Dissolved oxygen fluxes and ecosystem metabolism in an eelgrass (*Zostera marina*) meadow measured with the eddy correlation technique

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Abstract

Dissolved oxygen (DO) fluxes were measured by eddy correlation to estimate net ecosystem metabolism (NEM) during summer in a restored eelgrass (*Zostera marina*) meadow and a nearby, unvegetated sediment. This technique measures benthic fluxes under true in situ light and hydrodynamic conditions, integrates over a large area (typically > 100 m²), and captures short-term variations. DO fluxes measured through eight 24-h periods showed pronounced temporal variation driven by light and local hydrodynamics on multiple scales: hour-to-hour, within each daily cycle, and between deployments. The magnitude of variation between hours during single deployments equaled that between deployments, indicating that short-term variation must be included for metabolism estimates to be accurate. DO flux variability was significantly correlated to mean current velocity for the seagrass site and to significant wave height for the unvegetated site. Fluxes measured in low-flow conditions analogous to many chamber and core incubations underestimated those measured in higher-flow conditions typical of in situ conditions by a factor of 2–6. Rates of gross primary production (GPP), respiration (R), and NEM varied substantially between individual deployments, reflecting variations in light and hydrodynamic conditions, and daily values of GPP and R for individual deployments were tightly linked. Average daily NEM of the seagrass site was higher than that of the unvegetated site; the seagrass site was in metabolic balance, and the unvegetated site showed a tendency toward net heterotrophy during this midsummer period.

Benthic metabolism is the key component of overall system metabolism and nutrient cycling in shallow bays and lagoons where most of the seafloor lies within the photic zone (McGlathery et al. 2007). Benthic autotrophs, including seagrasses and micro- and macroalgae, fix substantial amounts of carbon, are a temporary sink for nutrients, and influence bacterial processes in the sediment (e.g., mineralization, nitrification–denitrification, anaerobic ammonium oxidation [Annamox]) by modifying redox conditions and competing with bacteria for limiting nutrients. The status of the benthos in shallow coastal systems as a source or a sink of carbon and nutrients is largely dependent on the metabolism of these primary producers and the associated heterotrophs in the community (Eyre and Ferguson 2005; Sundback and McGlathery 2005; McGlathery 2008). Seagrasses are the foundation of benthic communities in many shallow systems, and their metabolism often exceeds that of nearby unvegetated sediments by several fold (Barron et al. 2006; Stutes et al. 2007; Apostolaki et al. 2010). However, accurate estimates of metabolism are difficult to obtain using conventional methods because of the relatively large stature of seagrasses, their often dense root system, the complex hydrodynamics associated with the canopy structure, and the spatial heterogeneity characteristic of these communities.

There are many methods that have been used to determine benthic metabolism, including extrapolation from photosynthesis–irradiance curves (Kraemer and Alberte 1993), changes in primary producer biomass (Hasegawa et al. 2007), mass balance models (Kemp et al. 1997; Kaldy et al. 2002), diel changes in water-column concentrations of either dissolved oxygen (DO) or dissolved inorganic carbon ("open-water" method; D'Avanzo et al. 1996; Ziegler and Benner 1998), and direct benthic flux measurements of DO or dissolved inorganic carbon in either in situ chambers or laboratory incubations of substrate cores (McGlathery et al. 2001; Gazeau et al. 2005; Yarbro and Carlson 2008). These conventional methods all have significant limitations. For example, in situ chambers or laboratory-incubated cores may bias flux measurements, especially in permeable sediments, because they do not replicate natural hydrodynamic forcing and light levels (Cook and Røy 2006; Berg and Huettel 2008). Core incubations are typically done at a single irradiance level or in complete darkness to determine daytime and nighttime metabolism, and flow rates are generally very low compared to in situ conditions. The chamber incubation technique captures some of the natural variation in light, although the chamber itself reduces light penetration, and flows are also generally lower than in situ conditions (Tengberg et al. 2004). For the open-water method, there are inaccuracies associated with difficulties in correcting for oxygen exchange across the air–water interface and changes in oxygen concentrations due to horizontal flow (Ziegler and Benner 1998). Studies comparing the different methods have shown that the rates of ecosystem metabolism can vary significantly depending on the type of method used due to these problems (Kemp and Boynton 1980; Ziegler and Benner 1998).

In this study, we used the eddy correlation technique (Berg et al. 2003) to determine benthic DO fluxes for two benthic communities—a temperate seagrass meadow, and, for reference, a nearby "unvegetated" sediment that contained benthic microalgae. We used these fluxes to estimate gross primary production (GPP), community

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respiration (R), and net ecosystem metabolism (NEM) for these communities. In marine environments, the eddy correlation technique has thus far only been used to measure DO fluxes over cohesive and permeable unvegetated sediments (Berg et al. 2003; Kuwae et al. 2006; Berg et al. 2009).

The eddy correlation technique relies on measuring simultaneously the fluctuating vertical velocity and fluctuating oxygen concentration in the water column above the seafloor (5–30 cm) at a high temporal resolution (32–64 Hz). From these data, the DO flux between a benthic community and the overlying water can be extracted. While eddy correlation systems are more expensive and also are technically more challenging to use than traditional flux methods, they do have several significant advantages. Measurements can be made over vegetated or permeable sediments, and over hard surfaces such as coral reefs or mussel beds under true in situ conditions with no disturbance of the natural light and hydrodynamic conditions. Further, the "footprint" (the area of the sediment surface that contributes to the flux) covers a relative large area (typically > 100 m^2) and integrates more realistically the spatial heterogeneity inherent in benthic systems than replicate in situ chamber or core incubations (Berg et al. 2007). Finally, the high temporal resolution of the measurements allows us to capture short-term variations in benthic fluxes that are usually triggered by rapid variations in light and/or local hydrodynamic forcing (Berg and Huettel 2008).

The aims of this study were: (1) to use the eddy correlation technique for the first time and in multiple 24-h deployments over a seagrass vegetated sediment and a nearby unvegetated sediment, and (2) to compare the results to gain insight on the controls and dynamics of system metabolism at a higher temporal resolution than has been attempted before. The work was part of a larger study on the ecosystem-level consequences of a state change from unvegetated sediments to seagrass-vegetated sediments in Virginia coastal bays as a result of large-scale seagrass restoration.

