Vol. 244: 95–103, 2002 | MARINE ECOLOGY PROGRESS SERIES
Mar Ecol Prog Ser | Published November 29

Effects of benthic grazers on microalgal communities of morphologically different encrusting corallines: implications for abalone recruits

E. Day, G. M. Branch*

Marine Biology Research Institute, Zoology Department, University of Cape Town, Rondebosch 7701, South Africa

ABSTRACT: Recruits of the South African abalone *Haliotis midae* are more abundant on irregularly textured than on smooth encrusting corallines, and experimental removal of the urchin *Parechinus angulosus* leads to the virtual disappearance of *H. midae* recruits. Explanations include the following; (1) In the absence of larger grazers, prostrate diatoms that are preferentially consumed by abalone recruits become displaced by less suitable overstorey diatoms, filamentous algae or blue-green algae; (2) textured corallines protect pockets of suitable diatoms, whereas large grazers eliminate these from smooth corallines. We tested these possibilities in aquarium experiments in which microalgal communities were grown on artificial coralline mimics with different textures and then exposed to different grazers at densities simulating those in the field. Microalgal standing stocks (measured as chlorophyll *a*) increased in the absence of grazers, but this was reflected only in the amounts of filamentous and blue-green algae. Diatoms remained unchanged in quantity and community composition between different grazing treatments and substratum textures. Changes in diatom communities following urchin removal are thus unlikely to explain the ensuing disappearance of abalone recruits, although changes in filamentous and blue-green algae may do so. Textured surfaces did, however, consistently support higher stocks of microalgae than smooth surfaces (in either the presence or the absence of grazers), which may account for the preference of abalone recruits for irregular encrusting corallines in the field.

KEY WORDS: Encrusting corallines · Diatoms · Abalone · Grazers · Recruitment

INTRODUCTION

Benthic microflora, and diatoms in particular, are important for the early diet of post-larval abalone (Garland et al. 1985, Shepherd & Turner 1985, Norman-Boudreau et al. 1986, Kawamura et al. 1995, Matthews & Cook 1995). Veliger larvae of *Haliotis diversicolor* are stimulated to attach to diatom films, possibly because of the presence of particular bacteria within these films (Bryan & Qian 1998). Similarly, Johnson et al. (1991) suggested that settlement is elicited by chemicals released by bacteria in the diatomaceous films overlying coralline substrata. In either case, diatoms clearly play an important role in the settlement and early survival of abalone.

The particular composition of benthic diatom communities may also be of importance to these settlers. Work by Matthews & Cook (1995) on hatchery-reared *Haliotis midae* suggests that recent settlers have distinct, but not obligate, preferences for genera of prostrate or low-growing benthic diatoms. The abundance of these genera appears to be enhanced by grazing by larger juvenile abalone which remove loose, overstorey diatoms, thus maintaining populations of the smaller prostrate genera that would otherwise be overgrown. Nicotri (1977) has suggested that many prostrate diatoms have stronger adhesion than do overstorey diatoms, and may thus be less vulnerable to grazing. Low-growing diatoms are also likely to retain their dominance under conditions of strong wave action. Reisen & Spencer (1970) found both quantitative and qualitative differences in diatom communities grown under different current velocities.

*Corresponding author. Email: gmbranch@egs.uct.ac.za

Sommer (1997) demonstrated shifts in species dominance when diatoms were subjected to different densities of the grazing isopod *Idothea chelipes*. So-called 'well-edible' prostrate diatom taxa such as *Cocconeis costata* and *Navicula* spp. appeared to be the preferred diet of the isopods, and gave way to 'poorly edible', slower-growing and larger algae in the face of grazing. Kawamura et al. (1995) suggested that diatoms with either high adhesive strength or weakly silicified walls are favoured by many grazers, because they are most easily broken open, whereas less adhesive or stronger cells are easily ingested but often passed unharmed through the digestive system of the grazer.

The effects of grazing on diatom community composition obviously depend on the nature of the grazers themselves (reviewed by Underwood 1979, Hawkins & Hartnoll 1983). Even juvenile abalone consume different species to those favoured by abalone settlers (Matthews & Cook 1995). Underwood (1984) and Sharpe & Keough (1998) have shown that the gastropods *Nerita atramentosa* and *Cellana tramoserica* have different effects on microalgae. This may be due to differences in the size, behaviour or radular structure, or because different grazers deposit different amounts of mucus on the substratum, and this nutrient-rich material may fertilise microalgae (Connor & Quinn 1984). Andrew (1989) has shown that urchins and gastropod grazers respond differently to limitations in food supply, and this modifies their effects on algae.

It is not only the composition of the grazers that influences the composition of benthic microalgal communities. Shepherd & Daume (1996) suggest that the morphology of the settlement substratum itself plays a role. On the grounds that larger grazers apparently prefer smooth substrata, they hypothesised that biofilms will be more prevalent on uneven surfaces which, in turn, may thus be favoured by abalone recruits. In the case of the South African abalone *Haliotis midae*, recruits (defined here as post-settlers <3 mm shell length), exhibit a preference for knobbly corallines (Day & Branch 2000a). This pattern has been tentatively linked to avoidance of predation, because recruits may shelter in the cover provided by concavities on textured corallines. It may, however, also be attributable to greater food availability on these corallines. Juveniles of *H. midae* (3 to 35 mm in length) are closely associated with the urchin *Parechinus angulosus*, whereas recruits are only weakly associated with the urchin (Day & Branch 2000a,b) but nevertheless virtually disappeared from subtidal areas that were experimentally cleared of this urchin (Day & Branch 2002). Removal of *P. angulosus* had no effect on macroalgae, encrusting corallines or kelp sporelings, but reduced the accumulation of drift kelp and increased siltation. We believe that the disappearance of abalone recruits can be ascribed to the negative effects of increased siltation in urchin-cleared areas (see also Saito 1981, Tegner & Butler 1985). We recognise, however, that it might equally well have been due to the effects of urchin removal on the composition and abundance of microalgae. Potential effects include the alteration or loss of diatom communities that might provide chemical cues for settlement, or the reduction or loss of key diatom species favoured as a source of food by settlers.

