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The Microbial Ecology of Estuarine Ecosystems

Byron C. Crump[1] and Jennifer L. Bowen[2]

[1]College of Earth, Ocean, and Atmospheric Sciences, Oregon State University, Corvallis, Oregon, USA; email: byron.crump@oregonstate.edu

[2]Marine Science Center, Department of Marine and Environmental Sciences, Northeastern University, Nahant, Massachusetts, USA; email: je.bowen@northeastern.edu

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Abstract

Human civilization relies on estuaries, and many estuarine ecosystem services are provided by microbial communities. These services include high rates of primary production that nourish harvests of commercially valuable species through fisheries and aquaculture, the transformation of terrestrial and anthropogenic materials to help ensure the water quality necessary to support recreation and tourism, and mutualisms that maintain blue carbon accumulation and storage. Research on the ecology that underlies microbial ecosystem services in estuaries has expanded greatly across a range of estuarine environments, including water, sediment, biofilms, biological reefs, and stands of seagrasses, marshes, and mangroves. Moreover, the application of new molecular tools has improved our understanding of the diversity and genomic functions of estuarine microbes. This review synthesizes recent research on microbial habitats in estuaries and the contributions of microbes to estuarine food webs, elemental cycling, and interactions with plants and animals, and highlights novel insights provided by recent advances in genomics.

INTRODUCTION

Microbes are essential, inhabiting every estuarine environment and carrying out ecological functions that bridge the interface between ecology and chemistry. Microbes are simply defined as microscopic organisms and thus include bacteria, archaea, protists, fungi, viruses, and microalgae. Ecology is defined as the study of systems where individual organisms are considered as elements of interaction, either among themselves or with their environment (Margalef 1968). Prokaryotes like bacteria and archaea are the primary decomposers of organic matter, and they perform diverse metabolic functions that control the chemical state and biological availability of essential elements (nitrogen, phosphorus, sulfur, iron, manganese, silicon, etc.). These organisms also serve as prey for protists in microbial food webs that participate in organic matter decomposition, nutrient recycling, and the transformation of indigestible detrital materials into biomass for larger consumers.

This review focuses on interactions among microbes and between microbes and their environment, including the roles they play in biogeochemical cycling. The sections below describe where microbes are found in estuaries, how they interact in food webs, and their key ecological functions in the many environments that make up estuaries, including water, sediment, intertidal wetlands (marshes, mangroves, etc.), and subtidal stands of plants and animals (seagrasses, benthic algae, and coral and shellfish reefs). It also provides an update on the current state of knowledge concerning ecologically important microbial metabolic processes that influence biogeochemical cycling in estuaries. This review aims to strike a balance between broad concepts in estuarine microbial ecology that have been well studied by many scientists over the years and detailed examples from recent literature that provide granularity and specificity. Considering the limitations on the number of citations, our aim was to cite the most recent relevant work, from which further investigation would lead to the seminal work done by earlier researchers. This approach allows us to raise up the voices of a diverse portfolio of new scientists working in estuarine microbial ecology while still creating pathways to uncover, through their work, the foundational papers in the field.

ESTUARINE ENVIRONMENTS

Estuaries are remarkably variable in shape and size, and they feature many diverse interacting habitats (**Figure 1**). Many estuaries are shallow (average depth <10 m), so water and sediment environments are often intimately coupled, exchanging chemistry and biology via diffusion, trophic interactions, and particle settling and resuspension. Seagrass beds, macroalgal stands, and coral and shellfish reefs sit at the interface between water and sediment, influencing both environments. Tidal wetlands like marshes and mangroves are also closely coupled to the water environment via daily tidal inundation and material exchange. Moreover, all of these estuarine habitats occur across horizontal gradients of salinity that extend, for example, from tidal freshwater wetlands to salt marshes and from brackish submerged aquatic vegetation beds to marine seagrass beds (**Figure 1**). Salinity also varies vertically in many estuaries, creating density-stratified water columns that isolate bottom waters from exchange with the atmosphere and favor two-layer estuarine circulation with net flow directed up-estuary in the bottom layer (Snedden et al. 2023). Every estuary features a different combination of these environments, and every environment features a different assemblage of microbes and their associated ecological functions.

Microbes in estuarine plankton are well studied, and many publications provide detailed descriptions of their taxonomic and genomic diversity (Fortunato & Crump 2015, Wang et al. 2021). Planktonic microbes in estuaries are taxonomically distinct from those in freshwater and coastal oceans but can include organisms dispersed from these environments depending on the water residence time (e.g., Morency et al. 2022). Much of our understanding of microbial ecology in

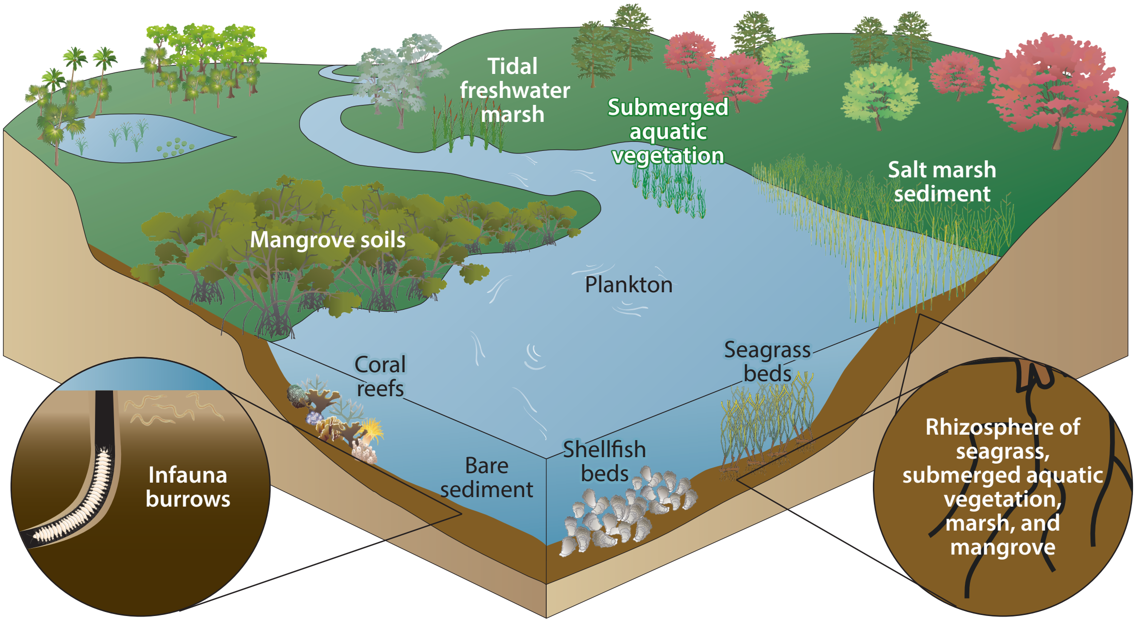

Figure 1

Schematic diagram identifying representative microbial habitats found in estuaries around the world. Insets indicate the important roles of bioturbating organisms (*left*) and the rhizospheres of estuarine macrophytes (*right*). Estuaries are defined by the mixing of coastal marine water with freshwater entering through rivers and groundwater, resulting in distinct salinity gradients and the unique habitats pictured here. Figure created with symbols from the Integration and Application Network (**https://ian.umces.edu/media-library**) (CC BY-SA 4.0). This figure is licensed under a Creative Commons Attribution-ShareAlike 4.0 International (CC BY-SA 4.0) license.

estuarine water comes from ocean research on microbial food webs (Azam et al. 1983) and measured rates of respiration, growth, and grazing. In estuarine water, microbial food webs include food chains of prokaryotes, small heterotrophic flagellates, and larger ciliates and heterotrophic dinoflagellates. In general, only the larger or particle-associated microbes are big enough to be grazed by abundant planktonic animals like copepods or benthic filter feeders like mussels (**Figure 2**). These planktonic microbial communities are responsive to phytoplankton blooms (Brussaard et al. 1996) and allochthonous organic matter inputs (Crump et al. 2017) and turn over quickly due to grazing and viral lysis (Kirchman 2016). Key ecological functions of planktonic estuarine microbes mirror those in oceans and are dominated by aerobic processes like aerobic respiration, nutrient recycling, and transformation of dissolved organic matter. In many estuaries, microbial regeneration of nutrients via mineralization of organic nitrogen and phosphorus fuels a portion of estuarine primary productivity annually (Wilkerson et al. 2006), and planktonic microbes carry out aerobic oxidation of reduced inorganic compounds like ammonium and sulfide, thus contributing to nitrogen and sulfur cycling in estuaries.

Sediments feature microbial food webs that are similar to those in plankton (Dietrich & Arndt 2000), except that benthic microbes are generally 1,000 times more concentrated than they are in water and feature a strikingly high diversity, likely due to the variety of microenvironments and redox conditions in sediments (Tee et al. 2021). Sediment microbes like those in water respire detritus carbon and recycle nutrients, and they form microbial food webs with prokaryotes, flagellates, ciliates, amoeba, foraminifera, and viruses (Lei et al. 2014, Middelburg 2018). But unlike in water, sediment microbes are grazed by larger organisms via direct consumption of sediment

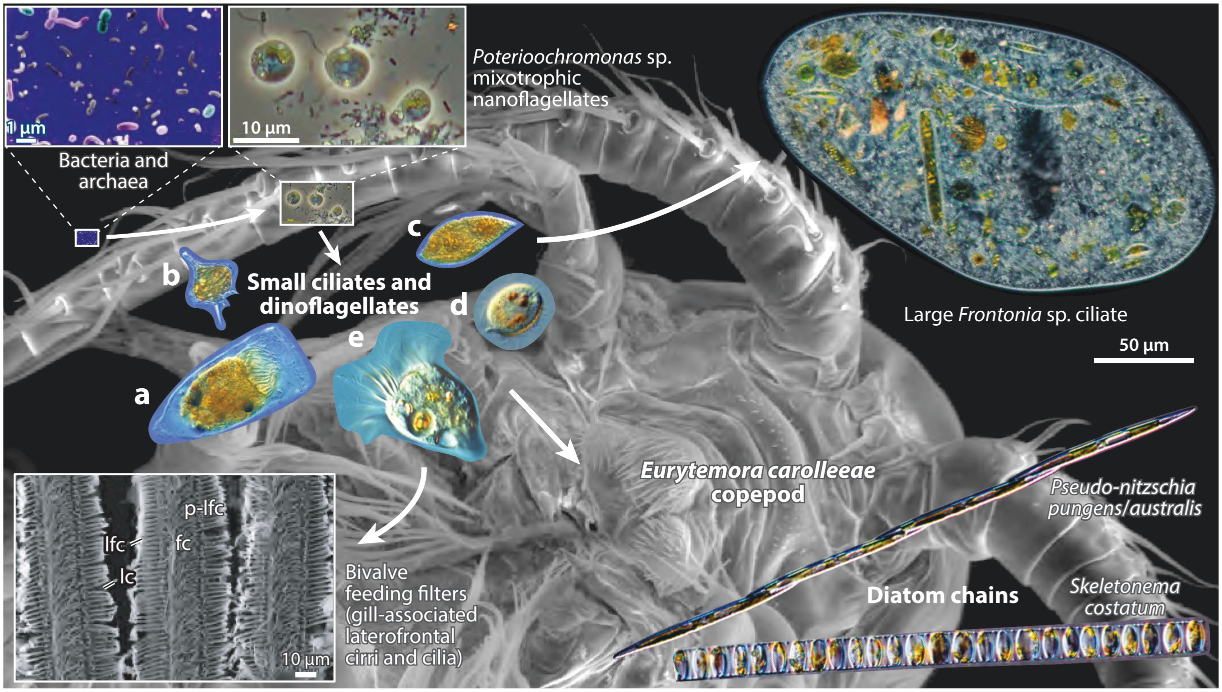

Figure 2

Composite images of microbes and predators with all organisms accurately scaled in and overlaid on an image of the head and antennae of the copepod *Eurytemora carolleeae*. The bacteria and archaea and the *Poterioochromonas* sp. mixotrophic nanoflagellates are shown both at the correct relative sizes and in magnified images (*top left*). The small ciliates and dinoflagellates are (*a*) a tintinnid ciliate (*Favella* sp.), (*b*) a dinoflagellate (*Gyrodinium* sp.), (*c*) a dinoflagellate (*Protoperidinium* sp.), (*d*) an oligotrich ciliate (*Strombidium* sp.), and (*e*) a ciliate (*Urotricha platystoma*). At the bottom right are the chain-forming diatoms *Skeletonema costatum* and *Pseudo-nitzschia pungens/australis*, and at the top right is a large *Frontonia* sp. ciliate that recently ingested some small diatoms. At the bottom left is the feeding apparatus of the mussel *Mytilus edulis*, showing the water-pumping frontal cilia (fc) that create a downward feeding current, while the upward stroke by the feather-like laterofrontal cirri (lfc) capture and transfer food particles to the frontal cilia with a retention efficiency of 100% for particles down to 4 µm [lateral cilia (lc) and pro-laterofrontal cirri (p-lfc) are also shown]. *Eurytemora carolleeae* image provided by Teresa Popp, Carol Eunmi Lee, and Catherine Lorin-Nebel. Bacteria and archaea image adapted from Giovannoni et al. (1990) with permission from the American Society for Microbiology. *Poterioochromonas* sp. image adapted with permission from Bob Andersen and D.J. Patterson. Protists *a–c* adapted from Tintinnidguy/Wikipedia (**https://commons.wikimedia.org/wiki/File:Dinoflagellates_and_a_tintinnid_ciliate.jpg**) (CC BY-SA 4.0). Protists *d* and *e* adapted with permission from Antonio Guillén/Proyecto Agua/Flickr (**https://www.flickr.com/photos/microagua/5229147099** and **https://www.flickr.com/photos/microagua/4892901955**, respectively) (CC BY-NC-SA 2.0). *S. costatum* and *P. pungens/australis* images adapted with permission from Phyto'pedia (**https://phytoplankton.eoas.ubc.ca**) (CC BY-NC-ND 3.0). *Frontonia* sp. image adapted with permission from Robert Berdan (**https://www.scienceandart.org**). *M. edulis* image adapted with permission from Riisgård et al. (2015). This figure is licensed under a Creative Commons Attribution-ShareAlike 4.0 International (CC BY-SA 4.0) license.

particles by infauna (Schratzberger & Ingels 2018). Sediment surfaces often feature microbial mats built by photosynthetic microbes that greatly narrow redox gradients (oxic above, anoxic below) and create fine-scale interactions between aerobic and anaerobic microbial communities. Grazing and burrowing activities by infauna (i.e., bioturbation) expand these redox gradients and modify redox conditions, which influences many microbial biogeochemical processes, including denitrification and sulfide oxidation (Schratzberger & Ingels 2018 and references therein).

