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Biological Approaches to the Treatment of Saline Oily Waste(waters) Originated from Marine Transportation

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Oily wastewater generated, in amounts of millions of tons per year, by ships mainly in engine-rooms (bilge waters) and by washing oil tanks (slops) create a major disposal problem throughout the world because of the persistence and accumulation of xenobiotic compounds in the environment. The high salinity levels (up to 25.000 p.p.m.) and the pollutants concentration limit the chances of discharge into the sewer systems and address the disposal of these waste(water)s to the sea. Tightening effluent regulations and consequent high energy and management costs has generated interest in the introduction of biological phases in the treatment of these wastewater. The objectives of this study were to evaluate the feasibility of using biological processes with purposely acclimated microorganism for the treatment of high salinity oily wastewaters (slops). Specifically both the bio-regeneration of the exhaust Granular Activated Carbons (GAC), loaded with a mixture of compounds occurring in slops, and a BioFilm Membrane BioReactor (BF-MBR) application were examined. Results proved the feasibility of using salt-adapted micro-organisms capable of degrading the main pollutants contained in slops.

1. Introduction

Analysis of the microbial communities that take part in hydrocarbons biodegradation activities has been a challenge for microbiologists (MacNaughton et al., 1999) and identification and subsequent isolation of these organisms could provide relevant candidate taxa for subsequent bioaugmentation studies as single organism inocula or consortia. Recently many marine hydrocarbonoclastic bacteria have been isolated and their degradation potential has been investigated (Yakimov et al., 2004).
Although bioremediation technologies are well established for the clean-up of chemical and/or petrochemical spills in marine environment, the role of these bacteria in high salinity and oily wastewater treatment plants remains unknown and the knowledge of the structure, composition and dynamic, as well as of the metabolic capability of the microbial community are still lacking. The

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selection of microorganisms able to break down the hydrocarbons in the sludge into harmless by-products can be carried out directly in the biological phase or from “natural” oily polluted environment. Specifically, in MBR secondary treatment, which is generally performed by indigenous, water-borne micro-organisms in a managed habitat, the selection and identification of microbial consortia with high capability to degrade hydrocarbons is a primary step for the optimization of the process. Also bio-regeneration of the exhaust GAC, treating high salinity oily waste(water)s can be performed from marine oil-degrading bacteria. The use of these micro-organisms has to be investigated according to their ability to degrade oil and survive to the extreme salinity conditions. Among these bacteria *Alcanivorax borkumensis*, showed to play a pivotal role in the oil-spill bioremediation (Schneiker et al., 2006), presumably for its capacity to grow on many saturated petroleum fraction constituents and biogenic hydrocarbons such as straight-chain and branched alkanes, isoprenoids, phytane, pristine and long sidechain alkyl compounds (Schneiker et al., 2006). Significantly, *Alcanivorax* was found in low number in unpolluted waters, but in high abundances in oil-polluted waters and coastlines, where it may comprise 80-90% of the oil-degrading microbial community (Kasai et al., 2002). It was however observed as the growth of *Alcanivorax,* on crude oil, was not particularly rapid in comparison with other marine oil-degrading bacteria (e.g. *Thalassolituus*). Abundance of *Alcanivorax* was also observed in field and microcosm and mesocosm experiments involving the addition of nitrose and phosphorus fertilizers to stimulate microbial degradation of oil (Cappello et al., 2007). Different hypotheses were proposed to explain the predominance of this strain in polluted environment (Kasai et al., 2002). Genome analysis yield unprecedented insights into the bacterium’s capacity for (i) n-alkane degradation, including metabolism, biosurfactant production and biofim formation, (ii) scaveging of nutrients and cofactors in the oligotrophic marine environment and (iii) coping with various habitat-specific stress factor (Schneiker et al., 2006). In particular, the presence of specifics multiple systems for saturated hydrocarbon catabolism, namely two alkane hydroxylase systems AlkB1 and AlkB2 and three P450 cytochromes (Schneiker et al., 2006) was detected.
Focusing on these issues the research work examined the feasibility of different treatments including:

- the bio-regeneration of the exhaust carbons (GAC), saturated by high-pre-treated slops, in order to reduce the GAC regeneration costs.
- An MBR process as secondary treatment for light-pre-treated slops.

Both the two biological phases were operated by specific high salinity acclimated microorganisms.

