Brunel UNIVERSITY LONDON

Occurrence and Removal of Emerging Contaminants in Wastewaters

A thesis submitted to Brunel University in fulfilment of the requirements for the degree of

Doctor of Philosophy

in

Environmental Engineering

by

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London, 2011

DECLARATION OF OWN WORK

I declare that this PhD dissertation:

Occurrence and Removal of Emerging Contaminants in Wastewaters

is entirely my own work and that where any material could be construed as the work of others, it is fully cited and references, and/or with appropriate acknowledgement given.

ABSTRACT

Over the past decade, the occurrence and removal of emerging contaminants in the environment has received much attention. Both natural and synthetic progestogens, which are hormones, and also benzotriazoles are two examples of such emerging contaminants. Sewage treatment works are recognised as one of the main routes of these compounds to the environment. Low concentrations (nanograms per litre) of biologically active chemicals may exhibit an impact on aquatic organisms and human health. This study was undertaken to determine the occurrence and removal of these two classes of chemicals at sewage treatment works, along with an evaluation of the performance of advanced treatment and also to investigate their fate in the aquatic environment. Therefore, field-based sampling campaigns were undertaken at a sewage treatment works, rivers and potable water to achieve these aims. Solid phase extraction and LC/MS/MS were used in order to analyse the samples from these different locations, along with catchment modelling and assessment of how the use of benzotriazoles may contribute to their presence in the environment.

The results have demonstrated that progestogens and benzotriazoles are in the sewage system; the natural hormone (progesterone) was the most predominant compound entering the sewage treatment work (46.9 ng/l) among the progestogens while concentrations of the benzotriazoles were two orders of magnitude higher than the progestogens. The conventional sewage treatment works were, to some extent, able to remove these compounds from wastewaters. However, this may not be adequate to afford protection to the environment. The investigation of advanced treatments, ozone, granular activated carbon and chlorine dioxide, indicated no further significant removal of progestogens, probably as a result of concentrations being close to method detection limits. However, there were indications that benzotriazoles were removed. A degradation study demonstrated that the natural hormone (progesterone) was degraded rapidly while benzotriazoles were not degraded. Catchment modelling indicated that high (up to 2,000 ng/l) concentrations of benzotriazoles would be present in surface waters used for potable supply, and consequently benzotriazoles were found in the tap water with mean concentrations of 30.9 ng/l (benzotriazole) and 15.1 ng/l for tolyltriazole. It is therefore apparent that although conventional treatment may be seen as effective, achieving over 90% removal, this may not be good enough. However, before investing in tertiary treatment, a number of factors, such as the effectiveness at different sites, the presence of degradation products and costs, both financial and in relation to energy use, need to be considered.

ACKNOWLEDGMENTS

First and foremost, I would like to present my thankfulness and greatest glory to ALLAH who gave me the courage and willingness to complete my thesis.

I am immensely grateful to my academic supervisor Dr. Mark D. Scrimshaw for all his support, help and guidance throughout this PhD; I could not have done this without him. I would like to thank my second supervisor Dr. A. J. Chaudhary for his scientific support and guidance during my research.

I am grateful for financial support obtained from sponsors that enabled me to achieve this study, the Ministry of Higher Education and Scientific Research (IRAQ), the Engineering and Physical Sciences Research Council (EPSRC) (UK) and Severn Trent Water Ltd. (UK). Without their financial support and help, the work presented here would not have been possible.

I also would like to take this opportunity to thank the staff at the wastewater treatment plant, John Churchly, Michael and Emilie Cope for their help and efforts during the sampling campaigns. Moreover, I am deeply in debt to Richard Williams at Centre for Ecology and Hydrology (CEH) for his help with modelling. Special thanks goes to all staff, friends and colleagues at Brunel, for creating an unforgettable working environment and friendly atmosphere and also being my best 'teachers' of the English language.

Finally, a great thank you to my family and relatives for their encouragement, love and support, especially my parents that have always been there for me. I would like to thank my wife for being patient, loving and caring since the second year of my PhD. I am grateful they gave me guidance and encouragement to be the person I should be.

Hussein

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LIST OF ABBREVIATIONS, UNITS AND SYMBOLS

AOP	Advanced oxidation process
APs	Alkylphenols
APCI	Atmospheric pressure chemical ionization
ASP	Activated sludge process
BNR	Biological nutrient removal
BOD	Biochemical oxygen demand
BT	Benzotriazole
CAD	Collision gas pressure
CEH	Centre for Ecology and Hydrology
C_{eff}	Concentration of the effluent
Cl_2O_2	Chlorate
ClO_2^-	Chlorite
ClO_2	Chlorine dioxide
CPA	Cyproterone acetate
COD	Chemical oxygen demand
CUR	Curtain gas
DBPs	Disinfectant by products
DCM	Dichloromethane
DDT	Dichlorodiphenyltrichloroethane
DES	Diethylstilbestrol
DHG	Dydrogesterone
DSP	Drosprinone
DO	Dissolved oxygen
DOC	Dissolved organic carbon
DP	Declustering potential
DWF	Dry weather flow
E1	Estrone
E3	Estriol
EBCT	Empty bed contact time
EE2	17α-ethinyl estradiol
EDCs	Endocrine disrupting chemicals

EP	Entrance potential
ESI	Electrospray ionisation
F/M	Food to microorganism ratio
FE	Final effluent
FP	Focusing potential
GAC	Granular activated carbon
GC	Gas chromatography
GC/MS	Gas chromatography mass spectrometry
GC/MS/MS	Gas chromatography tandem mass spectrometry
GPC	Gel permeation chromatography
H_c	Henery's law constant
$HgCl_2$	Mercury chloride
HLB	Hydrophilic-lipophilic balance
HPLC	High performance liquid chromatography
HRT	Hydraulic retention time
K_a	Dissociation coefficient
K_{biol}	Degradation constant
K_d	Distribution coeficient
K_{ow}	Octanol-water partition coefficient
$K_{tot.}$	Over all degradation rate
LC	Liquid chromatography
LC/MS	Liquid chromatography mass spectrometry
LC/MS/MS	Liquid chromatography tandem mass spectrometry
LOD	Limit of detection
LOEC	Lowest observed effect concentration
Log K_{ow}	Logarithm of octanol-water partition coefficient
LOQ	Limit of quantification
MCRT	Mean cell residence time
MDP	Medroxyprogesterone
MPA	Medroxyprogesterone acetate
MRM	Multiple Reaction Monitoring
MTA	Megestrel acetate
MTBE	Methyl tert-butyl ether

MeOH	Methanol
N_2	Nitrogen
N.D	Not detected
NEB	Nebulizer gas
NGL	Norgestrel
NOEC	No observed effect concentration
NTD	Norethindrone
N/AS	Nitrifying activated sludge
O_3	Ozone
PAC	Powdered activated carbon
PAHs	Polycyclic aromatic hydrocarbons
PE	Population equivalent
PGT	progesterone
PPCPs	Pharmacetical and personal care products
POPs	presistant organic pollutants
Q	Volumetric flow rate (m^3/d)
RSD	Relative standard deviation
RO	Reverse Osmosis
RT	Retention time
SPE	Solid phase extraction
SRT	Sludge retention time
STWs	Sewage treatment works
T	Temperature
TBL	Tibolone
TF	Trickling filter
TT	Tolyltriazole
UK	United Kingdom
USA	United State of America
USEPA	United States Environment Protection Agency
V	Volt

LIST OF PUBLICATIONS

Hussein Janna, Mark D. Scrimshaw, Richard J. Williams, John Churchley & John P. Sumpter. (2011) "From Dishwasher to Tap? Xenobiotic Substances Benzotriazole and Tolyltriazole in the Environment", *Environ. Sci. Technol,* vol. 45, no. 9, pp. 3858-3864.

