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# Bacterial diversity in oil-polluted marine coastal sediments

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Marine environments harbour a persistent microbial seed which can be shaped by changes of the environmental conditions such as contamination by petroleum components. Oil spills, together with small but continuous discharges of oil from transportation and recreational activities, are important sources of hydrocarbon pollution within the marine realm. Consequently, prokaryotic communities have become well pre-adapted toward oil pollution, and many microorganisms that are exposed to its presence develop an active degradative response. The natural attenuation of oil pollutants, as has been demonstrated in many sites, is modulated according to the intrinsic environmental properties such as the availability of terminal electron acceptors and elemental nutrients, together with the degree of pollution and the type of hydrocarbon fractions present. Whilst dynamics in the bacterial communities in the aerobic zones of coastal sediments are well characterized and the key players in hydrocarbon biodegradation have been identified, the subtidal ecology of the anaerobic community is still not well understood. However, current data suggest common patterns of response in these ecosystems.

## Addresses

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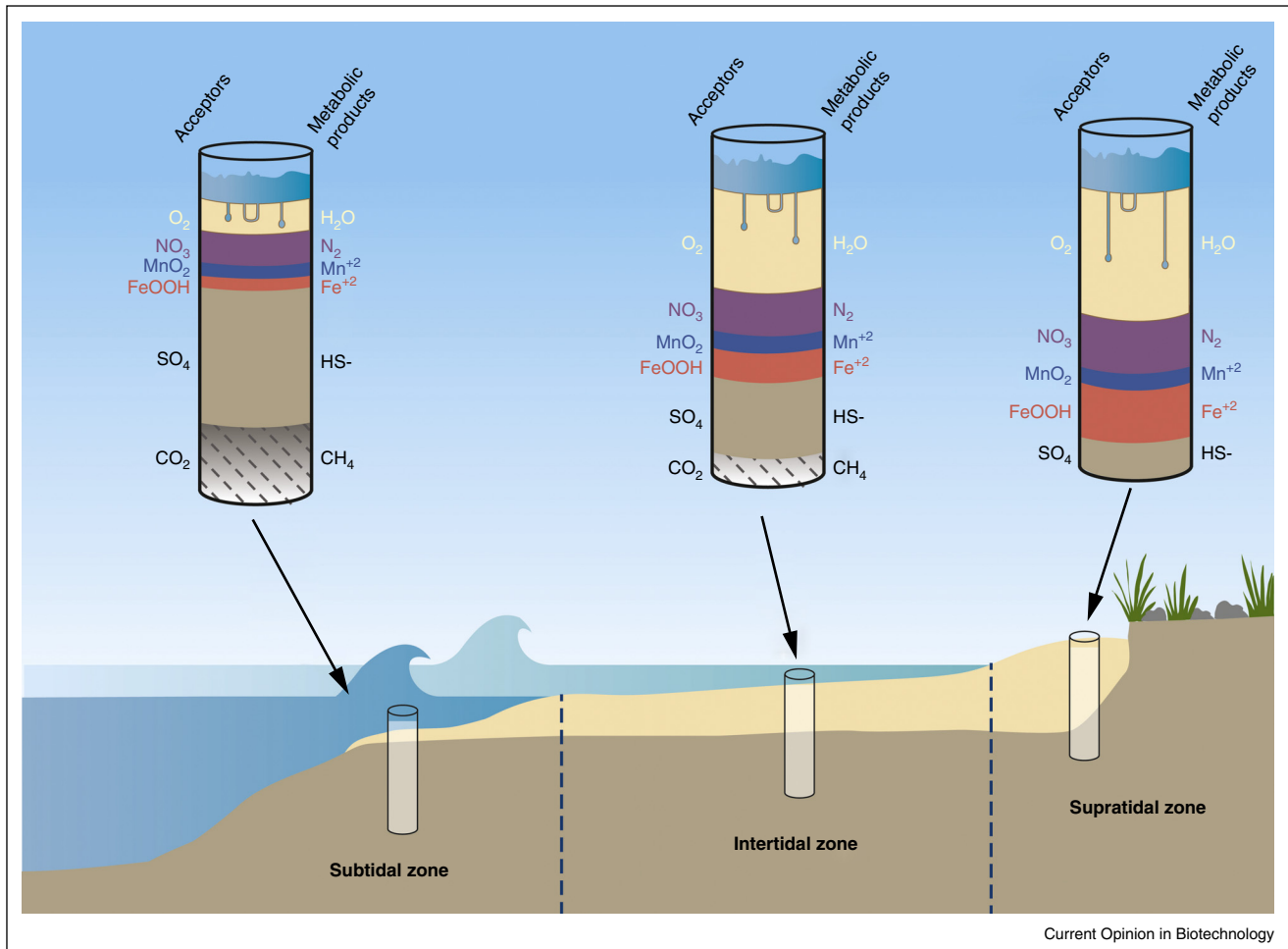
## Introduction

