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# Indigenous microbial communities along the NW Portuguese Coast: Potential for hydrocarbons degradation and relation with sediment contamination

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ABSTRACT

Hydrocarbon degradation (HD) potential by autochthonous microorganisms in the coastal sediments of the NW area of Portugal (coastal sandy beaches and estuaries of the rivers Minho and Douro) was evaluated, investigating if water and sediment contamination/characteristics influence it. Sediments were characterized for microbial abundance (by DAPI), HD microorganisms' abundance (by MPN), microbial community structure (by ARISA), hydrocarbons (by FTIR and SPME-GC–MS), hazardous and noxious substances (SPME-GC–MS) and metals (by AAS). To our knowledge, this is the first time all these pollutants, including the selected HNS, were measured simultaneously in sediments of the selected coastal area. Higher contaminants concentrations were, generally, registered in Douro samples. A clear differentiation of the microbial community structure between beaches and estuaries was observed, as well as, between Douro and Minho river estuaries. BIO-ENV analysis indicated both sediment characteristics (e.g. OM content) and contaminants presence/concentrations (e.g. tetrachloroethylene presence) affected the structure of the microbial community along the studied areas. In all the selected sites, the characterized autochthonous microbial communities showed potential for hydrocarbons degradation, with HD microorganisms being found in all collected sediments. These microorganisms can be a valuable asset to recover contaminated areas, but sediment characteristics and contaminants presence/levels need to be taken into account as they can affect their bioremediation potential and the success of their application as biotechnological tool.

## 1. Introduction

Accidental oil spills release tons of oil through pipelines leaks, shipping and anthropogenic uses. For instance, in 2016 the total amount of oil released in the environment due to incidents involving tanker was ca. 6000 tonnes (http://www.itopf.com/knowledge-resources/data-statistics/statistics/). The pollution caused by oil and its derivatives is considered a global problem, making it an increasingly prominent concern for the environment (Malik and Ahmed, 2012).

Petroleum hydrocarbons are organic pollutants of major concern because of their wide persistence, distribution, complex composition and toxicity. The most common ones include aliphatic hydrocarbons and polycyclic aromatic hydrocarbons (PAHs) (Fuentes et al., 2014). Pollution with hydrocarbons is of great concern due to its massive spread into the environment and their presence in marine and estuarine ecosystems provides serious risks to aquatic life and human health (Rocha et al., 2011).

Estuarine and coastal areas are ecologically very important, with a great diversity of species, offering numerous benefits to humans. These ecosystems are suffering from different anthropogenic pressures due to contaminants released by spills and/or discharged from domestic and industrial wastewaters (Mucha et al., 2011). This anthropogenic pressure, thus, leads to the loss of biodiversity, aquatic habitat destruction and therefore, compromises the entire environment and the associated ecosystem services. With additional pressure on biological systems from oil spills, understanding and protecting estuarine and coastal environments is crucial.

Consequently, the development of strategies to clean these contaminated ecosystems to facilitate their recovery is urgent. Bioremediation is presented as a promising alternative to restore

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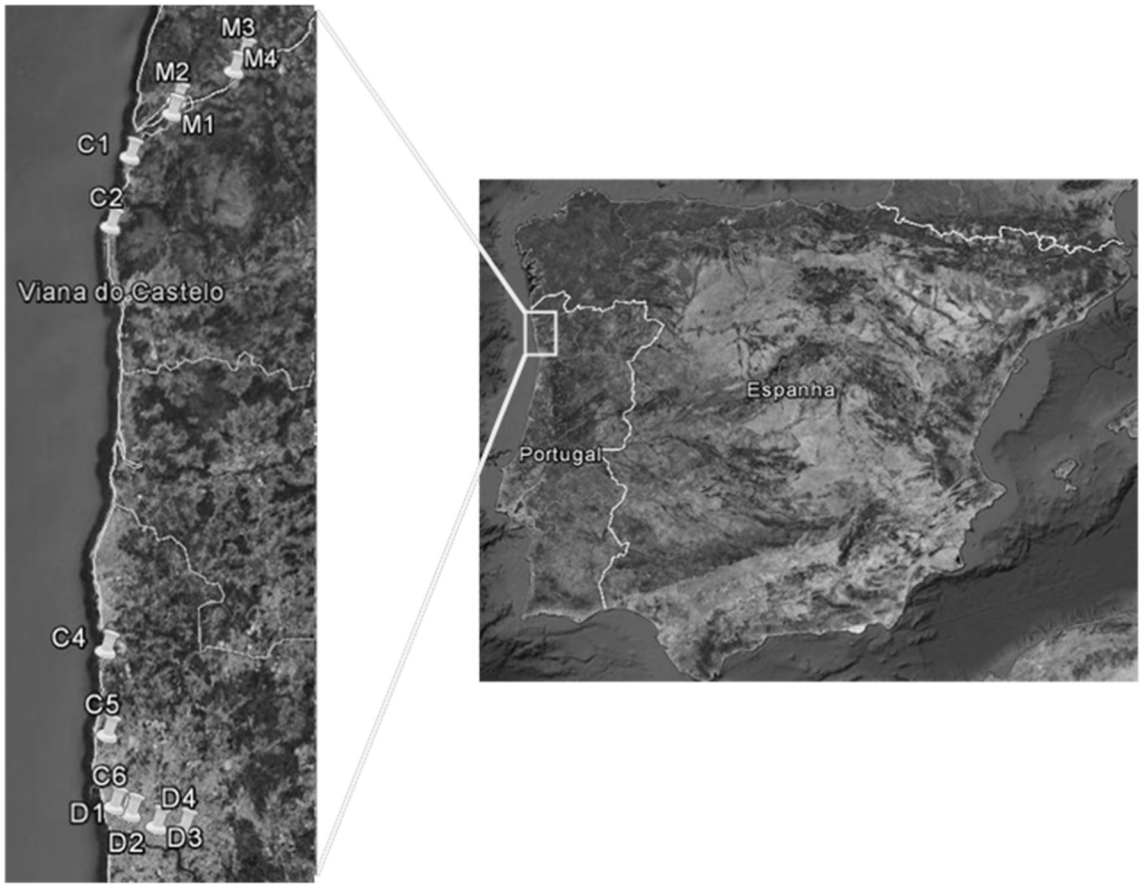