Methods

Study site—The study sites were located in South Bay (37°15′13.86″N, 75°48′43.70″W), a shallow coastal bay on the eastern shore of Virginia that is typical of coastal bays on the U.S. Mid-Atlantic coast. South Bay is located within the Virginia Coastal Reserve (VCR) Long Term Ecological Research (LTER) site and is bordered by the Delmarva Peninsula to the west and barrier islands to the east. The depth of the bay is 1–2 m at mean low water, and it has a semidiurnal tidal range of ~ 1.5 m (Oertel 2001). There are no river inputs to the VCR coastal bays, and there is limited exchange with the coastal ocean through narrow inlets between the barrier islands. Up until the early 1900s, the seafloor of the Virginia coastal bays was carpeted with dense seagrass meadows that supported a prosperous scallop fishery. The bays underwent a dramatic state change in 1933 when a powerful hurricane caused the eelgrass (*Zostera marina*) populations that were already weakened by a pandemic disease (the slime mold,

Labyrinthula zosterae) to become locally extinct. The bays remained devoid of seagrasses for over 60 yr, until the discovery of a small (< 10 m^2) patch in South Bay in the late 1990s spurred a large restoration effort by seeding starting in 2001 (Orth et al. 2006). Restored seagrass meadows now cover some 13.5 km^2 in the VCR coastal bays (R. Orth pers. comm.; http://web.vims.edu/bio/sav/maps). The seagrass meadows were 5-yr old at the time of sampling, and the seagrass density at the site sampled was ~ 400–500 shoots m^{-2}. This meadow was compared with an unvegetated sediment site colonized by benthic microalgae located ~ 200 m away. The sediments of both sites were composed of fine sands, with < 10 wt% mud (Lawson et al. 2007). Sediment permeability at a nearby site was determined to be 2.5 × 10^{-11} m^2 in a separate study (Lawson 2008). Sediment organic content of the seagrass meadows sampled was 0.8 ± 0.02 (SE) and 1.0 ± 0.06 (SE) wt% for the bare sediments and showed no legacy of the prior seagrass-vegetated period. We believe this is because of the high rates of sediment redistribution in these shallow coastal bays (Lawson et al. 2007).

Field measurements—The eddy correlation instrument (Berg et al. 2003; Berg and Huettel 2008) included an acoustic Doppler velocimeter (ADV) (Nortek AS) modified to interface with a Clark-type oxygen microelectrode through a custom-built pico-amplifier (Max Planck Institute for Marine Microbiology). These sensors were mounted on a stainless-steel frame with the ADV aligned vertically facing the sediment surface and the microelectrode positioned at approximately a 45° angle with the tip on the edge of the ADV's ~ 1-cm^3 measuring volume. This volume was ~ 15 cm above the sediment surface at the unvegetated sediment site and ~ 25 cm above the sediment surface at the seagrass site. This is above the maximum canopy height of ~ 20 cm at low current. Measurements were made in intervals or bursts consisting of 64-Hz data recordings for 14.5 min followed by a 0.5-min "sleep" period. In addition to these high-speed recordings of the three velocity components (two horizontal and one vertical) and O$_2$ concentration, the ADV recorded temperature, water depth, and the measuring volume's position above sediment surface with a coarser temporal resolution. The duration of each deployment was approximately 24 h. Ambient DO was determined with a handheld YSI 550A membrane probe DO meter (YSI Inc.) at the beginning and end of each deployment to calibrate the microelectrode. The DO meter was calibrated using a two-point calibration curve, and it has a precision of ± 2%. Water surface irradiance (PAR) data were taken from a LTER meteorological tower located ~ 1 km away from the study site using a LI-COR quantum sensor (LI-COR Biosciences) measuring every 5 s. In total, eight deployments were made between 20 June and 19 July 2007. Because we only had one eddy correlation system at that time, measurements were alternated between vegetated and unvegetated sites. The eddy correlation system was always deployed and recovered during low tide.

Data processing—DO fluxes, one for each 15-min burst, were extracted from the high-resolution data (Fig. 1).
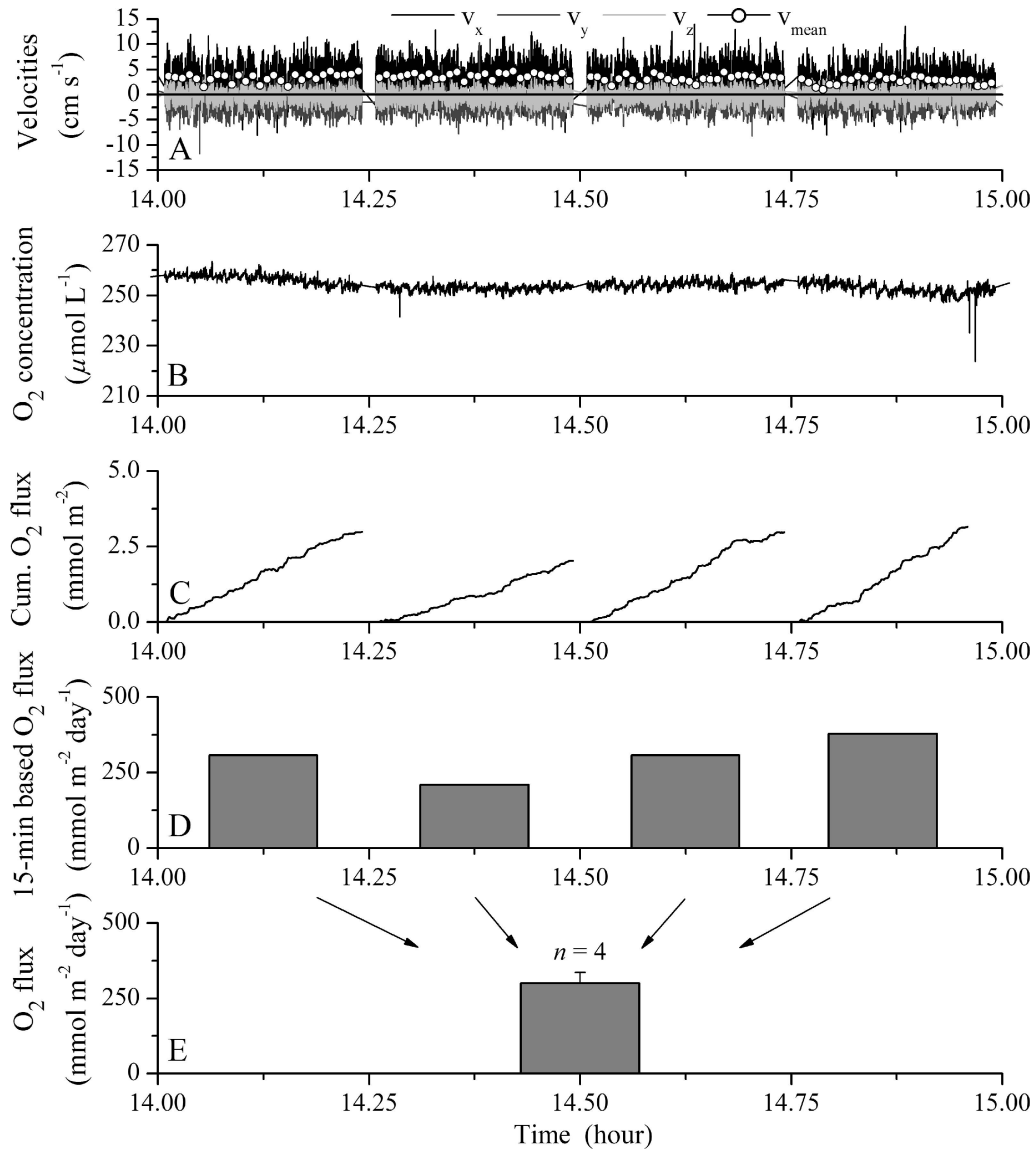
Fig. 1.   Raw eddy correlation data and derived fluxes through four 15-min-long bursts. (A) Three velocity components measured with high temporal resolution (64 Hz) and mean current velocity (30-s mean). The latter varied between 2 and 5 cm s^{-1}. (B) Measured high temporal resolution DO concentration. Noise is visible in both velocity and DO data. This noise is white (random and uncorrelated) and, hence, does not affect the flux calculation. The disturbance of the DO signal at the end of burst 4 was likely caused by debris temporarily attached to the sensor tip. Thus, the last part of the burst was not used in the flux calculation. (C) Calculated cumulative flux for each burst. The clear linear trend indicates a strong flux signal contained in the data and a statistically good representation of all eddy sizes that contribute to the flux. (D) Calculated flux for each burst. The positive values represent DO releases. (E) Averaged flux over the four bursts representing an hourly mean value (error bars represent SE). For each deployment, 24 hourly flux values were extracted and used in the further calculations.

