Marine and Freshwater Behaviour and Physiology

Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/gmfw20

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Published online: 21 Oct 2013.

To cite this article: Baoquan Li, John K. Keesing, Martin Lourey & James McLaughlin (2013) Feeding and bioturbation effects of the sand dollar Peronella lesueuri (L. Agassiz, 1841) (Echinodermata) on microphytobenthos and sediment fluxes, Marine and Freshwater Behaviour and Physiology, 46:6, 431-446, DOI: 10.1080/10236244.2013.850834

To link to this article: http://dx.doi.org/10.1080/10236244.2013.850834

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Feeding and bioturbation effects of the sand dollar Peronella lesueuri (L. Agassiz, 1841) (Echinodermata) on microphytobenthos and sediment fluxes

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(Received 25 April 2013; accepted 29 September 2013)

Respiration and excretion rates of a key bioturbating species, the sand dollar Peronella lesueuri, were measured in mesocosms at three different temperatures. Benthic oxygen and nutrient fluxes were additionally measured at winter and summer temperatures to assess the impact of P. lesueuri on ecosystem processes. Oxygen consumption by sand dollars increased significantly with wet weight at all three temperatures 16, 19, and 23 °C. Ammonia release also increased with body weight. The weight vs. oxygen uptake relationship was similar at 19 and 23 °C but oxygen uptake was significantly reduced at the lower exposure temperature. The bioturbation caused by sand dollar P. lesueuri reduced the photosynthetic rate of the microphytobenthos (MPBs) but had a much smaller and less obvious effect on nutrient fluxes across the sediment–water interface.

Keywords: Bioturbation; sediment; sand dollar; Peronella lesueuri; oxygen flux; nutrient flux; benthic metabolism

Introduction

Processes which act at the water–sediment interface can have an important influence on ecology and biogeochemistry of soft sediment habitats (Mermillod-Blondin & Rosenberg 2006). Key among these processes is bioturbation. Bioturbation is a combination of physical sediment displacement and irrigation activity (Mermillod-Blondin 2011). The mixing and displacement of sediment by benthic macrofauna can control rates of organic matter degradation and carbon burial (Lohrer et al. 2005). This kind of activity has significant effects on the fluxes of dissolved oxygen (DO) and allows oxygen to penetrate more deeply into the sediment. This increased oxic layer can lead to decreased anoxic processes such as denitrification and promote aerobic processes such as nitrification of ammonium to nitrate (Widdicombe & Austen 1998).

Macrobenthic invertebrates are a very important group of bioturbators in soft substrate benthic ecosystems where they exert a structuring influence on the habitat and sediment particle distribution (Gerino 1990). Echinoderms, in particular, are well known for the effect they have in structuring sedimentary benthic habitats by grazing (e.g. echi-
noids, Keesing 2001), predation (e.g. asteroids, Sloan 1980), or bioturbation (echinoids and holothuroids, Hammond 1982; Uthicke 1999). These effects can be particularly significant when the animal is highly abundant or when the population size fluctuates, as is a common feature of echinoderms (Uthicke, Schaffelke, & Byrne 2009).

Microphytobenthos (MPBs) are the main primary producers in most unvegetated shallow-water sediments and are usually dominated by epipelic diatoms (Smith & Underwood 2000). MPBs and macrofauna are considered the two key groups affecting benthic metabolism in shallow-water sediments because of their high abundance and level of activity in benthic ecosystems (e.g. Lohrer et al. 2004). The burrowing activities of macrofauna impact sediment in many ways, which could affect MPBs directly and indirectly. These include physical impacts such as burial, dispersal and resuspension and biogeochemical impacts such as irrigation (oxygenation) and nitrification (e.g. ammonia excretion). It is, however, not yet clear to which extent the bioturbation would affect the MPBs production in shallow coastal sediments (Tang & Kristensen 2007).

Irregular burrowing echinoids such as heart urchins and sand dollars can be very abundant in soft sediment habitats (e.g. Keesing & Irvine 2013) and can have an important influence on their soft sediment habitats in a variety of ways, for example, affecting sediment permeability and water content, destabilizing chemical gradients in pore water, subducting organic matter, and influencing rates of remineralization and inorganic nutrient efflux (Widdicombe & Austen 1998; Lohrer et al. 2004). The effects of animal activities on sediment turnover rates may be considerable. Hollertz and Duchène (2001) reported that Brissopsis lyrifera reworked sediment volume by 22 ml h⁻¹ at 13 °C and 14 ml h⁻¹ at 7 °C due to burrowing activities. Lohrer et al (2005) found that the volume of sediment displaced by Echinocardium populations reached 20,000 cm³ m⁻² d⁻¹, which means the surface sediment is reworked about every 3 days at sites where Echinocardium is abundant. By stimulating benthic bacterial metabolism, burrowing echinoids can enhance benthic oxygen consumption and may accelerate the process of oxygen depletion under increasingly eutrophic conditions (Osinga et al. 1995, 1997). Osinga et al. (1995) studied the effects of the presence of the sea urchin species Echinocardium cordatum on benthic oxygen uptake rates and oxygen penetration depths, and concluded that E. cordatum augments benthic oxygen uptake in addition to oxygen consumption by the animals themselves.

