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Distribution of selected antiandrogens and pharmaceuticals in a highly impacted watershed

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ABSTRACT

Endocrine disruption and high occurrences of intersex have been observed in wild fish associated with municipal wastewater treatment plant (WWTP) effluents in urbanized reaches of rivers around the globe. These reproductive effects have often been attributed to the presence of estrogen receptor agonists in effluents. However, recent studies have isolated a number of androgen receptor antagonists (antiandrogens) that may also contribute to the endocrine disruption observed at sites that are influenced by WWTP outfalls. This study aimed to characterize the spatial and temporal distribution of anti-androgenic personal care products (triclosan, chlorophene, dichlorophene, oxybenzone, 1-naphthol, and 2-naphthol), along with a herbicide (atrazine) and representative pharmaceuticals (carbamazepine, ibuprofen, naproxen, and venlafaxine) in the Grand River watershed in southern Ontario. Surface water sampling of 30 sites associated with six municipal WWTP outfalls was conducted during a summer low flow. Monthly samples were also collected immediately upstream and downstream of a major WWTP from August to November 2012.

Atrazine was consistently found in all surface water sampling locations. Many of the target pharmaceuticals and triclosan were detected in WWTP effluents, especially those that did not nitrify. Under low flow conditions, the concentrations of triclosan and several pharmaceuticals increased directly downstream of the WWTPs then decreased rapidly with distance downstream. Chlorophene was either found at trace levels or below detection limits in the effluents while dichlorophene, oxybenzone, 1-naphthol, and 2-naphthol were not detected in any samples. Chlorophene was detected in surface water during the low flow summer period and once during the monthly sampling from August to November. However, the primary source of chlorophene did not appear to be associated with WWTP effluent. This study documents the spatial and temporal occurrence of several anti-androgens and pharmaceuticals in a highly impacted Canadian watershed. It supports previous observations that there is a diversity of contaminants in wastewater effluents and other sources that have the potential to alter endocrine function in wild fish.

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1. Introduction

Municipal wastewater treatment plant (WWTP) effluent is a complex matrix with diverse aquatic environmental contaminants (Daughton and Ternes, 1999; Kolpin et al., 2002; Ternes et al., 2004). WWTP effluents and their receiving water bodies have been linked to endocrine disruption in wild fish (Jobling et al., 1998; Purdom et al., 1994; Vajda et al., 2008). The feminization of male fish in particular has generally been associated with environmental estrogens such as natural and synthetic hormones and industrial chemicals such as alkylphenols (Spengler et al., 2001; Tyler and Routledge, 1998). Recently, it has been suggested that endocrine effects may be associated not only with environmental estrogens, but also antiandrogens (Jobling et al., 2009; Johnson et al., 2007; Katsiadaki et al., 2012). An assessment of the final effluents from selected WWTPs in the United Kingdom (UK) has shown both antiandrogenic and estrogenic activities (Johnson et al., 2007). Furthermore, a statistical modeling showed a strong correlation of the co-occurrence of intersex and the presence of antiandrogens and estrogens in UK WWTPs (Jobling et al., 2009). In the United States (US), it was suggested (Anderson et al., 2012) that despite the reported prevalence of endocrine disruption in US surface waters (Barber et al., 2011; Hinck et al., 2009; Vajda et al., 2008; Woodling et al., 2006), risk of exposure to steroidal estrogens is relatively low for aquatic species. Hence, steroidal estrogens alone may not fully explain the endocrine effects observed.

In an Effects Directed Assessment using bile samples collected from caged rainbow trout exposed to municipal WWTP effluents, Hill et al. (2010) and Rostkowski et al. (2011) were able to isolate a number of antiandrogens using a Yeast Androgen Screen assay as the endpoint. Chemicals associated with the majority of the antiandrogen activity in high performance liquid chromatography fractions included antimicrobial agents (triclosan, chlorophene, dichlorophene, and chloroxylenol), resin acids, industrial chemicals (4-nonylphenol and bisphenol A), a sunscreen agent (oxybenzone), and PAH metabolites (1-hydroxypyrene and naphthols). Triclosan and chlorophene comprised 51% of the antiandrogenic activity found in the bile of the exposed fish (Rostkowski et al., 2011). Although a diversity of potential antiandrogens has been identified in effluents, the level of fish exposure to these chemicals in receiving waters and the mechanism of the responses and potential interactions with other chemicals remain poorly understood.