Specifically, the eddy flux was defined as $\overline{u'_z C'}$, where u'_z is the fluctuating vertical velocity away from its mean, C' is the fluctuating DO concentration away from its mean, and the bar symbolizes an averaging over time (Berg et al. 2003). The two fluctuating components were separated from their means as $u'_z = u_z - \overline{u_z}$ and $C' = C - \overline{C}$, where u_z and C represent the raw measurements in each 14.5-min period, and the means, $\overline{u}_z$ and $\overline{C}$, were defined as least-square linear fits to u_z and C. The DO concentration, velocity, and cumulative flux in each burst were checked carefully for unnatural variations that would typically be

caused by floating debris temporarily attached to or obstructing the sensors (*see* example in Fig. 1). Bursts, or parts of bursts, with any such abnormalities were not used in the further data processing. For more details on extraction of the eddy flux from raw measurements, *see* Berg et al. (2009).

Strictly speaking, the eddy flux represents the vertical flux across a horizontal plane that contains the measuring point. How well this flux approximates the production or respiration of the benthic community below can be evaluated from a DO mass balance for a control volume
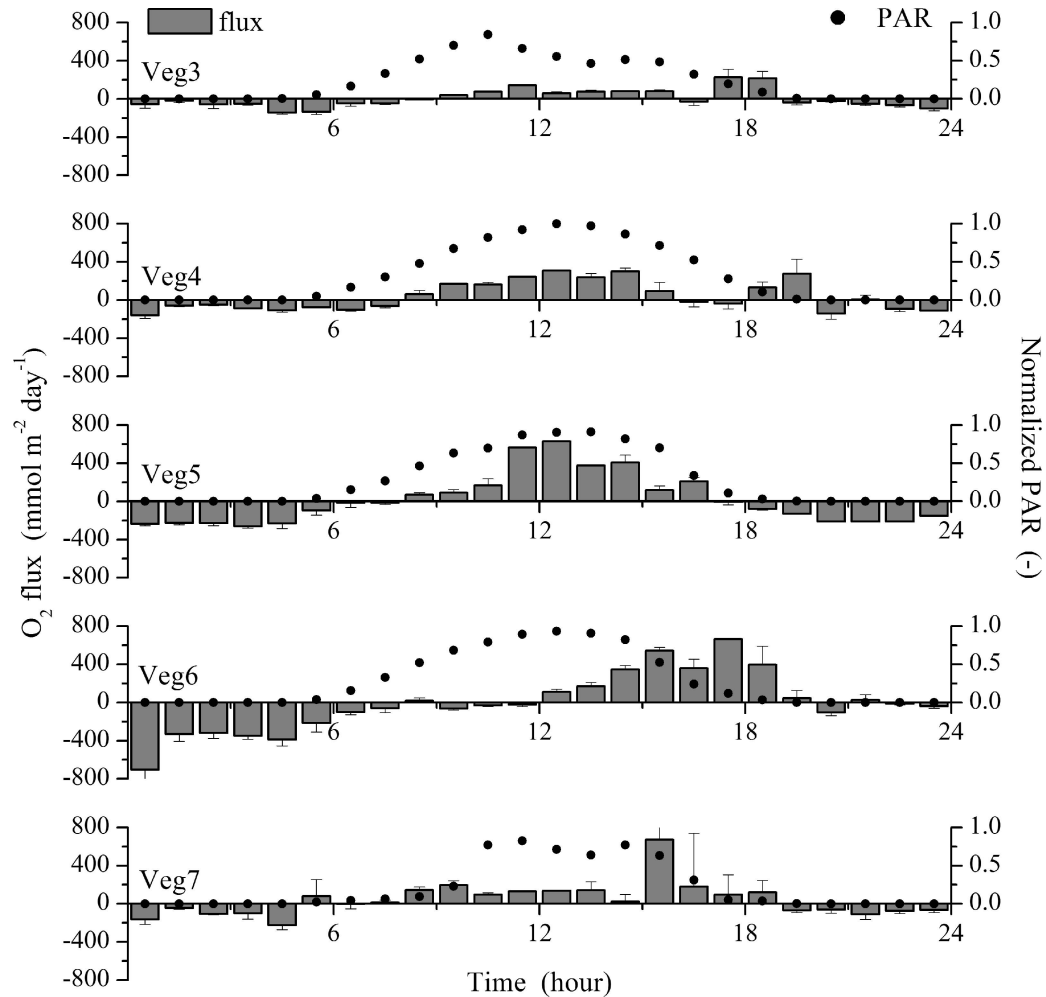
Fig. 2. Twenty-four hourly flux values and normalized PAR for each of the five deployments over the seagrass bed. Positive values represent DO release. Daylight exhibits a clear control on the flux, but other factors are clearly equally as important (*see* text for details).