Estuarine plants also modify sediment redox gradients and influence microbial communities in marshes, mangroves, submerged aquatic vegetation, and seagrass beds. Rhizosphere microbes living on and around roots and rhizomes of submerged and emergent plants are taxonomically similar to sediment microbes (Martin et al. 2020) but often include active populations that interact with plants directly (Crump et al. 2018). Plant roots modify redox conditions in sediments by injecting oxygen during the day and using oxygen at night (Scholz et al. 2021a and references therein). This enhances the activity of other microbial communities that use both aerobic and anaerobic metabolisms, such as coupled nitrification–denitrification, coupled sulfate respiration and sulfur oxidation, and methane cycling.

MICROBIAL FOOD WEBS

Microbial food webs capture the carbon and energy of detritus organic matter and in the process mineralize a huge fraction of the organic carbon, nitrogen, and phosphorus that cycle in estuaries (Williams 1981). These food webs are based on heterotrophic bacteria, archaea, and fungi, which are the principal consumers of detritus. These organisms are then grazed by protists or destroyed by viruses in an intricate microscopic food web that ultimately mineralizes most of the organic matter it consumes due to inefficient trophic transfers over multiple trophic steps (Azam et al. 1983). Microbial food webs also convert a small fraction of that material into living biomass for consumption by predators. In water, predators include many different planktonic animals—such as copepods, rotifers, and mysid shrimp, as well as benthic filter feeders such as mussels and sponges—that generally only consume larger microbes, like ciliates. In sediments, predators are infauna such as nematodes and polychaetes that can consume microbes of any size (Bergtold et al. 2005, Du et al. 2012). A diagram of carbon flow through microbial food webs (**Figure 3**) emphasizes three different modes of carbon transfer: (*a*) as dissolved organic carbon (DOC) that is consumed by bacteria and archaea, (*b*) as particulate organic carbon (POC) (i.e., detritus) that is colonized by prokaryotes and fungi and consumed by larger animals that directly ingest particles, and (*c*) as living phytoplankton and benthic microalgae that are consumed at many different trophic levels.

DOC is the main energy source for microbial food webs in estuarine waters. Bacteria and archaea recover DOC, and these organisms, in turn, are consumed primarily by heterotrophic and mixotrophic flagellates that are then consumed by ciliates and larger organisms. Viruses infect and destroy (lyse) microbes at all levels of this food web (Wommack & Colwell 2000), and when this happens, they release DOC that can subsequently be consumed by bacteria and archaea

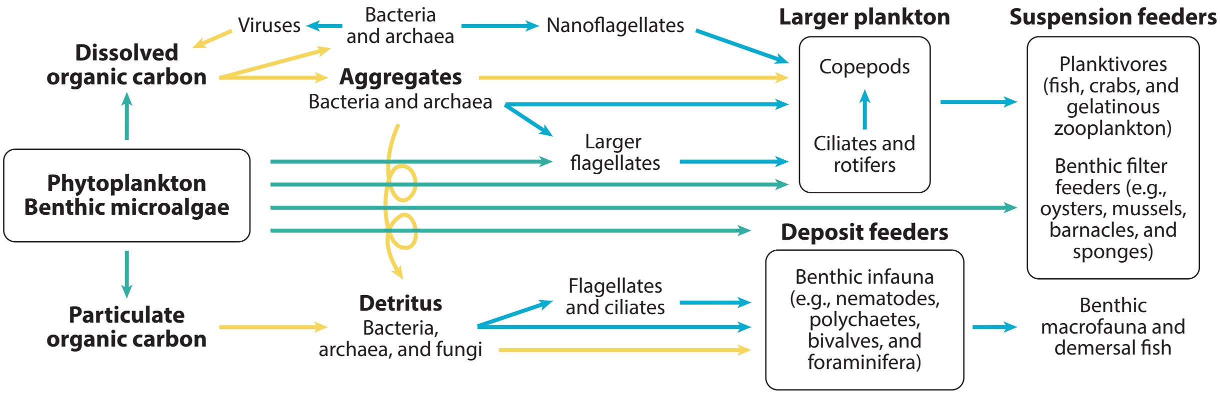

Figure 3

Microbial food web showing sources of organic matter and macroscopic consumers. Green lines represent primary production, yellow lines nonliving organic matter, and blue lines living organic matter.

again. DOC also coalesces in estuaries, forming aggregates with small particles and settling onto sediments as POC (Asmala et al. 2014).

POC feeds microbial food webs in estuarine sediments where infauna and other deposit-feeding animals consume microbes as they feed on detritus particles. Grazing by deposit feeders reduces bacterial and fungal biomass, but at the same time it stimulates microbial productivity and accelerates the flux of energy and nutrients through microbial food webs (Walters & Moriarty 1993) via bioturbation that oxidizes sediment and stimulates nutrient transformations such as coupled nitrification–denitrification and sulfide oxidation (Vasquez-Cardenas et al. 2016). Flagellates and ciliates also consume bacterial biomass in sediments, but their contribution is not well constrained due to methodological limitations (Tuorto & Taghon 2014 and references therein). A diverse community of deposit feeders, including nematodes, rotifers, foraminifera, copepods, and bivalves (Lei et al. 2014, Wieltschnig et al. 2003), consume ciliates, flagellates, and bacteria and thus institute top-down control on sediment microbial food webs (Middelburg 2018). As in estuarine water, sediment microbial food webs consume far more detritus organic matter than larger organisms do (see the section titled Microbial Respiration and Detritus Degradation, below), but the importance of living microbial biomass in the diet of deposit-feeding invertebrates remains uncertain.

Microbial food webs are also important consumers of primary production in estuaries, particularly in water where larger heterotrophic protists directly consume phytoplankton (Putland & Iverson 2007). For example, measurements over a year in the Matang Mangrove estuary in Malaysia indicated that approximately 82% ± 27% of phytoplankton production was consumed by protists (Yong et al. 2022). Similar results were found for the Zuari River estuary in India (58–97% of primary production; Gauns et al. 2015). Less is known about protist grazing of benthic microalgae, but early studies (e.g., Epstein 1997) suggested that benthic microbial food webs are dominant consumers of both detritus and benthic primary production. One recent study of several coastal and estuarine sites suggested that herbivory was the most common feeding strategy for benthic ciliates (Lei et al. 2014), although other studies in estuarine and freshwater sediments suggested that benthic ciliates share this trophic level with microscopic nematodes (Bergtold et al. 2005, Du et al. 2012).

In summary, estuarine microbial food webs feature a complete set of trophic levels, including autotrophic primary producers such as phytoplankton and benthic microalgae; heterotrophic detritivores such as bacteria, archaea, and fungi that consume dissolved and particulate organic matter; and a diverse assemblage of herbivorous and carnivorous protists. Trophic exchange among these microscopic organisms is responsible for much of the carbon cycling that occurs in estuaries.

MICROBIAL AUTOTROPHIC PRODUCTION

Photoautotrophy

Primary production in most estuaries is accomplished mainly by microbial photosynthesizers such as eukaryotic phytoplankton, cyanobacteria, and benthic microalgae (Pinckney 2018), which provide dissolved and particulate organic matter to support both microbial and macroscopic food webs. Intertidal and subtidal macrophytes can contribute to primary production in some estuaries, but even in salt marshes, benthic microalgae contribute a significant (Alongi 2020) and sometimes large fraction (approximately half) of total primary production (Zedler 1980). In these environments, macrophyte biomass and allochthonous detritus are thought to dominate carbon that is sequestered in sediments (Geraldi et al. 2019), likely due to its chemical complexity. However, isotopic studies have suggested that microbial photosynthesizers are often more important than macrophytes in the diets of estuarine planktonic and benthic invertebrates (Kim et al. 2020, Page 1997).

Besides contributing to estuarine productivity, benthic microalgae can physically and chemically modify estuarine sediments by forming microbial mats that bind sediment particles and limit sediment resuspension in subtidal and intertidal sediment (Cuadrado et al. 2011) and in nonvegetated areas of marshes and mangroves (Bolhuis & Stal 2011). The growth of organisms in these mats, a combination of photo- and chemoautotrophs and heterotrophs, creates sharp and intricate vertical layers in microbial communities and redox conditions (Stal et al. 2019) that undergo diel shifts, with daytime oxygen production and sulfide oxidation and nighttime oxygen consumption and sulfide production (Mandal et al. 2021). These layers typically consist of oxygenic phototrophic algae and cyanobacteria at the surface, followed by anoxygenic photosynthetic purple and green sulfur-oxidizing bacteria. Within and below these layers are aerobic and anaerobic heterotrophic taxa that consume photosynthate, including sulfate reducers that support chemoautotrophic sulfur-oxidizing bacteria by producing hydrogen sulfide (Armitage et al. 2012). Photosynthate can also support denitrifying bacteria (Morelle et al. 2022), although benthic microalgal growth can also inhibit denitrification by competing with denitrifiers for nitrate (Risgaard-Petersen 2003).

Chemoautotrophy

Chemoautotrophs, including nitrifiers and sulfur oxidizers, are important producers in some marine systems (Middelburg 2011), where they fix CO_2 by oxidizing reduced inorganic compounds. In nearshore systems, including estuaries, this light-independent dark carbon fixation was estimated at 0.19 Pg C y^{-1} (~5% of photoautotrophy) globally (Middelburg 2011), and several recent studies have argued that dark carbon fixation should be included in ocean carbon budgets (Baltar & Herndl 2019, Vasquez-Cardenas et al. 2020). However, dark carbon fixation is rarely included in estimates of estuarine primary productivity despite the importance of chemoautotrophs in estuarine nitrogen and sulfur cycling (see the section titled Nitrogen Cycle Processes and the section titled Sulfur and Methane Cycling, below). High rates of dark carbon fixation have been recorded in oxic–anoxic interfaces and particle-rich turbidity maxima regions of diverse estuaries, ranging from deep systems like the Baltic Sea (Glaubitz et al. 2009) and Saanich Inlet (Juniper & Brinkhurst 1986) to the shallower Rodrigo de Freitas Lagoon (Signori et al. 2020) and the Columbia River estuary (Bräuer et al. 2013).

The highest rates of dark carbon fixation in estuaries are typically found in surface sediments at the interface between oxygenated water and anoxic sediments (Vasquez-Cardenas et al. 2020). Here, sulfide oxidizers, principally members of the class Gammaproteobacteria, account for 70–86% of dark carbon fixation (Dyksma et al. 2016) and are estimated to fix 0.122 Pg C y^{-1} globally, which is similar to the rate of carbon burial in vegetated marine sediments (Duarte et al. 2005). These chemoautotrophic carbon fixation rates are enhanced by oxygen translocation into sediments via plant roots (Thomas et al. 2014) and bioturbation (Vasquez-Cardenas et al. 2016) that effectively expand the area of oxic–anoxic interfaces, where sulfur oxidizers thrive.

One recently characterized group of sulfur oxidizers, cable bacteria, are important members of chemoautotrophic communities in estuarine sediments, where they carry out electrogenic sulfur oxidation (Pfeffer et al. 2012). These colonial bacteria form centimeter-long, multicellular filaments that bridge redox gradients and shuttle electrons between sulfidic and oxygenated sediments, thereby extending the depths over which dark carbon fixation can occur (Boschker et al. 2014). Recent studies have found these organisms associated with roots of seagrass, submerged aquatic vegetation, marsh grass, and mangroves (Scholz et al. 2021b); the walls of animal burrows

(Li et al. 2020a); and sediments of bivalve reefs (Malkin et al. 2017). Cable bacteria and other sulfur oxidizers are thought to benefit plants and animals by detoxifying hydrogen sulfide and potentially serving as prey for burrowing infauna (Li et al. 2020a).

