Science of the Total Environment 650 (2019) 121–131

Contents lists available at ScienceDirect

Science of the Total Environment

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

Assessing the effects of Awba dam sediment (Nigeria) on the steroidogenesis of H295R cells using different extraction methods

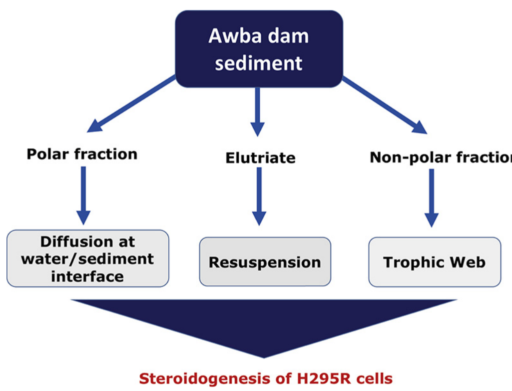
Luigi Natoli [a], Giacomo Luci [a], Elvira Mennillo [a], Aina O. Adeogun [b], Augustine Arukwe [a,*]

[a] Department of Biology, Norwegian University of Science and Technology (NTNU), Høgskoleringen 5, N-7491 Trondheim, Norway
[b] Department of Zoology, University of Ibadan, Ibadan, Nigeria

HIGHLIGHTS

- Extraction methods for screening of a tropical dam sediment for steroidogenesis
- In vitro (H295R cells) endocrine effects of a tropical dam sediment
- Endocrine disrupting chemicals in sediments of a tropical water supply dam
- Health and reproductive consequences of chemical contamination

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:
Received 27 May 2018
Received in revised form 26 August 2018
Accepted 28 August 2018
Available online 29 August 2018

Editor: Kevin V. Thomas

Keywords:
Aquatic sediment
Screening
Extraction methods
Endocrine disruption
H295R cells

ABSTRACT

In the present study, H295R human cells were used to investigate the endocrine disruptor potential of three different sediments extracts taken from a Nigerian tropical freshwater dam (Awba Dam), using three extraction methods that allowed a selective consideration of contaminants based on their binding affinity, which is mainly driven by polarity, to sediment particles. After exposure to different concentration of each extract, H295R cells were evaluated for the expression profiles of 10 steroidogenic enzyme genes and estradiol (E2) and testosterone (T) levels. Our results showed a comparable concentrated-related increase in the expression of *17β-hsd1*, *3β-hsd2* and *cyp21* in cells treated with the polar and non-polar extracts. The *star*, *hmgr*, *cyp11b2* and *17β-hsd4* were slightly decreased, in an apparent concentration-specific manner, after treatment with the polar extract and decreased in the non-polar treatment. The *cyp11a* and *cyp17* showed an opposite trend in the polar and non-polar treatments. E2 was significantly higher in cell treated with the non-polar extract. Elutriate exposure produced less pronounced variation in mRNA and hormones levels. Overall the extract with non-polar compounds produced the most severe effects in H295R cells. Thus, direct ingestion of detritus and mud from fishes and other benthonic organisms represent possible transfer of contaminants in the trophic web, and mainly account for alteration of the endocrine system previously observed in fish from the same study site.

1. Introduction

Aquatic sediments represent significant sink for contaminants such as persistent organic pollutants (POPs), pharmaceuticals and personal

* Corresponding author.
E-mail address: augustine.arukwe@ntnu.no (A. Arukwe).

https://doi.org/10.1016/j.scitotenv.2018.08.409

care products (PPCPs) and heavy metals (Beretta et al., 2014; Conkle et al., 2012; Fu et al., 2014; Martínez-Hernández et al., 2014). Several of these contaminants that are ultimately deposited in aquatic sediments are known endocrine disruptors with physiological, developmental, metabolic and reproductive effects in wildlife species (Arukwe, 2005; Arukwe et al., 2015; Arukwe et al., 2017; Bergman et al., 2013; Mrema et al., 2013; Sharma et al., 2017). EDCs are defined as compounds that possess the ability to alter the synthesis, secretion, transport, binding or elimination of natural hormones, that are responsible for the maintenance of homeostasis, reproduction, development and/or behavior (Dickerson and Gore, 2007; Kavlock et al., 1996; Lee et al., 2013). EDCs can occur naturally, but are also found in human and animal food such as phytoestrogens (genistein and coumestrol) or synthetic compounds (El-Kordy and Alshahrani, 2015). Synthetic EDCs belong to chemicals that are normally used as industrial solvents or lubricant and their by-product such as polychlorinated biphenyls (PCBs), polybrominated biphenyls (PBBs), dioxins, plastics components (e.g. bisphenol A: BPA), pesticides (e.g. dichlorodiphenyltrichloroethane: DDT), fungicide (e.g. vinclozolin) and some pharmaceutical agents (e.g. diethylstilbestrol: DES and 17α ethinylestradiol: EE2) (Diamanti-Kandarakis et al., 2009). EDCs can enter the aquatic environment through a wide variety of sources including as direct discharge of domestic and industrial wastewater, sewage treatment plant effluents (STPE), agricultural run-offs and overland flows after rainfall (Petrović et al., 2001).