Chapter One: Introduction

1.1 Introduction

One of the most essential components for the existence of life is water. As a result of the large increase in the population in the world, there is also an increasing demand for clean, safe water which either comes from the fresh water or as a result of reusing wastewater directly or indirectly. Therefore, the quality of the water plays a major role in the protection of wildlife and human health.

For many decades, heavy metals and persistent organic pollutants (POPs) were the area of interest for most of the researchers in the aquatic environment. However, these pollutants have had less awareness when a significant reduction in producing these chemicals was achieved and also when "new" or "emerging" unregulated contaminants emerged with some environmental problems (Barceló & Petrovic, 2008).

The U.S Geological survey defines emerging contaminants (ECs) as "any synthetic or naturally occurring chemical or any micro-organism that is not commonly monitored in the environment but has a potential to enter the environment and cause known or suspected adverse ecological or human health effects" (USGS, 2011). In addition, Wong (2006) has defined ECs as "chemicals (including veterinary and human pharmaceuticals) that currently are being used and released into the environment and are of special concern due to widespread occurrence and potential for toxic effects".

The concern about the presence of these compounds in the environment has led to the publication of complete editions of journals dedicated to the topic. For example, Environmental Science and Technology published a special issue in December 2006 about ECs (volume 40, issue 23), followed by Analytical and Bioanalytical Chemistry in February 2007 (volume387, issue 4) due the increased interest in the subject.

There is a large number of ECs in the environment with a myriad of sources and pathways. The USEPA have reported in August 2000 that about 87,000 chemicals need to be tested to determine if they have endocrine disrupter effects (USEPA, 2000). There are about 38,000 chemicals and some heavy metals that are reported as chemical substances suspected to have an endocrine disrupting action (Quan et al., 2005).

Emerging contaminants are released to the environment in different ways. There are a very large number of possible emerging contaminants, from industrial chemicals,

antiseptic and anti microbial agents, flame retardants, detergents and their derivatives, and plasticizers and their derivatives, to the category known as pharmaceuticals and personal care products (PPCPs), which are a diverse group of chemicals used in veterinary medicine, agricultural practices, human health and cosmetic care (e.g., cosmetics, cleaning products, and fragrances). Pharmaceuticals and personal-care products (PPCPs) have caught the scientific and public attention and therefore have seen a substantial increase in research over the past 10 years above the other emerging contaminants (Glassmeyer et al., 2008).

The presence of emerging contaminants in the aquatic environment represents a potential concern to wildlife (Daughton & Ternes, 1999; Pillard et al., 2001; Ferrari et al., 2003; Barceló & Kettrup, 2004) and they may also have an impact on human health (Daughton & Ternes, 1999; Barceló & Petrovic, 2008). Conversely, other scientists think that the risks from these substances to human health are negligible (Christensen, 1998; Fent, Weston & Caminada, 2006).

Although much has been published, there still remain gaps in understanding about the fate and effect of emerging contaminants in the environment. However, emerging contaminants are a wide range of substances, have many different sources and pathways, and may have a variety of effects on aquatic organisms and human health.

In order to identify compounds for inclusion in this study, we have used criteria based on biological activity and “new” or “emerging” contaminants which may be present in appreciable (µg/l) concentrations. It is also necessary to limit numbers (of chemicals) to ensure any work programme is deliverable. Therefore, two groups of compounds, progestogens and the benzotriazoles, which have relevance in terms of occurrence, removal processes and ecotoxicological effects, have been selected for study.

1.2 Aims and objectives

The overall aim of this study is to understand the occurrence and fate of progestogens and benzotriazoles with a focus on the effectiveness of conventional and advanced wastewater treatment techniques. This aim will be achieved by determining the occurrence of these chemicals in wastewaters and the aquatic environment in the UK. Therefore the overall aim of this study will be met through achieving the following specific objectives:

1.2.1 To determine the concentrations and removal efficiency of progestogens and benzotriazoles in conventional wastewater treatment with different biological treatment processes (activated sludge process and trickling filter).

1.2.2 To determine the concentrations and removal efficiency of progestogens and benzotriazoles during advanced wastewater treatment (ozone, granular activated carbon, and chlorine dioxide).

1.2.3 To determine the degradation rates and fate of progestogens and benzotriazoles in surface waters.

1.2.4 Analyse effluent samples from eight further wastewater treatment plants and specific points on the river to generate information on spatial distribution. Predict the wider occurrence of the progestogens and benzotriazoles in the UK by modelling the rivers at a catchment scale.

1.2.5 Analyse drinking water samples to find the concentration of benzotriazoles and also analyse different brands of detergents to justify the occurrence of these chemicals in the drinking water.

1.3 Thesis outline

This thesis is organized sequentially to address the above objectives, therefore chapter two provides a review to the available literature regarding the sources and possible pathways of emerging contaminants in the aquatic environment. In addition, an overview to the removal mechanisms, sewage treatment works and environmental impact of these contaminants will also conducted in this chapter.

One class of steroid hormones, the progestogens, represented by one natural hormone and nine synthetic compounds, were selected for this study due to their biological activity. Moreover, another group of chemicals, benzotriazoles, which are classified as emerging contaminants due to their extensive use in a range of products, represented by two compounds, were also chosen for this study. The physicochemical properties of all these compounds are illustrated in the Materials and Methods (chapter three), as are analytical methods to determine the concentrations of these compounds. In addition, a field study, the sampling regimes and quality control, are described in this chapter, along with conditions of the degradation study and also the software for modelling.

The concentrations and removal efficiencies of these 12 compounds in two different types of biological treatment, the activated sludge process (ASP) and trickling filter (TF) during wastewater treatment works are illustrated in chapter four. Chapter five presents the concentrations of these compounds and their removal efficiencies during advanced treatment with ozone (O_3), granular activated carbon (GAC) and chlorine dioxide (ClO_2).

Chapter six describes the behaviour and degradation of these 12 compounds during a laboratory test. Monitoring these compounds along the River Erewash with inputs from the existing sewage treatment works is also described, along with modelling the catchments of the Rivers Trent and Thames in chapter seven.

Chapter eight describes the occurrence of the benzotriazoles in drinking water and also investigated one of the possible sources of these contaminants. A discussion of the results and efforts from this study is undertaken in chapter nine, explaining the possible reasons for these results and comparing these results what other people has done.

Finally a summarization of the total results and major conclusions from these efforts about what was achieved in this study were described in chapter 10.

Chapter Two: Literature Review

Overview

During doing this research, a comprehensive review to the literature has been done in order to cover the emerging contaminants in sewage and aquatic environment. This chapter will talk about the history of emerging contaminants in the environment, and then discuss the possible sources of these compounds and their pathways to enter the aquatic environment. Usage of some chemicals with a high production volume (HPV) and also using pharmaceuticals in UK also will be described. An overview of sewage treatment works (STWs) and their role to remove these compounds in relation to the possible removal mechanisms which may occur will be also covered. Finally advanced treatment, represented by current technologies and their ability to remove emerging contaminants from effluents will be covered.