that has this plane as its top and stretches downward into the sediment. It is assumed that there is no horizontal gradient in the bottom-water mean DO concentration, which confines the mass balance to one dimension, the vertical direction (Stull 1988). From this mass balance, the measured eddy flux equals the net result of production and respiration by the benthic community, plus the net result of any production and respiration by organisms suspended in the water within the control volume, minus any DO being stored in the volume. For most benthic systems, the production and respiration by suspended organisms such as microalgae are insignificant relative to those of the benthic communities (Berg et al. 2003). With respect to storage of DO, a correction can be made because the mean DO concentration of water within the control volume can be extracted from eddy correlation measurements. This correction is usually very small and not applied in underwater eddy correlation applications (Berg et al. 2003; Berg and Huettel 2008; Lorrai et al. 2010). Since most of our calculations in this study integrate the eddy flux over relatively long periods of time, or many individual eddy flux estimates, these integrated values represent a good proxy for the net DO production or respiration of the

benthic communities investigated. In addition, from the description of the control-volume mass balance, it should be clear that the air–water exchange of DO will not bias or distort the eddy flux measurement itself or its approximation of the benthic community production or respiration. This is a common misconception of the eddy correlation technique.

The eddy fluxes for the 15-min bursts were lumped into groups to give hourly fluxes, each having an associated standard error. An example of our raw data in the form of velocity components and DO concentration, derived fluxes for each burst, and the final lumped hourly flux is given in Fig. 1. In the few cases in which hourly flux values were missing to cover a full 24-h period, the missing hourly fluxes were determined by interpolation. All PAR data were similarly lumped into hourly values to match the hourly DO fluxes. Corresponding hourly values of DO flux and normalized PAR for all deployments are shown in Figs. 2 and 3. Because PAR was measured above water at a nearby location, we report PAR as normalized values (based on the highest value recorded in the eight deployments) and only use these to distinguish between "light" and "dark" periods of time.
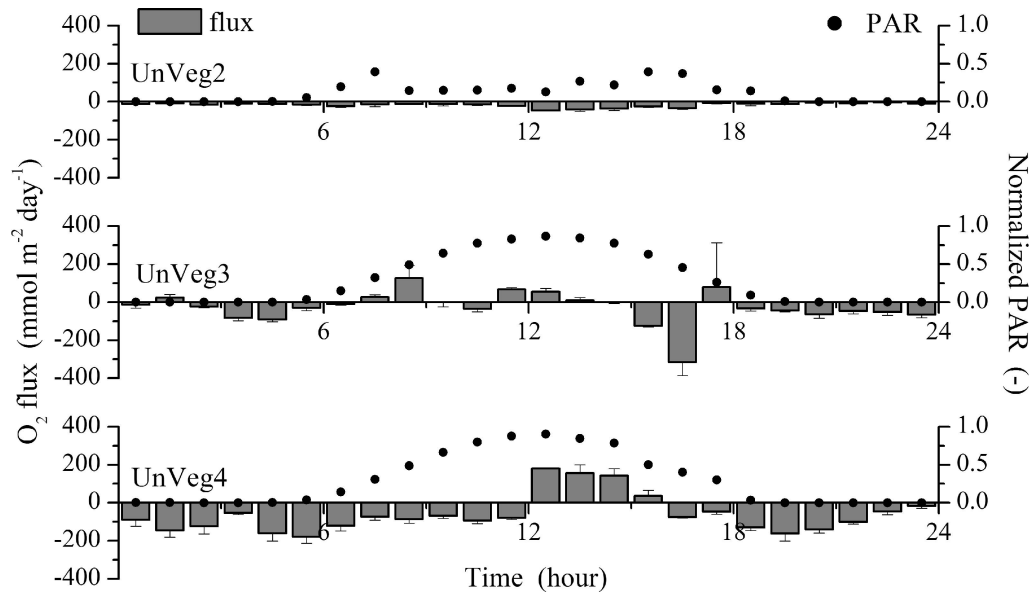
Fig. 3. Twenty-four hourly flux values and normalized PAR for each of the three deployments over the unvegetated sediment. Positive values represent DO release.

The effects of mean current velocity and significant wave height on the DO flux were examined by first grouping all DO fluxes into "seagrass" and "unvegetated" sediment data and then further subgrouping them into light and dark data. The subgrouping was done because different trends were anticipated for the different groups. For example, DO uptake during night (negative fluxes) was expected to be stimulated by currents over the sediment surface (Precht and Huettel 2003; Precht et al. 2004), while DO production

during daylight (positive fluxes) was expected to be stimulated by currents, at least for the vegetated site, due to a decrease in the diffusive boundary layer thickness over the leaves through which gas exchange occurs (Fonseca and Kenworthy 1987; Koch 1994). Finally, all DO fluxes in each subgroup were placed into bins for mean current velocity and significant wave height. Both unbinned and binned flux data were examined for any significant correlations to mean current velocity or significant wave height using simple linear regression. Significant wave
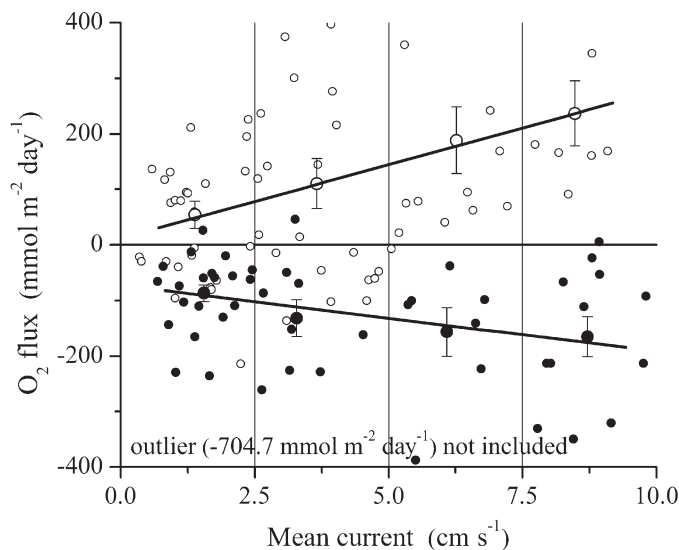

