

Identifying a link between cable bacteria and hydrocarbon degradation in polluted marine sediments

Dr Ugo Marzocchi's research focuses on the main factors that regulate macronutrient cycling in benthic zones, both in natural and artificial settings. His work concentrates on better understanding cable bacteria, which are microorganisms able to mediate electric currents in sediments. Dr Marzocchi is dedicated to understanding the way cable bacteria behave in nature, their metabolism, distribution and geochemical impact, and learn how to use cable bacteria as a bioremediation tool for the degradation of contaminants.

Crude oil, or unrefined petroleum, consists of a mixture of hydrocarbons. It maintains its liquid form both in underground geological formations and when extracted and brought to the surface. Human activities heavily rely on the use of petroleum for energy and transportation. Unfortunately, handling such large amounts of oil at a global scale has had serious repercussion on the environment. Major incidents, involving a transporter vessel capsizing and releasing large quantities of fuel into the ocean, plus spills/leakages arising from small and large maritime transport and from naval scrapping centres, add to the illegal disposal of waste rich in oils, affect the inhabitants of the ocean, the wildlife that preys on it, the fresh and saltwater bodies and the coastline that surround these. Over time, the petroleum hydrocarbons residues will accumulate and sink to the bottom of the waterbed, giving rise to a thick hydrocarbon-rich sediment. This affects the existing benthic community's (group organisms living together at the bottom of a seabed, river, lake, or stream) ecosystem, by altering the temperature, pH, light penetration, dissolved oxygen availability and nutrients concentration.

CURRENT CLEAN-UP METHODS

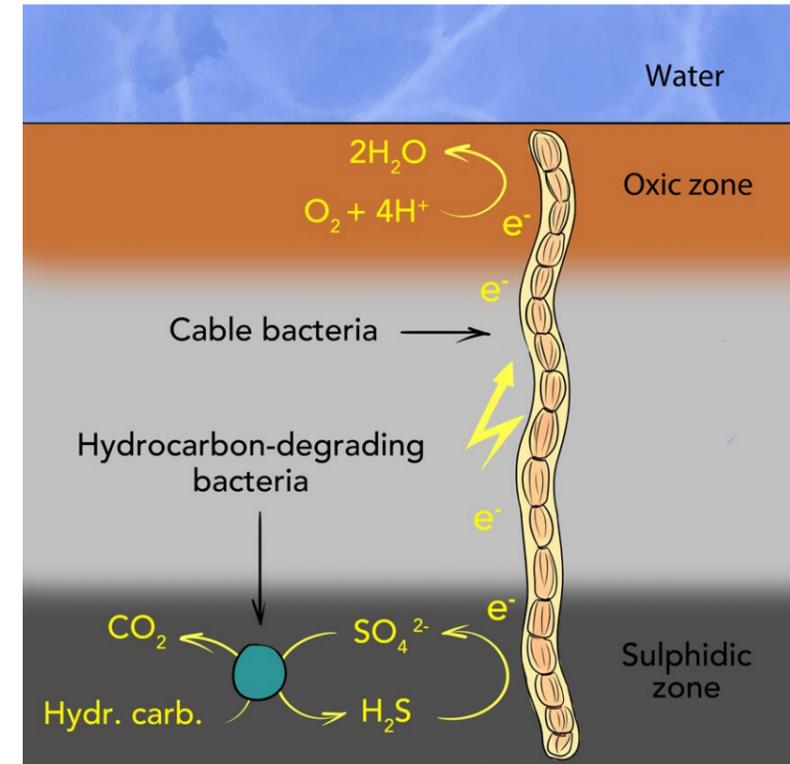
Conventionally, large pools of crude oil spills are contained using booms, long floating barriers which are placed

around the spill to stop it from spreading further afield. Different clean-up methods are used, including skimming the oil layer from the surface of the water, using for instance disks and floating drums, setting the oil alight, and using chemicals to break down the oil into smaller compounds to be later consumed by naturally occurring microbe. These methods, however, require calm waters in order to be somewhat successful, and they come with certain downsides. In the case of skimming, the procedure is time consuming and residues too small to be collected will remain dispersed in the waters. Burning the spill contributes to emissions and can be dangerous to the wildlife if out of control. Lastly, chemical dispersing introduces nasty and harmful chemicals to the environment which can be toxic to the wildlife and coastal inhabitants. Instead, degrading the hydrocarbons by environmentally friendly means, for example by using naturally occurring microorganisms, could be a viable solution.