2.1 Introduction

In the environment, emerging contaminants (ECs) are substances released from domestic, industrial, and agricultural sources (Yan et al., 2010). These chemicals are not routinely monitored but some of them are observed by researchers throughout our environment, these substances may have the ability to cause adverse affects in the environment (Sumpter & Jobling, 1995; Jobling et al., 1995; Witte, 1998). Emerging contaminants have been observed in the aquatic environment in wastewaters, surface waters, ground waters, and in some cases in the drinking water (Cancilla, Martinez & Van Aggelen, 1998; Stumpf et al., 1999; Ternes et al., 1999; Barnes et al., 2004; Bolong et al., 2009; Pojana, Fantinati & Marcomini, 2011). A wide spectrum of these ECs, both natural and synthetic chemicals, have the ability to mimic the natural hormones in the body, and may consequently interfere with the endocrine system or interact with the hormone receptor and activate or block a response and then could create an adverse influence in the body via affecting the reproductive, immune and neurological systems (Birkett & Lester, 2003; NIEHS, 2011). Such chemicals are known as endocrine disrupting chemicals (EDCs). The issue caught awareness when the environmental impacts of some of the ECs were linked with some effects like feminization of fish (Desbrow et al., 1998; Jobling et al., 1998), however, for many other substances, their

fate, behaviour, and eco-toxicological impact are still not understood. Conventional sewage treatment works cannot remove ECs completely from wastewater although these substances undergo different removal mechanisms during the STW processes based on their physicochemical properties and (bio) degradability. Advanced treatment is a promising technology to remove some of these contaminants, however, each technology has advantages and disadvantages in addition to cost/benefit criteria and energy consumption of operating these system. In general, ECs represent a challenge to the engineers, scientists, and also to society.

2.2 The history of emerging contaminants in the aquatic environment

During the past two decades, there was an increasing concern about the occurrences of emerging contaminants in the aquatic environment. However, one of the earliest preliminary reports about steroid hormones in wastewater was published in 1965 (Stumm-Zollinger & Fair, 1965). Then, other researchers published several studies about the presence of human hormones and pharmaceuticals in the aquatic environment between 1970s and 1980s (Tabak & Bunch, 1970; Hignite & Azarnoff, 1977; Aherne, English & Marks, 1985). In 1990s, it was feasible to detect these chemicals as the technology became more developed, and more attention to the issue began to be received from the environmental scientists when some studies linked the presence of these chemicals to those released to the environment from the STWs to a toxicological impact in aquatic organisms (Desbrow et al., 1998; Jobling et al., 1998; Kramer et al., 1998; Snyder et al., 2001). During the late 1990s and the past decade, there was an abundance of studies which have shown a spectrum of many emerging contaminants and their metabolites in the aquatic environment such as surface waters (Cancilla, Martinez & Van Aggelen, 1998; Stumpf et al., 1999; Kolpin et al., 2002; Metcalfe et al., 2003; Wiegel et al., 2004) , in ground water (Holm et al., 1995; Heberer, Schmidt-Baumler & Stan, 1998; Peterson, Davis & Orndorff, 2000; Sacher et al., 2001; Barnes et al., 2004), and in drinking water (Boyd et al., 2003; Loraine & Pettigrove, 2006; Kuster et al., 2008; Kleywegt et al., 2011; Pojana, Fantinati & Marcomini, 2011).

2.3 Emerging contaminants in the aquatic environment.

2.3.1 Classification of contaminants.

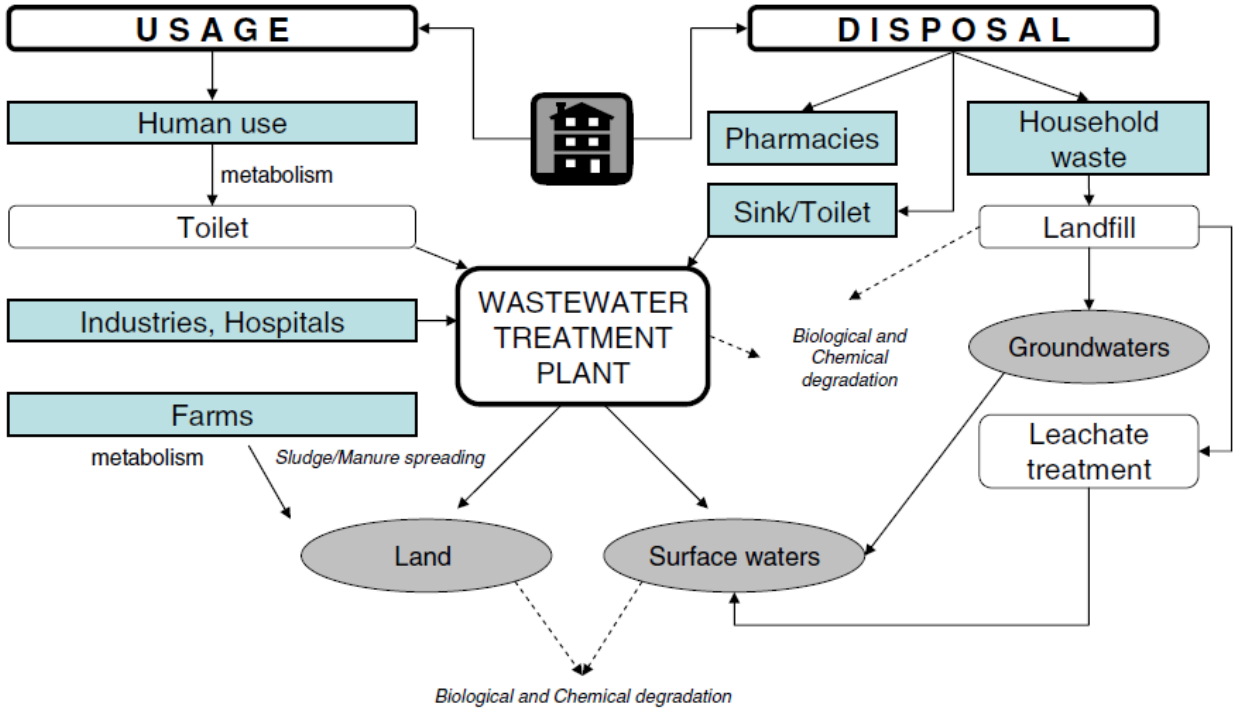
There are different types of environmental contaminants and these types are classified according to different criteria. Some of these classifications are according to their origin, environmental persistence, human toxicity and ecotoxicity. It is possible to say that there are three main types of emerging contaminants:

- Persistent organics: this group represents one of the major groups of emerging contaminants in the environment. These are often semi-volatile, halogenated compounds used as pesticides or other industrial compounds like flame retardants, and pesticide such as DDT. Some of these compounds are still produced and in large volumes therefore are classified as high production volume chemicals (USEPA, 2011).
- Pharmaceuticals and personal care products: this group includes all pharmaceuticals like antibiotics, antiseptics, anti-inflammatories, and antiepileptic drugs. Personal care products include UV protection screens and musks. Synthetic hormones such as 17αethinyletsradiol (EE2) are also part of this family.
- Inorganics: this group includes heavy metals like lead, mercury, and arsenic.