Fig. 4. Effect of mean current velocity on DO flux for the seagrass bed in the light (open circles) and in the dark (closed circles). Light and dark values of all hourly fluxes were binned in four binning intervals (error bars represent SE) and then fitted with a straight line. The binning was performed to emphasize the strong control that mean current velocity had on DO flux both during day and night. In both cases, the metabolic activity was stimulated several fold.
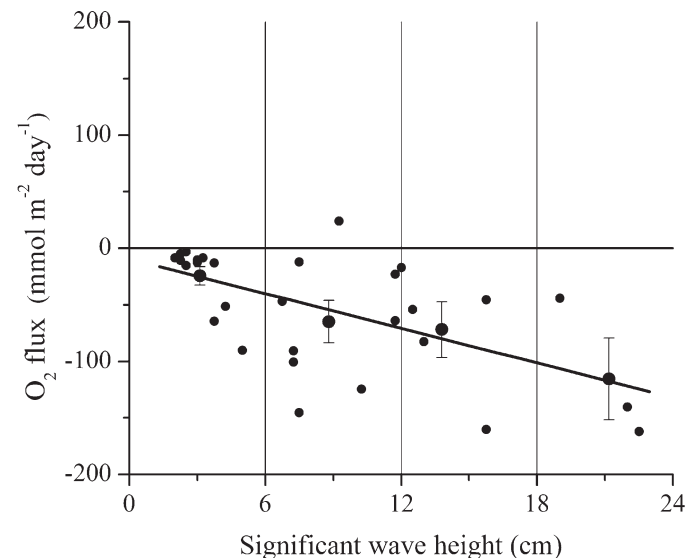

Fig. 5. Effect of significant wave height on DO flux for the unvegetated sediment in the dark. All hourly fluxes in the dark were binned in four binning intervals (error bars represent SE) and then fitted with a straight line. The binning was performed to emphasize the strong control that significant wave height had on DO flux. Sediment respiration was clearly stimulated by wave action.
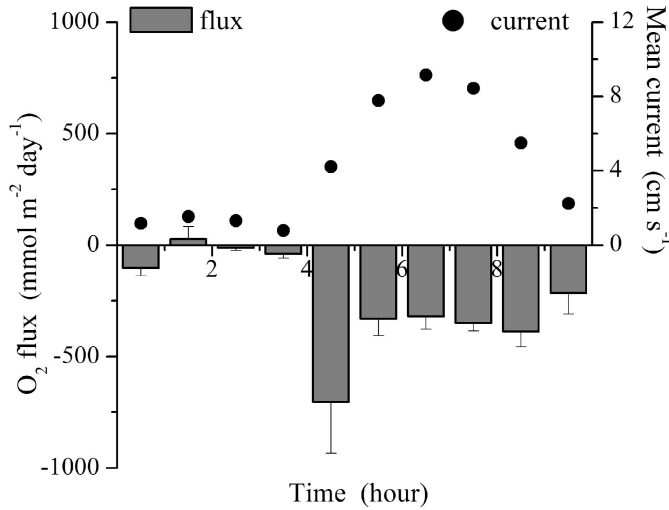
Fig. 6. An example for the seagrass meadow of the way in which a dynamic change, here a sudden increase in mean current velocity during nighttime, can strongly affect the DO flux. At the low current velocity (0–4 h), the respiration averaged -32 mmol m^{-2} day^{-1}, but at the onset (5 h) of the stronger current, respiration increased temporarily to -710 mmol m^{-2} day^{-1}, and then dropped and maintained (6–9 h) a stable level of -350 mmol m^{-2} day^{-1}. In addition to the static control current velocity can have on DO flux (Fig. 4), the peak value was likely the result of the sudden dynamic change in current velocity, prompting reduced compounds and water parcels with lower DO concentration to be released from the bottom of the seagrass canopy and the underlying permeable sediment. This temporarily enhanced the DO flux. Such dynamic events, in addition to the strong static controls depicted in Figs. 4 and 5, are obviously important for a realistic description of net DO flux and thus net ecosystem metabolism.

height was calculated with the QuickWave Software (Nortek AS). Unbinned and binned data and the significant correlations identified are shown in Figs. 4 and 5.

An example of the way in which a sudden change in local hydrodynamics, here a change in mean current velocity, can have an abrupt and pronounced effect on the DO flux for the seagrass bed meadow is shown in Fig. 6.

The 24 hourly DO fluxes for each deployment were used to calculate daily rates of GPP, R, and NEM, all reported in units of mmol DO m^{-2} d^{-1}. As a first step, the fluxes (Figs. 2, 3) were grouped into light and dark values using a normalized PAR value of 0.01 as a threshold. We justified this criterion by additional estimates of GPP, R, and NEM based on a threshold of 0.05, which gave practically the same result. Daily R was calculated as follows:

$$R = \frac{1}{24}\left(\sum flux_{DARK} + \frac{\sum flux_{DARK}}{h_{DARK}} h_{LIGHT}\right) \quad (1)$$

where $Flux_{DARK}$ is the flux shown in Figs. 2, 3, where PAR < 0.01, and h_{LIGHT} and h_{DARK} are the number of light and dark hours ($h_{LIGHT} + h_{DARK} = 24$). The calculation assumes that respiration in the light is equal to respiration in the dark. This may not be entirely accurate, because studies have shown that respiration rates are higher in the light than the dark for benthic microalgal-dominated sediments (Fenchel and Glud 2000; Glud et al. 2009), and

so R determined in this way may be a slight underestimate. Determining respiration in the light, however, is problematic, and the convention is to estimate light respiration from dark values as we do here (Cole et al. 2000). Daily GPP represents the gross oxygen production during photosynthesis and was calculated as:

$$GPP = \frac{1}{24}\left(\sum flux_{LIGHT} + \frac{|\sum flux_{DARK}|}{h_{DARK}} h_{LIGHT}\right) \quad (2)$$

where $Flux_{LIGHT}$ is the flux shown in Figs. 2, 3, where PAR > 0.01. As in the calculation of R, Eq. 2 assumes implicitly that light and dark respiration are equal, and so GPP determined this way also may be slightly underestimated. Daily NEM was calculated as:

$$NEM = \frac{1}{24}\left(\sum flux_{LIGHT} + \sum flux_{DARK}\right) \quad (3)$$

If GPP exceeds R on a sustained basis (NEM > 0), the system is storing or exporting carbon. If GPP is less than R (NEM < 0), the system is consuming more carbon than is being produced, and respiration must be subsidized by inputs of organic matter from external sources or carbon stored from previous periods. Values of $Flux_{LIGHT}$, $Flux_{DARK}$, h_{LIGHT}, GPP, R, and NEM for each deployment are given in Table 1, and the mean values of GPP, R, and NEM are shown in Fig. 7. Figure 8 shows daily values of R plotted against GPP and the derived model II regression line for each of the two sites.

Results

DO flux variability—The time series of individual hourly DO fluxes show substantial variation on several time scales: between days, over a diurnal cycle, and between hours within both the light and dark periods (Figs. 2, 3). The diel cycle, which is driven predictably by light, is evident during the majority of deployments, where hourly DO fluxes were mostly positive (DO release) during light hours and negative (DO uptake) during dark hours. It is also evident, especially from the nighttime data (Figs. 2, 3), that other factors affect the variations in DO flux markedly.