Our experiment tested the following hypotheses: (1) Benthic microalgal biomass should be greater in the absence of macro-grazers than in their presence; (2) different types of grazers will have different effects on microalgal biomass and community composition; (3) an increase in prostrate diatoms will occur in the presence of grazers, and a proliferation of loose, overstorey diatoms in their absence; (4) the impact of grazers on microalgae will be greatest on smooth surfaces and, as a consequence, grazing will lead to lower microalgal standing stocks on smooth than on irregularly textured coralline surfaces.

MATERIALS AND METHODS

Experimental conditions. Experiments were conducted in glass aquaria of 37 × 60 × 15 cm. Lighting in the aquarium room was set at a 12 h light/dark cycle. Temperature was maintained at 17°C, and each tank was supplied with a constant flow of recirculated gravel-filtered water. Artificial substrata were constructed to mimic 3 common morphological types of encrusting corallines, known to influence the distribution patterns of abalone recruits (Day & Branch 2000a), and termed 'velvet', 'knobbly' and 'paint'. Velvet corallines (represented almost solely by *Heydrichia woelkerlingii* in the area where we worked) are 2 to 5 mm thick and smooth in texture. Knobbly corallines are intermediate in thickness (1 to 2 mm) and have irregular knobs (0.5 to 1.5 mm high) that create a markedly uneven surface. Species included in this category are *Leptophytum foveatum*, *L. discrepans* and *Spongites discordias*. Paint corallines (notably *L. ascervatum*) are thin (<1 mm), with an irregular, rough surface. In recognising these 3 morphological types, we followed the practice advocated by Woelkerling et al. (1993), and our categories 'velvet', 'knobbly' and 'paint' correspond to their groups 'thick, smooth crusts', 'lumpy and warty' and 'thin, uneven', respectively.

The mimic coralline substrata were constructed from a mix of dry Polyfilla cement and fibreglass resin. Inverted pieces of real knobbly corallines were used as a template for the artificial knobbly mimics, while mimics of velvet corallines were made from slightly undulating sheets of smooth resin mixture. Paint coral-

lines were mimicked by a monolayer of fine beach sand overlaid with a thin coating of the resin mix. The resin mixes were set and supported on pre-cut flat glass bases, 17.5 × 18.5 × 0.5 mm, and were cured for 3 d in the sun to dissipate any toxic chemicals contained in the fibreglass resin. After curing, measurements were made of the ratios of total to planar areas by fracturing the plates and measuring the distance across the contours of the surface for standard linear distances. This yielded the following proportions: knobbly = 1.44; paint = 1.06; velvet = 1.0. The bases were then randomly arranged to cover the whole of the floor of each aquarium, in a matrix of 6 plates (2 replicates for each of the 3 coralline mimics).

Mimic coralline substrata were left in the tanks for 2 wk. By then, diatom growths were clearly visible on the plate surfaces, and the experiment was initiated. Twelve tanks were used, comprising 3 replicates of each of the following 4 grazer treatments, randomly interspersed: (1) no grazers; (2) urchins only (9 *Parechinus angulosus*); (3) mix of gastropods (4 *Oxystele sinensis* + 4 *Turbo sarmaticus* + 4 *T. cidaris*); (4) 'all grazer' mix of urchins and gastropods (4 *P. angulosus* + 2 *O. sinensis* + 2 *T. sarmaticus* + 4 *T. cidaris*). The water level in each tank was 5 cm below the top of the tank to prevent the grazers from climbing the walls and escaping. The shallow depth of the water (10 cm) largely restricted the grazers to feeding on the surfaces of the coralline mimics on the base of each tank. The densities of grazers in each tank simulated those in the field (Pulfrich & Branch 2002). Numbers were adjusted so that the total wet biomass of grazers in each treatment approximated 530 g per tank.

Grazers were habituated to aquarium conditions for 3 d before being introduced to the experimental tanks. The experiment ran for 10 d in late January, after which the grazers were removed and the microflora on the coralline mimics was sampled as described below.

Chlorophyll *a* analysis. Chl *a* was extracted and used as a measure of the total biomass of microalgae. This is a reliable but relative measure of microfloral abundance (Underwood 1984, Hill & Hawkins 1990) used in several studies (Nicotri 1977, Bustamante et al. 1995, Sharpe & Keough 1998).

Three plates, 1 of each type of coralline mimic, were removed from each tank and the chlorophyll on them extracted in the dark in 100 to 150 ml 90% acetone with periodic agitation for approximately 20 min (HMSO 1986). Samples were then centrifuged for 10 min, and 6 ml of acetone was added to 1 ml volumes extracted from the resultant supernatant. Fluorometric readings were taken using a Turner Designs fluorometer. After the initial readings, 2 drops of 10% HCl were added to each sample, and a second reading was taken.

Concentrations of chl *a* were calculated from standard fluorometric equations (HMSO 1986) and the data converted to mg chl *a* m^{-2} substratum. A 3-way ANOVA was run on the data, with fixed effects Grazers and Substratum, and replicate aquaria nested in the grazer treatments.