Nitrifying bacteria and archaea are another major group of chemoautotrophs in estuaries. These organisms use energy from the oxidation of ammonium and nitrite to fix inorganic carbon into biomass through various chemoautotrophic pathways, including the recently discovered archaeal 3-hydroxybutyrate 4-hydroxypropionate cycle (Könneke et al. 2014), through which they contribute substantially to the global carbon cycle (Meador et al. 2020). In estuaries, nitrifiers inhabit water and oxygenated surface sediments, where their abundance and growth depend on ammonium and oxygen availability and may be enhanced by temperature (Damashek & Francis 2018) and inhibited by light and competition with photoautotrophs for ammonium (Arun et al. 2021). The contribution of nitrifiers to dark carbon fixation rates in estuaries is not well studied but is thought to be low compared with their contribution to sulfide oxidation, particularly in estuarine sediments (Dyksma et al. 2016, Middelburg 2011). This idea is supported by a study of intertidal sediment in the Yangtze River estuary that showed correlations among dark carbon fixation rates, sulfide concentrations, and Calvin cycle *cbb*M genes from sulfur-oxidizing bacteria, as well as preferential labeling of these *cbb*M genes in incubations with ^{13}C-bicarbonate (Liu et al. 2022).

Heterotrophic Carbon Fixation and Anaplerosis

Dark carbon fixation is sometimes considered secondary production because it depends on energy from inorganic compounds released during organic matter oxidation (e.g., ammonium and hydrogen sulfide; Dyksma et al. 2016, Middelburg 2011). This categorization is somewhat convenient because measurements of dark carbon fixation using ^{14}C or ^{13}C incubations include both chemoautotrophic CO_2 fixation and nonautotrophic CO_2 fixation via heterotrophic metabolisms. The most important of these heterotrophic metabolisms is anaplerosis, which uses the enzyme pyruvate carboxylase to replenish intermediates like oxaloacetate in the tricarboxylic acid cycle (Braun et al. 2021). Heterotrophic carbon fixation has been documented in several marine systems (e.g., DeLorenzo et al. 2012, Molari et al. 2013). One early study in the freshwater Coromina Lagoon in Spain showed that dark CO_2 incorporation in water was dominated by heterotrophs and accounted for 31% of total carbon fixation (Llirós et al. 2011). In eutrophic Guanabara Bay, Brazil, dark carbon fixation was low compared with bacterial secondary production and was attributed to anaplerotic reactions (Signori et al. 2018). This form of dark carbon fixation is likely to be important in estuaries that receive a large amount of detritus that supports heterotrophic growth and associated anaplerosis and other forms of heterotrophic carbon fixation.

MICROBIAL RESPIRATION AND DETRITUS DEGRADATION

Estuaries are often net heterotrophic ecosystems where respiration of autochthonous organic carbon is enhanced by respiration of allochthonous material received from land or imported from coastal oceans (Caffrey 2004). Degradation and respiration of detritus carbon in estuaries are carried out by many organisms, including microbes and detritivorous invertebrates. These organisms cooperate to convert particulate detritus into dissolved organic matter, and microbes then respire DOC and mineralize organic nutrients. Rates of respiration vary depending on the bioavailability of the organic carbon and local environmental conditions (e.g., redox and temperature). The highest rates are associated with freshly produced organic carbon in oxic conditions, and the slowest are associated with older organic carbon in anoxic conditions.

The bioavailability of organic carbon for microbial respiration varies with source and with age in estuaries. Terrestrial DOC and POC are thought to be older and less bioavailable due to bio- and photodegradation during their transit to estuaries (Bianchi et al. 2018). This material includes dissolved organic matter from rivers and groundwater and sediment-associated particulate organic matter that accumulates in marshes, mangroves, and river delta environments. However, some allochthonous organic matter, particularly riverine and coastal phytoplankton, can enhance heterotrophic activity in estuaries (Crump et al. 2017, Smith & Hollibaugh 1997). Autochthonous organic matter sources in estuaries vary broadly and range from highly bioavailable material released by growing phytoplankton and benthic microalgae to recalcitrant lignin-based material produced by mangrove trees. The types of microbes that degrade these different materials are diverse, and seasonal variation in organic matter supply is associated with seasonally predictable synchronous changes in microbial communities of river and estuarine systems (e.g., Kellogg et al. 2019). For example, in the Baltic Sea during spring runoff, bacterial specialist taxa were favored by the influx of terrestrial DOC, whereas in summer and fall, bacterial generalist taxa capable of respiring either terrestrial or phytoplankton DOC were more abundant (Figueroa et al. 2021). Another study in the Altamaha–Doboy–Sapelo estuary, USA, used incubation experiments to show that microbes have a strong preference for marine DOC, leaving behind residual organic matter with a strongly terrestrial signature that was more resistant to decomposition (Medeiros et al. 2017).

Most respiration in estuaries is accomplished with either the most energetic electron acceptor, oxygen, or the most abundant electron acceptor, sulfate. In oxygenated water and sediments, respiration is accomplished by aerobic heterotrophs that outcompete anaerobes for organic matter resources. Aerobic respiration in water is assumed to dominate organic carbon oxidation in estuaries, although few studies compare respiration in water and sediment at the ecosystem scale. One meta-analysis estimated that most organic carbon respiration occurs in estuarine water, with sediments accounting for on average 24% of total respiration in estuaries (Hopkinson & Smith 2005). Recent studies have suggested that the relative contributions of benthic and pelagic respiration are more variable and depend on the system and season (e.g., Hetland & DiMarco 2008), although many of these studies restrict comparisons of sediment respiration to pelagic rates in subpycnocline bottom waters rather than the whole water column (Murrell & Lehrter 2011, Zhou et al. 2021).

In anoxic estuarine environments, sulfate reduction is the predominant decomposition pathway, accounting for an estimated 70–90% of decomposition in salt marshes (Howarth 1984) and 50–90% in other coastal sediments (Soetaert et al. 1996). This anaerobic respiratory process dominates despite the low energy of sulfate respiration because sulfate is the second most concentrated anion in seawater (Canfield & Farquhar 2009) and is orders of magnitude more concentrated than any other electron acceptor (e.g., Segarra et al. 2013).

In estuarine environments where primary production outpaces respiration, including coastal wetlands and seagrass beds, slow decomposition and respiration via sulfate reduction lead to high rates of carbon storage in anoxic sediments below the reach of plant roots (Chmura et al. 2003), leading to their designation as coastal blue carbon habitats (reviewed in Macreadie et al. 2019). Slow microbial decomposition and associated carbon storage also contribute to the ability of intertidal salt marshes and mangrove habitats to grow vertically and keep pace with sea level rise, although rates of vertical accretion are system specific and depend, in part, on organic matter content, sediment delivery, tide range, and distance from tidal creek channel (Chmura & Hung 2004).

These issues stimulated research into the impacts of eutrophication and climate change on rates of decomposition and respiration in blue carbon habitats. One recent study demonstrated eutrophication-enhanced respiration in marsh sediment incubations via the addition of more energetic electron acceptors, such as nitrate (Bulseco et al. 2019). Nitrate addition has the potential to diminish the carbon storage capacity of blue carbon habitats (Deegan et al. 2012, Palacios et al. 2021), although the effect may be offset in some systems by eutrophication-enhanced primary production (Moseman-Valtierra et al. 2022). The relative balance between enhanced decomposition and enhanced primary productivity may be a function of the form of nitrogen that is available (Bowen et al. 2020). Changes in salinity associated with sea level rise and shifting precipitation patterns may also influence degradation and respiration in these environments. In one study, soil respiration and organic carbon loss were enhanced by both increases and decreases in salinity (Chambers et al. 2013), suggesting that any deviation from average salinity increases decomposition. The mechanisms behind this effect remain unclear but may include shifting availability of electron acceptors used by microbes for anaerobic respiration.

In estuarine environments where respiration keeps pace with primary production, nutrient recycling (i.e., mineralization, regeneration, and ammonification) helps maintain continuous primary productivity. In fact, for many estuaries the fraction of phytoplankton productivity supported by recycled nitrogen is very high for most of the year. In surface oceans, the fraction of productivity supported by new nitrogen flux from deeper waters is referred to as the *f* ratio, where low ratios indicate that productivity is supported by recycled nutrients (Eppley & Peterson 1979). In estuaries, where new nitrogen flux is mainly from rivers and coastal oceans, the *f* ratio is often much less than 0.5 and increases only when external nitrogen inputs peak. This effect has been observed in many estuaries, such as during high freshwater flow in San Francisco Bay (Wilkerson et al. 2006) and Apalachicola Bay (Mortazavi et al. 2000) and during influx of nutrient-rich coastal water to Funka Bay, Japan, from the Oyashio current (Kudo et al. 2015). Thus, through nutrient recycling, microbial heterotrophic activity is essential for maintaining continuous primary productivity in estuaries.

Estuarine environments where respiration outpaces primary production are often light-limited habitats where allochthonous organic matter is respired, such as deeper water and sediments (Fennel & Testa 2019) and estuarine turbidity maxima (Abril et al. 1999). These environments are susceptible to hypoxia and anoxia, when oxygen consumption by aerobic respiration is greater than oxygen resupply by oxygenic photosynthesis, advection, and diffusion (Fennel & Testa 2019). The taxonomic composition of microbial communities in low-oxygen bottom waters is sometimes different from that of surface waters (e.g., Wu et al. 2019). However, temporal studies suggest that these communities are not highly sensitive to reductions in oxygen concentration (Crump et al. 2007, Zaikova et al. 2010), possibly because many aerobic bacteria can function at a very low oxygen concentration or switch to anaerobic respiration facultatively. One pair of studies used metatranscriptomics to show a gradual change in the expression of genes for anaerobic respiration following the formation of anoxia in Chesapeake Bay bottom waters (Eggleston et al. 2015, Hewson et al. 2014). Low-oxygen conditions are expanding in estuaries and oceans globally (Breitburg et al. 2018), and more research is required to understand microbial taxonomic and functional succession during seasonal hypoxia and anoxia.

NITROGEN CYCLE PROCESSES

The essential nutrient nitrogen is a component of diverse organic and inorganic compounds in estuaries, and most chemical transformations of these compounds are only carried out by microbes (**Figure 4**). The main external sources of nitrogen to estuaries are freshwater (rivers and groundwater) and ocean water that carry nitrogen into estuaries, mainly in the form of nitrate and organic nitrogen. Other potentially important sources are atmospheric deposition of inorganic nitrogen and autochthonous nitrogen fixation. As described above, organic nitrogen (e.g., urea, nucleotides, and amino acids) is mineralized to ammonium primarily by heterotrophic microbes in estuarine water and sediments. Ammonium can be taken up by primary producers or oxidized by nitrifying bacteria and archaea. Once oxidized, nitrogen becomes useful for a suite of anaerobic microbial

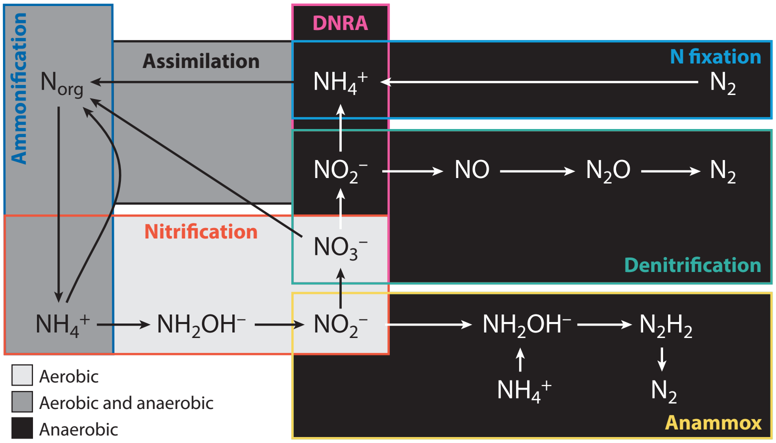

Figure 4

Nitrogen cycle processes in estuaries, showing the major oxidation and reduction reactions that are performed by microbial communities. Typical oxygen conditions associated with these processes are indicated by background shading. Abbreviations: anammox, anaerobic ammonium oxidation; DNRA, dissimilatory nitrate reduction to ammonium; N_{org}, organic nitrogen. Figure adapted from Philippot et al. (2007) with permission from Elsevier.

processes, many of which produce gaseous nitrogen compounds, including inert N_2 gas and the greenhouse gas N_2O (**Figure 4**). The primary sinks for nitrogen in estuaries are burial, uptake by primary producers, export to coastal oceans, and loss to the atmosphere. Recent metagenomic studies have shed new light on the microbial nitrogen cycle in estuaries and coastal waters.