CABLE BACTERIA: STIMULATING HYDROCARBON DEGRADATION

A good example of this is the research conducted by Dr Ugo Marzocchi and his collaborators at Aarhus University in Denmark and at the National Research Council of Italy in Rome, who have taken up the challenge of looking for bio-based methods to increase the degradation rate of hydrocarbons using microorganisms.

The research team first had to understand what basic principles needed to be addressed in order to



Cable bacteria metabolism in surface sediment.

degrade the concentration of hydrocarbons within the sediment. It is important to note that hydrocarbon degradation is limited by the availability of oxidants, whose concentration is higher in layers of the sediment closer to the water compared to deeper within the sediment. By increasing the level of oxidising agents (such as sulphate), it is possible to speed up the degradation process, perhaps by means of a natural catalyst or an expeditor, such as cable bacteria.

Let's take a quick step back and get to know cable bacteria, the protagonists of this article. Cable bacteria are generally found in reduced sediments within freshwater, saltwater lakes, and marine habitats. They were first discovered in 2010, within sediments of the Aarhus Bay, and subsequently detected in other marine and freshwater sediments throughout the world. Thanks to their unusual constitution, they have been able to adapt and survive in marine sediments rich in hydrocarbons, where their presence led to investigating their possible applications in bioremediation.

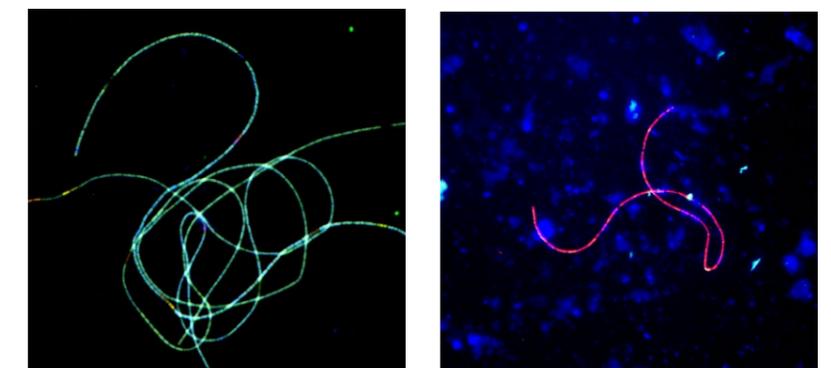
These microorganisms get their name from their filament-like shape. They

are electrically conductive multicellular filaments and can effectively re-generate sulphate in the sediment by promoting sulphide oxidation. We humans like to think that we were the first to invent electricity, with the advent of the first battery in the 1800s. However, not

surprisingly, bacteria have mastered this trick over thousands to millions years ago. Scientist have found that the ridges (thin fibres that make up the body of the bacteria) are conductive structures. In fact, cable bacteria carry out long-distance electron transport, which involves transferring electrons from one end of their body to the other (thus generating electricity), travelling distances of over 2 to 3 cm in sediments and groundwater aquifers. To put this in perspective, think of transporting a package over a distance of 1 km at incredible speed – this is what the cable bacteria do in the blink of an eye. This is quite remarkable as not all, if any, biological organisms are good at conducting, yet alone producing electricity.

So, why have these microorganisms evolved in such a way? The simple answer is, to survive. A way to think about this is by picturing a world where on one end food is widely available (no air), the other where air is widely available (no food). In this world the air and the food cannot coexist in the same location. However, in order for the population at both ends to survive, both air and food are needed. To ensure the survival of both populations, a network where one end consumes the food whilst the other respire would need to be established. Cable bacteria have done such feat: they have evolved and split their body into

Dr Marzocchi and his collaborators assessed the role of cable bacteria by simulating a hydrocarbon degradation environment and compared the results to a method called Snorkel.



Microscopic images of cable bacteria.

Cable bacteria are multicellular filaments that conduct electricity.



Set-up of Dr Marzocchi's Snorkel and Cable bacteria experiment.

two dedicated reaction centres, which are centimetres apart and which feed into each other. Thus, hydrogen sulphide, present within the sediment, is oxidised to sulphate in the deeper sediment layers, and the resulting electrons are conducted through the body of the cable bacterium, like an electric wire, to the oxic sediment (richer in oxygen, at the interface with the water), and used to reduce molecular oxygen or, alternatively, nitrate.