Analysis of diatom community structure. The remaining 3 plates from each tank were preserved in 4% seawater formalin for diatom cell counts. Microalgal material was removed from the plates by brushing, and subsamples were diluted in distilled water and allowed to settle for 40 min in a 2 ml chamber slide before being examined with an inverted microscope at 400× magnification, and counted according to the Utermöhl technique (Hasle 1978). Dilution varied between samples: mixtures were diluted until only a single layer of diatoms collected on the chamber bottom, and counts were standardised to allow for the dilution.

Diatoms were initially classified into 20 gross morphological 'types' whilst being counted under the inverted microscope. For each sample, 300 to 400 individuals were counted. Lund et al. (1958) estimated that counts of 400 individuals provide a 10% variance at the 95% Critical Level, assuming a random distribution of cells.

Identification of diatoms with Scanning Electron Microscopy (SEM). A subsample of the extracts from each substratum was washed repeatedly with distilled water, diluted to achieve uniform spread, and settled on a photographic plate covering an individual SEM stub. As the diatoms were not highly silicified, pretreatment was limited to washing and the samples were air-dried as recommended by MacLulich (1986) and Hill & Hawkins (1990). SEM samples were coated with gold-palladium and examined at an accelerated voltage of 10 kV at magnifications of up to 50 000×. Micrographs were used to identify the taxa corresponding to the morphological categories into which the samples had previously been grouped on the basis of light microscopy. This resulted in the initial 20 types being reduced to 11 taxa. For each of these, the diatom counts conducted by light microscopy were converted into cells per cm^2 of substratum. A 3-way ANOVA was run on total counts of diatoms in each sample, with fixed effects Grazers and Substratum, with replicate aquaria nested in the grazer treatments. Normality (Kolmogorov-Smirnov test) and homogeneity of variance (Levene's test) were tested prior to running ANOVA and, where necessary, data were transformed to meet the assumptions of ANOVA.

A multivariate statistical analysis package, PRIMER (Ver. 4; Plymouth Marine Laboratory), was used to test for differences in diatom communities between treatments. Data on the abundance of each diatom taxon

were 4th-root-transformed, and the Bray-Curtis coefficient was employed because it is not affected by joint absences of species (Field et al. 1982) and gives more weight to abundant species than to rare ones. A dendrogram was derived from the data, based on hierarchical agglomerative clustering (Field et al. 1982, Clarke & Warwick 1990). Diatom types that accounted for less than 5% of the total number were not included in the latter analyses.

Filamentous and blue-green algal growth on substrata. Larger 'non-diatomaceous' microalgae, notably filamentous and blue-green algae, could not be quantified in the same way as diatoms. Instead, an undiluted sample of each substratum extract from each treatment was pipetted onto a glass slide, and covered with a glass cover slip. The base of the slide was placed over a grid of 1 × 1 mm squares. Using a stereo microscope, the percentage area occupied by mats of this 'non-diatomaceous' algal material was measured. ANOVA was run on these data after transformation using the formula $p^1 = \sqrt{(\arcsin p)}$, where p^1 is the transformed value and p is the original value, with fixed effects Grazers and Substratum, and replicate aquaria nested in the grazer treatment.

RESULTS

Chlorophyll *a* analysis

Chl *a* concentrations were consistently and significantly lower on velvet coralline mimics than on either of the other 2 substrata (Fig. 1). Significant differences were also observed between different grazer treatments (ANOVA: substratum effect, $F = 14.69$, $df_{2,16}$, $p < 0.001$; grazer effect, $F = 3.82$, $df_{3,8}$, $p < 0.05$; interaction of grazing and substratum not significant: $F = 1.83$, $df_{6,16}$, $p > 0.05$). Tukey *a posteriori* tests showed that at a significance level of $p < 0.05$, the velvet coralline mimic had consistently lower chl *a* than either of the other 2 substrata, which did not differ from each other. They also showed that only the 'mixed gastropod' and 'no grazer' treatments were significantly different from each other, with ungrazed substrata having higher concentrations of chl *a*.

Structure of diatom communities

Analysis of the diatoms under an inverted microscope and by SEM revealed large numbers of very small diatoms (2 to 35 µm). On the whole, the diatoms were not highly silicified, suggesting a state of rapid growth. In total, 20 morphological types were identified under the light microscope, but these were reduced to 11 taxa after analysis under the SEM, 7 of which were subsequently identified to generic level. The remaining 4 types represented either minor taxa, or unrecognised views of more common taxa, and were excluded from detailed analyses, although they were used to generate total counts of diatoms.

The total diatom counts and the relative numbers of the 6 most abundant identified taxa are shown in Fig. 2. Contrary to the hypotheses advanced, no significant differences in the total numbers of diatoms were found between different grazer treatments, or between different substrata (ANOVA: substratum effect, $F = 1.76$, $df_{2,16}$, $p > 0.1$; grazer effect, $F = 0.22$, $df_{3,8}$, $p > 0.1$; no significant interaction: $F = 0.32$, $df_{6,16}$, $p \gg 0.1$). Knobbly textures generally had the highest densities of diatoms, followed by paint and then velvet textures. This ranking applied to all but the gastropod grazer treatments (Treatment B) and a marginally higher value for velvet than paint corallines in the urchin grazer treatment (Treatment C).

The diatoms were prostrate taxa with the single exception of an unidentified type of overstorey diatom, which constituted 0.02% of the total numbers and displayed no pattern with respect to grazer combinations or substratum types.