2. Materials and methods

2.1 Format

Grab samples were collected from a floating tank of an oil costal deposit in the Augusta harbour (Sicily). A simple gravity separation (2 h) was carried out on site to separate floating oil. About 1000 litres of the clarified slops were stored in a fridge at 4°C, in order to inhibit any biological activity. Analytical characterization and most of the experiments were carried out in the Environmental Engineering laboratory of the University Kore of Enna (Sicily) according to APHA (2005). Water for GAC saturation were pre-treated by coagulation-precipitation with Ferric Chloride (Mancini et al. 2010).

2.2 GAC saturation and Bioregeneration

Saturation of GAC was carried out through continuous flow column filtration. The standard experiment utilized a column filled with GAC (23 cm of length) with an empty bed contact time (EBCT) ranging from 18 to 21.7 minutes in the different experiments. In each saturation run the 3.2-cm diameter column was packed with 20-30 mesh PicaHydro S23 GAC or Picabiol GAC. The flow rate ranged from 50.1 to 62.8 ml min^{-1}. With the respect to TOC values the GAC was considered saturated after about twelve litres. Bach bioregeneration tests were carried out through a combination of slops, buffered nutrient (urea and biphasic ammonium <30% and vegetable substances <65%), activated sludge sampled from the biological section of the oily contaminated groundwater treatment plant of Gela Refinery (Sicily) and the bacterium *Alcanivorax borkumensis* strain SK2 (DSMZ 11573), which was added to the high salinity sludge (conductivity ≈14 mS cm^{-1}). Loaded GAC was directly subjected to the recirculation of the mixture pre-emptively filtered on sand. Layout of the experimental setup is shown in Figure 1.

2.3 BF-MBR Pilot Plant

The pilot plant was fed with the light-pre-treated slop. The biological phase was operated inside a bench-scale reactor designed to allow both the development of suspended activated sludge and biofilm on free floating plastic carriers followed by a submerged membrane unit as solid-liquid separation phase. Such configuration is usually referred as hybrid moving bed biofilm reactor (HMBBR), coupled to a final ultrafiltration stage, characterized by an hollow fibers module, in the following referred to as BF-MBR (BioFilm Membrane BioReactor).

Further details of the bench-scale plant can be found Torregrossa et al. (2010). For the start-up phase, 20 L of halophilic activated sludge withdrawn from the salty (≈14 mS cm^{-1}) groundwater treatment plant were used as inoculum for the BF-MBR pilot plant. However, the seed was previously acclimatized to the new wastewater (slops) salinity. Initially the MLSS (mixed liquor suspended solids) concentration in the bioreactor was 7 gTSS·L^{-1}, later it was maintained between 8 and 9 gTSS·L^{-1} throughout the experiment. The reactor was fed with slops stored in a 50 L tank that was refilled 3 times a week.

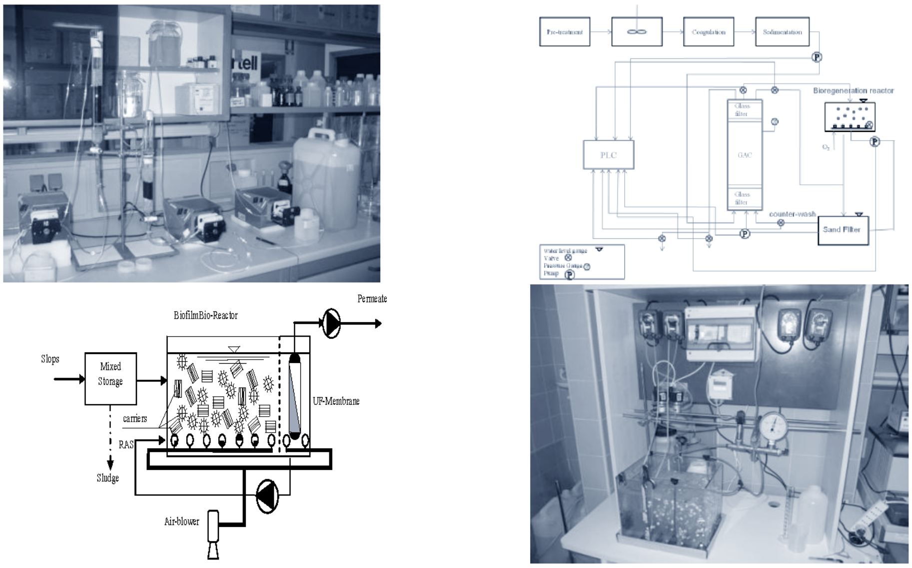

Figure 1: GAC bioregeneration and MBR bench scale plants.