Among the irregular urchins, there are far fewer studies on sand dollars than on heart urchins, and the influence of the former (which do not burrow as deeply as heart urchins) on sediment community respiration and biogeochemical fluxes is unknown. The sand dollar, Peronella lesueuri, is a large (to 15 cm) conspicuous urchin common in marine and estuarine soft sediment habitats along much of the western Australian coastline from at least the Dampier Archipelago in the north (Marsh & Morrison 2004; Keesing et al. 2011) to Albany in the south (Clark 1938). In Cockburn Sound, where we collected specimens for this study, it may occur in densities of up to 6 individuals m⁻² (personal observation using SCUBA). P. lesueuri had a greater movement rate in the summer with mean of 5.3 cm h⁻¹ at day and 2.0 cm h⁻¹ at night, and it was estimated that a single P. lesueuri can bioturbate 0.1 m² day⁻¹ of sediment habitat (Yeo, Keesing & van Keulen 2013). As P. lesueuri is the dominant macroinvertebrate in the habitats they occupy, they are likely to be important ecologically but this is yet to be demonstrated.

The aim of this study was to determine the interactions among the behavior (feeding and burrowing activities), physiology (respiration and excretion) of P. lesueuri and MPBs photosynthesis, and sediment metabolic rates, and to clarify the connection
between its behavior and physiology. We performed mesocosm studies at three different temperatures to test the null hypothesis that bioturbation activity by *P. lesueuri* has no effect upon MPBs productivity.

**Materials and methods**

**Collection and acclimation**

Sand dollars, *P. lesueuri* ranging from 62 to 211 g, and the native sandy sediment were collected by SCUBA from 4 to 5 m depth in Cockburn Sound 32.1536 S, 115.75083 E, southwest of Perth on the west coast of Australia on 6 March 2009. Divers collected sediment by scooping the top 2–3 cm from areas of the seabed showing visible evidence of a surface layer of benthic microalgae or MPBs. After collection, the sand dollars were immediately placed into 1 m³ plastic holding tanks with flowing seawater. Within 1 h, they were transferred to (400 l) temperature acclimation tanks with a 3 cm layer of sandy sediment. The tanks were supplied with recirculated seawater passed through a biological filter. Acclimation and experimental temperatures (23 and 16 °C) were similar to summer and winter water temperatures in Cockburn Sound. Healthy and more active individuals were used for the experiments by selecting the individuals who burrowed immediately into the sediment. A photoperiod of 12 h light and 12 h dark was maintained at all times. The light intensity was $\sim$450 μmol m⁻² s⁻¹ during the simulated daylight experimental periods.

**Experimental measurements**

The experiments were conducted in two 400 l tanks with a bottom covering of sediment to 3 cm depth containing MPBs that had been acclimating for at least 24 h before experiments were conducted. Each tank had four respiration chambers equipped with a magnetic stirring bar (Volume = 6283 ml, Diameter = 20 cm, Height = 20 cm, Base surface area = 314 cm²). Pairs of 12 h experiments were conducted on consecutive days, first in light and the second in dark. The same sediment was used for each light/dark pair of experiments. In each 12 h experiment, there were three 4 h periods. In an initial ‘Before’ period, empty respiration chambers were carefully inserted into the sediment so as not to disturb the MPBs controls and used to measure the background oxygen and nutrient fluxes of sediment and MPBs. In the second 4 h period (‘Experiment’), a single sand dollar was introduced to two of the chambers in each tank to measure the interaction between sand dollars and MPBs (and the other two chambers in each tank serve as controls). The control chambers were treated in the same manner as the experimental treatments and because the experimental chambers had to be lifted from the sediment to insert the sand dollar, the controls were lifted as well. In the final 4 h ‘Recovery’ period, sand dollars were removed to measure the ongoing effect of the bioturbation by the sand dollars. Again, the same level of disturbance from handling the chambers was applied to both treatments and controls. During each 12 h experiment, hourly measurements of oxygen consumption rate (OCR, in mg h⁻¹) and ammonia –N excretion rate (AER, in μg h⁻¹) were conducted by taking a sample with a syringe from each chamber and the tanks containing the chambers.