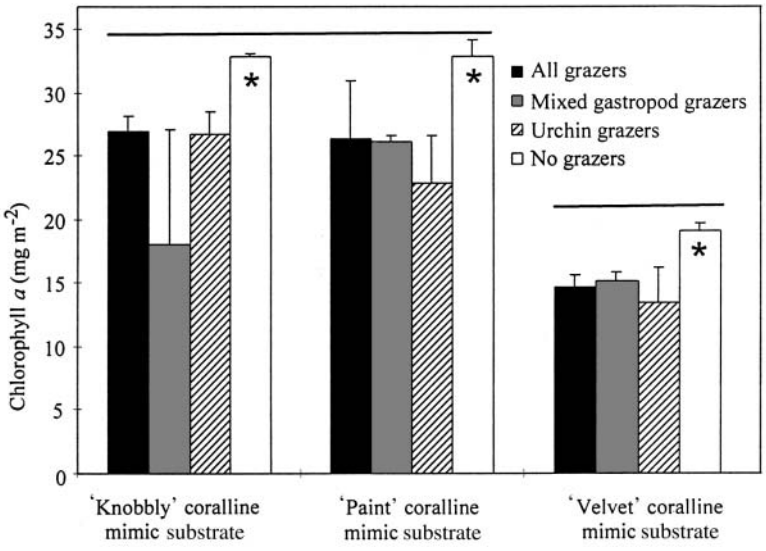

Fig. 1. Concentration of chl *a* (mean + SE) from different grazer treatments and on different substrata. Horizontal bars: substrata are not significantly different. *: Significantly different from other treatments of the same substratum

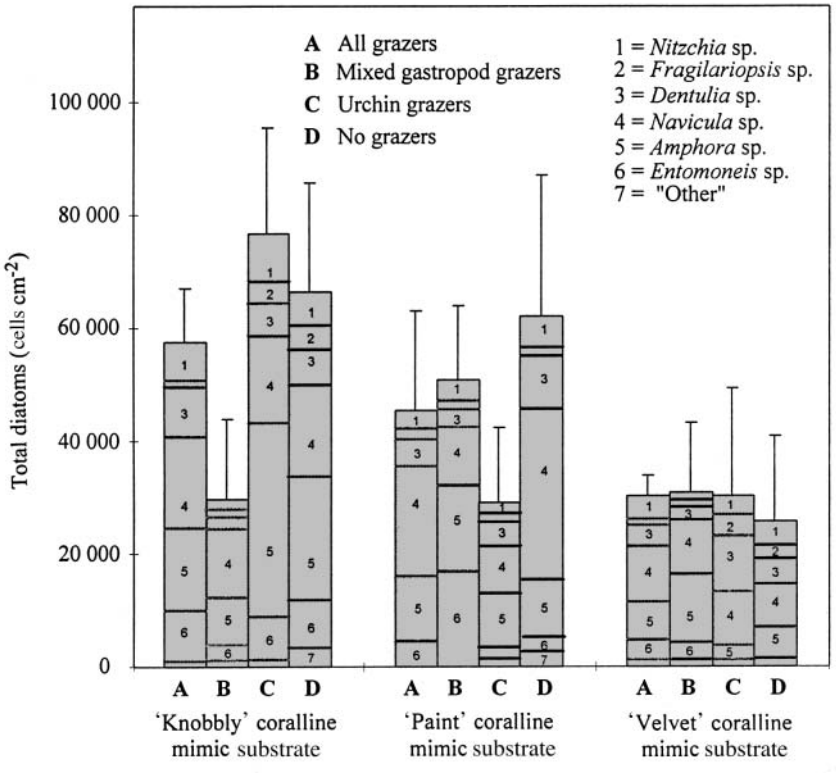

Fig. 2. Effects of grazer treatments and coralline-mimic substratum types on diatom densities (mean + SE) and composition

Analysis of diatom community structure

Bray-Curtis similarities based on the relative numbers of different diatom taxa were high between all samples, exceeding 85% similarity in most cases (Fig. 3). Three clusters separated out at the 75% level (Groups A, B and C). The dendrogram did not, however, suggest any clear links between diatom community composition and either substratum type or grazer treatments.

Filamentous and blue-green algal growth on substrata

Significantly more 'non-diatomaceous' growth was evident on all ungrazed treatments, regardless of substratum type (Fig. 4). In addition, 'paint' substrata had significantly larger amounts of material than did 'velvet' substrata, but did not differ from 'knobbly' substrata (ANOVA: substratum effect, $F = 3.41$, $df_{2,16}$, $p < 0.05$; grazer effect, $F = 15.99$, $df_{3,8}$, $p < 0.001$; interaction effect not significant: $F = 1.06$, $df_{6,16}$ $p > 0.1$; Tukey *a posteriori* tests, $p < 0.05$). Although differences in scale and methods of measurement made it difficult to compare the amounts of diatoms and non-diatomaceous algae quantitatively, we estimated that diatoms accounted for only 5 to 10% of the volumes occupied by non-diatomaceous algae.