The Grand River in southern Ontario, Canada is a highly agricultural and urbanized watershed that receives effluents from 30 municipal WWTPs (Anderson, 2012). Effects in fish associated with wastewater effluents in the watershed have been observed including influences in gene expression, physiology, population endpoints, and community assemblages (Ings et al., 2011, 2012; Tetreault et al., 2011, 2013). Intersex occurring at high incidences has also been observed in several sentinel species in the areas downstream of two major WWTPs, Waterloo and Kitchener (Tanna et al., 2013; Tetreault et al., 2011). These effluents have been shown to be estrogenic (Tanna et al., 2013) and likely contain several key environmental estrogens (Smith, 2013). However, the specific compounds and mechanisms responsible for endocrine disruption in fish is yet undetermined. The distribution and fate of potential antiandrogens in effluents and surface waters of the Grand River watershed also remain largely unknown. In this paper, the occurrence and distribution of several antiandrogens in the watershed were examined (Table 1). In addition, several frequently detected pharmaceuticals and the herbicide, atrazine were examined to provide a general pattern of the spatial distribution of trace organic compounds coming from both point and nonpoint sources. The spatial and temporal distribution patterns of these compounds can further enhance the overall understanding of the distribution of target contaminants in watersheds and help direct future water management and infrastructure investments.

2. Materials and methods

2.1. Reagents and chemicals

Chlorophene, dichlorophene, 1-naphthol, 2-naphthol, oxybenzone, ibuprofen, naproxen, carbamazepine, and

Table 1 – Physical and chemical properties of selected contaminants included in the study (United States National Library of Medicine, 2010). pKa = acid-dissociation constant. K_{ow} = octanol–water partitioning coefficient; MDL = method detection limit; SW = surface water; WW = wastewater.

Compound	Major use	pKa	Log K_{ow}	Henry's Law constant (atm m^3/mol)	Solubility (mg/L)	MDL in SW (ng/L)	MDL in WW (ng/L)
Triclosan	Antibacterial	7.9, 8.1[a]	4.76	2.10×10^{-8}	10 at 20 °C	1	46
Chlorophene	Antibacterial	10.8 at 20 °C[b]	3.60	2.70×10^{-8}	149 at 25 °C	3	25
Dichlorophene	Antibacterial	$pKa_1 = 7.6$ $pKa_2 = 11.6$	4.26	1.20×10^{-12}	30 at 25 °C	9	41
Oxybenzone	Sunscreen agent	7.6	3.79	1.50×10^{-8}	69 at 25 °C	20	127
1-Naphthol	PAH metabolite	9.3	2.90	6.00×10^{-8}	866 at 25 °C	15	86
2-Naphthol	PAH metabolite	9.5	2.70	4.60×10^{-8}	756 at 25 °C	12	50
Atrazine	Pesticide	1.9	2.61	2.60×10^{-9} at 25 °C	35 at 26 °C	10	125
Carbamazepine	Anti-epileptic	13.9	2.45	1.08×10^{-7} at 25 °C	18 at 25 °C	3	66
Ibuprofen	Anti-inflammatory	5.2	3.97	1.50×10^{-7}	21 at 25 °C	2	74
Naproxen	Anti-inflammatory	4.2	3.18	3.39×10^{-10} at 25 °C	16 at 25 °C	3	85
Venlafaxine	Antidepressant	10.1	3.20	2.04×10^{-11} at 26 °C	267 at 25 °C	1	302

[a] Perron et al. (2012).
[b] United States Environmental Protection Agency (1995).