During the ‘Recovery’ period, the four sand dollars that had been removed from the chambers were placed in 4 l beakers, which were sealed with plastic film. OCR and AER in the absence of the sediment were calculated for each sand dollar from the rate
of change of oxygen and ammonia in the beaker. The paired light/dark experiments were conducted at 16 and 23 °C. These could not be conducted at different temperatures simultaneously, so they were repeated one after the other. The wet weight, body size (maximum length along oral/anal axis), and area of sand dollars were recorded after all experiments.

Feeding and burrowing behavior of *P. lesueuri*

During the experiments, feeding behavior of *P. lesueuri* was observed by disturbance of the sediment resulting in a change in color to gray inside the experimental chambers (containing a sand dollar) compared to sediment surfaces having a greenish brown coloration in the control chambers. After the experiments finished, sediment samples both inside and outside of the chamber were analyzed for chlorophyll *a* and phaeopigments to determine the effect of feeding activities on MPBs biomass. Observations of the burrowing behavior of *P. lesueuri* were also made during all experiments. However, it was hard to measure the distance and movement rates of *P. lesueuri* due to the relatively small respiration chambers used in the present work (Diameter = 20 cm, Base surface area = 314 cm²).

Sand dollar excretion and respiration rates

In order to measure the background oxygen and nutrient fluxes of sand dollar’s OCR and AER, 4–5 h experiments without sand sediment were conducted. Sand dollars were collected at the same place and time and handled as previously described. To simulate seasonal variations in temperature of the study area, south-west Australia, acclimation temperatures and experimental temperatures were 16, 19, and 23 °C, respectively. One sand dollar was added to each bucket (4.27 l) brim full of fresh seawater and carefully sealed with plastic film so as not to trap air bubbles. According to the different number of sand dollars available, three controls (without sand dollars) and 12 different body-sized individuals were incubated at 16 °C, 5 controls and 32 different body-sized individuals were incubated at 19 °C, and 3 controls and 30 different body-sized individuals were incubated at 23 °C.

Oxygen consumption rate (OCR, in mgO₂ h⁻¹) and ammonia –N excretion rate (AER, in μmoles NH₄ l⁻¹ h⁻¹) were determined from oxygen and ammonium samples collected at the beginning and end of each incubation. Samples were collected with a syringe after gently shaking the bucket to mix its contents. DO was measured using a YSI 5100 DO meter. Duplicate samples of incubation chamber and tank water taken for analysis of ammonia and nitrate were frozen immediately. A fluorescence detector coupled to a flow injection analyzer was used to analyze ammonium.

OCR and AER were calculated for each sand dollar from the before and after conditions of the water in the bucket. The wet weight and body size (maximum length of oral/anal axis) were recorded after all experiments. NO₃ + NO₂ and phosphate fluxes were also measured during the experiments to test the bioturbation activity by *P. lesueuri* upon MPBs productivity.

Data analysis

The relationships between sand dollar respiration, sand dollar size, and temperature were analyzed using analysis of covariance (ANCOVA) and principal component analysis (PCA). Each experiment was analyzed separately. Analysis of variance and
Kruskal–Wallis H test were used to determine the significance of variation among means of all measured variables. Multiple-regression analysis was used to identify the importance of the variables on oxygen consumption. A significance level of 5% was used in all analyses. OCR and AER were determined using the following formulae.

\[
OCR = \frac{[(DO_0 - DO_t)_{\text{treatments}} - (DO_0 - DO_t)_{\text{controls}}] \times V}{(W \times t)}(1)
\]

where OCR: oxygen consumption rate; DO₀: the oxygen concentration in water at the start of experiment; DOₜ: the oxygen concentration in water at the end of experiment; \( V \): the water volume of experiment; \( W \): the wet weight of sand dollar; \( t \): the experimental time duration.

\[
AER = \frac{[(N_t - N_0)_{\text{treatments}} - (N_t - N_0)_{\text{controls}}] \times V}{(W \times t)}(2)
\]

where AER: ammonia excretion rate; N₀: the ammonia concentration in water at the start of experiment; Nₜ: the ammonia concentration in water at the end of experiment; \( V \): the water volume of experiment; \( W \): the wet weight of sand dollar; \( t \): the experimental time duration.