3. Result and discussion

3.1 Slops characterization

The slops collected from the oil deposit were found to have the general characteristics summarized in Table 1. The waters have Total Petroleum Hydrocarbon and COD values well above the discharge limits to the sea (5 and 160 mg L^{-1} respectively). Of particular concern was the high concentration of total dissolved solids (TDS) approx. 1.5 times higher than seawater. At such elevated conditions, current analytical methods for organic content analysis can show some limits.

Table 1: Slops characterization

Parameter	M. U.	Value	Parameter	M. U.	Value
pH	-	7.5	Sulphate SO_4^{-2}	mg L^{-1}	1.970
COD	mg L^{-1}	1,100±500	Conductivity	mS cm^{-1}	18.7
TSS	mg L^{-1}	26	TOC	mg L^{-1}	65.7
TDS	mg L^{-1}	55.000	Total carbon	mg L^{-1}	161.7
TPH	mg L^{-1}	50	Inorganic carbon	mg L^{-1}	95.9
Chloride (Cl^{-1})	mg L^{-1}	17.760	Anionic surfactant	mg L^{-1}	2.65

3.2 GAC bio-regeneration results

GAC renewal of adsorptive capacity consisted in re-circulating the mixture of acclimated bacteria, nutrients and dissolved oxygen in the described closed batch system. According to Goeddertz et al. (1988) the offline process for activated carbon bioregeneration was preferred to the bioregeneration process occurring during the BAC treatment because of the limitations of the BAC process for availability of nutrients and dissolved oxygen, the persistence of many organic compounds and operational difficulties including hydraulic short circuiting and excessive head loss.

In the case of offline systems, where pre-loaded activated carbon is consecutively biologically treated, it could appear easier to determine the extent of bioregeneration. However only very few studies, reporting quantitative measurements of bioregeneration, are described in the literature (Aktas and Cecen, 2007) and no one deals with salty wastewater. Bioregeneration was here directly quantified through the comparison of the equilibrium adsorption capacities of fresh and bio-regenerated GAC.

Results from the bioregeneration tests (a typical trial is reported in Figure 2) showed a good recovery of the adsorptive capacity of the biologically treated GAC.

Two mechanisms were considered involved in the bioregeneration of granular activated carbon (GAC): a) the concentration gradient mechanism (Aktas and Cecen, 2007) involves organic compounds released from the activated carbon following desorption due to a concentration gradient between bulk liquid and activated carbon surface; organic compound released into the liquid phase are then degraded by microbial activities, causing a lowering of the organic compound concentration in the liquid phase; b) extracellular enzyme reactions mechanism involves exoenzymes excreted by microorganisms diffuse into activated carbon pores and react with adsorbed substrates; hydrolytic decay of the substrate may occur or desorption of the resulting enzyme metabolite may take place due to the weaker absorbability of this metabolite.

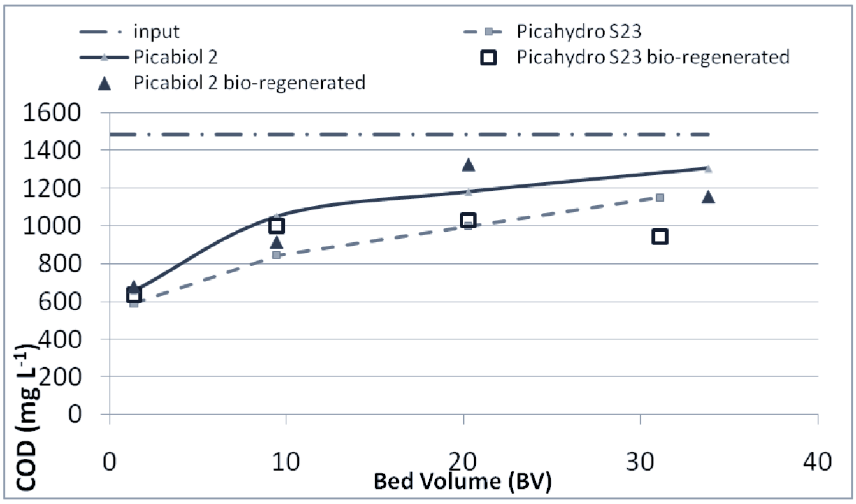

Figure 2: Comparison of the virgin and bio-regenerated GACs adsorption behaviour.