Nitrification

Nitrification, the oxidation of nitrogen from NH_4^+ to NO_3^-, is a critical transformation in the nitrogen cycle of estuaries. The process has two steps: the conversion of NH_4^+ to NO_2^- followed by the conversion of NO_2^- to NO_3^-. These two steps, ammonia oxidation and nitrite oxidation, respectively, have been recognized as important steps in nitrogen management since their discovery in the late 1800s (Winogradsky 1890); however, it is only with recent genomic insights that we recognize the breadth of organisms that carry out these processes (Zou et al. 2020 and references therein). Traditionally, ammonia oxidation was thought to be restricted to clades of Betaproteobacteria and Gammaproteobacteria, but early shotgun metagenomics suggested that the ammonia monooxygenase gene, encoding an essential protein in ammonia oxidation, might exist within the archaea (Venter et al. 2004). This was later confirmed when the first ammonia-oxidizing archaea were isolated (Könneke et al. 2005), dramatically expanding our list of nitrifying taxa. Ammonia oxidation is considered the rate-limiting step in nitrification and has thus received considerably more attention than nitrite oxidation; however, new insights into nitrite oxidation are also expanding our understanding of nitrification. Of particular interest is that nitrite oxidation, long considered to be oxygen dependent, appears to be possible in both oxic and anoxic waters (Sun et al. 2021), greatly expanding the number of estuarine environments where nitrification can occur. Lastly, even the long-held paradigm that the two steps of nitrification are facilitated by two distinct syntrophic microbes has fallen with the recent discovery of complete ammonia oxidizers (comammox bacteria) that can perform both steps within a single cell (Daims et al. 2015). Sedlacek (2020) published a summary of these bacteria and other paradigm shifts in nitrification over the last 150 years.

In estuaries, nitrification forms a bridge between regeneration of NH_4^+ from organic nitrogen and removal of NO_3^- via denitrification (**Figure 4**), thereby helping to prevent eutrophication of coastal waters. Nitrification requires NH_4^+ and so can occur in estuarine waters and surface sediments in systems with NH_4^+ inputs both from anthropogenic sources and from autochthonous production via mineralization (also called ammonification). Rates of nitrification in estuaries are generally higher than those found in adjacent ocean systems. However, they are highly variable in space and time, with the highest rates of nitrification typically associated with estuarine turbidity maxima and at oxic–anoxic interfaces (Damashek & Francis 2018). Damashek et al. (2016) reported average rates of nitrification in the ocean water column to be 50 nM d^{-1}, whereas estuaries can have rates in excess of 10,000 nM d^{-1}. Water column nitrification is also a source for the potent greenhouse gas N_2O, though a recent meta-analysis indicated that benthic denitrification dominates the N_2O flux from estuaries (Murray et al. 2015). The relative importance of ammonia-oxidizing archaea (AOA) versus ammonia-oxidizing bacteria (AOB) also varies from system to system; in some systems (e.g., San Francisco Bay; Mosier & Francis 2008), AOB were more abundant than AOA, and in other systems (e.g., Plum Island Sound; Bernhard et al. 2010), the opposite was true. Caffrey et al. (2007) quantified the abundance of AOA and AOB in six estuaries and showed that in five of the six, AOA were up to 80 times more abundant than AOB, and that measured rates of potential nitrification more closely tracked the abundance of AOA than that of AOB (Caffrey et al. 2007). Comammox can also play a role in estuaries, with rates accounting for up to 27% of measured nitrification in tidal flats of the Yangtze River estuary (Sun et al. 2022). Additional research is needed to disentangle controls on the relative importance of these different diverse nitrifying groups in estuarine systems.

Coupled Nitrification and Denitrification

Since NO_3^- produced by nitrification serves as a highly valued electron acceptor in the decomposition of organic matter via denitrification, the two reactions are often very tightly coupled, particularly in oligotrophic estuaries where ambient NO_3^- concentrations are low (Vieillard & Thrush 2021). Coupled nitrification–denitrification occurs largely along oxic–anoxic interfaces since nitrification requires a supply of oxygen and denitrification requires its absence. Thus, coupled nitrification–denitrification tends to be the dominant mechanism of nitrogen loss in vegetated estuarine sediments, where oxygen can penetrate further into anoxic sediments than would be possible via diffusion alone. For example, coupled nitrification–denitrification was the predominant nitrogen loss pathway in Florida seagrass beds (Hoffman et al. 2019), New England salt marshes (Mason et al. 2021), and mangrove sediments in Vietnam (Comer-Warner et al. 2022), to list just a few of the many examples. Unvegetated sediments can also support high rates of coupled nitrification–denitrification. Rates of nitrification can be further enhanced by benthic microalgal oxygen production and infaunal bioturbation (reviewed in Moulton et al. 2016). Seasonal hypoxia, which is increasingly common in estuaries, can inhibit coupled nitrification–denitrification and promote nitrogen retention, which may exacerbate estuarine eutrophication over time, indicating that controls on coupled nitrification–denitrification may be highly sensitive to future changes in climate and oxygen supply.

Dissimilatory Nitrate Reduction Processes

Microbes use three distinct processes for dissimilatory nitrate reduction: denitrification, where NO_3^- is reduced to N_2 or N_2O; dissimilatory nitrate reduction to ammonium (DNRA); and anaerobic ammonium oxidation (anammox), which also produces N_2 (**Figure 4**). It was once widely thought that denitrification was performed predominantly by heterotrophs, anammox by autotrophs, and DNRA by a combination of the two. However, recent evidence indicates that autotrophic denitrification coupled to oxidation of hydrogen sulfide can also be important in estuarine systems (Bulseco et al. 2020). Furthermore, denitrification is now recognized to be a highly modular process, with some taxa having only a subset of the genes needed for the complete denitrification pathway (Roco et al. 2017). The capacity for denitrification is widespread phylogenetically and is typically facultative, with many taxa capable of using alternative electron acceptors in the absence of nitrate (Bowen et al. 2015). The genetic capacity for DNRA is also phylogenetically widespread (Welsh et al. 2014), and DNRA frequently co-occurs with the genetic capacity for denitrification and occasionally co-occurs with the genetic capacity for nitrogen fixation. In contrast, anammox is highly phylogenetically constrained, with the genetic capacity thought to belong solely to a monophyletic group of Planctomycetes (Woebken et al. 2008).

In estuaries, understanding the controls on denitrification, DNRA, and anammox is important because anammox and denitrification remove fixed nitrogen from the system, whereas DNRA retains fixed nitrogen that can fuel additional primary production. Furthermore, the modularity of the denitrification pathway can lead to incomplete denitrification, resulting in the production of the potent greenhouse gas N_2O (Bakken et al. 2012), which does not appear to be produced via anammox. Delineating where and when these three processes dominate is essential for predicting both primary productivity and the production of greenhouse gas from estuaries. Rates of anammox in estuaries appear to be low, relative to coupled nitrification–denitrification and direct denitrification, across a range of habitats (Chen et al. 2021), including subtidal and vegetated sediments (Koop-Jakobsen & Giblin 2009). DNRA is much less well studied in estuaries than denitrification, though it can account for substantial NO_3^- reduction (reviewed in Burgin & Hamilton 2007, Giblin et al. 2013). Modeling results suggest that denitrification is favored in systems where the ratio of bioavailable carbon to NO_3^- is low and DNRA is favored when this ratio increases (Algar & Vallino 2014) and when salinity and sulfide concentrations are high (Domangue & Mortazavi 2018). These complex patterns of nitrogen retention and loss make it challenging to scale up rates of nitrogen cycling to the ecosystem level, rendering cross-system comparison and prediction of future rates a challenge.

Nitrogen Fixation

Nitrogen fixation is the microbially mediated conversion of N_2 gas to NH_4^+, making it available for biological use. Historically, microbes capable of nitrogen fixation, collectively referred to as diazotrophs, were thought to be dominated by colony-forming cyanobacteria (e.g., *Trichodesmium* spp.) in marine systems (Zehr & Capone 2020). However, recent discoveries of divergent cyanobacterial clades and noncyanobacterial heterotrophic nitrogen fixers have expanded our understanding of the phylogenetic breadth of nitrogen fixation (Delmont et al. 2018) and raised new questions about the distribution and function of these diazotrophs in estuaries. For example, the small unicellular cyanobacteria UCYN-A, -B, and -C, some of which are obligate symbionts with extensive genome streamlining, appear to be globally distributed (Thompson et al. 2012) and function even under nitrogen-replete conditions, where nitrogen fixation is thought to be inhibited (Mills et al. 2020). Additional recent discoveries of nitrogen-fixing symbionts associated with lucinid clams and seagrasses (Mohr et al. 2021) that are widespread in coastal habitats further expanded the possible role of diazotrophic microbes in estuaries.

The prevailing wisdom, prior to these discoveries, was that nitrogen fixation rates in estuaries were low, perhaps because of apparent molybdate inhibition in estuarine waters and because of high ambient NH_4^+ concentrations in estuarine sediments (Howarth et al. 1988). Fixing nitrogen is energetically costly, requiring 16 moles of ATP per mole of N_2 fixed, fueling the expectation that nitrogen fixation rates would be low in nutrient-rich estuaries. However, recent studies suggest otherwise. As just one of many examples, in one Australian estuary, rates of nitrogen fixation (10–64 nmol L^{-1} d^{-1}) were 5–30 times higher than rates measured in nearby coastal waters and were attributed to a combination of UCYN-A cyanobacteria and noncyanobacteria diazotrophs related to the cluster 3 Deltaproteobacteria (Messer et al. 2021). These rates were comparable to those observed in the water of other tropical (Jabir et al. 2020) and temperate (Li et al. 2020b) estuaries. Benthic nitrogen fixation rates in estuaries are also substantial (Oczkowski et al. 2020), and resuspended sediments can contribute significantly to nitrogen fixation in estuarine waters (Pedersen et al. 2018). While countless studies have predicted anthropogenic inputs of nitrogen to estuaries, surprisingly few have quantified both allochthonous nitrogen fluxes and autochthonous nitrogen fixation, perhaps because net nitrogen fixation is so variable in space and time (Marcarelli et al. 2022). Thus, it is important that we assess the contributions of the broad diversity of diazotrophs in these systems to better constrain their contributions to estuarine nitrogen cycling.

SULFUR AND METHANE CYCLING

Sulfur and methane metabolisms are important aspects of estuarine biogeochemistry. Biogenic sulfur compounds, including dimethylsulfide, dimethylsulfoniopropionate, and dimethylsulfoxide, are derived from algae and phytoplankton and from some estuarine macrophytes, including the salt marsh grass *Spartina alterniflora* (Husband & Kiene 2007). These trace gases can serve as cloud condensation nuclei and thus play important roles in the climate system. They can also be decomposed by estuarine microbes (Kiene & Service 1991) and thus contribute to estuarine food webs. The estuarine sediment sulfur cycle is closely coupled to the methane cycle, with well-defined zones of sulfate-rich and methane-rich sediment pore water and a transition zone where concentrations overlap. These concentrations are influenced by several oxidation and reduction reactions (**Figure 5**), including the reduction of SO_4^{2-} to sulfide and its subsequent reoxidation back to SO_4^{2-} and the reduction of CO_2 to CH_4 (i.e., methanogenesis) and its subsequent reoxidation by methanotrophs. Broadly, estuarine organic matter decomposition follows the supply of energy-yielding electron acceptors, with SO_4^{2-} and CO_2 reduction having lower energy yields than O_2 and NO_3^- (Canfield et al. 2006). Despite the lower energy yield, the ubiquity of SO_4^{2-} in seawater results in SO_4^{2-} reduction being the predominant detrital decomposition pathway in many coastal systems (Howarth 1984).