Continental shelves, which represent 15% of the total volume of marine sediments [1], are areas constantly exposed to pollution caused by human activities, and are particularly threatened by oil spills. Three zones,

with specific geochemical and ecological characteristics, can be distinguished within these habitats: (i) the supratidal zone, above the high tide shoreline, where sediment deposits are exposed to subaerial conditions most of the time; (ii) the intertidal zone (littoral) that comprises the area covered by the highest tide and is exposed during the lowest tide, and which includes wetlands (i.e., sandy, silty, and muddy sediments) and rocky cliffs; and (iii) the subtidal zone that remains permanently submerged (Figure 1). Studies on the microbial ecology of sediments have mainly focused on muddy sediments because of their high organic matter (OM) content, which is at least 10-fold higher than the OM in sandy sediments, and of the higher cell density inherent to the surface of the sediment's smallest particles [2]. However, black tides mainly impact sandy sediments, which cover >50% of coastal sediments worldwide (<200 m water depth) [3]. Many decades ago, pioneering works on microbial physiology established the role of microorganisms in carbon, nitrogen, sulphur, and iron cycles within coastal environments, and described the nexus to geochemical profiles throughout the sediment depth (from a thin superficial oxic layer, through an oxygen/redox 'transition' zone, to a deep anoxic zone, see Figure 1) [4]. Initial taxonomic studies were limited by the impossibility of cultivating the majority of prokaryotes but this was partially solved recently, when the emergence of powerful sequencing technologies provided the possibility to address the bacterial uncultured fraction in depth [5]. The massive spill resulting from the blowout of the Deepwater Horizon (DWH) platform generated a number of significant 'omics-based' studies, which have recently been reviewed [6\*\*]. Research initially focused on the water column, where the microbial response to the spill was thoroughly described [7,8\*]. Nevertheless, these techniques have not been extensively applied to polluted marine sediments, which remained insufficiently studied. However, the information gathered during recent years, largely thanks to metagenomic (16S rRNA and shotgun) and metatranscriptomics approaches, provides the first clues for understanding the response of such complex bacterial communities to the presence of hydrocarbon pollution. Linking changes in microbial diversity to biodegradation should be an important goal in this research, providing new biotechnological opportunities for the efficient recovery of polluted coastal areas [9].

Two main strategies have been used to address the microbial response to oil pollution: *in situ* studies at

Figure 1



Redox processes present in marine coastal sediments. Schematic representation of redox processes present in marine coastal sediments. The depth profiles show the stratification of different zones depending on the availability of terminal electrons acceptors (inherent to each site). A well-defined succession of respiratory metabolisms is evidenced and explained by the tendency of these compounds to accept electrons: microorganisms consume the oxygen that penetrates the surface influenced by the type of matrix (sand, mud, etc.), mixing effect by waves (few millimetres/centimetres in the case of subtidal sediments) or bioturbation. Once the oxygen consumption exceeds its supply, anoxic conditions are established below the zone of oxygen-influence. Nitrate, manganese and iron are used for anaerobic respiration if present. Sulphate reduction is dominant as sulphate is not a limiting factor in marine environments. Aerobic and anoxic processes are stimulated if biodegradable OM is available for oxidation, converting the use of each terminal electron acceptor as the driving force for microbes to use a specific metabolism. These redox zones formed in the coastal sediments can be visualized by eye inspection especially if a carbon input (such as oil) is present: (i) an upper oxidized brown layer that can range between a few millimetres to several centimetres; (ii) an anoxic but oxidized transition zone where nitrate, manganese and/or iron oxides are reduced; and (iii) finally, a dark grey to black reduced sulphidic zone, in which sulphate reduction predominates. However, in many cases more than one terminal electron acceptor can be used in the same redox zone. Methanogenesis develops in the sediment depending on the interaction between methanogens and sulphate reducers, generally described as mutually exclusive. In parallel, both biotic (chemolithotrophs) and abiotic processes can oxidize the inorganic compounds reduced by chemoorganotrophs, thus recycling elements in the sediments.

the polluted sites, and simulation experiments in microcosms and mesocosms under controlled laboratory conditions. The results from hydrocarbon-polluted coastal sediments of the recent years will be revised here, with the exception of wetland sediments, which have recently been addressed in an excellent review [10<sup>\*</sup>].

### Coastal marine sediments are heterogeneous and sustain highly diverse bacterial communities

Recent estimates reveal that coastal sediments harbour the highest values of microbial abundance [11<sup>\*</sup>] and diversity (alpha and beta indexes) within the marine realms [12], which is due in part to the stratification of

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