**Results**

*Background oxygen and nutrient fluxes of sand dollar OCR and AER*

There was a statistically significant increase in OCR with wet weight at all temperatures. The slope of the OCR–wet weight relationships was similar at 16 °C (0.92) and 19 °C (0.89), but lower at 23 °C (0.58). OCR at 16, 19, and 23 °C was significantly different \((F = 7.76, p < 0.05)\). Further post hoc tests showed that the OCR was not significantly different between 16 and 19 °C \((\text{LSD, least-significant difference, } p > 0.05)\), but was significantly lower at 23 °C \((\text{LSD, } p < 0.05)\). The relationship between OCR and wet weight as well as temperature can be expressed by the following multiple linear regression equation:

\[
OCR = -0.593 + 0.004 W + 0.040 \text{ temperature } (p < 0.05)
\]

with the multiple \( R \) of 0.79 explaining 98.7% of all variation (PCA). There was no interaction effect between temperature and body weight.

Oxygen uptake also increased with body length \((0.46 < R^2 < 0.84)\) and was significantly lower at the lower exposure temperature \((F = 33.47, p < 0.05)\). The results of component score coefficient matrix (factor analysis) showed that the score of length (0.50) was similar to weight (0.51) which indicated that there were no differences between body size and weight in explaining variation of OCR. The multiple linear regression equation of OCR to body length and temperature is as follows:

\[
OCR = -0.91 + 0.009 L + 0.14 \text{ temperature } (p < 0.05)
\]

with the multiple \( R \) of 0.79, explaining 98.7% of all variation (PCA). There was also no interaction effect between temperature and body length.

AER–wet weight relationship was similar at 16 and 23 °C with AER increasing slightly with wet weight (slope of 0.50 at 16 °C and 0.56 at 23 °C). At 19 °C, AER decreased slightly with increasing wet weight (slope of −0.33). Mean AER at this temperature was 3.7 μmoles NH₄₁⁻h⁻¹ and significantly higher than 23 °C treatment (2.16 μmoles NH₄₁⁻h⁻¹) and 16 °C treatment (1.36 μmoles NH₄₁⁻h⁻¹) \((F = 29.62, p < 0.05)\).
Effect of the presence of sand dollars on benthic respiration and primary production

In the dark at 23 °C, the DO fluxes in treatment groups (with animals) were all negative but increased slightly between the ‘before’ (−4.55 mmol m⁻² h⁻¹) and ‘experiment’ periods (−4.70 mmol m⁻² h⁻¹) and the ‘recovery’ period (−3.65 mmol m⁻² h⁻¹). The difference between ‘experiment’ vs. ‘recovery’ periods was significant (\( F = 6, p < 0.05 \)), while ‘before’ vs. ‘recovery’ and ‘before’ vs. ‘experiment’ were not significant (\( p > 0.05 \), Wilcoxon rank sum test) (Figure 1(a)). In the dark at 16 °C, the results in the treatment group were significantly lower in the ‘experiment’ periods (\( F = 18.15, p < 0.05 \)); however, the flux rates were higher and less variable among three periods than the status at 23 °C (\( F = 58.79, p < 0.05 \)) (Figure 4(c)). DO flux value in control group (without animals) had a similar distribution to the treatments with animals but in this case, the differences between the three periods were not significant at both 23 °C (\( F = 0.07, p > 0.05 \)) and 16 °C (\( F = 1.1, p > 0.05 \)) (Figure 1(a) and (c)).

![Figure 1](image.png)

Figure 1. The oxygen production and consumption interaction between sand dollar’s activities and microphytobenthic production response.
Note: Panels (a) and (b), (c) and (d) are from experiments in March 2009 at different temperatures of 16 and 23 °C, respectively. Positive values indicate efflux of materials out of the sediment; negative values indicate influx into sediment. Before, experimental treatment and recovery are the three periods of each 4 h set up in sequence. In Before period, without animals; in experimental treatment, four individuals introduced and four controls; in recovery period, four individuals removed, without animals again. Error bars show the standard deviations for measurements that we made in duplicate chambers. With animals-treatment groups; without animals-control groups.
In the light, the DO flux in experimental groups was very different between the three periods. At 23 °C, the presence of the animals reduced DO production from 3.50 mmol m$^{-2}$ h$^{-1}$ in the ‘before’ the introduction of animals to −1.29 mmol m$^{-2}$ h$^{-1}$ in the ‘experiment’ period. There was a slight increase in DO flux after the animals were removed (to −0.45 mmol m$^{-2}$ h$^{-1}$ in the ‘recovery’ period) but well short of the production rates encountered in the before treatment. The difference between the three periods was statistically significant ($F = 41.10, p < 0.05$) (Figure 4(b)). At 16 °C, the differences between the periods were similar to 23 °C but the flux values were lower and with less variability between replicates ($F = 6.28, p < 0.05$) (Figure 4(d)). In the control tanks (without animals), the oxygen fluxes were positive during all irradiated periods at both 23 and 16 °C (Figure 4(b) and (d)). Slightly reduced oxygen flux in the recovery treatment was not different to both the ‘Before’ and ‘Experiment’ periods at both 23 °C (chi-square = 1.12, $p > 0.05$) and 16 °C (chi-square = 1.85, $p > 0.05$) (Figure 1(b) and (d)).