Methanogenic organisms often compete for substrates (e.g., hydrogen and acetate) with SO_4^{2-} reducers, so it was once thought that methanogenesis is hindered by substrate availability in the presence of SO_4^{2-}, with methanogens only gaining the upper hand in regions of estuaries where both salinity and SO_4^{2-} concentrations are low (Weston et al. 2014). However, recent evidence for both a cryptic sulfur cycle and a cryptic methane cycle in coastal systems makes assessing gross fluxes in methane and sulfide production across estuaries a challenge (Kevorkian et al. 2022). Nonetheless, there is extensive evidence from incubation experiments and sediment pore water chemistry that SO_4^{2-} reduction and CH_4 production can co-occur, in part because methanogens can use methanol or trimethylamine for CH_4 production, avoiding the need to compete with SO_4^{2-} reducers for hydrogen or acetate (Oremland & Polcin 1982). The relative proportions of these rates tend to vary seasonally (Jørgensen & Sørensen 1985), and although CH_4 production can be measured throughout estuarine habitats, the rates are typically orders of magnitude lower than those of SO_4^{2-} reduction (Sela-Adler et al. 2017). Methane production rates often appear low because the methane that is produced can be rapidly reoxidized (**Figure 5**), either by reversal of the metabolic pathway in the same organisms or by syntrophic interactions with others (Kevorkian et al. 2022). In one study, up to 25% of SO_4^{2-} reduction was coupled via electron transfer with CH_4

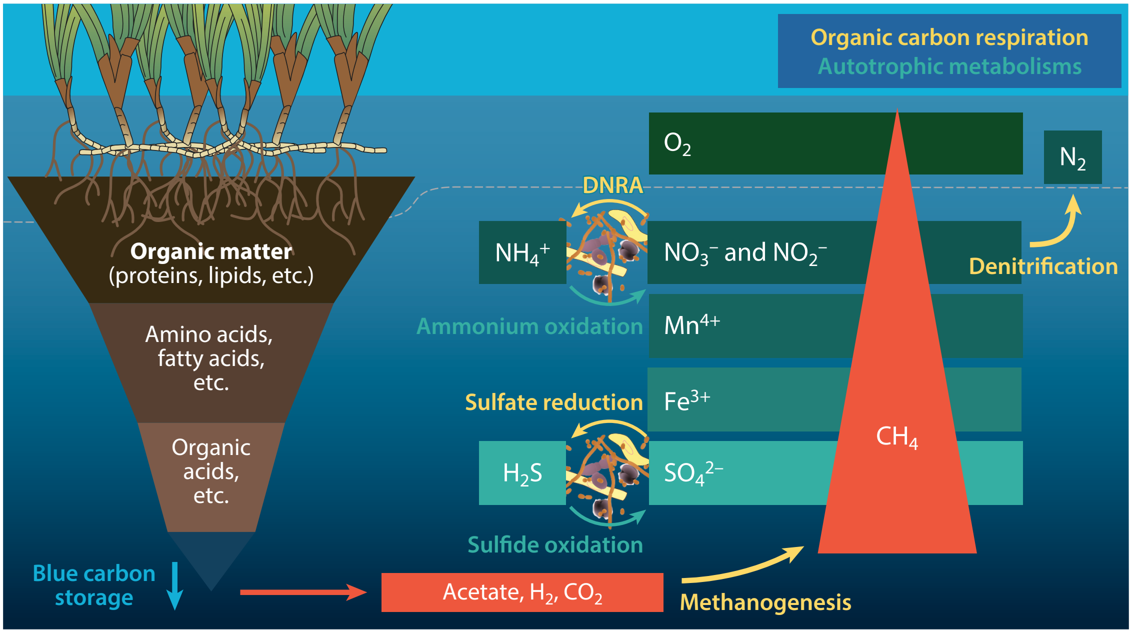

Figure 5

Microbial respiratory processes in sediments. Newly derived organic material at the surface of sediments is selectively decomposed, ultimately producing CO_2, acetate, and H_2 that can be used to support methanogenesis. The remaining organic material that is resistant to decomposition is stored as blue carbon. The methane that is produced via methanogenesis can then be reoxidized through microbial transformations using several electron acceptors, including SO_4^{2-}, NO_3^-, and O_2. Abbreviation: DNRA, dissimilatory nitrate reduction to ammonium. Figure created with symbols from the Integration and Application Network (**https://ian.umces.edu/media-library**) (CC BY-SA 4.0). This figure is licensed under a Creative Commons Attribution-ShareAlike 4.0 International (CC BY-SA 4.0) license.

oxidation, demonstrating the importance of the syntrophic interaction between SO_4^{2-} reducers and the supply of CH_4 produced by methanogens (Sawicka & Brüchert 2017).

A diverse collection of microbes are responsible for methane and sulfur cycles in estuaries. As with denitrification, the genetic capacity for SO_4^{2-} reduction is not phylogenetically constrained and can be carried out by at least 13 microbial phyla (Anantharaman et al. 2018), including Proteobacteria, Nitrospirae, and Firmicutes (among the bacterial lineages) as well as Archaeoglobus from the Euryarchaeota and Thermoproteaceae from the Crenarchaeota (among the archaeal lineages) (Chernyh et al. 2020). In contrast, the capacity for sulfide oxidation is performed mainly by a small number of clades within the Proteobacteria, with members of the Gammaproteobacteria being particularly common in estuarine systems (Baker et al. 2015, Thomas et al. 2014), along with members of the Chlorobia (Gregersen et al. 2011). Oxidation of reduced sulfur compounds can occur through two different pathways, mediated either by the SOX complex or through a reverse-acting dissimilatory sulfite reductase pathway (Thomas et al. 2014), and is typically, though not always (Xia et al. 2017), autotrophic, using either O_2 or NO_3^- as terminal electron acceptors. Some taxa capable of sulfur oxidation feature unique metabolic strategies, such as storage of NO_3^- in vacuoles for later oxidation of sulfide, as is done by the Gammaproteobacteria *Beggiatoa*, and transmission of electrons from oxic sediment surfaces to suboxic depths by cable bacteria belonging to the Desulfobulbaceae (Malkin et al. 2014).

The phylogeny of methanogens continues to shift as genomic insights reveal greater methanogen diversity than previously recognized (Sorokin et al. 2017). All known methanogens belong to the Archaea and until recently were thought to be part of the Euryarchaeota; however, recent discoveries indicate that the genetic capacity for methanogenesis may also exist in the Bathyarchaeota and the Verstraetearchaeota (Evans et al. 2019). Euryarchaeota also perform anaerobic methane oxidation, or methanotrophy, through partial reversal of methanogenesis by the Methanophagales and one clade of Methanosarcinales (Evans et al. 2019). Methanotrophy is also widespread among bacterial lineages and can involve a diverse array of electron acceptors (**Figure 5**) to reduce the vast majority of methane produced in coastal sediments (Kevorkian et al. 2022).

PLANT–MICROBE AND ANIMAL–MICROBE INTERACTIONS

Although there have been numerous studies on plant–microbe and animal–microbe mutualisms in estuaries, research on host-associated microbiomes in estuaries still lags far behind our understanding of these interactions in humans and other model organisms. Host-associated microbiomes provide concrete benefits to hosts through nutrient provisioning, defense, and chemical signaling, while perturbations to the host microbiomes can lead to decreased fitness and disease. Understanding the specific nature of host–microbe interactions and how they will respond to environmental change is critical for predicting the sustainability of estuarine habitats and commercially important organisms.

Microbe–Plant Interactions

Microbes associated with seagrasses, submerged aquatic vegetation, salt marshes, and mangroves are important in host physiology, contributing to pathogen inhibition and influencing plant growth directly through the release of plant growth-promoting compounds and the removal of potential toxins such as hydrogen sulfide. There has long been evidence that nitrogen-fixing microbes promote the success of seagrasses in oligotrophic waters, such as in the Mediterranean Sea (Welsh 2000). An exciting recent finding, however, suggests that seagrasses may have endosymbiotic nitrogen fixers in their roots, akin to those found in leguminous land plants, providing further evidence for this important mutualism (Mohr et al. 2021). Seagrass roots release oxygen into sediments and create oxic microzones (**Figure 1**) that lead to an increased abundance of sulfur-oxidizing bacteria compared with those found in bulk sediment and decreased concentrations of toxic hydrogen sulfide (Miranda et al. 2022 and references therein). Initially, the symbiosis responsible for reducing hydrogen sulfide was thought to require a three-way relationship in which sulfur-oxidizing bacteria lived in association with the gills of lucinid clams living in seagrass sediments (Van Der Heide et al. 2012). More recent genomic work, however, has shown that sulfur-oxidizing microbes are present on root surfaces of seagrasses worldwide, even in the absence of lucinid clams (Martin et al. 2020, Crump et al. 2018). It is evident that both nitrogen-fixing and sulfur-oxidizing microbes support the global distribution of seagrasses in estuarine and coastal waters, and further research on their role in facilitating successful seagrass restoration is warranted.

As with seagrass systems, the microbiomes of salt marsh and mangrove vegetation contribute to several critical ecosystem functions. For example, fungi are important primary decomposers of standing dead biomass in salt marshes (Buchan et al. 2003) and are important in facilitating salt marsh denitrification (Kearns et al. 2019, Starr et al. 2022). Many of the microbe–plant interactions described above for seagrass rhizospheres, including high rates of nitrogen fixation and release of oxygen from roots stimulating an active sulfur cycle, are also important in salt marsh (Rolando et al. 2022, Thomas et al. 2014) and mangrove (Zhuang et al. 2020) sediments. Recent research in salt marshes indicates, however, that rhizosphere microbiomes are strongly influenced by plant genetics. For example, there is a greater diversity of microbes in regions colonized by the tall variation of the marsh plant *S. alterniflora* compared with regions colonized by the short variation, with the tall form being dominated by iron oxidizers and the short form being dominated by sulfur oxidizers (Kolton et al. 2020). Not only is there a difference between tall and short forms of *S. alterniflora*, but there is also an effect of plant genotype, with evidence of unique microbial signatures in the rhizospheres of different genotypes within the same species (Hanley et al. 2021), differences that can mediate the effect of flooding on soil microbial community structure (Tang et al. 2021). Lastly, the microbiome found on the blades of macroalgae also plays important roles in defense, nutrient exchange, reproduction, and settlement (Weigel et al. 2022), and environmental changes can disrupt the host–microbe symbiosis, leading to disease.

Microbe–Animal Interactions

Many animals in estuaries rely on microbiomes for a range of different processes. Microbes can provide reproductive and settling cues for marine invertebrates and can produce key nutrients and vitamins for their hosts. However, much of the research on animal-host-associated microbes in estuaries focuses on the role of microbes in diseases of marine organisms. Dysbiosis, a shift in microbiomes that is associated with disease, can be induced by environmental change (King et al. 2019) and can cause disease of commercially important estuarine organisms, including oysters (Kunselman et al. 2022) and lobsters (Feinman et al. 2017). Animal-host microbiomes are sensitive to both environmental change and stress-induced changes to host physiology (Unzueta-Martínez et al. 2022), so understanding microbial shifts that result from global change drivers is critical for protecting commercially important species.

Recent reviews have addressed the role of microbe–animal associations in marine systems (Apprill 2017, Moulton et al. 2016), corals (van Oppen & Blackall 2019), and sponges (Lurgi et al. 2019). One of the most important of these microbe–animal associations for estuarine ecosystem function is found in reef-building bivalves and benthic macrofauna. Reef-building bivalves couple pelagic and benthic systems by filtering phytoplankton from the water and shuttling waste to the sediments in the form of biodeposits (Kellogg et al. 2013). This filtration improves water quality and decreases the symptoms of eutrophication, in part because the biodeposits promote microbially mediated benthic denitrification (Ayvazian et al. 2021). The bioreactivity and denitrification potential of those deposits and the microbes that decompose them vary by bivalve species, with consequences for estuarine nitrogen removal (Murphy et al. 2019), making predicting effects challenging. Benthic macrofauna also affect estuarine microbial ecology both directly through their own symbiotic relationships and through engineering sediments, which leads to greater oxygen penetration and increased coupled nitrification–denitrification (Stief 2013). However, little is known about the direct effect of host-associated microbes for many species of benthic invertebrates. As climate change continues to shift the geographic ranges of important estuarine taxa (e.g., Johnson 2015) and their associated microbes, novel communities will form, with potential implications for transmission of microbial disease.

CONCLUSION

Estuaries are centers of human civilization, and many of these ecosystems have been impacted by anthropogenic activities for decades or centuries. Estuaries are now facing a new set of anthropogenic impacts associated with climate change, including sea level rise, ocean acidification and deoxygenation, altered hydrological cycles, and shifting geographic ranges of estuarine species. Predicting the impacts of these changes in estuaries requires a sound understanding of estuarine ecology. Microbes are intimately involved in this ecology through their contributions to estuarine food webs, control of elemental cycling, and interactions with plants and animals. New molecular techniques are expanding our understanding of estuarine microbial taxa and the microbial processes that underlie the ecosystem services provided by estuaries. Considering the outsized roles that microbes play in the functioning of estuaries, continued research on microbial diversity and function is important for the health and resilience of estuarine ecosystems.

DISCLOSURE STATEMENT

The authors are not aware of any affiliations, memberships, funding, or financial holdings that might be perceived as affecting the objectivity of this review.

