Journal of Hazardous Materials xxx (xxxx) xxx–xxx

Contents lists available at ScienceDirect

Journal of Hazardous Materials

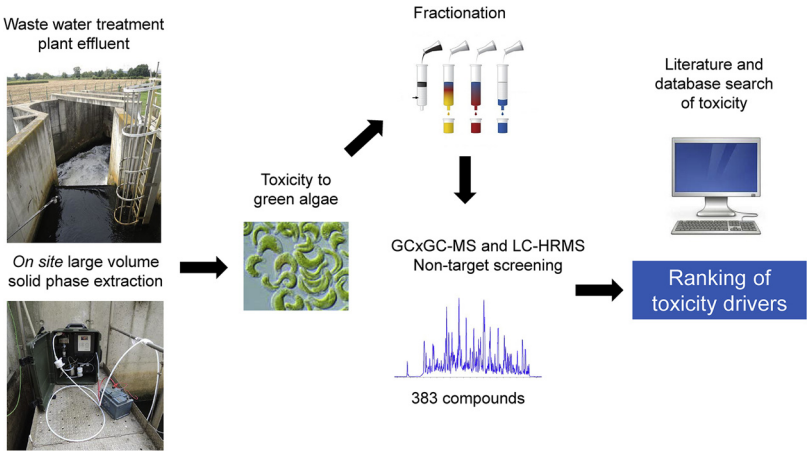
journal homepage: www.elsevier.com/locate/jhazmat

Identification of algal growth inhibitors in treated waste water using effect-directed analysis based on non-target screening techniques

Zuzana Tousova[a,b], Jean Froment[c,d], Peter Oswald[a], Jaroslav Slobodník[a], Klara Hilscherova[b], Kevin V. Thomas[d,e], Knut Erik Tollefsen[d], Malcolm Reid[d], Katherine Langford[d], Ludek Blaha[b,*]

[a] Environmental Institute (EI), Okruzna 784/42, 972 41 Kos, Slovak Republic
[b] Masaryk University, Faculty of Science, RECETOX, Kamenice 753/5, 625 00 Brno, Czech Republic
[c] Helmholtz Centre for Environmental Research (UFZ), Permoserstraße 15, 04318 Leipzig, Germany
[d] Norwegian Institute for Water Research (NIVA), Gaustadallèen 21, NO-0349 OSLO, Norway
[e] Queensland Alliance for Environmental Health Sciences (QAEHS), University of Queensland, 39 Kessels Road, Coopers Plains, Queensland, 4108 Australia

GRAPHICAL ABSTRACT



ARTICLE INFO

Keywords:
Contaminants of emerging concern
Effect directed analysis
Fractionation
Non-target screening
Toxicity drivers

ABSTRACT

Growth inhibition of freshwater microalga *Pseudokirchneriella subcapitata* caused by a waste water treatment plant (WWTP) effluent extract was investigated using an effect directed analysis (EDA) approach. The objective was to identify compounds responsible for the toxicity by combining state-of-the-art sampling, bioanalytical, fractionation and non-target screening techniques. Three fractionation steps of the whole extract were performed and bioactive fractions were analysed with GC (xGC)-MS and LC-HRMS. In total, 383 compounds were tentatively identified, and their toxicity was characterized using US EPA Ecotox database, open scientific literature or modelled by ECOSAR. Among the top-ranking drivers of toxicity were pesticides and their transformation products, pharmaceuticals (barbiturate derivatives and macrolide antibiotics e.g. azithromycin), industrial

Abbreviations: CEC, contaminants of emerging concern; DMSO, dimethylsulfoxide; EC50/20, concentration which causes 50% (20%) growth rate inhibition of algae compared to solvent control; EDA, effect directed analysis; EtOAc, ethylacetate; GC(xGC)-MS, (two dimensional) gas chromatography coupled to mass spectrometry; LC-HRMS, liquid chromatography coupled to high resolution mass spectrometry; LVSPE, large volume solid phase extraction; MeOH, methanol; MTV, minimum toxicity value; PCPs, personal care products; PPP, plant protection product; REF, relative enrichment factor; RP-HPLC, reverse phase - high performance liquid chromatography; (RP)-SPE, (reverse phase) solid phase extraction; WWTP, waste water treatment plant

* Corresponding author.
E-mail address: blaha@recetox.muni.cz (L. Blaha).

https://doi.org/10.1016/j.jhazmat.2018.05.031
Received 19 January 2018; Received in revised form 11 May 2018; Accepted 15 May 2018

compounds or caffeine and its metabolites. Several of the top-ranking pesticides are no longer registered for use in plant protection products or biocides in the Czech Republic (e.g. prometryn, atrazine, acetochlor, resmethrin) and some are approved only for use in biocides (e.g. terbutryn, carbendazim, phenothrin), which indicates that their non-agricultural input into aquatic environment via WWTPs should be carefully considered. The study demonstrated a functional strategy of combining biotesting, fractionation and non-target screening techniques in the EDA study focused on the identification of algal growth inhibitors in WWTP effluent.