Each group of these three classes has some chemicals that have an impact on the endocrine system, therefore some of these chemical are named EDCs. Some of the pharmaceutical compounds are biologically active and can affect environmental and human health. Because of these interpretations between these chemical in their characteristics and then in their impact on human and wild life, it is possible to say that these contaminants can belong to more than one group according to the criteria of classification. Emerging contaminants may be in any other group, but they are emerging only once we become aware of them.

2.3.2 Sources and pathways of emerging contaminants.

Emerging contaminants are released into the environment via different sources; Figure 2.1 shows the possible ways of releasing these contaminants.



Adapted from Nikolaou, Meric & Fatta, 2007

Figure 2.1 Sources and fate of emerging contaminates in the environment showing the range of possible inputs and receptors.

There are many different sources of ECs into the environment because of the variety in the classes of the ECs. Emerging contaminants can be divided into natural and synthetic compounds. The natural compounds which are hormones are excreted by vertebrates and by some other invertebrates groups (Oehlmann & Schulte-Oehlmann, 2003), while synthetic compounds or man-made compounds can be produced by manufacture. Factories and manufacture are the main sources of these contaminants where ECs are either every day products or used as raw materials to produce other products (Marttinen, Kettunen & Rintala, 2003; Moon et al., 2007). Effluents are discharged from these manufacture, for example the pharmaceuticals and personal care products industry, mining, rubber manufacturing, corrosion inhibitors and also from pesticides industry, although these discharges are mostly controlled by legislation, for example IPPC directive (EUROPA, 2011). Therefore, different chemicals were found in the

wastewater like flame retardants (Peng et al., 2009), alkyphenols (APs) and polycyclic aromatic hydrocarbons (PAHs) (Vogelsang et al., 2006; Loos et al., 2007), phthalates (Balabanic & Klemencic, 2011) and pesticides (Kahle et al., 2008).

Wastewater treatment plants are also a well known source. Many natural and synthetic emerging contaminants from households, hospitals, industrial use, and sometimes from storm water enter the sewage treatment works. Because these contaminants are not fully or partially removed during the chemical, physical, and biological treatment processes (Jones, Voulvoulis & Lester, 2005; Sarmah et al., 2006; Nakada et al., 2006; Jones, Voulvoulis & Lester, 2007; Koh et al., 2008; Bolong et al., 2009), a considerable amount of these ECs, like natural hormones and benzotriazoles, are released to the receiving water (Desbrow et al., 1998; Weiss & Reemtsma, 2005). In addition, in some cases (such as storm events), the untreated or partially treated wastewater may reach the receiving water and could also have significant amounts of these contaminants. Thus, wastewater treatment plants seem to be an important source of emerging contaminants (Tan et al., 2007).

Another possible source for emerging contaminants to enter into the environment is storm water runoff. Many types of ECs were found in storm water samples, such as flame retardants, plasticizers, pesticides, PPCPs, and heavy metals (Hurst & Sheahan, 2003). Miltner et al. (1989) found that many emerging contaminants in the influent of three STWs came from storm runoff. Boyd et al. (2004) presented results about the occurrences of PPCPs and endocrine disrupters in three sites of storm water samples.

The runoff from the agricultural area is also a source of emerging contaminants especially with pesticides to improve crop productivity, antibiotic from animal feed process, steroids hormones and antimicrobial from animals and livestock (Birkett, 2003; Matthiessen et al., 2006; Song et al., 2007; Snow et al., 2010). Emerging contaminants released from agriculture practices contribute to loading the aquatic environment such as antibiotics and hormones as they were detected in a watershed in USA which contains 62% of the area as an agriculture land (Arikan, Rice & Codling, 2008). Hormones were also found in the soil and runoff grasslands in the USA (Finlay-Moore, Hartel & Cabrera, 2000). In addition fields irrigated with treated wastewater also contribute to provide many emerging contaminants and their metabolites to the receiving waters (Pedersen, Yeager & Suffet, 2003). The reuse of the wastewater for agricultural purposes may

transfer some of these ECs to the land again, therefore, an environmental and hazards risk assessment studies have achieved to evaluate the effect of these ECs and also developing the tools or approaches to assess their impact (Dominguez-Chicas & Scrimshaw, 2010; Gros et al., 2010).

The other source is the leakage from landfills, industrial wastes systems, sewer, and leakage from sewage treatment facilities. Many different ECs such as pharmaceuticals, plasticizers, and pesticides were found in the seepage and leakage water from waste landfill (Schwarzbauer et al., 2002). Due to the leakage in the sewage pipes in Germany, iodinated contrast x-ray media were found in the groundwater samples (Wolf, Eiswirth & Hotzl, 2006). Percolating the rainwater through the domestic landfill and leakage from septic tanks are also classified as one of ECs sources (Birkett, 2003).

2.3.3 Analytical methods to determine emerging contaminants in aquatic environment.

After the technology has become more developed to detect trace chemical in the aqueous phase, many studies have focused on the methods for determination these substances in the environment. Different techniques have been applied in order to improve the accuracy of detection these compounds and consequently find the actual concentrations.

The first step of analysis is sample preparation and begins from sample filtration after collecting the sample, this is especially for wastewater samples because they contain a high loading of organic material and suspended particles, and additionally the purpose of this step is to protect the subsequent extraction step. The target chemicals (estrogenic components) were not retained by the filter material and they were present in the dissolved phase of the sample effluent (Desbrow et al., 1998). Also 99% recovery was obtained when wastewater samples were spiked with 17β-estradiol, this means that sorption of hormones on filter was negligible (Huang & Sedlak, 2001).

The next step is extraction of the sample which could be achieved by either liquid/liquid separation or solid phase extraction (SPE). The type of separation depends on the type

and properties of the target compound and also on the sample matrix. There are two commonly used types of SPE, cartridge or disk. Although disks reduce the clogging of sample due to their large surface area, however larger amount of solvent are required when using disks in order to elute the analyte and that will increase the method duration and consequently time taken to concentrate the eluted sample (Gomes, Scrimshaw & Lester, 2003). Further clean up might be required to purify the extracts from non target chemicals which is known as purification, which can be achieved by separating the interested molecular from other molecular masses based on size by using gel permeation chromatography prior to their analysis (Ternes et al., 2002). In some cases, there is a need to improve the stability and enhance the detectability, of the compounds in order to increase the sensitivity during the use of gas chromatography to analyse the target compound, which is known as derivatization. Derivatization is achieved by using some agents to derivatise the extracted sample; nevertheless, these advantages are sometimes offset by loss of sample during the additional manipulation (Desbrow et al., 1998; de Alda & Barcelo, 2001).

There are many different methods to quantify the ECs in the aquatic environment. Biological techniques such as immunoassay are among the most sensitive analytical techniques; however, they are limited by the availability of the specific antisera and subject to cross-reactivity (de Alda & Barcelo, 2001). Other two methods which are commonly used for quantification of compound of interest after extraction are gas chromatography (GC), and liquid chromatography (LC) coupled to mass spectrometry (MS) or tandem mass spectrometry (MS/MS). Despite the fact that chromatographic methods are not as sensitive as biological techniques, however chromatographic methods are widely used due to their ability to screen steroids and their conjugate simultaneously (de Alda & Barcelo, 2001).