To test for potential bias due to differences between the individual sand dollars used in the experiments, DO fluxes of the eight individuals used were measured individually, with the average value of DO flux $-1.51 \pm 0.599$ mmol O$_2$ m$^{-2}$h$^{-1}$ (Table 1). To test the effect of sand dollars on DO flux (23 °C, dark), the difference between treatment groups and control group of three periods was analyzed yielding the following results: ‘before’ period ($F = 0.004, p > 0.05$); ‘experiment’ period ($F = 4.56, p < 0.05$); ‘recovery’ period ($F = 0.18, p > 0.05$). In the light at 23 °C, the DO flux was also significantly different between treatment and control groups during the ‘experiment’ period ($F = 9.34, p < 0.05$). The results demonstrated that the sand dollar decreased the DO flux in the experiment period. At 16 °C, the sand dollar also lowered the DO fluxes during the experiment period (in the dark: $F = 13.8, p < 0.05$; light: $F = 31.8, p < 0.05$).

### Table 1. DO flux of eight sand dollars used in experiments a & b, c & d (refer Figure 4).

<table>
<thead>
<tr>
<th>Experiment/sand dollar chamber</th>
<th>Body area (shading area) (mm$^2$)</th>
<th>DO flux (mmol O$_2$ m$^{-2}$h$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>a &amp; b/1</td>
<td>92.5</td>
<td>−2.05</td>
</tr>
<tr>
<td>a &amp; b/2</td>
<td>114</td>
<td>−2.36</td>
</tr>
<tr>
<td>a &amp; b/5</td>
<td>95</td>
<td>−2.05</td>
</tr>
<tr>
<td>a &amp; b/6</td>
<td>96</td>
<td>−1.59</td>
</tr>
<tr>
<td>c &amp; d/1</td>
<td>93</td>
<td>−1.31</td>
</tr>
<tr>
<td>c &amp; d/2</td>
<td>93.5</td>
<td>−1.10</td>
</tr>
<tr>
<td>c &amp; d/5</td>
<td>109.5</td>
<td>−0.70</td>
</tr>
<tr>
<td>c &amp; d/6</td>
<td>91.7</td>
<td>−0.93</td>
</tr>
</tbody>
</table>

### Effect of the presence of sand dollars on the nutrient fluxes

#### Ammonia

In the dark at 23 °C, the ammonia fluxes were highest during the experimental period in both the control ($F = 4.30, p < 0.05$) and treatment groups ($F = 3.91, p < 0.05$) (Figure 2(a)). At 16 °C, the ammonia fluxes were less variable. Ammonium flux was highest when the animals were present and reduced during the recovery period relative to both the initial and treatment periods ($F = 11.57, p < 0.05$) whereas, in the dark, the control group there was a gradual but significant reduction in ammonium flux across
the periods from 0.89 µmol m$^{-2}$ h$^{-1}$ in the ‘before’ which decreased to −17 µmol m$^{-2}$ h$^{-1}$ in the ‘experiment’ period and −48 µmol m$^{-2}$ h$^{-1}$ in the ‘recovery’ period ($F = 15.88, p < 0.05$ (Figure 2(c))).

In light at 23 °C, the ammonia flux in three periods was lowest during the experimental period in both the control ($F = 42.47, p < 0.05$) and treatment groups ($F = 128.27, p < 0.05$) (Figure 2(b)). At 16 °C, absolute differences were much less but the ammonium flux was highest during the experimental period in both the control ($F = 6.74, p < 0.05$) and treatment groups ($F = 9.87, p < 0.05$) (Figure 2(d)).

At 16 °C, the sand dollar also increased the ammonia fluxes during the experimental period (in the dark: $F = 12.6, p < 0.05$; light: $F = 9.45, p < 0.05$). At 23 °C, however, the effect of sand dollar on the ammonia fluxes was not significant (in the dark: $F = 0.64, p > 0.05$; light: $F = 0.56, p > 0.05$).