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LITERATURE CITED

Abril G, Etcheber H, Le Hir P, Bassoullet P, Boutier B, Frankignoulle M. 1999. Oxic/anoxic oscillations and organic carbon mineralization in an estuarine maximum turbidity zone (The Gironde, France). *Limnol. Oceanogr.* 44:1304–15

Algar CK, Vallino JJ. 2014. Predicting microbial nitrate reduction pathways in coastal sediments. *Aquat. Microb. Ecol.* 71:223–38

Alongi DM. 2020. Carbon balance in salt marsh and mangrove ecosystems: a global synthesis. *J. Mar. Sci. Eng.* 8:767

Anantharaman K, Hausmann B, Jungbluth SP, Kantor RS, Lavy A, et al. 2018. Expanded diversity of microbial groups that shape the dissimilatory sulfur cycle. *ISME J.* 12:1715–28

Apprill A. 2017. Marine animal microbiomes: toward understanding host-microbiome interactions in a changing ocean. *Front. Mar. Sci.* 4:222

Armitage DW, Gallagher KL, Youngblut ND, Buckley DH, Zinder SH. 2012. Millimeter-scale patterns of phylogenetic and trait diversity in a salt marsh microbial mat. *Front. Microbiol.* 3:293

Arun S, Ramasamy S, Pakshirajan K. 2021. Mechanistic insights into nitrification by microalgae-bacterial consortia in a photo-sequencing batch reactor under different light intensities. *J. Clean. Prod.* 321:128752

Asmala E, Bowers DG, Autio R, Kaartokallio H, Thomas DN. 2014. Qualitative changes of riverine dissolved organic matter at low salinities due to flocculation: riverine DOM flocculation. *J. Geophys. Res. Biogeosci.* 119:1919–33

Ayvazian S, Mulvaney K, Zarnoch C, Palta M, Reichert-Nguyen J, et al. 2021. Beyond bioextraction: the role of oyster-mediated denitrification in nutrient management. *Environ. Sci. Technol.* 55:14457–65

Azam F, Fenchel T, Field J, Gray J, Meyer-Reil L, Thingstad F. 1983. The ecological role of water-column microbes in the sea. *Mar. Ecol. Prog. Ser.* 10:257–63

Baker BJ, Lazar CS, Teske AP, Dick GJ. 2015. Genomic resolution of linkages in carbon, nitrogen, and sulfur cycling among widespread estuary sediment bacteria. *Microbiome* 3:14

Bakken LR, Bergaust L, Liu B, Frostegård Å. 2012. Regulation of denitrification at the cellular level: a clue to the understanding of N_2O emissions from soils. *Philos. Trans. R. Soc. B* 367:1226–34

Baltar F, Herndl GJ. 2019. Ideas and perspectives: Is dark carbon fixation relevant for oceanic primary production estimates? *Biogeosciences* 16:3793–99

Bergtold M, Gunter V, Traunspurger W. 2005. Is there competition among ciliates and nematodes? *Freshw. Biol.* 50:1351–59

Bernhard AE, Landry ZC, Blevins A, De La Torre JR, Giblin AE, Stahl DA. 2010. Abundance of ammonia-oxidizing archaea and bacteria along an estuarine salinity gradient in relation to potential nitrification rates. *Appl. Environ. Microbiol.* 76:1285–89

Bianchi TS, Cui X, Blair NE, Burdige DJ, Eglinton TI, Galy V. 2018. Centers of organic carbon burial and oxidation at the land-ocean interface. *Org. Geochem.* 115:138–55

Bolhuis H, Stal LJ. 2011. Analysis of bacterial and archaeal diversity in coastal microbial mats using massive parallel 16S rRNA gene tag sequencing. *ISME J.* 5:1701–12

Boschker HTS, Vasquez-Cardenas D, Bolhuis H, Moerdijk-Poortvliet TWC, Moodley L. 2014. Chemoautotrophic carbon fixation rates and active bacterial communities in intertidal marine sediments. *PLOS ONE* 9:e101443

Bowen JL, Giblin AE, Murphy AE, Bulseco AN, Deegan LA, et al. 2020. Not all nitrogen is created equal: differential effects of nitrate and ammonium enrichment in coastal wetlands. *BioScience* 70:1108–19

Bowen JL, Weisman D, Yasuda M, Jayakumar A, Morrison HG, Ward BB. 2015. Marine oxygen-deficient zones harbor depauperate denitrifying communities compared to novel genetic diversity in coastal sediments. *Microb. Ecol.* 70:311–21

Bräuer SL, Kranzler K, Goodson N, Murphy D, Simon HM, et al. 2013. Dark carbon fixation in the Columbia River's estuarine turbidity maxima: molecular characterization of red-type cbbL genes and measurement of DIC uptake rates in response to added electron donors. *Estuaries Coasts* 36:1073–83

Braun A, Spona-Friedl M, Avramov M, Elsner M, Baltar F, et al. 2021. Reviews and syntheses: heterotrophic fixation of inorganic carbon – significant but invisible flux in environmental carbon cycling. *Biogeosciences* 18:3689–700

Breitburg D, Levin LA, Oschlies A, Grégoire M, Chavez FP, et al. 2018. Declining oxygen in the global ocean and coastal waters. *Science* 359:eaam7240

Brussaard C, Gast G, van Duyl F, Riegman R. 1996. Impact of phytoplankton bloom magnitude on a pelagic microbial food web. *Mar. Ecol. Prog. Ser.* 144:211–21

Buchan A, Newell SY, Butler M, Biers EJ, Hollibaugh JT, Moran MA. 2003. Dynamics of bacterial and fungal communities on decaying salt marsh grass. *Appl. Environ. Microbiol.* 69:6676–87

Bulseco AN, Giblin AE, Tucker J, Murphy AE, Sanderman J, et al. 2019. Nitrate addition stimulates microbial decomposition of organic matter in salt marsh sediments. *Glob. Change Biol.* 25:3224–41

Bulseco AN, Vineis JH, Murphy AE, Spivak AC, Giblin AE, et al. 2020. Metagenomics coupled with biogeochemical rates measurements provide evidence that nitrate addition stimulates respiration in salt marsh sediments. *Limnol. Oceanogr.* 65(S1):S321–39

Burgin AJ, Hamilton SK. 2007. Have we overemphasized the role of denitrification in aquatic ecosystems? A review of nitrate removal pathways. *Front. Ecol. Environ.* 5:89–96

Caffrey JM. 2004. Factors controlling net ecosystem metabolism in U.S. estuaries. *Estuaries* 27:90–101

Caffrey JM, Bano N, Kalanetra K, Hollibaugh JT. 2007. Ammonia oxidation and ammonia-oxidizing bacteria and archaea from estuaries with differing histories of hypoxia. *ISME J.* 1:660–62

Canfield DE, Farquhar J. 2009. Animal evolution, bioturbation, and the sulfate concentration of the oceans. *PNAS* 106:8123–27

Canfield DE, Rosing MT, Bjerrum C. 2006. Early anaerobic metabolisms. *Philos. Trans. R. Soc. B* 361:1819–36

Chambers LG, Osborne TZ, Reddy KR. 2013. Effect of salinity-altering pulsing events on soil organic carbon loss along an intertidal wetland gradient: a laboratory experiment. *Biogeochemistry* 115:363–83

Chen J, Erler DV, Wells NS, Huang J, Welsh DT, Eyre BD. 2021. Denitrification, anammox, and dissimilatory nitrate reduction to ammonium across a mosaic of estuarine benthic habitats. *Limnol. Oceanogr.* 66:1281–97

Chernyh NA, Neukirchen S, Frolov EN, Sousa FL, Miroshnichenko ML, et al. 2020. Dissimilatory sulfate reduction in the archaeon '*Candidatus* Vulcanisaeta moutnovskia' sheds light on the evolution of sulfur metabolism. *Nat. Microbiol.* 5:1428–38

Chmura GL, Anisfeld SC, Cahoon DR, Lynch JC. 2003. Global carbon sequestration in tidal, saline wetland soils. *Glob. Biogeochem. Cycles* 17:1111

Chmura GL, Hung GA. 2004. Controls on salt marsh accretion: a test in salt marshes of Eastern Canada. *Estuaries* 27:70–81

Comer-Warner SA, Nguyen ATQ, Nguyen MN, Wang M, Turner A, et al. 2022. Restoration impacts on rates of denitrification and greenhouse gas fluxes from tropical coastal wetlands. *Sci. Total Environ.* 803:149577

Crump BC, Fine LM, Fortunato CS, Herfort L, Needoba JA, et al. 2017. Quantity and quality of particulate organic matter controls bacterial production in the Columbia River estuary. *Limnol. Oceanogr.* 62:2713–31

Crump BC, Peranteau C, Beckingham B, Cornwell JC. 2007. Respiratory succession and community succession of bacterioplankton in seasonally anoxic estuarine waters. *Appl. Environ. Microbiol.* 73:6802–10

Crump BC, Wojahn JM, Tomas F, Mueller RS. 2018. Metatranscriptomics and amplicon sequencing reveal mutualisms in seagrass microbiomes. *Front. Microbiol.* 9:388

Cuadrado DG, Carmona NB, Bournod C. 2011. Biostabilization of sediments by microbial mats in a temperate siliciclastic tidal flat, Bahia Blanca estuary (Argentina). *Sediment. Geol.* 237:95–101

Daims H, Lebedeva EV, Pjevac P, Han P, Herbold C, et al. 2015. Complete nitrification by *Nitrospira* bacteria. *Nature* 528:504–9

Damashek J, Casciotti KL, Francis CA. 2016. Variable nitrification rates across environmental gradients in turbid, nutrient-rich estuary waters of San Francisco Bay. *Estuaries Coasts* 39:1050–71

Damashek J, Francis CA. 2018. Microbial nitrogen cycling in estuaries: from genes to ecosystem processes. *Estuaries Coasts* 41:626–60

Deegan LA, Johnson DS, Warren RS, Peterson BJ, Fleeger JW, et al. 2012. Coastal eutrophication as a driver of salt marsh loss. *Nature* 490:388–92

Delmont TO, Quince C, Shaiber A, Esen ÖC, Lee ST, et al. 2018. Nitrogen-fixing populations of Planctomycetes and Proteobacteria are abundant in surface ocean metagenomes. *Nat. Microbiol.* 3:804–13

DeLorenzo S, Bräuer SL, Edgmont CA, Herfort L, Tebo BM, Zuber P. 2012. Ubiquitous dissolved inorganic carbon assimilation by marine bacteria in the Pacific Northwest coastal ocean as determined by stable isotope probing. *PLOS ONE* 7:e46695

Dietrich D, Arndt H. 2000. Biomass partitioning of benthic microbes in a Baltic inlet: relationships between bacteria, algae, heterotrophic flagellates and ciliates. *Mar. Biol.* 136:309–22

Domangue RJ, Mortazavi B. 2018. Nitrate reduction pathways in the presence of excess nitrogen in a shallow eutrophic estuary. *Environ. Pollut.* 238:599–606

Du Y, Xu K, Warren A, Lei Y, Dai R. 2012. Benthic ciliate and meiofaunal communities in two contrasting habitats of an intertidal estuarine wetland. *J. Sea Res.* 70:50–63

Duarte CM, Middelburg JJ, Caraco N. 2005. Major role of marine vegetation on the oceanic carbon cycle. *Biogeosciences* 2:1–8

Dyksma S, Bischof K, Fuchs BM, Hoffmann K, Meier D, et al. 2016. Ubiquitous Gammaproteobacteria dominate dark carbon fixation in coastal sediments. *ISME J.* 10:1939–53

Eggleston EM, Lee DY, Owens MS, Cornwell JC, Crump BC, Hewson I. 2015. Key respiratory genes elucidate bacterial community respiration in a seasonally anoxic estuary. *Environ. Microbiol.* 17:2306–18

Eppley RW, Peterson BJ. 1979. Particulate organic matter flux and planktonic new production in the deep ocean. *Nature* 282:677–80

Epstein SS. 1997. Microbial food webs in marine sediments. I. Trophic interactions and grazing rates in two tidal flat communities. *Microb. Ecol.* 34:188–98

Evans PN, Boyd JA, Leu AO, Woodcroft BJ, Parks DH, et al. 2019. An evolving view of methane metabolism in the Archaea. *Nat. Rev. Microbiol.* 17:219–32

Feinman S, Unzueta Martínez A, Bowen J, Tlusty M. 2017. Fine-scale transition to lower bacterial diversity and altered community composition precedes shell disease in laboratory-reared juvenile American lobster. *Dis. Aquat. Org.* 124:41–54

Fennel K, Testa JM. 2019. Biogeochemical controls on coastal hypoxia. *Annu. Rev. Mar. Sci.* 11:105–30

Figueroa D, Capo E, Lindh MV, Rowe OF, Paczkowska J, et al. 2021. Terrestrial dissolved organic matter inflow drives temporal dynamics of the bacterial community of a subarctic estuary (northern Baltic Sea). *Environ. Microbiol.* 23:4200–13

Fortunato CS, Crump BC. 2015. Microbial gene abundance and expression patterns across a river to ocean salinity gradient. *PLOS ONE* 10:e0140578

Gauns M, Mochemadkar S, Patil S, Pratihary A, Naqvi SWA, Madhupratap M. 2015. Seasonal variations in abundance, biomass and grazing rates of microzooplankton in a tropical monsoonal estuary. *J. Oceanogr.* 71:345–59

Geraldi NR, Ortega A, Serrano O, Macreadie PI, Lovelock CE, et al. 2019. Fingerprinting blue carbon: rationale and tools to determine the source of organic carbon in marine depositional environments. *Front. Mar. Sci.* 6:263

Giblin A, Tobias C, Song B, Weston N, Banta G, Rivera-Monroy V. 2013. The importance of dissimilatory nitrate reduction to ammonium (DNRA) in the nitrogen cycle of coastal ecosystems. *Oceanography* 26(3):124–31

Giovannoni SJ, DeLong EF, Schmidt TM, Pace NR. 1990. Tangential flow filtration and preliminary phylogenetic analysis of marine picoplankton. *Appl. Environ. Microbiol.* 56:2572–75

