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Short communication

Use of biomarkers to investigate toxicological effects of produced water treated with conventional and innovative methods

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Abstract

The aim of this study was to develop and apply a multi-biomarker system to assess the toxicological effects of produced water (PW) from a Mediterranean off-shore oil platform. The selected bioindicator organism, mosquitofish (*Gambusia affinis*), was exposed in the laboratory to high concentrations of different PW: PW before treatment (BT), after conventional treatment (ACT) and after innovative treatment with zeolites in a prototype system (AIT). A set of biomarkers (benzo-(α)pyrene monooxygenase, ethoxyresorufin-*o*-deethylase, vitellogenin, porphyrins, PAH bile metabolites, esterases, catalase, micronuclei) and PAH concentrations were measured in the experimental organism. The methodology proved to be appropriate and biomarker responses (CYP 1A1, PAH bile metabolites, micronuclei, esterases, porphyrins) affected by BT were less affected by ACT. PW treated with zeolites (AIT) had the lowest toxicological impact. The results obtained applying this multi-biomarker approach suggest that the system using zeolites is effective for treating produced water.

Keywords: Off-shore produced water; Mosquitofish; Biomarkers; PAHs; Zeolites

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Produced water (PW) is formation (and injection) water from which oil and gas is recovered. It is generated in large quantities during oil and gas extraction. PW is a complex mixture containing residual hydrocarbons, metals, naturally occurring radioactive material and potentially toxic treatment chemicals, such as biocides, corrosion inhibitors, dispersant, emulsion breakers, detergents and scale inhibitors, used in oil production. Since only water with an oil content less than 40 mg/l can be discharged from Mediterranean off-shore installations, produced water is treated to reduce its hydrocarbon and metal content before discharge into the sea. It is nevertheless necessary to investigate the toxicological properties of PW in order to better understand their effects on the marine environment. Some studies have been conducted into the acute toxic effects of PW on test species (Strømgren et al., 1995), and others into the sub-lethal effects on the fish fauna (Stephens et al., 2000).

Oil companies are continuously exploring new and more efficient techniques for PW treatment. The new treatment system considered by ENI S.p.A. and investigated in the present study is based on the use of zeolites (microporous crystalline solids containing silicon, aluminium and oxygen in their lattice, with high selective adsorption capacities) and photooxidation by acidification, ozonization and UV irradiation.

The aims of the present study were: (a) to develop a multi-biomarker system using mosquitofish (*Gambusia affinis*) as bioindicator species to study the toxicological potential of PW, (b) to compare the toxicological effects of produced water from a Mediterranean offshore oil platform, before and after conventional treatments and after treatment with zeolites in a laboratory prototype system, (c) to explore the effectiveness of the system using zeolites in reducing the toxic effects of PW, as a first indication about the possibility of using this technique on off-shore platforms.

Produced water before treatment (BT) and after conventional (active carbon) treatment (ACT) was collected from the off-shore oil platform (Floating Production Storage and Offloading Unit ''Firenze'', production field ''Aquila'', (south Adriatic sea). Approximate concentration of the main chemical constituents of PW were provided by ENI S.p.A. BT: total hydrocarbons 2000 mg/l, total aromatic hydrocarbons 10 mg/l, paraffins 500 mg/l. ACT: total hydrocarbons <10 mg/l, total aromatic hydrocarbons <5 mg/l, paraffins <5 mg/l. Trace element levels did not differ between BT and ACT: Pb and Cu 1 mg/l; Cd 0.1 mg/l; Cr, As, Ni 0.5 mg/l; Hg 0.005 mg/l. Additives used in the platform are corrosion inhibitors, emulsion breakers and biocides, most of them having aromatic components. PW sampled at the same time and treated with zeolites (after innovative treatment – AIT) was provided by EniTecnologie S.p.A. (Milan, Italy). After acclimatization in the laboratory, mosquitofish ($n = 30$ per group, two tanks per group), were exposed to the different PW in a static system. Male specimens were exposed for 8 days to BT, ACT and AIT at 25% and 50%. Female specimens were exposed for 8 days to 50% BT, ACT and AIT. Control groups were maintained in tap water adjusted to the same salinity as the exposure groups (18‰). Water was continuously oxygenated in all tanks. Mortality was found to be $\leqslant$20%, with no remarkable differences among groups, including controls. A set of biomarkers was measured in the experimental organisms. CYP1A1 enzyme activities (benzo(α)pyrene monooxygenase (BPMO), ethoxyresorufin-*o*-deethylase (EROD)) (Kurelek et al., 1977; Lubet et al., 1985), porphyrin concentration (copro, uro and protoporphyrins) (Grandchamp et al., 1980), acetylcholinesterase (AChE) activity (Ellmann et al., 1961), and catalase (CAT) activity (Aebi, 1986) were measured using whole body homogenates. Vitellogenin was measured semi-quantitatively in plasma (Goksoyr, 1991) using a