1. Introduction

Microalgae as primary producers are a key functional group in aquatic food webs and possible adverse effects on algal communities may lead to changes at multiple trophic levels and ultimately impair ecosystem health [1]. Toxic effects of anthropogenic contaminants to phytoplankton have been previously reported [2,3,4,5], and methods to test toxicity of compounds or their mixtures to microalgae have therefore been developed and standardized [6,7].

Despite great progress, WWTP effluents entering surface waters still present a major source of toxic pollutants [8,9]. Studies combining biological and chemical analytical approaches reported that between one half to two thirds of phytotoxic effects in surface or waste waters could be explained by herbicides and their metabolites [3,10].

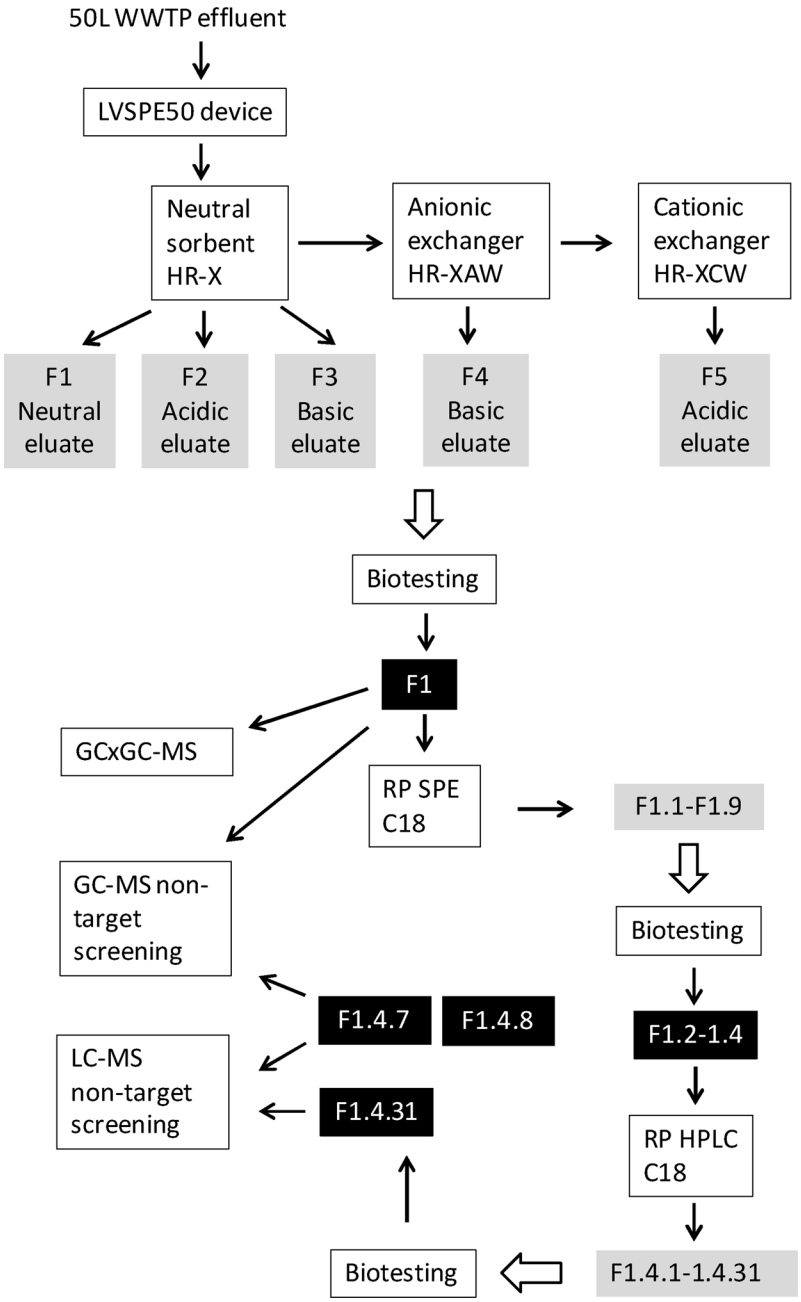


Fig. 1. Overview of the fractionation strategy, biotesting and chemical analyses workflow used for the identification of phytotoxic compounds in the WWTP effluent. The fractions identified as phytotoxic (black boxes) were further fractionated and analyzed.