In surface water, EDCs undergo series of chemical and biological processes such as dilution, photolysis and biodegradation, allowing a partial elimination from the water column (Błędzka et al., 2010; Chen et al., 2006; Rosenfeldt and Linden, 2004; Ying et al., 2008). However, given the low polarity of many EDCs, sorption to sediment bed (including particles) is probably the main process responsible for the removal of these compounds from the aqueous matrix (Petrović et al., 2001; Sun et al., 2010; Xu et al., 2008; Ying et al., 2003). In the sediment, the anaerobic or low oxygen conditions, normally present in sub-surface layers and low light intensity slow down the photodecomposition and biodegradation process, allowing the persistence of these EDCs for long periods of time (Petrović et al., 2001; Wu et al., 2015). Consequentially, sediment is considered an alternative source for these types of contaminants in the aquatic environment (Fig. 1). Associated with the sediment, EDCs can be transported to other areas and eventually re-released in the aquatic environment by diffusion across the sediment-water interface or through resuspension after strong underwater perturbation events (Casatta et al., 2015). Furthermore, many organisms that spend part or their entire lifespan in association with the benthic domain, can directly ingest detritus/mud or sand grains, and transfer the associated contaminants through the trophic web providing an additional route of exposure (Kelly et al., 2007; Kidda et al., 1998; Kucklick et al., 1996; Wu et al., 2009).

Coastal and inland water of developing countries such as Nigeria, are vulnerable ecosystems due to high fluctuation in water flow rates and contamination pressure from extensive urbanization, industrial and agricultural activities, and waste disposal (Eruola et al., 2012; Onyeike et al., 2002). Some studies, have reported higher sensitivity of tropical freshwater fish species to toxicants exposure when compared with their temperate counterpart (Daam and Van Den Brink, 2010; Kwok et al., 2007). Furthermore, the release of EDCs in these environments will most likely affect pivotal activities such as fisheries which represent a major source of food for the population of these areas leading to major human and wildlife health concerns. A significant part of in vivo and in vitro (e.g. using H295R cell line) studies is oriented towards the effect of contaminants with the ability to interact directly with hormone receptors such as estrogen (ER) and androgen receptors (AR) by mimicking the natural endogenous compounds (Wilson et al., 2004). However, it has also been reported that different EDCs can affect the endocrine system through alternative pathways that does not involve direct binding to hormones receptors, but through modulation of activity and synthesis of enzymes and proteins that control the production, transformation and transport of steroid hormones (Hanet et al., 2008; Rubin, 2011; Tabb and Blumberg, 2005). The H295R cell line is derived from a human adrenocarcinoma and has been widely used (Bláha et al., 2006; Gracia et al., 2008; Grund et al., 2011; Hilscherova et al., 2004; Huang et al., 2011) in evaluating contaminants modulation of steroidogenesis (Fig. 2) through the alteration of enzymes and proteins that regulate steroid hormone synthesis. Thus, representing a unique in vitro system able to produce the steroid hormones found in the adult adrenal cortex and gonads, and allowing the test of both corticosteroids and sex steroid hormones synthesis (Hecker and Giesy, 2008).

To assess the toxicity of sediments, various extraction protocols, permit a selective consideration of different groups of chemicals in relation to physical and chemical properties such as polarity, dimension, shape and density (Fig. 3). The same characteristics also determine the fate of the toxicants in the natural environment with regard to binding to specific routes of exposure to biota (Holt, 2000). The elutriation of sediment has been widely used in ecotoxicological studies (Haring et al.,

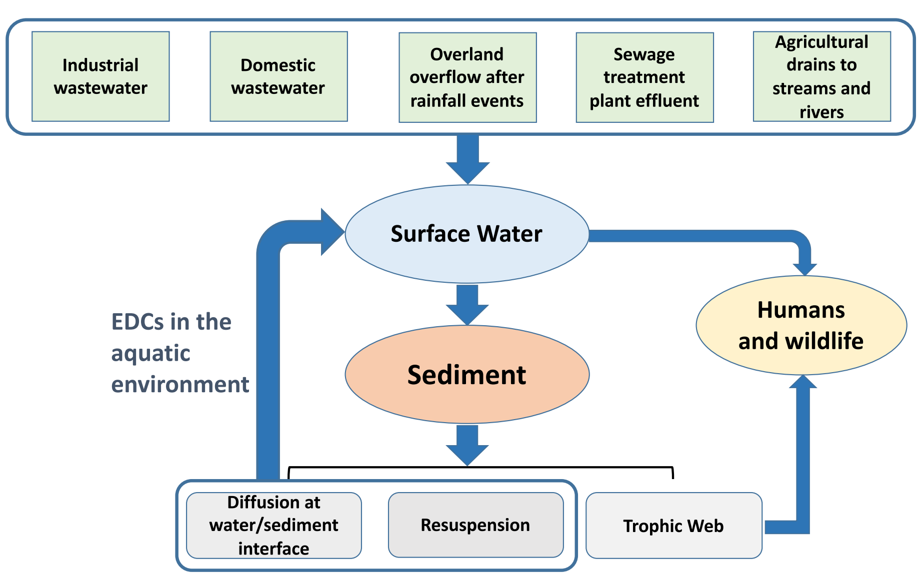


Fig. 1. Contaminants dynamics in aquatic sediment-water interface.