There are several studies achieved to determine the concentration of many EDCs in the aquatic environment using biological techniques (Huang & Sedlak, 2001; Xin et al., 2009), and chemical methods, for LC/MS/MS (Vanderford et al., 2003; D'Ascenzo et al., 2003; Voutsa et al., 2006; Gros, Petrovic & Barcelo, 2006; Hernando et al., 2006; Gomez et al., 2006; Vega-Morales, Sosa-Ferrera & Santana-Rodriguez, 2010; Sapozhnikova et al., 2011), for GC/MS (Daughton & Ternes, 1999; Labadie & Budzinski, 2005a; Labadie & Budzinski, 2005b), and for GC/MS/MS (Huang & Sedlak, 2001; Kolodziej, Gray & Sedlak,

2003; Ternes et al., 2003; Kolodziej, Harter & Sedlak, 2004). Extraction and purification were involved in most of these studies in order to quantify the concentrations of target compounds.

GC/MS and GC/MS/MS are unlike LC/MS/MS, in that they are limited to the volatility property and molecular weight of the chemical of interest. In addition, LC/MS could screen the conjugated and unconjugated chemicals without need from derivatization (de Alda & Barcelo, 2001). Several of these methods demonstrated a high limit of detection from the wastewater samples compared to the other samples collected from clean river or lake water or drinking water and this is due to the matrix of the sewage samples. For example, the limit of detection was from 0.3-600 ng/l in the study of steroids in the urine and final effluent sample (D'Ascenzo et al., 2003), also from 0.6-35 ng/l during the observation of hormones and antibiotic in the influent and effluent wastewater in Germany (Schlüsener & Bester, 2005).

2.3.4 The occurrence of emerging contaminants in the aquatic environment.

There are a large number of emerging contaminants that are potentially present in the environment. Emerging contaminants include PPCPs, hormones and veterinary medicines, household compounds, and some chemicals used in industry. There is a wide spectrum of studies from a number of countries focused on these contaminants in terms of their occurrence, behaviour in the aquatic environment and also their impact on humans and aquatic organisms. Therefore this section will provide a very brief review of the literature which demonstrates their presence in wastewaters (influent and effluent), river waters, ground waters, and in drinking waters.

Worldwide, the high usage of chemicals in industry and households, and also the use of therapeutic drugs, is likely to make the presence of these substances in the environment unavoidable. In addition, unused medicines and the surplus chemicals used for domestic use might be discharged through the sewerage system. As a result of leakage from septic tanks or throughout the sewer systems, some of these chemicals are then transmitted to the ground waters. There are several studies which have reported the occurrence of different chemicals in ground waters (Heberer et al., 1997; Sacher et al., 2001;

Schwarzbauer et al., 2002; Drewes et al., 2003; Wolf, Eiswirth & Hotzl, 2006; Vulliet et al., 2008; Barnes et al., 2008; Schulz et al., 2008; Silvia Diaz-Cruz & Barcelo, 2008; Teijon et al., 2010).

However, the bulk of contaminants will flow to the STWs and therefore it is very likely to find these compounds in the influent of the STWs at concentrations ranging from ng/l to μg/l. Due to incomplete removal of ECs in the STWs (Nakada et al., 2006; Koh et al., 2008; Bolong et al., 2009; Kusk et al., 2011), this means that these contaminants will also be present in the effluent of the STWs, and that the residual concentrations will be discharged to the receiving water bodies. Many studies have illustrated the occurrence of ECs in the wastewater treatment processes (Stumpf et al., 1999; Petrovic et al., 2002; Andersen et al., 2003; Joss et al., 2004; Weiss & Reemtsma, 2005; Carballa, Omil & Lema, 2005; Carballa et al., 2005; Sarmah et al., 2006; Nakada et al., 2006; Reemtsma et al., 2006; Voutsa et al., 2006; Choi et al., 2008; Kuster et al., 2008; Chang et al., 2008; Reemtsma et al., 2010; Gros et al., 2010; Sodre, Locatelli & Jardim, 2010; Jelic et al., 2011; Chang et al., 2011; Fan et al., 2011) and also in surface waters (Aherne, English & Marks, 1985; Kolpin et al., 2002; Boyd et al., 2003; Moldovan et al., 2007; Nakada et al., 2007; Vieno et al., 2007; Choi et al., 2008; Kim et al., 2009; Benotti et al., 2009; Jover, Matamoros & Bayona, 2009; Chang, Wan & Hu, 2009; Reemtsma et al., 2010).

Because some of these chemicals are recalcitrant, they will persist in the environment and it is also possible that they will be present in the potable water when the surface or ground waters are contaminated and used as a drinking water supply (Aherne, English & Marks, 1985; Vieno, Tuhkanen & Kronberg, 2005; Loraine & Pettigrove, 2006; Richardson et al., 2007; Kuster et al., 2008; Sodre, Locatelli & Jardim, 2010; Benotti et al., 2009; Pojana, Fantinati & Marcomini, 2011).

2.3.5 The impact of emerging contaminants in the aquatic environment.

In history, for about 33 years since 1938, diethylstilbestrol (DES) was given to avoid miscarrying of pregnant women (CDC's DES Update, 2009). Afterwards, it was revealed that DES can alter the fertility and reproductive performance of the children of mothers who were directly exposed to this synthetic hormone (CDC's DES Update, 2009). Due to

the variety in the types of ECs particularly EDCs and each compound has a different properties and modes, thus each chemical behaves differently in the environment. Although there is a vast number of substances that are released to the environment have not been studied yet in terms of their potential effect to the environment, however there is a large number of studies demonstrating the effects of the ECs on the environment and therefore it would be difficult to cover the impact of studied compounds on the environment especially on wildlife and human. The endocrine system can be affected by many natural and synthetic chemicals such as natural and synthetic estrogens, natural androgens, pesticides, phthalates, and bisphenol A (Richardson, 2009). Organisms in the surface waters are exposed to the contaminants and most of them affected at concentrations as low as a few ng/l due to the continuous release of ECs (Daughton & Ternes, 1999; Barcelo & Kettrup, 2004). Some chemicals have the ability to accumulate in the fat tissue and cause adverse effect to the organisms, such as the feminization and reproductive problems of fish in the surface water due to uncompleted removal of estrogens from the STWs (Purdom et al., 1994; Jobling et al., 1998; Kramer et al., 1998). The declination in population of bald eagle was correlated with some ECs in Chesapeake Bay (Wiemeyer et al., 1984). Invertebrates can be also influenced by ECs and may be more sensitive than vertebrates (Ferrari et al., 2003; Kusk et al., 2011). Humans can be exposed to the ECs via the drinking water or food chain and consequently might be affected. Although there are some studies have shown no substantial concern about the effect of ECs on human and they are unlikely to harm the human (Christensen, 1998; Webb et al., 2003) , however there is a suspicion about declining the number of sperm in human due to the impact of ECs even though not confirmed yet (Richardson, 2009).

In conclusion, the impact of ECs on the environment and wild life represented by some problems in reproductive systems in fish, birds, and mammals in addition to the breakage of eggs of fish, birds and turtles, additionally feminization of male fish and finally some changes in the immunologic system of marine mammals. The ECs can influence the human also represented via reduction in sperm count and increase the incidence of breast or endometriosis cancer for women, testis and prostate cancer from men (Esplugas et al., 2007).