$NO_3 + NO_2$ fluxes
In the dark at 23 °C, the $NO_x$–N flux of treatment groups was negative during all three periods, with the lowest values in the ‘experimental’ period (Figure 3(a)). In this case, the
control group also had the lowest flux during the ‘experimental’ period but the NO$_x$–N efflux during the ‘Before’ and ‘Recovery’ period was positive. At 16 °C, the NO$_x$–N efflux in both groups significantly increased throughout the experiment ($F = 43.59$, $p < 0.05$ for treatment; $F = 104.0$, $p < 0.05$ for control group); however, there were no differences between the treatment or the controls ($F = 0.34$, $p > 0.05$) (Figure 3(c)).

In the light, the NO$_x$–N flux was different to the status in darkness, with lowest flux value in the ‘experimental’ period than that of in the ‘before’ and ‘recovery’ period (Figure 3(b) and (d)).

The sand dollar had less obvious effect on NO$_x$–N fluxes across sediment–water interface at both 23 and 16 °C (23 °C, dark: $F = 0.7$, $p > 0.05$; light: $F = 0.18$, $p > 0.05$. 16 °C, dark: $F = 1.12$, $p > 0.05$; light: $F = 5.4$, $p > 0.05$).

**Phosphate fluxes**

In the dark at 23 °C, the phosphate flux in the presence of the animal was higher than the ‘before’ period and was higher again during the ‘recovery’ period after the animals were removed. In the control groups, phosphate flux was lower during the ‘experimental’ period (phosphate was consumed) than the ‘before’ and ‘recovery’ periods (Figure 4(a)). In the light, both the treatment and control groups decreased steadily from ‘before’ period to ‘recovery’ period, namely, from a positive flux to a negative flux in the treatments and a negative flux to a lower negative flux in the control group.
Figure 4. Sand dollar *P. lesueuri* effect on phosphate fluxes. Note: Panels (a) and (b), (c) and (d) are from experiments in March 2009 at different temperatures of 16 and 23 °C, respectively.

(Figure 4(b)). In the dark at 16 °C, the phosphate flux in two groups was less variable, except for the high positive flux in the treatment group during the ‘experimental’ period (Figure 4(c)). In the light, two groups showed a similar steady decrease in phosphate flux during the ‘experimental’ period to the status at 23 °C with the exception of a large decrease in phosphate flux in the control during the experimental period. Except for the negative flux in the control during the experimental period, phosphate efflux was positive (Figure 4(d)).

**Feeding and burrowing behavior of *P. lesueuri***

Chlorophyll *a* in sediments was not different between ‘inside’ and ‘outside’ the chambers (*p* > 0.05, paired-samples test), between treatments ‘with’ and ‘without’ sand dollars (*p* > 0.05, paired-samples test) and between light and dark incubations (*p* > 0.05, independent-samples t test) at 23 and 19 °C (Figure 5(a)–(h)).

At 23 °C, phaeopigments ‘inside’ chambers were always lower than ‘outside’ chambers. However, the large range of values means this difference cannot be regarded as being of ecological significance (Figure 5(i)–(p)).

All the sand dollars burrowed immediately into sediment to a depth of 1–3 cm inside the chambers before the beginning of experiments and showed an obvious moving trace on the sandy sediment surface.
The relationship of metabolic rates of sand dollar with body weight, body size, and temperature

In all invertebrates, metabolic rates are one of the most important components of the energy budget and are affected by numerous internal and external factors. Two factors are animal size (Kleiber 1932) and temperature (Sabourin & Stickle 1981; Marsden 1999; Zhuang & Liu 2006). As expected, oxygen uptake for *P. lesueuri* increased significantly with wet weight and body size at all temperatures and overall oxygen consumption was significantly reduced at the lower exposure temperatures. In contrast, mean AER was not linearly related to temperature and was higher at 19 °C than at 23 and 16 °C. In general, we would expect both OCR and AER to vary with temperature in a similar manner as higher metabolism generates more waste products. While the causes of the difference between the temperature dependence of metabolism (as indicated by respiration/oxygen consumption) and ammonium excretion are not immediately clear it may be related to processes that are independent of the organisms themselves. Of which, the feeding condition of the previous day might be very important in the

Figure 5. Results of Chlorophyll *a* and Phaeopigments in sediment at the end of experiment (23 & 16 °C).

Note: Panels (a)–(b) are the results of Chlorophyll *a* from experiments in March 2009 at different temperatures of 16 and 23 °C; panels (i)–(p) are the results of Phaeopigments from experiments at different temperatures of 16 and 23 °C, respectively. Error bars show the standard deviations for measurements that we made in duplicate chambers. With animals-treatment groups; without animals-control groups.

