnol. Oceanogr.* 52:1853–64.