2.4 Overview of STW.

The main aim of the wastewater treatment works is to produce treated effluent that is suitable and safe to discharge to the environment and increasingly to reuse it. Therefore, the role of wastewater treatment is to convert the waste material present in the wastewater to stable oxidised end products (Gray, 2004). The sewage treatment works normally consists of physical, chemical and biological processes in order to remove the physical, chemical and biological contaminants. Therefore STWs are assembled from a combination of unit processes. In general, there are five stages during sewage treatment (Gray, 2004):

- Preliminary treatment: gross solid and grit are removed and sometime oil and grease as well if they present in large amount.
- Primary (sedimentation) treatment: the settable solids are removed in the sedimentation tanks.
- Secondary (biological) treatment: in this stage, the organic matter (dissolved and colloidal) with presence of microorganisms is removed.
- Tertiary treatment: the residual suspended solid and nutrient are removed.
- Sludge treatment: sludge collected from previous stage are treated by dewatering, stabilization in order to disposal it.

Many studies have focused on the removal of emerging contaminants (ECs) during secondary treatment (biological treatment). This is because other studies have shown that the major removal of these compounds is by biodegradation and adsorption. Therefore, this section will focus on the fate and behaviour of these compounds during the treatment processes, but mainly during the biological treatment which is usually consists of activated sludge process (ASP) or trickling filter (TF).

2.4.1 Preliminary treatment.

The function of screens, shredders, grit removal, and flow equalization in the head of sewage treatment works is to protect the downstream equipments and to provide a homogenous feed to subsequent processes facilities (Davis, 2011). Large floating objects and small amount of organic matter are removed via screens. Grit and dense material

solids are removed by grit removal. A small amount of emerging contaminants were removed during preliminary treatment (Lester & Edge, 2000; Fan et al., 2011).

2.4.2 Primary treatment.

In this stage, the raw sewage enters the primary sedimentation tanks and in this process the settable solids (suspended solids) are removed from wastewater by allowing the particles to gravitate to settle in the bottom of the tank. The most significant mechanism in primary sedimentation tanks is adsorption onto solids, which is under the impact of the gravity settle from the primary sludge. Many factors could affect the degree of pollutant removal, one of the important factors is the hydrophobicity of the compound, and also suspended solids which is controlled by settling characteristics of the particle and also the operation specification of the settling tank such as retention time and surface loading rate play an important role (Langford & Lester, 2003; Carballa et al., 2005).

2.4.3 Secondary treatment.

In this stage, the settled sewage enters the secondary treatment which is biological treatment. The aspect of this process depends on the presence of aerobic bacteria and other microorganisms to oxidize or incorporate into cells the organic matter. This treatment occurs by providing a sufficient amount of oxygen to the bacteria (Langford & Lester, 2003). Trickling filter where the large population of microorganisms are attached to the fixed surface and the ASP where the large population of microorganisms mix with the wastewater are the two main kinds of biological treatment. Each one of them is normally followed by a secondary sedimentation, the purpose of secondary sedimentation tank is to separate the dense microbial biomass that formed during the biological conversion from the water by settlement (Gray, 2004).

Activated sludge process (ASP)

The most common and widely used type of biological treatment at large STW is ASP, treating both industrial and domestic wastewater. The name derived from the process itself where air is injected to the reactor and biomass is recirculated to the aeration tank continuously. The activated sludge system consists of three main divisions (Metcalf & Eddy, 2003):

- Reactor in which the microorganisms are responsible for treatment are kept in suspension and aerated.
- Liquid solid separation system which is sedimentation tank usually.
- Recycle system which is responsible for feeding solids that settled in the sedimentation tank to the aeration tank.

The performance of activated sludge process to remove estrogens in Brazilian STW was better than that of the trickling filter (Ternes et al., 1999). There are many advantages and disadvantages for using activated sludge process and these depend on the type of the process. For example, the advantages of some types are less space, higher level of ammonia removal and odour free. However, the required energy for aeration tank is high, and also the flexibility to change the effluent characteristics is limited (Metcalf & Eddy, 2003). There are many factors that could affect the process such as the characteristics of the wastewater being treated, foaming due to detergents, temperature, return rate, and oxygen availability (Gray, 2004).

Trickling filter (TF)

Fixed film or attached growth systems, trickling filter, has been used for about 100 years (Metcalf & Eddy, 2003). In trickling filter, wastewater is sprinkled over the media continuously where the microbial biomass is present as a film which grows on the surface of an inert or solid medium (Gray, 2004). The conventional trickling filter using circular rock as the packing materials and now days have been converted to plastic packing in order to increase the treatment capacity.

There are many advantages of using this kind of biological treatment like easy to operate, low energy required, less equipment maintenance, and better sludge

thickening. However, the effluent quality from fixed- film system are relatively poorer than suspended growth systems in terms of biochemical oxygen demand (BOD) and suspended solids (SS) (Metcalf & Eddy, 2003).

There are many factors that could affect the performance of a trickling filter like hydraulic and organic matter, media type and depth, retention time, and temperature (Metcalf & Eddy, 2003; Gray, 2004). Therefore, there is a relatively variation in the removal efficiency of some ECs like estrogens in trickling filter (Svenson, Allard & Ek, 2003).

2.5 Removal mechanisms.

Contaminants undergo to different mechanisms in order to be removed during their presence in the sewage. There are different types of removal mechanisms that might occur during the sewage treatment which are mainly biodegradation, sorption, photolysis and volatilization. The first two types represent the main mechanisms that have the potential to remove pollutant from the sewage water (Daughton & Ternes, 1999; Vader et al., 2000; Ternes et al., 2004; Onesios, Yu & Bouwer, 2009; Chang et al., 2011). Below is a brief definition of each mechanism.

2.5.1 Biodegradation.

Biodegradation is one of the most significant removal mechanisms for emerging contaminants. One of the main objectives of the biological treatment in the STW is to reduce the concentrations of the organic matter. This could be achieved by transform the biodegradable constituents into acceptable end product. Another object is also to remove the nutrients such as phosphorus and nitrogen. Therefore nitrification and denitrification are the main processes that occurred during the biological treatment. The biological conversion of ammonia to nitrate is known as nitrification which is technically occurred into two steps, where nitrosomonas bacteria oxidize the ammonia to nitrite (NO_2-N) and nitrobacter bacteria oxidize nitrite to nitrate (NO_3-N) (Langford & Lester, 2003). Many factors can affect the nitrification rate such as dissolved oxygen level, pH,

temperature, metals, acids and free ammonia (Metcalf & Eddy, 2003). However nitrification requires a high retention time, low food to microorganism ratio (F/M), and high mean cell residence time (MCRT) or sludge age. The integral step to get a suitable end product is denitirfication which is a biological reduction of nitrate to nitrogen gas (N_2) occurred and that achieved by a facultative heterotrophic bacteria that get the oxygen either from the dissolved oxygen or from the nitrate molecules. This process of denitrification only occurs when the oxygen level are very low (anoxic condition) and bacteria use nitrate-oxygen (Gray, 2004).

Biological degradation occurs in two phases, aerobic degradation and that happen in activated sludge, trickling filter, or anaerobiclly in the sewage system or at sludge digesters (Langford & Lester, 2003). Halling-Sorensen et al. (1998) reviewed pharmaceuticals as emerging contaminants in the environment; they stated that the contaminants in the sewage treatment works could undergo one or more of these three fates:

- Mineralization which the substance eventually mineralized to CO_2 and H_2O.
- The contaminant may transform to become more hydrophobic and a portion of the substance partitions onto the solid portion.
- Transformation to more hydrophilic persistent compounds, which are discharged to the receiving water body.