Discussion

The relationship of metabolic rates of sand dollar with body weight, body size, and temperature

In all invertebrates, metabolic rates are one of the most important components of the energy budget and are affected by numerous internal and external factors. Two factors are animal size (Kleiber 1932) and temperature (Sabourin & Stickle 1981; Marsden 1999; Zhuang & Liu 2006). As expected, oxygen uptake for *P. lesueuri* increased significantly with wet weight and body size at all temperatures and overall oxygen consumption was significantly reduced at the lower exposure temperatures. In contrast, mean AER was not linearly related to temperature and was higher at 19 °C than at 23 and 16 °C. In general, we would expect both OCR and AER to vary with temperature in a similar manner as higher metabolism generates more waste products. While the causes of the difference between the temperature dependence of metabolism (as indicated by respiration/oxygen consumption) and ammonium excretion are not immediately clear it may be related to processes that are independent of the organisms themselves. Of which, the feeding condition of the previous day might be very important in the
process. It presumably needs more than one day for the process from feeding to discharging, so that the present amount of waste products would not correlate with the present metabolism rate. Normally, the bacterial nitrification (the conversion of ammonium to nitrate) is most efficient at the higher temperature (Frederick 1956; Myers 1975). However, in the present work, the mean AER attained the peak value at 19 °C rather than at 23 °C, which seems to be contradictory to the above studies. The reason might be complicated due to many environmental variables and biological factors involved during the experiment period. Combined factors might play more import roles under this condition rather than a single one alone. Efficient conversion of ammonium to nitrate would act to reduce the observed ammonium excretion rate. However, identifying such a process is beyond the scope of this survey and will require a dedicated investigation.

Effect of the presence of sand dollars on the MPBs primary production and respiration
In the present paper, the DO flux is obviously different in dark and light ($F = 36.4, p < 0.05$), which was mostly due to the net primary production by the MPBs in the light treatments. In dark, the DO flux of the MPBs, sand dollar, and sediment is all negative; while in the light without sand dollar, the MPBs produce enough O2 to compensate for the O2 consumed by the heterotrophs in the sediment, resulting in a positive DO flux.

In the treatments with animals, burrowing and shading as well as O2 consumption by the animals themselves yielded a significantly negative DO flux across the sediment–water interface both at 23 and 16 °C.

In the light treatment, the flux in the control during the ‘experiment’ period was 4.2 mmol O2 m$^{-2}$ h$^{-1}$ at 23 °C and 1.6 mmol O2 m$^{-2}$ h$^{-1}$ at 16 °C. The average consumption by the animals used in each experiment was $-2.0$ and $-1.0$ mmol O2 m$^{-2}$ h$^{-1}$ at 23 and 16 °C, respectively. The animals themselves shade around 30% of the sediment in each chamber. Assuming this shading reduces the DO produced by 30%, we would expect a further reduction of 1.3 mmol O2 m$^{-2}$ h$^{-1}$ at 23 °C and 0.49 mmol O2 m$^{-2}$ h$^{-1}$ at 16 °C. Therefore, the combined contribution of animal respiration and shading to the difference observed between the control and animal treatments amounts to $-3.3$ and $-1.5$ mmol O2 m$^{-2}$ h$^{-1}$ at 23 and 16 °C, respectively. This amounts to around 68% of the observed difference between the control and the treatment at 23 °C and 48% at 16 °C. The difference between the temperatures reflects the greater importance of respiration relevant to production at the higher temperature. Among factors regulating MPBs production in silty sediment, temperature has been shown to play an important role in determining the rate of productivity (Barranguet et al. 1998).

Effect of the presence of sand dollars on the nutrient recycling and fluxes
Ammonium (NH$_4^+$) is an animal excretory product which can be used as a nutrient by marine MPBs at the sediment surface. Echinocardium has positive effect on NH$_4$–N efflux from sediments in darkened chambers and provides an important ecosystem service by supplying overlying waters with a readily utilizable form of inorganic nitrogen (Lohrer et al. 2004). In light at 23 °C, the ammonia flux was highly negative in the ‘treatment’ period and much lower than the ‘before’ period. However, this large reduction between the ‘before’ and ‘treatment’ periods was observed in both treatment and
control groups suggesting it is an experimental artifact. As a result, we must be cautious about how we interpret these results. There is, however, an indication of a positive effect of sand dollar activity from sediments in the ‘recovery’ period in light at 23 °C. The NH$_4$–N flux rates in the treatment recovering from the presence of sand dollars were significantly higher than the control during that period. In the light at 16 °C, the NH$_4$–N effluxes were negative in all three periods, and there was little variation between the controls across the three periods. In the treatment group, the NH$_4$-N efflux in the ‘experiment’ period was significantly higher than the ‘before’ period and corresponding control suggesting that ammonia excretion by the animal was greater than uptake of NH$_4$–N by the microphyte community and nitrification by bacteria at this lower temperature.

