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The assessment of WWTP performance: Towards a jigsaw puzzle evaluation?

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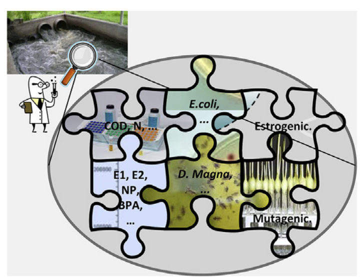
HIGHLIGHTS

- A framework for an integrated monitoring of WWTPs is proposed as a puzzle of assays.
- It is applied to a full-scale WWTP for a holistic evaluation of its performances.
- Chemical (4 EDCs) and biological (3 assays) tests are presented.
- The promotion of a multitiered approach is crucial for a real integrated evaluation.

GRAPHICAL ABSTRACT



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ABSTRACT

A chemical and bio-analytical protocol is proposed as a holistic monitoring framework for the assessment of WWTPs (Wastewater Treatment Plants) performance. This combination of tests consists of: i) an analysis of emerging contaminants, to be added to the established physico–chemical parameters in order to understand the causes of (new) pollution phenomena and ii) some of the bio-analytical tools most widely applied in the field of wastewater research, which provide information on groups of chemicals with a common mode of toxic action (baseline toxicity, estrogenicity and mutagenicity/genotoxicity, selected as the most representative for human health). The negative effects of the discharge can thus be highlighted directly and used to assess the global environmental impact of WWTPs.

As a validation, this multi-tiered approach was applied to a full-scale WWTP (150,000 p.e.), where different measurements were carried out: EDCs (Endocrine Disrupting Compounds) detection; algal growth inhibition, bioluminescence inhibition and acute toxicity test (for baseline toxicity); an E-Screen-like assay (for estrogenic activity); Ames, *Allium cepa* and Comet tests (for mutagenic/genotoxic activity). As a result, the WWTP showed good performance for all these issues, displaying its ability to enhance effluent quality, except for residual mutagenic behaviour, probably due to the by-products generated by the tertiary ozonation.

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1. Introduction

WWTPs (Wastewater Treatment Plants) usually undergo “conventional” monitoring, which consists of determining several traditional physico–chemical parameters – solids, COD, nutrients – (Vasquez and Fatta-Kassinos, 2013), in order to monitor the quality of discharged effluent, so as to protect the receiving water bodies from contamination, e.g. eutrophication. Nevertheless, pollution phenomena are increasing nowadays, becoming more diversified and complicated, due to the high level of “anthropization” and related contamination processes, with new synthetic substances being continuously released into the environment: as an example of new pollution mechanisms, impaired reproductive performance in aquatic organisms, and even feminization of shellfish, have been recorded in the past decade (see, *inter alia*, Niemuth and Klaper, 2015), as a consequence of exposure to hormone-like compounds. For these reasons, information provided by physico–chemical parameters alone is no longer sufficient to describe the real nature of WWTP effluents and their hidden perils, which can only be revealed by means of a more in-depth analysis aimed at improving knowledge of the nature of wastewater.

On one hand, this up-grade can be performed by analytical chemistry, increasing the number of detected substances. For instance, studies on the presence and fate of unconventional pollutants such as pharmaceuticals and EDCs (Endocrine Disrupting Compounds: molecules that can mimic, block or interfere with hormonal activities in living organisms – Castillo et al., 2013) are indeed one of the hot topics in the field of wastewater research (Luo et al., 2014), as they represent a fundamental step in identifying the causes of new pollution phenomena (e.g. EDCs are the main chemicals responsible for shellfish feminization). Even if chemical analysis allows us to detect single priority pollutants of concern, it is not clear which fraction of the overall pollutant burden can be covered: transformation products and unknown compounds are often missing, for instance (Escher et al., 2008).

On the other hand, the global effects of WWTP effluents can only be assessed with the application of biological assays, which can directly measure the activity exerted by this matrix: its adverse impacts can be revealed, overcoming the constraints and limitations of the chemical “single substances” approach (Gartiser et al., 2010). Only in this way can a holistic evaluation of WWTPs be achieved, and the real efficiency of wastewater treatments can be determined with an “effects-based”, rather than a “compounds-based”, approach.