For the seagrass site, mean current velocity significantly affected DO fluxes for both light and dark periods (Fig. 4), but different mechanisms were likely responsible for the two observed relationships. For the light data, DO flux was positively related to current velocity (linear regression; unbinned data: $p = 0.0018$; binned data: $p = 0.0009$, $r^2 = 1.00$), demonstrating that the net DO release due to photosynthetic production was stimulated when current velocity increased. The regression for the binned data (Fig. 4) can be used to estimate the average increase in DO flux that would result from an increase in mean current velocity. For example, an increase in mean current velocity from 1.25 cm s^{-1} to 8.75 cm s^{-1} gives a 5.5-fold increase in DO release from the seagrass community. For the dark data, DO flux was negatively related to current velocity (linear regression; unbinned data: $p = 0.048$; binned data: $p = 0.049$, $r^2 = 0.90$), indicating that DO uptake due to respiration was stimulated by increased current velocities.

Table 1. Values of daylight hours (h_{LIGHT}), sum of DO fluxes in the light ($\Sigma flux_{LIGHT}$), sum of DO fluxes in the dark ($\Sigma flux_{DARK}$), rates of respiration (R), gross primary production (GPP), and net system metabolism (NEM) for 24-h deployments over unvegetated and vegetated sediment (Figs. 2, 3). Daylight hours are defined as the hours where normalized PAR (Figs. 2, 3) exceeds 0.01, and R, GPP, and NEM are calculated from Eqs. 1, 2, and 3.

Deployment	h_{LIGHT} (h)	$\Sigma flux_{LIGHT}$ (mmol m^{-2} d^{-1})	$\Sigma flux_{DARK}$ (mmol m^{-2} d^{-1})	R (mmol m^{-2} d^{-1})	GPP (mmol m^{-2} d^{-1})	NEM (mmol m^{-2} d^{-1})
UnVeg2	15	−335.3	−86.4	−9.6	−8.0	−17.6
UnVeg3	14	−184.2	−456.8	−45.7	19.0	−26.7
UnVeg4	14	−442.5	−1040.8	−104.1	42.3	−61.8
Veg3	14	728.0	−610.0	−61.0	65.9	4.9
Veg4	15	1680.7	−805.2	−89.5	126.0	36.5
Veg5	14	2423.3	−2101.4	−210.1	223.6	13.4
Veg6	14	2117.1	−2173.6	−217.4	215.0	−2.4
Veg7	14	2021.6	−1021.0	−102.1	143.8	41.7

An increase in mean current velocity from 1.25 cm s^{-1} to 8.75 cm s^{-1} would result in a 2.0-fold increase in DO uptake.

For the unvegetated sediment, significant wave height was a strong driver of dark DO fluxes (Fig. 5). The DO flux was negatively related to significant wave height (linear regression; unbinned data: $p = 0.00076$; binned data: $p = 0.019$, $r^2 = 0.96$), demonstrating that DO uptake due to respiration was stimulated by increased wave height. An increase in significant wave height from 3.0 cm to 21.0 cm would result in a 4.5-fold increase in DO uptake.

For the seagrass site, no clear correlation was found between DO fluxes and significant wave height, and for the unvegetated sediment site, no correlation was found between DO fluxes and mean current velocity.

The effect of a change in local hydrodynamics on net DO flux can be significant and immediate. In the example of the way in which a sudden change in current velocity during nighttime can affect the DO exchange (Fig. 6), the flux varied many fold over a 10-h period. A significant portion of the variability between the individual data points in Figs. 2, 3, 4, and 5 is caused by such dynamic changes.
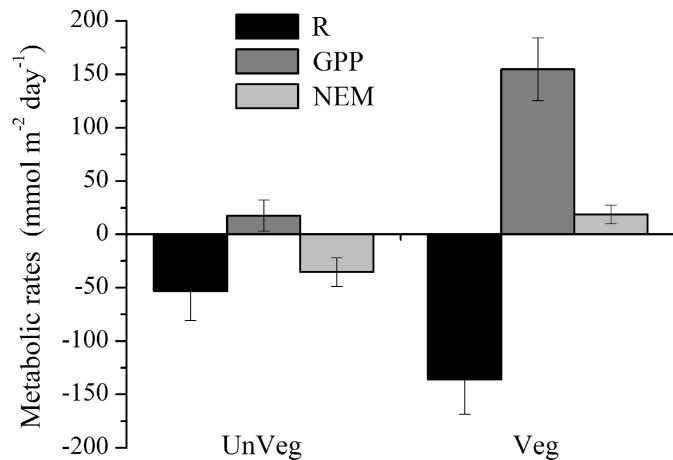

Fig. 7. Averages of estimated daily rates of respiration (R), gross primary production (GPP), and net system metabolism (NEM) for the seagrass bed and the unvegetated sediment (error bars represent SE).
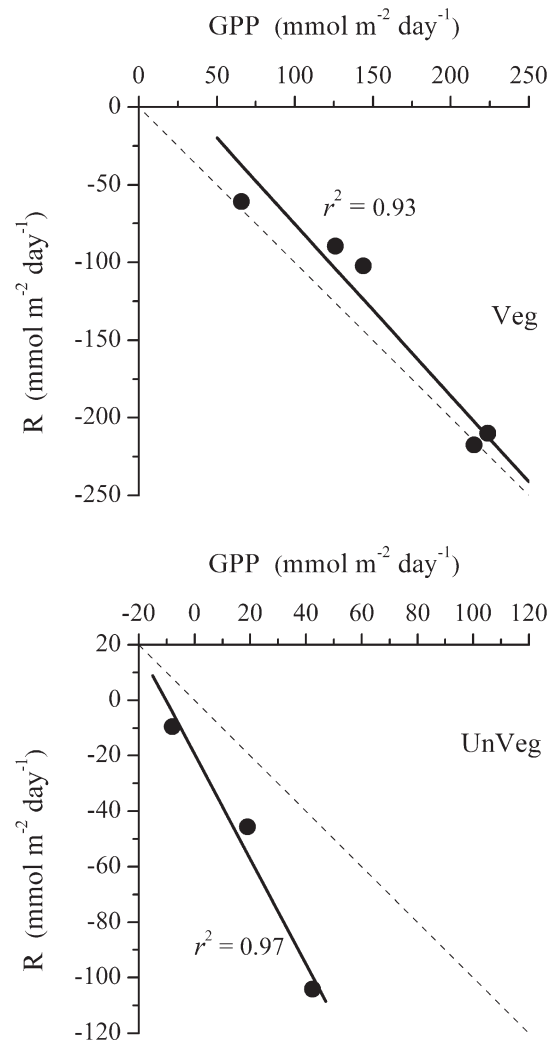

Fig. 8. Daily rates of respiration (R) plotted as a function of gross primary production (GPP) for the seagrass bed and the unvegetated sediment. For both sites, a clear increase is seen in R when GPP increases. The close proximity of the seagrass data to the 1:1 line represents a system in metabolic balance at this time of the year, while the unvegetated sediment likely consumed more carbon than it produced.