DISCUSSION

Effects on chl *a* concentrations

Chl *a* concentrations provided 1 measure of the effects of grazers on different substratum types. As predicted, ungrazed treatments did have higher levels of chl *a* than grazed treatments (Fig. 1). Beyond this level of analysis, however, interpretation becomes more complex. Statistically significant differences in chl *a* existed only between ungrazed and gastropod-grazed treatments. The reasons for this may lie in the grazing mechanisms employed by the different suites of grazers involved. The gastropods used in these experiments, for example, have powerful radulae adapted towards effective grazing close to the substratum surface (Steneck 1982). The urchin *Parechinus angulosus*, by contrast, has been shown to feed under natural conditions predominantly by trapping drift kelp (Day & Branch 2002). In the absence of drift kelp, however, we expected these urchins to feed effectively on microalgae. We also expected that, as a consequence, they would influence the abundance of microalgae, but they did not. At face value, thus, the results indicate that the gastropods had a significant impact on the microalgal biomass, whereas the urchins did not. This partially upholds the second hypothesis advanced in the introduction, that different types of grazers will have different effects on microalgal biomass.

A consideration of substratum textures introduces another aspect of the data. Concentrations of chl *a* were consistently higher on knobbly or paint coralline mimics than on smooth surfaces, regardless of the grazer treatment. In part, this reflects the fact that the surface area available for microalgal growth is greater on irregular than on smooth surfaces. However, even if the differences between total and planar areas are considered (see 'Materials and methods'), substratum texture still has a significant effect on chl *a* concentrations (ANOVA, $F = 3.62$, $df_{2,16}$, $p < 0.05$; Tukey *a posteriori* tests show significant differences between 'velvet' and 'paint' substrata at $p < 0.05$). Thus, the ratios of total to planar areas cannot alone explain the differences. Differences in the ease of grazing on different

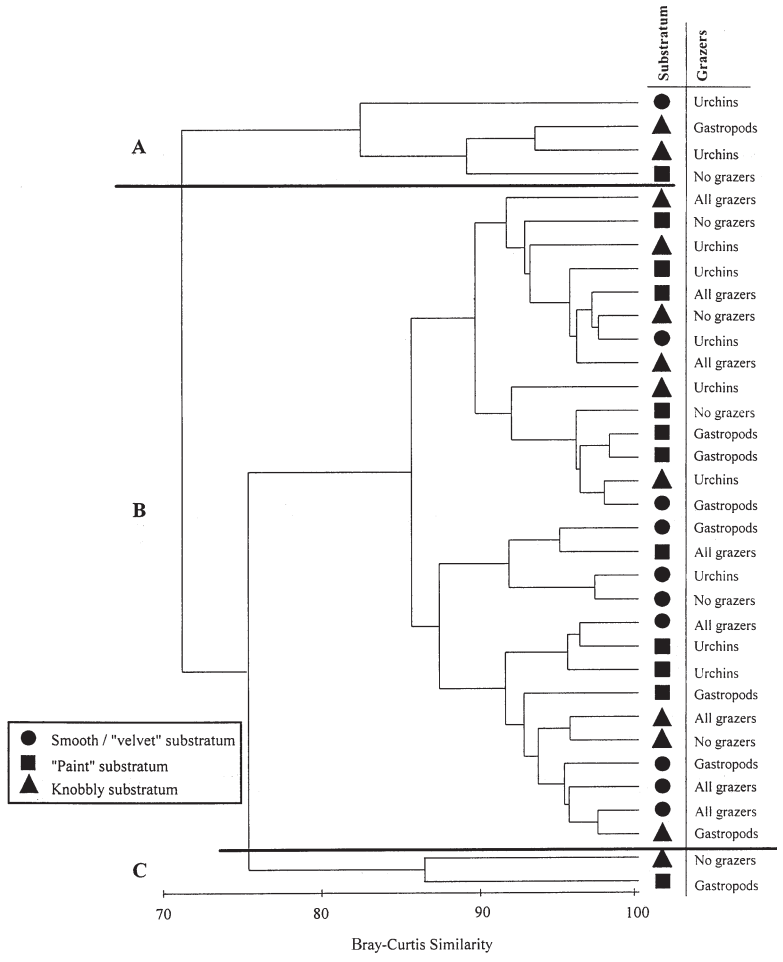

Fig. 3. Similarity of diatom samples from different grazer treatments and different substratum types

substrata might, however, play a role in explaining these data, at least for the grazed treatments.

In general, however, the most obvious outcome was that the concentration of chl *a* was lowest on the smooth coralline-mimics. This upholds the hypothesis that microalgal biomass should be higher on textured rather than smooth surfaces, and this pattern was maintained in both the presence and the absence of the grazers. In terms of the implications for abalone settlers, occupation of an irregular coralline is likely to provide both protection from bulldozing by larger grazers (McShane 1992) and an enhanced food supply (Shepherd and Daume 1996).

The differences between grazed and ungrazed treatments were not as dramatic as expected, possibly due to experimental conditions (see Peterson & Black 1994). Under laboratory conditions, the availability of light in a shallow tank is higher than that on the floor of a kelp bed, where shading reduces light penetration (Reed & Foster 1984). In the experiment, therefore, microalgal growth may have been above natural rates. This could have dampened the effects of grazer treatments. The use of artificial plates standardised the substrata and allowed isolation of texture as the only variable, but may not have reflected diatom densities or succession on natural corallines in the field (cf. Hudon & Bourget 1981).

Concentrations of chl *a* do not, of course, necessarily indicate the quantities of diatoms themselves (Hill & Hawkins 1990). Taxa may vary in the amount of chl *a* they contain, so that differences in community structure may not be reflected by differences in chl *a* concentrations (Sharpe & Keough 1998). In the present context, measures of chl *a* will have been strongly affected by the abundance of 'non-diatomaceous' microalgae, particularly filamentous or blue-green algae, obscuring differences between grazing and substratum treatments.