The main aim of this work was to illustrate a multi-tiered testing approach for assessing WWTP performance. The parameters were: selected emerging contaminants (tier I: analytical chemistry), and biological assays for the relevant modes of toxic action (tier II: ecotoxicological effects, faecal contamination, estrogenic and mutagenic activity). This multidimensional monitoring was then validated by application to a full-scale WWTP ($\approx$150,000 p.e.) in order to assess its performance under the conceived puzzle: the application to a real case study allows an increase in the amount of data and experience, a requirement if further steps in terms of legislation and policy are to be pursued.

2. The puzzle framework: description of the assays

This section illustrates the assays adopted, in order to highlight the meaning and importance of each measurement. Proposed framework does not contain extremely novel concepts, as a lot of work has been published in recent years on Whole Effluent Toxicity, Effects-Directed Analysis and Toxicity Identification Evaluation approaches (see, for instance, Burgess et al., 2013). But, as a peculiarity of this work, a meaningful sub-set of tests was selected, as we were aware that applying all the existing chemical and biological assays is not techno-economically feasible for WWTP managers. This sub-set of assays was decided for two main reasons: i) first of all, the “ideal” test battery consists of a small number of indicator bioassays that are able to cover a wide range of cellular toxicity pathways and take into account each mode of action: the non-specific (i.e. baseline), the specific (e.g. estrogenicity) and the reactive (e.g. genotoxicity) toxicity, as recommended by Escher et al., 2014. Moreover, ii) selected assays are in accordance with the model for wastewater treatment evaluation proposed by the authors in a previous work (Papa et al., 2013), which in brief calculates the damage on human health as key factor for the evaluation of different wastewater treatment processes. The damage due to wastewater discharge is linked to specific waterborne diseases (according to World Health Organization: WHO, 2008) that are weighted by means of appropriate bioassays. For this reason, two specific modes of toxic action are considered: the estrogenicity and the mutagenicity; indeed, they represent the indicators of the main diseases linkable to wastewater discharge (endocrine disorders and malignant neoplasms, respectively), next to the diarrhoeal diseases (measured by means of conventional microbiological assays, e.g. *E. coli* determination).

2.1. Chemical monitoring: from conventional to emerging contaminants

Over the last few decades, the occurrence of emerging contaminants in the aquatic environment has become a worldwide issue of increasing environmental concern. They consist of a vast and expanding array of anthropogenic as well as natural substances, including pharmaceuticals, personal care products, steroid hormones, industrial chemicals, pesticides and many others, which are commonly present in waters at trace concentrations, ranging from a few ng/L to several μg/L, and therefore called micropollutants (Luo et al., 2014).

Although the occurrence of micropollutants in the aquatic environment has been frequently associated with a number of negative effects (Fent et al., 2006), precautions and monitoring actions for micropollutants have not been well established, nor standardized at a legislative level: to date, guidelines on WWTP discharges do not exist (Luo et al., 2014), although they represent the main source for the release of micropollutants into the environment. Water policy, on the other hand, has started to propose environmental quality standards for several emerging contaminants: e.g. the EU sets threshold concentrations of micropollutants in water, sediment or biota, which should not be exceeded in order to protect human health and the environment (European Directive 2013/39/EU). The detection of micropollutants in wastewater plays an important role from the standpoint of policy alone. Indeed, it can provide useful information for addressing the control of pollution at source (i.e., green chemistry), by limiting and/or banning the most dangerous substances, as already happens for phosphorus in surfactants in order to prevent eutrophication phenomena (see, for instance, the latest EU Regulation 259/2012).

This leads straight away to a very important question: is the detection of this cocktail of substances necessary, possible and techno-economically feasible for WWTPs? Clearly the answer is no! Therefore, the choice of emerging contaminants to be detected should be made *a posteriori*: if specific negative effects are evidenced by the bioassays, analytical chemistry can be properly oriented in the search of the causes, i.e. the detection of a specific group of compounds (see also Table 1). In this way, it is possible to properly steer the choice of micropollutants, in order to detect only those that are supposed to be actually found. Moreover, the upcoming European monitoring list, “Watch List” (Carvalho et al.,