These dynamic events, in addition to the strong static controls depicted in Figs. 4 and 5, are clearly important for a complete and accurate description of the DO flux and net ecosystem metabolism.

Net ecosystem metabolism—There was considerable variation in GPP and R between individual deployments for both the seagrass and unvegetated sites (Table 1). For all deployments, the daily rates of GPP varied from 65.9 to 223.6 mmol m^{-2} d^{-1} for the seagrass site and from -8.0 to 42.3 mmol m^{-2} d^{-1} for the unvegetated site. The mean GPP for all deployments was significantly higher for the seagrass site than for the unvegetated site (Fig. 7; 154.9 $\pm$ 29.3 mmol m^{-2} d^{-1} vs. 17.8 $\pm$ 14.5 mmol m^{-2} d^{-1}, $p =$ 0.015). Daily rates of R for the seagrass site varied from -61.0 to -217.4 mmol m^{-2} d^{-1}, and for the unvegetated site, values varied from -9.6 to -104.1 mmol m^{-2} d^{-1}. The mean R values for the seagrass and unvegetated sites were not significantly different (Fig. 7; -136.0 ± 32.4 mmol m^{-2} d^{-1}vs. -53.1 ± 27.5 mmol m^{-2} d^{-1}, $p = 0.13$). These average values include the temporal variation over this 5-week period, a midsummer period of high seagrass biomass. The net result of the higher rates of GPP and similar rates of R is that NEM was significantly higher at the seagrass site than at the unvegetated site (Fig. 7; 18.8 $\pm$ 8.7 mmol m^{-2} d^{-1} vs. -35.4 ± 13.5 mmol m^{-2} d^{-1}, $p =$ 0.012).

Daily values of NEM also varied considerably between deployments at both sites. This was particularly evident in the data for the seagrass site (Table 1), which showed values ranging from a net heterotrophic (-2.4 mmol m^{-2} d^{-1}) to a net autotrophic (41.7 mmol m^{-2} d^{-1}) state. Averaged over all deployments, calculated NEM for the seagrass community was not significantly different from zero, suggesting that the site was in metabolic balance (Fig. 7; 18.8 $\pm$ 8.7 mmol m^{-2} d^{-1}, $p = 0.09$) during this midsummer period. Calculated daily NEM for the unvegetated site suggested similarly that this sediment community was in metabolic balance (Fig. 7; $-35.4 \pm$ 13.5 mmol m^{-2} d^{-1}, $p = 0.12$).

The estimated daily values of R and GPP from the individual deployments for both the seagrass site and the unvegetated site showed a strong linear correlation (Fig. 8, $r^2 = 0.93$ and 0.97). This allows us to determine the trophic status of the system in a way that takes into account the natural variation between individual deployments, and thus it is more sensitive than testing if the mean of NEM is statistically different from zero. Specifically, we tested if each of the linear fits in Fig. 8 were significantly different from the 1:1 line, assuming that a fit not significantly different from the 1:1 line indicates a system in metabolic balance. A heterotrophic system would be characterized by a fitted line located below the 1:1 line, and an autotrophic system would be characterized by a fitted line above the 1:1 line. Statistically, we tested to see if the two "populations," consisting of the distances from the data points to the fitted line and to the 1:1 line, were significantly different. This alternative approach revealed that the seagrass site was very clearly in metabolic balance ($p = 0.57$) at this time of the year, while the unvegetated site

showed a tendency toward a net heterotrophic state ($p =$ 0.083). A similar approach was used by Duarte and Agusti (1998) and Gattuso et al. (1998) to compare net ecosystem metabolism among diverse systems. The approach we use here (Fig. 8) can be more precise than calculating an average daily ratio of GPP:R, particularly for values of GPP and R around zero, where the ratio becomes highly variable and uncertain.

Discussion

DO flux variability—The high temporal resolution of the eddy correlation technique reveals large short-term variability in DO fluxes (Figs. 2, 3) driven by changes in light and local hydrodynamics. Because light (PAR) was measured above the water at a nearby location and not at the sediment surface, we did not investigate a direct linkage between light and benthic production. With respect to changes in hydrodynamics, the high variability in DO flux was significantly correlated to mean current velocity for the seagrass site (Fig. 4) and to significant wave height for the unvegetated site (Fig. 5). These strong correlations with changes in local hydrodynamics, with increases in DO flux of up to a factor of six (Fig. 4), highlight the importance of capturing these mechanisms in any realistic determinations of benthic DO fluxes.

For the seagrass-vegetated sediments (Fig. 4), the strong correlation between DO flux and mean current velocity for the light data is consistent with the effects of increasing flow in enhancing seagrass photosynthesis. This can occur by several mechanisms. First, numerous studies have shown that the mass transfer of nutrients and inorganic carbon is flow-dependent, and that decreasing the thickness of the diffusive boundary layer of the plant with increasing flow enhances photosynthesis (Fonseca and Kenworthy 1987; Koch 1994; Enriquez and Rodriguez-Roman 2006). This assumes that nutrients and/or dissolved inorganic carbon are limiting photosynthesis and that diffusion, rather than uptake kinetics, controls mass transfer. However, a recent study has shown that for a freshwater macrophyte (*Vallisneria americana*) with a morphology similar to *Z. marina*, uptake kinetics of dissolved inorganic carbon rather than diffusive transport controlled mass-transfer processes under moderate current velocities (0.5–6.6 cm s^{-1}) and at saturating light (Nishihara and Ackerman 2009), indicating that biological processes can also control mass transfer. A second mechanism that has been proposed recently to explain flow-enhanced photosynthesis is an apparent increase in oxygen release across the diffusive boundary layer of the plant. Mass et al. (2010) showed that this is a nearly instantaneous response and increases the affinity of RuBisCO to CO_2, presumably because internal oxygen concentrations within the mesophyll are lowered by the oxygen release, which reduces photorespiration and enhances photosynthetic efficiency.