3.3 MBR performance

In order to check the treatment efficiency of the BF-MBR pilot plant, quality parameters at the inlet and at the outlet of the system were determined. These parameters included: COD and TOC, (this last was considered to verify the total organic load of waters before and after treatment), TSS and conductivity, that is related to the total dissolved ions in the samples. It should be considered that these experiments were carried out with almost raw wastewater, thus maximizing the carbon source, in order to increase the chance of acclimatizing the bacteria consortium to the high salinity slops.

Figure 3 shows COD concentrations in the feed and in the BF-MBR permeate versus time throughout the experiment. COD in the permeate was generally less than 150 mg L^{-1} with two exceptions (217 mg L^{-1}) only. The results suggested that COD in the BF-MBR permeate can be well within the discharge limit of 160 mg L^{-1}, especially if a well-pretreated influent is used instead of a raw one. TSS values of

BF-MBR effluent showed little variation with an average value of 19.9 ± 2.3 mg L^{-1} and an average reduction of 72%.

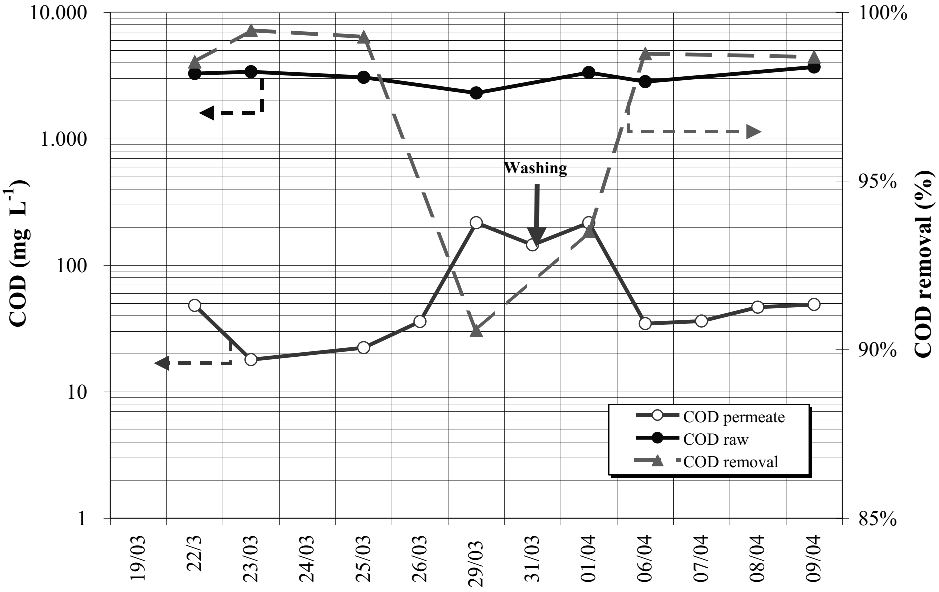

Figure 3: COD concentrations in the permeate and COD removal efficiency.

4. Conclusions

In this paper the feasibility of treating an emulsified oily wastewater (slop) of high salinity, TPH and COD values was verified through GAC and BF-MBR systems. The selection of salt-tolerant micro-organisms involved an adaptation of a specifically selected sludge joined to *Alcanivorax borkumensis* SK2 to high salt concentrations. Tests on bioregeneration of exhaust GAC, carried out in batch units gave encouraging results in terms of TPH and COD removal from saturated GAC. Results proved the biological mixture capability of removing efficiently the organic matter adsorbed on the GAC thus increasing the service-life of the GAC without removing GAC from the filter (with consequently reduced management costs).

The results of the reported short experimental campaign during which the BF-MBR pilot plant was directly fed with almost raw wastewater (without chemical-physical pre-treatment), showed that this technology represents a reliable process for slop treatment. The high sludge retention time and high filtration efficiency in the system enabled a large part of TOC to be removed, allowing the limits imposed for discharge into the sea to be met.

Some key issues however remain and need to be addressed in the application of GAC bioregeneration and BF-MBR processes to the treatment of slops including the reduction of membrane fouling by proper pre-treatment and the identification of the role of the specifically added Alcanivorax borkumensis. Although biological treatment is usually inhibited by high salt concentrations, results from the present research proved the feasibility of using salt-adapted micro-organisms consortia capable of degrading the main pollutants contained in these oily and salty contaminated waters.

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