In dark at 23 and 16 °C, NH$_4$–N efflux in the sand dollar treatment was higher in the ‘experiment’ period than the ‘recovery’ period. In the controlled experiment, there was a gradual decrease in NH$_4$–N efflux from the ‘before’ to ‘recovery’ period at 16 °C but a similar trend to the treatment groups at 23 °C. Differences in the range of the ammonium fluxes between the temperatures in both light and dark reflect the higher metabolism of the sand dollars and more rapid remineralization of organic carbon at higher temperatures. NO$_3$ + NO$_2$ and phosphate efflux were variable across treatments and periods, and the animal treatments tended to vary in concert with the controls. As the fluxes were small (an order of magnitude lower than the ammonium fluxes) and did not indicate a consistent animal effect they probably reflect patchiness in the system rather than the influence of 

The impact from the actions of urchins on primary production is different depending on the urchin species. In the present study, the sand dollar 

Echinocardium excavates the front sediment and moves it backwards, accumulating it on its back, which results in mixing of the sediment, and because of the globular test shape the spatangoid must excavate much larger cross area than sand dollars to move through sediment. Thus, there is a remarkable difference in burrowing between sand dollars and spatangoids. Secondly, the urchins also affect the primary production by shading the surface MPBs with different body shape. The sand dollar has a flat shape with bigger shade area than globular shape of spatangoid heart urchins.
Another burrowing heart urchin, *B. lyrifera*, is a non-selective, infaunal deposit feeder. By ‘bulldozing’ through the sediment, *B. lyrifera* has a major impact on sediment structure and stability (Widdicombe et al. 2000).

Burrowing behavior of sand dollars (locomotion) could mix the sediments and cause bioturbation, which has major impact on biogeochemical cycling at the seabed (Lohrer et al. 2004). In the present work, due to the fact that the sand dollars had a negative effect on the productivity of the MPBs and that the sediment chlorophyll *a* and phaeopigment concentrations were not different between ‘inside’ and ‘outside’ the chambers and treatments ‘with’ and ‘without’ sand dollars, this negative effect is most likely due to the sand dollars’ bioturbation on MPBs rather than as a result of consuming them. Lohrer et al. (2004) also found no correlation between *Echinocardium* density and chlorophyll *a* in surface sediments.

Nevertheless, *P. lesueuri* has the potential to significantly impact benthic primary productivity given their high density, rates of movement, and large area of sediment reworked (0.1 m² day⁻¹ per urchin, Yeo et al. 2013). The relationship between wet weight and temperature suggests that this effect may be dependent both upon seasonal variation and animal density; however, further tests, preferably under field conditions are required to confirm this hypothesis. The relationship between bioturbation by *P. lesueuri* and nutrient fluxes is unclear; however, it is suggested that *P. lesueuri* affects microbial redox processes, which could alter the exchange of nutrients across the benthic–pelagic interface.

**Conclusions**

The respiration and excretion rates of the sand dollar *P. lesueuri*, as well as their influence on benthic metabolism, were studied in this work. The following conclusions have been made:

1. Oxygen consumption by sand dollars increased significantly with wet weight at all three temperatures 16, 19, and 23 °C. Ammonia release also increases with body weight.
2. The weight vs. oxygen uptake relationship was similar at 19 and 23 °C but oxygen uptake was significantly reduced at the lower exposure temperature.
3. The bioturbation caused by sand dollar *P. lesueuri* reduced the photosynthetic rate of the MPBs but had a smaller influence on nutrients fluxes across the sediment-water interface.
4. The effect of the bioturbation caused by sand dollars was different from spatangoid heart urchins as a result of its unique burrowing and feeding behavior, body shape, and physiology.

**Acknowledgements**

We thank Stelios Kondylas, Douglas Bearham, and Ryan Crossing for assistance with sand dollar and sediment collections and Jennelle Ritchie and Tennille Irvine for assistance with respiration measurements. The manuscript benefited from a critical review by Dr Bonnie Laverock.
Funding

This work was funded by the CSIRO Wealth from Ocean’s National Research Flagship, the Western Australia Marine Science Institution, and the Yantai Institute of Coastal Zone Research, Chinese Academy of Science.

References