Effects on microalgal densities and cover

No significant differences were found in the total diatom densities of communities from different treatments and samples. Even grazed versus ungrazed treatments did not differ significantly (Fig. 2). Analyses of the relative abundance of blue-green and filamentous algae showed that grazing by all suites of grazers substantially and significantly decreased the quantities of these 'non-diatomaceous' algae on all substrata, and also repeated the pattern that 'velvet' coralline mimics sustained the lowest levels (Fig. 4). These results suggest that differences in chl *a* concentrations between gastropod-grazed and ungrazed treatments were due to the effect of grazing on non-diatomaceous algal material. Thompson et al. (2000) similarly showed that the abundance of blue-green algae (but not that of diatoms) increased after grazers were excluded in intertidal field experiments.

Hargrave (1970), Calow (1973) and Nicotri (1977) argued that since blue-green algae tend to be toxic or indigestible, they are seldom consumed by grazers that are small enough to select between different types of microalgae. Larger grazers such as those employed in our experiments are, however, likely to graze non-selectively. If blue-green algae thrive in the absence of

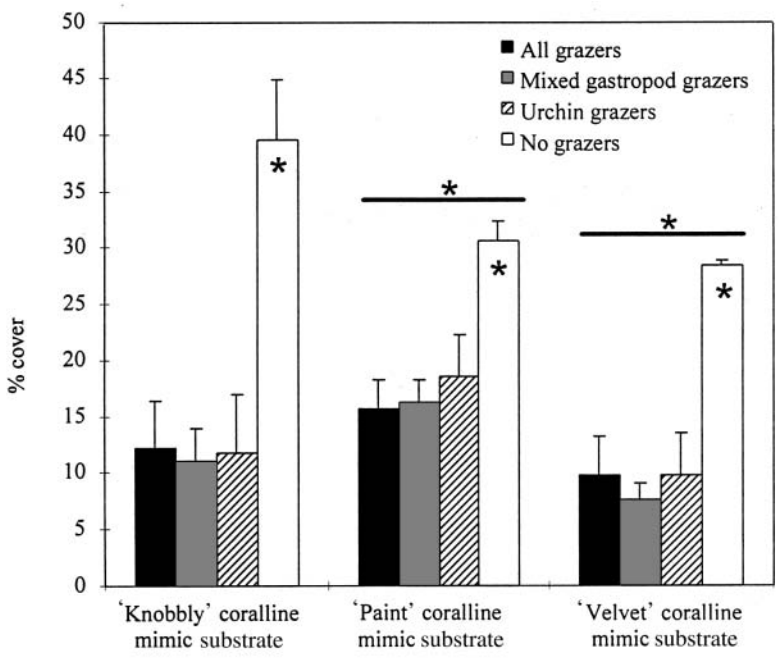

Fig. 4. Abundance of filamentous and blue-green ('non-diatomaceous') algae on different substrata exposed to different suites of grazers (% cover; mean + SE). * over horizontal lines: significant differences between substrata; * within data bars: significantly different from other treatments of the same substrate

large grazers, their development might negatively affect the survival of small abalone settlers, by reducing their access to the more nutritious diatoms preferred by abalone recruits. This might account for the disappearance of *Haliotis midae* settlers from areas from which the urchin *Parechinus angulosus* had been experimentally removed (Day & Branch 2002). Enhancement of the growth of blue-green and filamentous microalgae following a reduction of grazing pressure may also have contributed to the entrainment of sediment in mucilage threads. This has been suggested by Kennelly (1983) and may account for the increase in sedimentation observed in urchin-removal areas (Day & Branch 2002). In the absence of firm field-based experimental evidence, however, these suggestions remain hypothetical.

Our work focused on whether differences would develop in the composition and abundance of diatom assemblages on corallines with different textures exposed to different types of grazing. It is, however, difficult to extrapolate our results, because other factors may come into play under field conditions, including variations in illumination, temperature, and the supply of microbiota from the plankton. In particular, removal of urchins may have strong effects on various components of the ecosystem, not just microflora. Elsewhere, exclusion of urchins causes profound increases in foliose macroalgae (e.g. Estes et al. 1974, Harrold & Reed 1985, Fletcher 1987, Sala et al. 1998). This is unlikely in the present context, however, as experimental removals of *Parechinus angulosus* had no influence on foliose algae or kelp sporelings, probably because this urchin traps drift kelp rather than acting as a grazer (Day & Branch 2002). Its removal did lead to an accumulation of silt and diminished the build-up of drift kelp. The disappearance of abalone recruits could thus have been a consequence of changes in the quality of the habitat that were unrelated to microalgae, increased siltation being the most likely explanation. Our field experiments on urchin removal did not, however, include observations on microfloral composition, which is why we undertook the present work.

Diatom community composition

In terms of the composition of the diatom communities, SEM and light microscopy showed that all the diatoms comprised small, low-growing or prostrate forms, with one poorly represented exception. The prostrate forms included genera described by Matthews & Cook (1995) as being preferred food of abalone recruits. Among them, *Amphora* and *Entomoneis* were dominant elements of the diatom communities and present in all samples (Fig. 2). *Amphora* was a common component of benthic microfloral communities in experiments by Hudon & Bourget (1981) and Matthews & Cook (1995).

There was no evidence in our experiments of a succession leading to overstorey diatoms of genera such as *Delphineis* sp., which Matthews & Cook (1995) reported as forming a loose overstorey in ungrazed treatments. If any succession occurred at all, it was probably towards one dominated by blue-green or filamentous algae.