In the dark, the increase in DO uptake for the seagrass bed with higher current velocities (Fig. 4) is likely caused by a more efficient transport of oxygen into the dense bottom canopy and probably also into the permeable sediment below, combined with an increased release of low-

oxygen water from the canopy and the sediment. Several recent studies have highlighted the importance of hydro-dynamically driven exchange of pore water and water-column water for permeable sediments (Huettel et al. 2003; Precht and Huettel 2003; Berg and Huettel 2008). This causes oxygen and both solid and dissolved organic matter to be brought into the permeable sediments and enhances microbial mineralization, and, in turn, anoxic pore water is flushed out of the sediments and into the overlying water (Precht and Huettel 2003; Precht et al. 2004).

For the unvegetated sediment (Fig. 5), the strong correlation between the dark DO flux and significant wave height is consistent with previously published studies showing that respiration-driven DO uptake in permeable sediments is stimulated by wave-induced pore-water flushing (Precht and Huettel 2003; Precht et al. 2004). Our result is also in line with the findings of a previous eddy correlation study over an intertidal sand flat in Japan (Kuwae et al. 2006), which observed a relationship between DO flux and vertical velocity at a frequency band consistent with that of wind-driven waves. The effect of significant wave height on the daytime DO flux for both the seagrass and unvegetated sediment is likely more complex. For seagrass meadows, wave action should have the same effect on the DO flux as we found for current velocity (Fig. 4). However, sediment resuspension caused by wave action is common in shallow coastal bays (Lawson et al. 2007), and this would have a negative effect on photosynthesis of both seagrass and benthic microalgae by reducing light availability. Berg and Huettel (2008) showed that a local turbidity event could result in a pronounced and rapid decrease in DO flux within minutes in at shallow-water sandy sediment site in Apalachicola Bay, Florida.

The variability in benthic DO flux driven by local hydrodynamics is clearly important (Figs. 4, 5) because it can alter the flux many fold. This variability must be included in any realistic determination of benthic DO fluxes—a result that we believe can be generalized beyond our study sites. These effects are not captured in chamber and core incubations, and we believe that results from these flux methods should be interpreted cautiously, at least for the kind of permeable sediment systems studied here. For example, the relationship between mean current velocity and DO flux (Fig. 4) shows that the fluxes measured in low-flow conditions (< 2.5 cm s^{-1}) analogous to many chamber and core incubations (Tengberg et al. 2004) may underestimate higher-flow conditions (7.5–10 cm s^{-1}) that are common in shallow coastal bays (Lawson et al. 2007) by a factor of 2–6. Further, the response of the DO flux to a sudden change in mean current velocity is very rapid and pronounced, as is shown in the example in Fig. 6. Here, the DO flux increased by an order of magnitude with the first hourly flux measurement when the current increased from < 2 cm s^{-1}, and was sustained at half this level in the following hours with a mean current speed of 4–8 cm s^{-1}. Dramatic events like this one, and even more subdued ones, would be excluded in traditional chamber and core incubations (Tengberg et al. 2004). Similarly, the effects of wave action on DO fluxes are not included in chamber

and core incubations, which our data show can enhance DO uptake 5-fold (Fig. 5). Combined, these results suggest that core and chamber incubations may underestimate the DO flux for permeable sediments.

Net ecosystem metabolism—The substantial variation in daily GPP, R, and NEM between individual deployments (Table 1; Fig. 8) underlines both the importance of capturing variations in natural conditions and also the fact that any single 24-h deployment would not likely provide a trustworthy estimate of NEM that represents a given time of year. Gacia et al. (2005) also showed variation in daily NEM calculated from consecutive chamber deployments in a Philippine seagrass community, with one deployment showing net autotrophy and one net heterotrophy. They attributed this variation to a close balance between GPP and R, where presumably a small change in either of the two could result in a different trophic status determined for that time period, rather than to a specific environmental factor that would influence metabolism.

Seagrass meadows are sites of high metabolism, and the higher average GPP and NEM values measured in the seagrass meadow during midsummer compared to the unvegetated sediment (Fig. 7) are in agreement with previous studies that have compared vegetated and nearby unvegetated sediments (Gazeau et al. 2005; Barron et al. 2006; Apostolaki et al. 2010). The high GPP is related to the larger biomass of primary producers in seagrass-vegetated sediments than in unvegetated sediments, where benthic microalgae dominate. Even though benthic micro-algae have faster turnover times than seagrass, this higher production potential is outweighed by the larger ($\sim$ 400-fold) areal biomass in seagrass-vegetated sediments (Gat-tuso et al. 1998). However, average rates of R in the seagrass meadow and the unvegetated sediment were not significantly different (Fig. 7), probably because of the relatively moderate shoot densities (400–500 m^{-2}) in the 5-yr-old *Z. marina* meadow compared to that of more mature seagrass meadows (> 1000 m^{-2}; Olesen and Sand-Jensen 1994). As seagrass biomass increases with colonization time, it is expected that respiration rates would also increase (Barron et al. 2004), in part because of the high rates of bacterial metabolism fueled by increased organic matter (detritus, phytoplankton) deposition and decreased particle resuspension (Koch 1999; Gacia and Duarte 2001; Gacia et al. 2002). Some previous studies and literature reviews have suggested that seagrass meadows are generally net autotrophic (Gattuso et al. 1998; Hemminga and Duarte 2000; Gazeau et al. 2005); however, it is also common for seagrass meadows to be either in metabolic balance as we found or net heterotrophic (Hemminga and Duarte 2000; Barron et al. 2004; Yarbro and Carlson 2008). This variation is likely related to differences in environ-mental factors (light, nutrients, hydrodynamics), organic matter inputs, and self-shading within the seagrass canopy, and it is clearly important to capture these natural variations in order to understand patterns in trophic status correctly.

The variation between daily values of GPP and R among individual deployments shows that GPP and R are tightly

linked for both the seagrass and unvegetated site (Fig. 8). This covariation for individual sites has been reported previously from chamber incubations (Santos et al. 2004; Barron et al. 2006; Stutes et al. 2007), and it has been used to argue that daily rates of seagrass metabolism in many systems are nearly balanced. D'Avanzo et al. (1996) showed that GPP and R were correlated over the year for a shallow bay using the open-water DO technique, but they related this to seasonal coupling of GPP and R rather than to close day-to-day metabolic coupling. On a larger scale, comparisons of daily (Duarte and Agusti 1998) and annual (Gattuso et al. 1998) rates for a number of individual systems show a coupling of GPP and R and provide insight into the carbon balance in these systems. For individual sites, these comparisons give a refined picture of the trophic status of the system because they take into account the natural variation between daily values for the system, and thus they are more sensitive than testing if the mean NEM is statistically different from zero. Capturing the natural variability in DO fluxes for these benthic communities is critical to making accurate estimates of metabolism. We show here that variations in mean current velocity and significant wave height, beyond what is represented in the more static and confined core and chamber incubations, can alter the DO flux by 2–6 times and are well represented in eddy correlation measurements.

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