**Fig. 1.** Sampling locations in estuaries of the Minho (M1-M4) and Douro (D1-D4) rivers and in sandy coastal beaches (C1-Vila Praia de Âncora, C2-Carreço, C3-Bartolomeu, C4-Vila Chã, C5-Cabo do Mundo and C6-Foz) (courtesy of http://earth.google.com).

impacted areas. Bioremediation involves the use of biological agents, particularly microorganisms, that catabolize specific molecules, destroying hazardous contaminants or transforming them into less harmful forms (Fuentes et al., 2014). Microbial consortia with wide enzymatic capacity are, in general, needed to remove organic contaminants (Malik and Ahmed, 2012). But bioremediation with bacteria exogenous to the impacted site can fail because these bacteria may not be able to compete with the natural microbial community or adapt to the site environmental conditions, and so, display a lower oil degradation rate. Besides, their use implies the introduction of non-native microorganisms to the site which can cause ecosystems disturbances. The use of autochthonous microorganisms can be advantageous, because they are better adapted to the site, which can result in an increased efficiency in oil degradation.

A few studies, in which authors have participated, have explored the potential of autochthonous microorganisms for the bioremediation of contaminants in coastal and estuarine areas, namely petroleum hydrocarbons. For example, Reis et al. (2014) evaluated the bioremediation potential of autochthonous microorganisms from intertidal sediment of a sandy beach affected by an old oil spill, observing that microorganisms still had the ability to degrade up to 85% of total aliphatic hydrocarbons and 70% of total PAHs. Almeida et al. (2013a) investigated the potential of the microbial communities present in an unimpacted beach sediment to degrade hydrocarbons, showing these microbial communities could respond to an oil spill, degrading hydrocarbons. Pontes et al. (2013) showed autochthonous bioaugmentation contributed to a faster oil elimination, achieving 80% of hydrocarbons removal in 60 days, when applied to oil buried in a sand column in a laboratory controlled experiment. Moreover, Almeida et al. (2013b) observed hydrocarbon degradation rates between 15 and 30% by indigenous microbial communities of two estuarine sediments.

In short, bioremediation using indigenous microorganisms can be an effective alternative for removing petroleum hydrocarbon and their derivatives from contaminated sediments. However, prior knowledge of the ability of autochthonous microbial communities from different locations to biodegrade petroleum hydrocarbons is necessary for a proper implementation of this biotechnology. Moreover, microbial communities can be influenced by the presence of other contaminants, conditioning their response to oil spills. For example, Almeida et al. (2013b) investigated the potential effect of metals on the biodegradation of petroleum hydrocarbons in estuarine sediments, reporting metals changed the microbial community structure and that copper displayed significant deleterious effects on hydrocarbon degradation processes. Therefore, characterization of the different locations should be considered.

This study aimed to characterize the indigenous microbial communities present in sediments collected along the Northwest (NW) Portuguese Coast and evaluate their potential for hydrocarbon degradation. Water contamination (nutrients and fecal indicators) and sediment contamination, not only in terms of hydrocarbons (aliphatic and aromatic hydrocarbons) but also of other compounds (metals and hazard and noxious substances (HNS)) was correlated with this potential. If these microbial communities present hydrocarbon biodegradation potential, they can be preserved in a georeferenced microbial consortia bank and in the future, after an oil spill, be applied at the native geographic region from which the microbial consortia were obtained through bioremediation technologies.