Glaubitz S, Lueders T, Abraham W-R, Jost G, Jürgens K, Labrenz M. 2009. ^{13}C-isotope analyses reveal that chemolithoautotrophic *Gamma*- and *Epsilonproteobacteria* feed a microbial food web in a pelagic redoxcline of the central Baltic Sea. *Environ. Microbiol.* 11:326–37

Gregersen LH, Bryant DA, Frigaard N-U. 2011. Mechanisms and evolution of oxidative sulfur metabolism in green sulfur bacteria. *Front. Microbiol.* 2:116

Hanley TC, Bowen JL, Kearns PJ, Hughes AR. 2021. Short- and long-term effects of nutrient enrichment on salt marsh plant production and microbial community structure. *J. Ecol.* 109:3779–93

Hetland RD, DiMarco SF. 2008. How does the character of oxygen demand control the structure of hypoxia on the Texas-Louisiana continental shelf? *J. Mar. Syst.* 70:49–62

Hewson I, Eggleston EM, Doherty M, Lee DY, Owens M, et al. 2014. Metatranscriptomic analyses of plankton communities inhabiting surface and subpycnocline waters of the Chesapeake Bay during oxic-anoxic-oxic transitions. *Appl. Environ. Microbiol.* 80:328–38

Hoffman DK, McCarthy MJ, Newell SE, Gardner WS, Niewinski DN, et al. 2019. Relative contributions of DNRA and denitrification to nitrate reduction in *Thalassia testudinum* seagrass beds in coastal Florida (USA). *Estuaries Coasts* 42:1001–14

Hopkinson CS, Smith EM. 2005. Estuarine respiration: an overview of benthic, pelagic, and whole system respiration. In *Respiration in Aquatic Ecosystems*, ed. P del Giorgio, P Williams, pp. 122–46. Oxford, UK: Oxford Univ. Press

Howarth RW. 1984. The ecological significance of sulfur in the energy dynamics of salt marsh and coastal marine sediments. *Biogeochemistry* 1:5–27

Howarth RW, Marino R, Cole JJ. 1988. Nitrogen fixation in freshwater, estuarine, and marine ecosystems. 2. Biogeochemical control. *Limnol. Oceanogr.* 33:688–701

Husband JD, Kiene RP. 2007. Occurrence of dimethylsulfoxide in leaves, stems, and roots of *Spartina alterniflora*. *Wetlands* 27:224–29

Jabir T, Vipindas PV, Jesmi Y, Valliyodan S, Parambath PM, et al. 2020. Nutrient stoichiometry (N:P) controls nitrogen fixation and distribution of diazotrophs in a tropical eutrophic estuary. *Mar. Pollut. Bull.* 151:110799

Johnson DS. 2015. The savory swimmer swims north: a northern range extension of the blue crab *Callinectes sapidus*? *J. Crustac. Biol.* 35:105–10

Jørgensen BB, Sørensen J. 1985. Seasonal cycles of O_2, $NO_3{}^-$, and $SO_4{}^{2-}$ reduction in estuarine sediments: the significance of an $NO_3{}^-$ reduction maximum in spring. *Mar. Ecol. Prog. Ser.* 24:65–74

Juniper SK, Brinkhurst RO. 1986. Water-column dark CO_2 fixation and bacterial-mat growth in intermittently anoxic Saanich Inlet, British Columbia. *Mar. Ecol. Prog. Ser.* 33:41–50

Kearns PJ, Bulseco-McKim AN, Hoyt H, Angell JH, Bowen JL. 2019. Nutrient enrichment alters salt marsh fungal communities and promotes putative fungal denitrifiers. *Microb. Ecol.* 77:358–69

Kellogg CTE, McClelland JW, Dunton KH, Crump BC. 2019. Strong seasonality in Arctic estuarine microbial food webs. *Front. Microbiol.* 10:2628

Kellogg M, Cornwell J, Owens M, Paynter K. 2013. Denitrification and nutrient assimilation on a restored oyster reef. *Mar. Ecol. Prog. Ser.* 480:1–19

Kevorkian RT, Sipes K, Winstead R, Paul R, Lloyd KG. 2022. Cryptic methane-cycling by methanogens during multi-year incubation of estuarine sediment. *Front. Microbiol.* 13:847563

Kiene RP, Service SK. 1991. Decomposition of dissolved DMSP and DMS in estuarine waters: dependence on temperature and substrate concentration. *Mar. Ecol. Prog. Ser.* 76:1–11

Kim C, Kang HY, Lee Y-J, Yun S-G, Kang C-K. 2020. Isotopic variation of macroinvertebrates and their sources of organic matter along an estuarine gradient. *Estuaries Coasts* 43:496–511

King WL, Jenkins C, Seymour JR, Labbate M. 2019. Oyster disease in a changing environment: decrypting the link between pathogen, microbiome and environment. *Mar. Environ. Res.* 143:124–40

Kirchman DL. 2016. Growth rates of microbes in the oceans. *Annu. Rev. Mar. Sci.* 8:285–309

Kolton M, Rolando JL, Kostka JE. 2020. Elucidation of the rhizosphere microbiome linked to *Spartina alterniflora* phenotype in a salt marsh on Skidaway Island, Georgia, USA. *FEMS Microbiol. Ecol.* 96:fiaa026

Könneke M, Bernhard AE, De La Torre JR, Walker CB, Waterbury JB, Stahl DA. 2005. Isolation of an autotrophic ammonia-oxidizing marine archaeon. *Nature* 437:543–46

Könneke M, Schubert DM, Brown PC, Hügler M, Standfest S, et al. 2014. Ammonia-oxidizing archaea use the most energy-efficient aerobic pathway for CO_2 fixation. *PNAS* 111:8239–44

Koop-Jakobsen K, Giblin AE. 2009. Anammox in tidal marsh sediments: the role of salinity, nitrogen loading, and marsh vegetation. *Estuaries Coasts* 32:238–45

Kudo I, Hisatoku T, Yoshimura T, Maita Y. 2015. Primary productivity and nitrogen assimilation with identifying the contribution of urea in Funka Bay, Japan. *Estuar. Coast. Shelf Sci.* 158:12–19

Kunselman E, Minich JJ, Horwith M, Gilbert JA, Allen EE. 2022. Variation in survival and gut microbiome composition of hatchery-grown native oysters at various locations within the Puget Sound. *Microbiol. Spectr.* 10:e01982-21

Lei Y, Stumm K, Wickham SA, Berninger U. 2014. Distributions and biomass of benthic ciliates, foraminifera and amoeboid protists in marine, brackish, and freshwater sediments. *J. Eukaryot. Microbiol.* 61:493–508

Li C, Reimers C, Chapman J. 2020a. Microbiome analyses and presence of cable bacteria in the burrow sediment of *Upogebia pugettensis*. *Mar. Ecol. Prog. Ser.* 648:79–94

Li D, Jing H, Zhang R, Yang W, Chen M, et al. 2020b. Heterotrophic diazotrophs in a eutrophic temperate bay (Jiaozhou Bay) broadens the domain of N_2 fixation in China's coastal waters. *Estuar. Coast. Shelf Sci.* 242:106778

Liu B, Hou L, Zheng Y, Zhang Z, Tang X, et al. 2022. Dark carbon fixation in intertidal sediments: controlling factors and driving microorganisms. *Water Res.* 216:118381

Llirós M, Alonso-Sáez L, Gich F, Plasencia A, Auguet O, et al. 2011. Active bacteria and archaea cells fixing bicarbonate in the dark along the water column of a stratified eutrophic lagoon: dark C fixation by planktonic bacteria and archaea. *FEMS Microbiol. Ecol.* 77:370–84

Lurgi M, Thomas T, Wemheuer B, Webster NS, Montoya JM. 2019. Modularity and predicted functions of the global sponge-microbiome network. *Nat. Commun.* 10:992

Macreadie PI, Anton A, Raven JA, Beaumont N, Connolly RM, et al. 2019. The future of Blue Carbon science. *Nat. Commun.* 10:3998

Malkin SY, Rao AM, Seitaj D, Vasquez-Cardenas D, Zetsche E-M, et al. 2014. Natural occurrence of microbial sulphur oxidation by long-range electron transport in the seafloor. *ISME J.* 8:1843–54

Malkin SY, Seitaj D, Burdorf LDW, Nieuwhof S, Hidalgo-Martinez S, et al. 2017. Electrogenic sulfur oxidation by cable bacteria in bivalve reef sediments. *Front. Mar. Sci.* 4:28

Mandal A, Dutta A, Das R, Mukherjee J. 2021. Role of intertidal microbial communities in carbon dioxide sequestration and pollutant removal: a review. *Mar. Pollut. Bull.* 170:112626

Marcarelli AM, Fulweiler RW, Scott JT. 2022. Nitrogen fixation: a poorly understood process along the freshwater-marine continuum. *Limnol. Oceanogr. Lett.* 7:1–10. Corrigendum. 2022. *Limnol. Oceanogr. Lett.* 7:450

Margalef R. 1968. *Perspectives in Ecological Theory*. Chicago: Univ. Chicago Press

Martin BC, Alarcon MS, Gleeson D, Middleton JA, Fraser MW, et al. 2020. Root microbiomes as indicators of seagrass health. *FEMS Microbiol. Ecol.* 96:fiz201

Mason OU, Chanton P, Knobbe LN, Zaugg J, Mortazavi B. 2021. New insights into the influence of plant and microbial diversity on denitrification rates in a salt marsh. *Wetlands* 41:33

Meador TB, Schoffelen N, Ferdelman TG, Rebello O, Khachikyan A, Könneke M. 2020. Carbon recycling efficiency and phosphate turnover by marine nitrifying archaea. *Sci. Adv.* 6:eaba1799

Medeiros PM, Seidel M, Gifford SM, Ballantyne F, Dittmar T, et al. 2017. Microbially-mediated transformations of estuarine dissolved organic matter. *Front. Mar. Sci.* 4:69

Messer LF, Brown MV, Van Ruth PD, Doubell M, Seymour JR. 2021. Temperate southern Australian coastal waters are characterised by surprisingly high rates of nitrogen fixation and diversity of diazotrophs. *PeerJ* 9:e10809

Middelburg JJ. 2011. Chemoautotrophy in the ocean. *Geophys. Res. Lett.* 38:L24604

Middelburg JJ. 2018. Reviews and syntheses: to the bottom of carbon processing at the seafloor. *Biogeosciences* 15:413–27

Mills MM, Turk-Kubo KA, van Dijken GL, Henke BA, Harding K, et al. 2020. Unusual marine cyanobacteria/haptophyte symbiosis relies on N_2 fixation even in N-rich environments. *ISME J.* 14:2395–406

Miranda K, Weigel BL, Fogarty EC, Veseli IA, Giblin AE, et al. 2022. The diversity and functional capacity of microbes associated with coastal macrophytes. *mSystems* 7:e00592-22

Mohr W, Lehnen N, Ahmerkamp S, Marchant HK, Graf JS, et al. 2021. Terrestrial-type nitrogen-fixing symbiosis between seagrass and a marine bacterium. *Nature* 600:105–9

Molari M, Manini E, Dell'Anno A. 2013. Dark inorganic carbon fixation sustains the functioning of benthic deep-sea ecosystems. *Glob. Biogeochem. Cycles* 27:212–21

Morelle J, Roose-Amsaleg C, Laverman AM. 2022. Microphytobenthos as a source of labile organic matter for denitrifying microbes. *Estuar. Coast. Shelf Sci.* 275:108006

Morency C, Jacquemot L, Potvin M, Lovejoy C. 2022. A microbial perspective on the local influence of Arctic rivers and estuaries on Hudson Bay (Canada). *Elem. Sci. Anthr.* 10:00009

Mortazavi B, Iverson R, Huang W, Lewis F, Caffrey J. 2000. Nitrogen budget of Apalachicola Bay, a bar-built estuary in the northeastern Gulf of Mexico. *Mar. Ecol. Prog. Ser.* 195:1–14

Moseman-Valtierra SM, Szura K, Eagle M, Thornber CS, Wang F. 2022. CO_2 uptake offsets other greenhouse gas emissions from salt marshes with chronic nitrogen loading. *Wetlands* 42:79

Mosier AC, Francis CA. 2008. Relative abundance and diversity of ammonia-oxidizing archaea and bacteria in the San Francisco Bay estuary. *Environ. Microbiol.* 10:3002–16

Moulton OM, Altabet MA, Beman JM, Deegan LA, Lloret J, et al. 2016. Microbial associations with macrobiota in coastal ecosystems: patterns and implications for nitrogen cycling. *Front. Ecol. Environ.* 14:200–8

Murphy AE, Kolkmeyer R, Song B, Anderson IC, Bowen J. 2019. Bioreactivity and microbiome of biodeposits from filter-feeding bivalves. *Microb. Ecol.* 77:343–57

Murray RH, Erler DV, Eyre BD. 2015. Nitrous oxide fluxes in estuarine environments: response to global change. *Glob. Change Biol.* 21:3219–45

Murrell MC, Lehrter JC. 2011. Sediment and lower water column oxygen consumption in the seasonally hypoxic region of the Louisiana continental shelf. *Estuaries Coasts* 34:912–24