An intensive study by Joss et al. (2006) investigated the biodegradation of 35 different emerging contaminants like pharmaceuticals, personal care products and hormones. According to the degradation constant (K_{biol}) value, target compounds were categorised into three groups. The first group which included four compounds (hormones and pharmaceuticals), more than 90% was transformed when (K_{biol}>10/gss/d). Partial degradation was achieved when (0.1< K_{biol}<10/gss/d), and no degradation was observed for 17 compounds (<20% transformed) when K_{biol} <0.1/gss/d). In the wastewater treatment processes, biodegradation is the main removal mechanism to remove progestogens (Fan et al., 2011).

Emerging contaminants have a wide range of degradation rates and therefore their half lives are different. Some of these are biodegraded readily like 17β-estradiol (E2) (Joss et al., 2004), or some of them have a half-life for more than 100 days (Benotti &

Brownawell, 2009) , while other compounds are more resistant and not degraded. Although many studies have shown the importance of this process, however, little is known about the biodegradation of many emerging contaminants and the factors that can affect their degradation rate.

2.5.2 Sorption.

This is the second of the most significant process that control the fate of emerging contaminants in the aquatic environment. It is a physical process where the more lipophilic (hydrophobic) contaminants partition onto the settled sewage solids in the primary sedimentation tank or to biomass in the biological stage. Rogers (1996) divided the sorption potential for the compounds according to their octanol-water coefficient value to:

- Low sorption potential when Log K_{ow} less than (2.5).
- Medium sorption potential when log K_{ow} between (2.5-4.0).
- High sorption potential when log K_{ow} more than (4.0).

The ratio between the concentrations in the solid and liquid phases at equilibrium conditions which is solid-water distribution coefficient (K_d) can also give a good prediction for sorption potential and therefore have become common approach to determine the partitioning of ECs, particularly PPCPs (Suarez et al., 2008). According to Ternes et al. (2004), there are two sorption mechanisms:

- **Absorption** which is hydrophobic interaction of the aliphatic and aromatic groups of a compound with the lipophilic cell membrane of the micro-organisms and the lipid fractions of the sludge. Therefore, the sorption potential in this phenomenon depends on the octanol-water partition coefficient (K_{ow}).

- **Adsorption** which are electrostatic interactions of positively charged groups of chemicals with the negatively charged surfaces of the micro-organisms. In this case the dissociation coefficient (K_a) for the compounds plays an important role to determine the sorption potential for chemicals.

In general, therefore, these two coefficients (K_{ow}) and (K_a) are guide to predict the sorption potential of emerging contaminants particularly PPCPs.

2.5.3 Photolysis.

This mechanism may be responsible for complete or partial removal of some ECs. Yamamoto et al. (2009) showed that some emerging contaminants were relatively easily photodegraded, while other compounds were relatively stable against sun light. There is a wide range of half lives for emerging contaminants, some of these compounds were easily photolyzed and some were more slowly to degrade depending on conditions (Gomez et al., 2008). Therefore, many factors could affect the removal rate positively or negatively such as pH, oxygen concentration, structural properties of the compound, and occurrence of organic matter (Lin & Reinhard, 2005; Neamtu & Frimmel, 2006). In addition, the STW configuration plays a role in photolysis, for example large aeration tanks or polishing lagoons allow for some photolysis to occur (Metcalf & Eddy, 2003).

2.5.4 Volatilization.

The removal of volatile organic compounds from wastewater surfaces to the atmosphere is named volatilization (Metcalf & Eddy, 2003). According to the physiochemal properties of the compounds like Henry's law constant (H_c) and the octanol-water partition coefficient (K_{ow}), the volatilisation potential can be predicted (Rogers, 1996):

- The compound has a low volatilization potential when Hc $<1x10^{-4}$ and (H_c/K_{ow}) $<1x10^{-9}$.
- The compound has a high volatilization potential when Hc $>1x10^{-4}$ and (H_c/K_{ow}) $>1x10^{-9}$.

Therefore, this mechanism seems to be negligible for most of the emerging contaminants especially with compounds that have a hydrophilic behaviour. Pharmaceutical compounds and hormones have low H_c and consequently have low potential volatilization. Conversely, musk fragrances have H_c and H_c/K_{ow} higher than the limit mentioned by Rogers (1996), therefore, a high potential volatilization may be expected especially with the aerated biological treatment due to the abundance of air (Ternes et al., 2004).

2.6 Advanced treatment.

Many ECs are only partially removed in conventional wastewater treatment (Nakada et al., 2006; Kuster et al., 2008; Bolong et al., 2009; Gros et al., 2010; Jelic et al., 2011). Thus, one approach to improve the removal efficiency of the STWs for ECs is by adding a further technology as one of the alternative solutions. Advanced treatment is defined as the additional treatment required to remove suspended, colloidal and dissolved substances remaining after conventional secondary treatment (Metcalf & Eddy, 2003). There is a wide variety of treatment technologies have been applied and developed. Advanced treatment technologies can be classified according to their operation types or according to the residual compounds that are required to be removed and some of these are micro-and ultra-filtration, reverse osmosis, electro dialysis, adsorption, ion exchange, advanced oxidation processes and chemical precipitation (Metcalf & Eddy, 2003; Gray, 2004). An overview of some available technologies such as ozone (O_3), granular activated carbon (GAC), and chlorine dioxide (ClO_2) are described below.

2.6.1 Treatment by ozone (O_3).

In order to decrease the concentrations of the organic compounds in the sewage water, ozonation and particularly advanced oxidation processes (AOPs), therefore, are used (Metcalf & Eddy, 2003). Ozone is an extremely reactive oxidant; unstable and decomposed to oxygen quickly after generation, therefore it must be generated onsite. Ultraviolet (UV) irradiation of air or oxygen and corona discharge are used to generate O_3 (Gomes & Lester, 2003). Many studies have shown that ozonation is a very effective technique to decrease the concentrations or remove some hormones and PPCPs from wastewater (Ternes et al., 2003; Jasim et al., 2006; Esplugas et al., 2007; Zhang, Yamada & Tsuno, 2008; Gagnon et al., 2008; Giri et al., 2010;). For specific ECs like bisphenol-A, nonylphenol, and also 17-β estradiol, ozonation was found very useful to reduce the concentrations of these chemicals and their estrogenicity in secondary effluents (Kim et al., 2008). In addition, the removal efficiency of estrogens could be affected by pH, since the removal efficiency at pH 3 has been observed to be higher than that one at pH 11

(Maniero, Bila & Dezotti, 2008), although changing the pH of sewage may not be practical.

A study by Huber et al. (2005a), presented that ozone could oxidize more than 90% of PPCPs and estrogens in wastewater samples at a dose of 3.5 mg/l. Similar results were achieved by Hashimoto, Takahashi & Murakami (2006), showing that ozonation with 1mg/l could remove more than 90% of estrogens, and to below the limit of detection for estrogens when the ozone dose was increased to 3 mg/l. Another study by Ternes et al. (2003), found that in municipal STW in Germany that by applying 10-15mg/l of O_3 and 18 minutes as a contact time, it was able to decrease the concentration of pharmaceutical compounds, hormones and musk fragrances in his study to be below the limit of detection. However other compounds like iodinated contrast media were still detected with that level of ozone dose. There was a correlation between the dose of O_3 applied and the removal efficiency to remove some estrogenic compounds in bench scale for effluent from STW in Denmark (Hansen, Andersen & Ledin, 2010). Additionally to ozone dose, contact time also plays an important role in removing ECs as it can be seen in the study achieved to remove nonylphenol (NP) and bisphenol (BPA) from wastewater in Italy, hence 30% of NP were removed during 15 minutes at a dose of 8 mg/l and about 60% were removed at the same dose with 30 minutes (Bertanza et al., 2010). As a result, many factors could affect the performance of the ozonation such as ozone dose, contact time, the characteristics of the target compounds and the wastewater, pH, and temperature (Yargeau & Leclair, 2008).