The most important result of our experiment is the fact that no significant differences were observed in diatom community structure between grazed and ungrazed treatments. One of the queries raised by our urchin removal experiments (Day & Branch 2002) was whether the failure of abalone settlers following removal of urchins might have been due to changes in the composition of the (unmonitored) diatom community associated with encrusting corallines. Matthews & Cook (1995) have shown that grazing by juvenile abalone can alter the nature of diatom communities under aquaculture conditions.

The data presented here, however, provide no support for this line of thought. Neither the total amount of diatoms (Fig. 2) nor the composition of the diatom community (Fig. 3) showed any significant response to grazing. Our experiments simulated the range and densities of grazers typical of field conditions, whereas the experiments of Matthews & Cook (1995) were based on grazing by abalone juveniles at substantially greater densities than under natural circumstances. Matthews & Cook (1995) tested the effect of grazing by juvenile abalone, whereas our experiment examined the effects of other species of subtidal grazers, all of which were larger than juvenile abalone. Juvenile abalone might have generated microalgal communities suitable for abalone settlement and recruit survival simply because they were smaller and grazed more selectively. Finally, Bryan & Qian (1998) showed that larvae of the abalone *Haliotis diversicolor* settle and survive better on a combination of diatom film and conspecific mucus than on diatom film alone.

In sum, our experiments employed those gastropod and urchin grazers that are most abundant in the field, and they were held close to natural densities. Under these conditions, they failed to alter the diatom communities, compared with those that developed in the absence of grazing. In terms of the original finding that prompted this work—the decline in abalone recruits in urchin exclusion areas—we conclude that differences in diatom communities between areas with and without urchins are very unlikely to explain why abalone recruitment collapsed in the experimental urchin exclusion plots (Day & Branch 2002).

Returning to the original hypotheses outlined in the Introduction, (1) benthic microalgal standing stocks (as measured by chl *a*) did increase in the absence of grazers, but (2) only gastropods, and not sea urchins, evoked this response. (3) In the absence of grazers, larger microalgae (filamentous and blue-green algae) benefited, but diatoms failed to increase in density or to show any pattern of change in community structure. Specifically, there was no increase in prostrate diatoms in the presence of grazers, nor a proliferation of overstorey species in their absence. (4) Textured substrata did consistently support greater microalgal stocks than smooth ones, although this difference occurred in both the presence and absence of grazers. The existence of elevated microalgal stocks may be a central reason why textured corallines appear to be the preferred habitat for abalone recruits (Shepherd & Duame 1996, Day & Branch 2000a).

Acknowledgements. Funding for the research was provided by the National Research Foundation (NRF), the Mellon Foundation, the University of Cape Town (UCT), and the South African Network for Coastal and Oceanographic Research (SANCOR). E.D. received a bursary from the NRF. Johnny Day devoted his garage and his time to many trial runs constructing coralline mimics. Thanks to 4 anonymous referees for their improvements to the paper.

LITERATURE CITED

Andrew NL (1989) Contrasting ecological implications of food limitation in sea urchins and herbivorous gastropods. Mar Ecol Prog Ser 51:189–193

Bryan PJ, Qian PY (1998) Induction of larval attachment and metamorphosis in the abalone *Haliotis diversicolor* (Reeve). J Exp Mar Biol Ecol 223:39–51

Bustamante RH, Branch GM, Eekhout SE, Robertson B, Zoutendyk P, Schleyer M, Dye A, Hanekom N, Keats D, Jurd M, McQuaid C (1995) Gradients of intertidal primary productivity around the coast of South Africa and their relationships with consumer biomass. Oecologia 102:189–201

Calow P (1973) The food of *Ancylus pluviatilis* (Mull.), a littoral stone–dwelling herbivore. Oecologia 12:113–133

Clarke KR, Warwick RM (1990) Statistical analysis and interpretation of marine community data. Unpubl rep. Intergovernmental Oceanographic Commission, UNESCO, Paris

Connor VM, Quinn JF (1984) Stimulation of food species growth by limpet mucus. Science 225:843–844

Day E, Branch GM (2000a) Field evidence for an association between juveniles of the South African abalone *Haliotis midae* and the sea urchin *Parechinus angulosus*. S Afr J Mar Sci 22:137–144

Day E, Branch GM (2000b) Relationships between recruits of the abalone *Haliotis midae*, encrusting corallines and the sea urchin *Parechinus angulosus*. S Afr J Mar Sci 22: 145–156

Day E, Branch GM (2002) Effects of sea urchins (*Parechinus angulosus*) on recruits and juveniles of abalone (*Haliotis midae*). Ecol Monogr 72:133–149

Estes JA, Palmisano JF (1974) Sea otters: their role in the structuring of nearshore communities. Science 185:1058–1060

Field JG, Clarke KR, Warwick RM (1982) A practical strategy for analyzing multispecies distribution patterns. Mar Ecol Prog Ser 8:37–52

Fletcher WJ (1987) Interactions among subtidal Australian sea urchins, gastropods and algae: effects of experimental removals. Ecol Monogr 57:89–109

Garland CD, Cook SL, Grant JF, McMeekin TA (1985) Ingestion of the bacteria on the cuticle of crustose (nonarticulated) coralline algae by post-larval and juvenile abalone (*Haliotis ruber* Leach) from Tasmanian waters. J Exp Mar Biol Ecol 91:137–149

Hargrave BT (1970) The utilisation of benthic microflora by *Hyalella azteca* (Amphipoda). J Anim Ecol 39:327–437

Harrold C, Reed DC (1985) Food availability, sea urchin grazing and kelp-forest community structure. Ecology 66: 1160–1169