SEDIMENT BIOREMEDIATION VIA HYDROCARBON DEGRADATION

Based on these findings, Dr Marzocchi and his collaborators investigated cable bacteria further, and set up a simple experiment that would prove or disprove if cable bacteria played a role in hydrocarbon degradation, and perhaps surpass other bio-based methods. The group quantitatively assessed the role of cable bacteria by simulating a hydrocarbon degradation environment. They then compared the results to a method called Snorkel, a Bio-Electrochemical System, which implements a Microbial Fuel Cell (battery powered by a group of electrically conducting microbes) to degrade

hydrocarbons. Snorkel takes its name from its set-up: in a given soil sample, an electrode is placed upwards. At the bottom we find the microbes – unlike cable bacteria, these organisms are unicellular. Microbes consume the organic compounds within the soil and respire via the electrode, which acts like a snorkel; comparable to a diver breathing air through a snorkel when underwater. This system is similar to a battery as it consists of both a cathode and an anode, and in principle, works in a similar way to the cable bacteria method, with the main difference being that the electric current runs through the electrode.

The experiment consisted of incubating Snorkel, the cable bacteria separately

Cable bacteria may play an important role in the self-healing capacity of hydrocarbon-contaminated sediments.

and together, in sediments for 7 weeks. Cable bacteria showed 25% higher rates of hydrocarbon degradation, similar to the results obtained by Snorkel. The combination of Snorkel and cable bacteria showed the highest degradation rates of 46%. The results indicate that cable bacteria alone may

play an important role in the self-healing capacity of hydrocarbon-contaminated sediments, and may inspire novel remediation treatments upon hydrocarbon spillage. Dr Marzocchi's encouraging results require further testing to better attest the efficacy of this method; still, they represent a turning point for decontaminating various water bodies from the presence of unwanted hydrocarbons in a low-impact and sustainable manner.

WHAT DOES THE FUTURE HOLD?

For a long time, cable bacteria have been an overlooked player in the self-healing capacity of crude oil contaminated sediments. They may inspire novel remediation treatments upon hydrocarbon spillage. Nevertheless, the electric properties of the microorganisms may not just limit them to their bioremediation activities; due to cable bacteria's unique fibre structure, their application could be extended to their use in the biomedical field, where metallic compounds are substituted by these filaments and perhaps to be implemented in battery packs for small portable devices.



Behind the Research

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Bio

Dr Ugo Marzocchi is an Assistant Professor at Aarhus University. He is interested on the main factors that regulate the cycling of macronutrients (nitrogen, phosphorous, sulphate, iron) in benthic systems. He is particularly interested on the geochemical impact and ecology of electro-active bacteria, and their potential in technological applications.

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Collaborators

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Research Objectives

Dr Marzocchi identified a link between cable bacteria and hydrocarbon degradation in polluted marine sediment.

Personal Response

Do you envisage the fabrication of large benthic mats using cable bacteria, that could be used to treat medium to large oils spills?

“ The possibility to fabricate benthic mats is fascinating, but surely challenging. As an alternative, the growth of cable bacteria could be triggered *in situ* via providing favourable conditions (e.g., supplying suitable electron acceptors such as oxygen or nitrate to the bottom water). Thanks to their unique metabolism cable bacteria will extend the effect of the treatments to centimetres depth into the sediment. Further, an early diagnose of the presence of cable bacteria in contaminated sites could help decision makers to evaluate, within cost/benefit analysis, whether to implement expensive mitigation measures or let the system recover naturally.

The results showed that a combination of Snorkel and cable bacteria yielded the highest degradation rate. Could this be attributed to the two working together in a synergist way?

Within the timespan of our incubation, the rate of alkane degradation of the combined cable bacteria and Snorkel treatment equalled the sum of the treatments where the two ‘systems’ were applied individually, therefore suggesting a mere additive effect. However, our geochemical data suggest that in the presence of the snorkel, cable bacteria expand their volume of influence and possibly their life span. The question on the synergy therefore remains open. A recent study reported the ability of cable bacteria to attach to solid electrodes. Although we could not find evidence of this phenomenon in our experiment, we cannot exclude this possibility. ”



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