Oczkowski AJ, Santos EA, Martin RM, Gray AB, Hanson AR, et al. 2020. Unexpected nitrogen sources in a tropical urban estuary. *J. Geophys. Res. Biogeosci.* 125:e2019JG005502

Oremland RS, Polcin S. 1982. Methanogenesis and sulfate reduction: competitive and noncompetitive substrates in estuarine sediments. *Appl. Environ. Microbiol.* 44:1270–76

Page HM. 1997. Importance of vascular plant and algal production to macro-invertebrate consumers in a southern California salt marsh. *Estuar. Coast. Shelf Sci.* 45:823–34

Palacios MM, Trevathan-Tackett SM, Malerba ME, Macreadie PI. 2021. Effects of a nutrient enrichment pulse on blue carbon ecosystems. *Mar. Pollut. Bull.* 165:112024

Pedersen JN, Bombar D, Paerl RW, Riemann L. 2018. Diazotrophs and N_2-fixation associated with particles in coastal estuarine waters. *Front. Microbiol.* 9:2759

Pfeffer C, Larsen S, Song J, Dong M, Besenbacher F, et al. 2012. Filamentous bacteria transport electrons over centimetre distances. *Nature* 491:218–21

Philippot L, Hallin S, Schloter M. 2007. Ecology of denitrifying prokaryotes in agricultural soil. *Adv. Agron.* 96:249–305

Pinckney JL. 2018. A mini-review of the contribution of benthic microalgae to the ecology of the continental shelf in the South Atlantic Bight. *Estuaries Coasts* 41:2070–78

Putland J, Iverson R. 2007. Microzooplankton: major herbivores in an estuarine planktonic food web. *Mar. Ecol. Prog. Ser.* 345:63–73

Riisgård HU, Funch P, Larsen PS. 2015. The mussel filter-pump – present understanding, with a re-examination of gill preparations. *Acta Zool.* 96:273–82

Risgaard-Petersen N. 2003. Coupled nitrification-denitrification in autotrophic and heterotrophic estuarine sediments: on the influence of benthic microalgae. *Limnol. Oceanogr.* 48:93–105

Roco CA, Bergaust LL, Bakken LR, Yavitt JB, Shapleigh JP. 2017. Modularity of nitrogen-oxide reducing soil bacteria: linking phenotype to genotype. *Environ. Microbiol.* 19:2507–19

Rolando JL, Kolton M, Song T, Kostka JE. 2022. The core root microbiome of *Spartina alterniflora* is predominated by sulfur-oxidizing and sulfate-reducing bacteria in Georgia salt marshes, USA. *Microbiome* 10:37

Sawicka JE, Brüchert V. 2017. Annual variability and regulation of methane and sulfate fluxes in Baltic Sea estuarine sediments. *Biogeosciences* 14:325–39

Scholz VV, Brodersen KE, Kühl M, Koren K. 2021a. Resolving chemical gradients around seagrass roots—a review of available methods. *Front. Mar. Sci.* 8:771382

Scholz VV, Martin BC, Meyer R, Schramm A, Fraser MW, et al. 2021b. Cable bacteria at oxygen-releasing roots of aquatic plants: a widespread and diverse plant-microbe association. *New Phytol.* 232:2138–51

Schratzberger M, Ingels J. 2018. Meiofauna matters: the roles of meiofauna in benthic ecosystems. *J. Exp. Mar. Biol. Ecol.* 502:12–25

Sedlacek CJ. 2020. It takes a village: discovering and isolating the nitrifiers. *Front. Microbiol.* 11:1900

Segarra KEA, Comerford C, Slaughter J, Joye SB. 2013. Impact of electron acceptor availability on the anaerobic oxidation of methane in coastal freshwater and brackish wetland sediments. *Geochim. Cosmochim. Acta* 115:15–30

Sela-Adler M, Ronen Z, Herut B, Antler G, Vigderovich H, et al. 2017. Co-existence of methanogenesis and sulfate reduction with common substrates in sulfate-rich estuarine sediments. *Front. Microbiol.* 8:766

Signori CN, Felizardo JPDS, Enrich-Prast A. 2020. Bacterial production prevails over photo- and chemosynthesis in a eutrophic tropical lagoon. *Estuar. Coast. Shelf Sci.* 243:106889

Signori CN, Valentin JL, Pollery RCG, Enrich-Prast A. 2018. Temporal variability of dark carbon fixation and bacterial production and their relation with environmental factors in a tropical estuarine system. *Estuaries Coasts* 41:1089–101

Smith SV, Hollibaugh JT. 1997. Annual cycle and interannual variability of ecosystem metabolism in a temperate climate embayment. *Ecol. Monogr.* 67:509–33

Snedden GA, Cable JE, Kjerfve B. 2023. Estuarine geomorphology, circulation, and mixing. In *Estuarine Ecology*, ed. BC Crump, JM Testa, KH Dunton, pp. 16–35. Hoboken, NJ: Wiley & Sons. 3rd ed.

Soetaert K, Herman PMJ, Middelburg JJ. 1996. A model of early diagenetic processes from the shelf to abyssal depths. *Geochim. Cosmochim. Acta* 60:1019–40

Sorokin DY, Makarova KS, Abbas B, Ferrer M, Golyshin PN, et al. 2017. Discovery of extremely halophilic, methyl-reducing euryarchaea provides insights into the evolutionary origin of methanogenesis. *Nat. Microbiol.* 2:17081

Stal LJ, Bolhuis H, Cretoiu MS. 2019. Phototrophic marine benthic microbiomes: the ecophysiology of these biological entities. *Environ. Microbiol.* 21:1529–51

Starr SF, Mortazavi B, Tatariw C, Kuehn KA, Cherry JA, et al. 2022. Poor recovery of fungal denitrification limits nitrogen removal capacity in a constructed Gulf Coast marsh. *Soil Biol. Biochem.* 170:108692

Stief P. 2013. Stimulation of microbial nitrogen cycling in aquatic ecosystems by benthic macrofauna: mechanisms and environmental implications. *Biogeosciences* 10:7829–46

Sun D, Tang X, Li J, Liu M, Hou L, et al. 2022. Chlorate as a comammox *Nitrospira* specific inhibitor reveals nitrification and N_2O production activity in coastal wetland. *Soil Biol. Biochem.* 173:108782

Sun X, Frey C, Garcia-Robledo E, Jayakumar A, Ward BB. 2021. Microbial niche differentiation explains nitrite oxidation in marine oxygen minimum zones. *ISME J.* 15:1317–29

Tang H, Liebner S, Reents S, Nolte S, Jensen K, et al. 2021. Plant genotype controls wetland soil microbial functioning in response to sea-level rise. *Biogeosciences* 18:6133–46

Tee HS, Waite D, Lear G, Handley KM. 2021. Microbial river-to-sea continuum: gradients in benthic and planktonic diversity, osmoregulation and nutrient cycling. *Microbiome* 9:190

Thomas F, Giblin AE, Cardon ZG, Sievert SM. 2014. Rhizosphere heterogeneity shapes abundance and activity of sulfur-oxidizing bacteria in vegetated salt marsh sediments. *Front. Microbiol.* 5:309

Thompson AW, Foster RA, Krupke A, Carter BJ, Musat N, et al. 2012. Unicellular cyanobacterium symbiotic with a single-celled eukaryotic alga. *Science* 337:1546–50

Tuorto SJ, Taghon GL. 2014. Rates of benthic bacterivory of marine ciliates as a function of prey concentration. *J. Exp. Mar. Biol. Ecol.* 460:129–34

Unzueta-Martínez A, Scanes E, Parker LM, Ross PM, O'Connor W, Bowen JL. 2022. Microbiomes of the Sydney Rock Oyster are acquired through both vertical and horizontal transmission. *Anim. Microbiome* 4:32

Van Der Heide T, Govers LL, De Fouw J, Olff H, Van Der Geest M, et al. 2012. A three-stage symbiosis forms the foundation of seagrass ecosystems. *Science* 336:1432–34

van Oppen MJH, Blackall LL. 2019. Coral microbiome dynamics, functions and design in a changing world. *Nat. Rev. Microbiol.* 17:557–67

Vasquez-Cardenas D, Meysman FJR, Boschker HTS. 2020. A cross-system comparison of dark carbon fixation in coastal sediments. *Glob. Biogeochem. Cycles* 34:e2019GB006298

Vasquez-Cardenas D, Quintana C, Meysman F, Kristensen E, Boschker H. 2016. Species-specific effects of two bioturbating polychaetes on sediment chemoautotrophic bacteria. *Mar. Ecol. Prog. Ser.* 549:55–68

Venter JC, Remington K, Heidelberg JF, Halpern AL, Rusch D, et al. 2004. Environmental genome shotgun sequencing of the Sargasso Sea. *Science* 304:66–74

Vieillard AM, Thrush SF. 2021. Ecogeochemistry and denitrification in non-eutrophic coastal sediments. *Estuaries Coasts* 44:1866–82

Walters K, Moriarty DJW. 1993. The effects of complex trophic interactions on a marine microbenthic community. *Ecology* 74:1475–89

Wang H, Chen F, Zhang C, Wang M, Kan J. 2021. Estuarine gradients dictate spatiotemporal variations of microbiome networks in the Chesapeake Bay. *Environ. Microbiome* 16:22

Weigel BL, Miranda KK, Fogarty EC, Watson AR, Pfister CA. 2022. Functional insights into the kelp microbiome from metagenome-assembled genomes. *mSystems* 7:e01422-21

Welsh A, Chee-Sanford JC, Connor LM, Löffler FE, Sanford RA. 2014. Refined NrfA phylogeny improves PCR-Based *nrfA* gene detection. *Appl. Environ. Microbiol.* 80:2110–19

Welsh DT. 2000. Nitrogen fixation in seagrass meadows: regulation, plant-bacteria interactions and significance to primary productivity. *Ecol. Lett.* 3:58–71

Weston NB, Neubauer SC, Velinsky DJ, Vile MA. 2014. Net ecosystem carbon exchange and the greenhouse gas balance of tidal marshes along an estuarine salinity gradient. *Biogeochemistry* 120:163–89

Wieltschnig C, Fischer UR, Kirschner AKT, Velimirov B. 2003. Benthic bacterial production and protozoan predation in a silty freshwater environment. *Microb. Ecol.* 46:62–72

Wilkerson FP, Dugdale RC, Hogue VE, Marchi A. 2006. Phytoplankton blooms and nitrogen productivity in San Francisco Bay. *Estuaries Coasts* 29:401–16

Williams PJLB. 1981. Microbial contribution to overall marine plankton metabolism: direct measurements of respiration. *Oceanol. Acta* 4:359–64

Winogradsky S. 1890. Recherches sur les organisms de la nitrification. *Ann. Inst. Pasteur* 4:213–31

Woebken D, Lam P, Kuypers MMM, Naqvi SWA, Kartal B, et al. 2008. A microdiversity study of anammox bacteria reveals a novel *Candidatus* Scalindua phylotype in marine oxygen minimum zones. *Environ. Microbiol.* 10:3106–19

Wommack KE, Colwell RR. 2000. Virioplankton: viruses in aquatic ecosystems. *Microbiol. Mol. Biol. Rev.* 64:69–114

Wu D-M, Dai Q-P, Liu X-Z, Fan Y-P, Wang J-X. 2019. Comparison of bacterial community structure and potential functions in hypoxic and non-hypoxic zones of the Changjiang Estuary. *PLOS ONE* 14:e0217431

Xia Y, Lü C, Hou N, Xin Y, Liu J, et al. 2017. Sulfide production and oxidation by heterotrophic bacteria under aerobic conditions. *ISME J.* 11:2754–66

Yong YL, Lee CW, Bong CW, Chew LL, Chong VC. 2022. The role of microzooplankton grazing in the microbial food web of a tropical mangrove estuary. *Estuar. Coast. Shelf Sci.* 275:107969

Zaikova E, Walsh DA, Stilwell CP, Mohn WW, Tortell PD, Hallam SJ. 2010. Microbial community dynamics in a seasonally anoxic fjord: Saanich Inlet, British Columbia. *Environ. Microbiol.* 12:172–91

Zedler JB. 1980. Algal mat productivity: comparisons in a salt marsh. *Estuaries* 3:122

Zehr JP, Capone DG. 2020. Changing perspectives in marine nitrogen fixation. *Science* 368:eaay9514

Zhou J, Zhu Z-Y, Hu H-T, Zhang G-L, Wang Q-Q. 2021. Clarifying water column respiration and sedimentary oxygen respiration under oxygen depletion off the Changjiang Estuary and adjacent East China Sea. *Front. Mar. Sci*. 7:623581

Zhuang W, Yu X, Hu R, Luo Z, Liu X, et al. 2020. Diversity, function and assembly of mangrove root-associated microbial communities at a continuous fine-scale. *npj Biofilms Microbiomes* 6:52

Zou D, Wan R, Han L, Xu MN, Liu Y, et al. 2020. Genomic characteristics of a novel species of ammonia-oxidizing Archaea from the Jiulong River Estuary. *Appl. Environ. Microbiol.* 86:e00736-20