Hasle GR (1978) Using the inverted microscope. In: Sournia A (ed) Phytoplankton manual. UNESCO, Paris, p 469–474

Hawkins SJ, Hartnoll RG (1983) Grazing of intertidal algae by marine invertebrates. Oceanogr Mar Biol Annu Rev 21: 195–282

Hill AS, Hawkins SJ (1990) An investigation of methods for sampling microbial films on rocky shores. J Mar Biol Assoc UK 70:77–88

HMSO (1986) The determination of chlorophyll-*a* in aquatic environments. In: Methods for the examination of water and associated materials, Vol 4, Sec 2, HMSO, London, p 6–13

Hudon C, Bourget E (1981) Initial colonisation of artificial substrate: community development and structure studied by scanning electron microscopy. Can J Fish Aquat Sci 38:1371–1384

Johnson CR, Muir DG, Reysenbach AL (1991) Characteristic bacteria associated with surfaces of coralline algae: a hypothesis for bacterial induction of marine invertebrate larvae. Mar Ecol Prog Ser 74:281–294

Kawamura T, Saido T, Takami T, Yamashita Y (1995) Dietary value of benthic diatoms for the growth of post-larval abalone *Haliotis discus hannai*. J Exp Mar Biol Ecol 194: 189–199

Kennelly SJ (1983) An experimental approach to the study of factors affecting algal colonisation in a sublittoral kelp forest. J Exp Mar Biol Ecol 68:257–276

Lund JWG, Kipling C, Le Cren ED (1958) The inverted microscope method of estimating algal numbers and the statistical basis of estimations by counting. Hydrobiology 11: 143–170

MacLulich JH (1986) Experimental evaluation of methods for sampling and assaying intertidal epilithic microalgae. Mar Ecol Prog Ser 34:275–280

Matthews I, Cook PA (1995) Diatom diet of abalone post-larvae (*Haliotis midae*) and the effect of pre-grazing the diatom overstorey. Mar Freshw Res 46:545–548

McShane PE (1992) Early life history of abalone: a review. In: Shepherd SA, Tegner MJ, Guzman SA (eds) Abalone of the world: biology, fisheries and culture. Fishing News Books, Oxford, p 120–138

Nicotri ME (1977) Grazing effects of four marine intertidal herbivores on the microflora. Ecology 58:1020–1032

Norman-Boudreau K, Burns D, Cook CA, Austin A (1986) A simple technique for detection of feeding in newly-metamorphosed abalone. Aquaculture 51:313–317

Peterson CH, Black R (1994) An experimentalist's challenge: when artifacts of intervention interact with treatments. Mar Ecol Prog Ser 111:289–297

Pulfrich A, Branch TA (2002) Population dynamics and potential yield of three species of giant winkles in the western Cape, South Africa. S Afr J Mar Sci 24:161–183

Reed DC, Foster MS (1984) The effects of canopy shading on algal recruitment and growth in a giant kelp forest. Ecology 65:937–948

Reisen WK, Spencer DJ (1970) Succession and current demand relationships of diatoms on artificial substrates in Prater's Creek, South Carolina. J Phycol 6:117–121

Saito K (1981) The appearance and growth of 0-year Ezo abalone. Bull Jpn Soc Sci Fish 47:1393–1400

Sala ECF, Boudouresque CF, Harmelin-Vivien M (1998) Fishing, trophic cascades, and the structure of algal assemblages: evaluation of an old but untested paradigm. Oikos 83:425–439

Sharpe AK, Keough MJ (1998) An investigation of the indirect effects of intertidal shellfish collection. J Exp Mar Biol Ecol 223:19–38

Shepherd SA, Daume S (1996) Ecology and survival of juvenile abalone in a crustose coralline habitat in South Australia. In: Watanabe Y, Yamashita Y, Oozeki Y (eds) Survival strategies in early life stages of marine resources: proceedings of an international workshop. Balkema, Rotterdam, p 297–313

Shepherd SA, Turner JA (1985) Studies on southern Australian abalone (Genus *Haliotis*). VI. Habitat preference, abundance and predators of juveniles. J Exp Mar Biol Ecol 93:285–298

Sommer U (1997) Selectivity of *Idothea chelipes* (Crustacea: Isopoda) grazing on benthic microalgae. Limnol Oceanogr 42:1622–1628

Steneck RS (1982) A limpet-coralline alga association: adaptations and defences between a selective herbivore and its prey. Ecology 63:507–522

Tegner MJ, Butler RA (1985) The survival and mortality of seeded and native red abalones, *Haliotis rufescens*, on the Palos Verdes Peninsula. Calif Fish Game 71(3):150–163

Thompson RC, Norton TA, Roberts MF, Hawkins SJ (2000) Feast or famine for intertidal grazing molluscs: a mismatch between seasonal variations in grazing intensities and abundance of microbial resources. Hydrobiol 440:357–366

Underwood AJ (1979) The ecology of intertidal gastropods. Adv Mar Biol 16:111–210

Underwood AJ (1984) Vertical and seasonal patterns in competition for microalgae between intertidal gastropods. Oecologia 64:211–222

Woelkerling WJ, Irvine LM, Harvey AS (1993) Growth-forms in non-geniculate coralline red algae (Corallinales, Rhodophyta). Aust Syst Bot 6:277–293

Editorial responsibility: Roger Hughes (Contributing Editor), Bangor, Wales, United Kingdom

Submitted: March 19, 2001; Accepted: June 7, 2002
Proofs received from author(s): November 11, 2002