

# SEASONAL VARIATIONS OF DIOXIN-LIKE ACTIVITY IN SEDIMENTS OF THE BIZERTA LAGOON (TUNISIA) DETECTED WITH IN VITRO CELLULAR BIOASSAYS

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## Abstract

In order to evaluate sediment contamination in six stations from Bizerta lagoon, a combination of *in vitro* bioassays and chemical analysis was applied to sediment organic extracts. By using 7-ethoxyresorufin-O-deethylase (EROD) induction in the fish hepatoma cell line PLHC-1 after 4 h of cell exposure, dioxin-like activities were detected in all analysed samples. Dioxin-like activities were higher in winter than in summer. A highly significant correlation was observed between bioassay- and chemical analyses-derived toxic equivalents (TEQs), but PAHs accounted for only a small part of the detected biological activities. Our results suggest the presence of unknown readily metabolised EROD inducing compounds, which will need to be further characterized.

**Keywords:** Lagoons, Pollution, Sediments, Ecotoxicology

## Introduction

Bizerta lagoon, situated in northern Tunisia, is an important ecological area implicated in various activities as fishing and aquaculture development. Unfortunately, this lagoon is submitted to many anthropic pressures, which led to chemical contaminations by various toxic compounds that may be biologically active. The aim of this study was (i) to evaluate the seasonal variations of dioxin-like chemicals, using *in vitro* bioassays, (ii) to compare bioassay data with PAHs analyses by using a toxic-equivalents (TEQ) approach.

## Material and Methods

Superficial sediments were collected in January and July 2006 at six stations in Bizerta lagoon and at a reference station (GH) in the seawards entrance of Ghar el Melh lagoon. The sediment extracts were subjected to chemical and bioassays analyses. Then, the PAHs in sediment extract were quantified by gas chromatographic analysis. The EROD activity assay in PLHC-1 cells line was processed, as previously described by (1).

Dose-response curves were fitted by Hill equation model and the EC50 were calculated. Bioassay-derived benzo[a]pyrene- (BaP-EQ) and instrumentally derived toxic equivalents (TEQs) for PAHs were determined as described by (1).

## Results and Discussion

As shown in Figure 1, all sediment extracts were able to induce EROD activity. The extract from Menzel Bourguiba (MB) site, which is subjected to intensive industrial activity, was the most active sample.

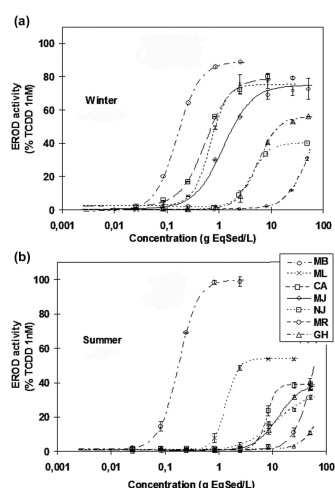


Fig. 1. EROD induction in PLHC-1 cells exposed for 4 h to serial dilution of sediment extracts sampled in winter (a) and summer (b). Results are expressed as percentage of maximal EROD activity induced by TCDD 1 nM. Values are means of triplicates  $\pm$  SD.

Regarding seasonal variations, it is noteworthy that, although similar site ranking was observed between summer and winter, higher activities were observed in winter in all stations, except for MB site, which was still highly potent in summer. This result suggests that the chemicals responsible for activation are metabolized in hot season, and support the hypothesis for a major role of PAH-like chemicals in EROD induction.

The regression analyses (Figure 2) showed highly significant positive correlation between biological- and instrumental-derived TEQs ( $R^2 = 0.94$ ). However, when comparing toxic-equivalent values the TEQ-chem values explained only a small part of the biological activities detected by the bioassay, suggesting that other EROD-inducing compounds than the 16 analysed PAHs are present in the extracts.

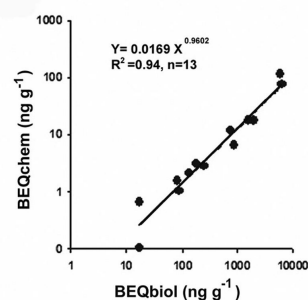


Fig. 2. Correlation between chemical analyses- and bioassays-derived BEQs determined in sediments extracts.

## Conclusion

Seasonal variations were observed by both *in vitro* dioxin-like activities and PAHs concentrations in sediment extracts, which were lower in summer. Such variations probably reflect a higher abiotic and biological degradation of organic chemicals in the hot season. Furthermore, the dioxin-like responses observed in our study were thus likely to have been caused by other ubiquitous biogenic and anthropogenic PAH-like AhR ligands that were not taken into account by the chemical analyses.

## References

1 - Louiz I., Kinani S., Gouze M.-E., Ben-Attia M., Menif D., Bouchonnet S., Porcher J.M., Ben-Hassine O.K., Aït-Aïssa S., 2008. Monitoring of dioxin-like, estrogenic and anti-androgenic activities in sediments of the Bizerta lagoon (Tunisia) by means of *in vitro* cell-based bioassays: Contribution of low concentrations of polynuclear aromatic hydrocarbons (PAHs). *Sci. Total Environ.* 402 (2-3): 318-329.