These studies have shown an evidence of the role of ozone in removing wide spectrum of ECs in pilot and full scale experiments in both water and wastewater treatment plants. However, there are some drawbacks in using this technology such as the formation of disinfection by products (DBPs). Because of the O_3 dose applied in the wastewater treatment is higher than the dose in drinking water treatment, therefore, that will lead to generate more DBPs in wastewater effluent than in the drinking water (Wert et al., 2007). Many types of DBPs like aldehydes (formaldehyde, acetaldehyde, glyoaxl, and methyl glyoxal), various acids (acetic acid, formic acid, oxalic acid, and succinic acid) and ketones are formed during ozonation in the absence of bromide, while other DBPs may also generated in the presence of bromide such as bromated ion, bromoform, and brominated acetic acid and in some occasion hydrogen peroxide is also

produced (Metcalf & Eddy, 2003). A direct relationship was found between the dose of O_3 applied to remove pharmaceuticals and bromide ion concentration as DBPs (Kim & Tanaka, 2010).

In general, ozone is very powerful oxidant and ozonation is the most studied oxidation processes with best expectations; the performance of ozonation is a function of some parameters like dose, contact time, and properties of the substances present in the aqueous phase.

2.6.2 Treatment by chlorine dioxide (ClO_2).

Due to the variability in the performance of the conventional STWs in eliminating emerging contaminants (Koh et al., 2008), therefore there may be a need to improve the removal efficiency and that can be achieved by installing additional treatment. Chlorine dioxide is one of these additional technologies to enhance the effluent quality, it is powerful oxidant that oxidises the organic compounds presented in water, it unstable, therefore it has to be generated onsite (Metcalf & Eddy, 2003). Because of many pharmaceuticals have phenolic moieties and /or amino group in their structure, thus a large number of these pharmaceuticals are expected to be oxidised with ClO_2 (Huber et al., 2005b; Lee & von Gunten, 2010).

There are a few studies focused on the performance of ClO_2 in the aquatic environment, for example (Huber et al., 2005b; Filby et al., 2010). However the study by Huber et al (2005b) demonstrated that ClO_2 was effective to remove some classes of emerging contaminants like antibiotics and estrogens although it reacts more slowly than ozone with these compounds in water treatment samples. Another study showed that ClO_2 is highly effective to oxidize the estrogens (more than 90% removal) with the wastewater samples (Filby et al., 2010).

Although it is a very effective oxidant it also produces residuals, however, and one of the main disadvantages of using this technology is formation of disinfection by products (DBPs), the principle DBPs formed are chlorite (ClO_2^-) and chlorate (Cl_2O_2) which are potentially toxic (Metcalf & Eddy, 2003). Additionally, ClO_2 decompose in sunlight, is highly corrosive and moderately expensive (Metcalf & Eddy, 2003).

2.6.3 Treatment by granular activated carbon (GAC).

Activated carbon is another effective technology used to eliminate a wide spectrum of contaminants. Initially, activated carbon was used to remove taste and odour from drinking water. Nowadays it has been used in the sewage treatment and water works in order to remove the organic pollutants. There are two types of activated carbon that are usually used in the advanced treatment, powder activated carbon (PAC) and granular (GAC) are widely used. A number of studies have reported the performance of these processes to remove contaminants from the aqueous phase (Filby et al., 2010; Zhang & Zhou, 2005; Kim et al., 2007). Pojana, Fantinati & Marcomini (2011) found GAC was more efficient than other technologies (ozone treatment & sand filtration) in his study to remove pharmaceutical compounds in drinking water treatment plants. However, PAC were tested to remove EDCs like 17α-ethynylestradiol and 17β-estradiol from raw drinking water and achieved a range of removal efficiency from 31-99% based on many parameters like type of PAC, dose of PAC and the presence of organic materials in the water, it also paralleled to the physiochemical properties of the EDCs particularly log K_{ow}, where the high removal correspond to the high log K_{ow} (Yoon et al., 2003). In addition, in an extensive study by Westerhoff et al. (2005) to understand the fate of endocrine disrupters in simulated drinking water treatment process, showed that adding 5mg/l of PAC with four hours of contact time could remove between 10 to >98% of the compounds depending on their physicochemical properties, and it also demonstrated that a high dose (20 mg/l) of PAC at the same contact time (4 hours) enhanced the EDCs removal to more than 90%. Therefore the dose of the PAC and the contact time may play an important role in the removal efficiency.

The effectiveness of GAC as an adsorbent was shown in the removal of 17α-ethynylestradiol from wastewater by sand, GAC, and manganese oxide (de Rudder et al., 2004), where the results demonstrated removal efficiencies were 17.3% by sand, 99.8% by GAC, and 81.7% by manganese oxide. In another study (Ifelebuegu et al., 2006), the removal efficiency of 17α-ethynylestradiol from wastewater by coal based GAC was 98.6%, by coconut based GAC 99.3%, and by wood based GAC 96.4%. This shows that the source of GAC may have some influence on removal efficiency. Therefore, activated carbon is a successful technology to remove emerging contaminants from aqueous

phase (Chang et al., 2009); however, there are some disadvantages such as saturation of the GAC which then requires regeneration.

In general, among these advanced treatments, the main principle of GAC is to remove ECs from the aqueous phase depending on adsorption removal mechanism, while the function of O_3 and ClO_2 treatment is to oxidise or transform these ECs depending on the chemical oxidation, but not remove them and consequently that may create, transformation products or new compounds (DBPs) which may have an adverse effect on the environment. It also been noticed that a series or combination of these technologies provides a significant control to remove contaminants from wastewater (Gray, 2004).

2.7 Selection of emerging contaminants in this study.

There is a large number of emerging contaminants are released to the environment. However limited information is available about ECs in terms of their occurrence, fate, and toxicity associated to the humans and wildlife. Pharmaceuticals and personal care products (PPCPs) is a group of large number of compounds, and they are widely used in society. This group includes many different types such as hormones (natural and synthetic), drugs (antibiotic, anti-epileptics), and musk fragrances. There are adverse effects on the aquatic and wild life when these compounds are released to the environment due to their bioactivity (Jobling et al., 1998; Barcelo & Kettrup, 2004). There is large number of steroid hormones and each group of hormones consists of a range of compounds with different properties, therefore it is necessary to choose a particular group for this investigation. Progestogens (natural and synthetic) are one group of steroid hormones of emerging contaminants, represented by progesterone (natural hormone) and nine other synthetic compounds were chosen in this study.

Nowadays many chemicals are extensively used in the industry, some of these chemicals are persistent, and could have an impact on humans and wildlife by disrupt the endocrine system due to their accumulation and bioactivity. One of these chemicals group is benzotriazoles which are produced in large amount and therefore are classified as HPV according to USEPA (USEPA, 2011). Thus, benzotriazole and tolyltriazole were

also selected in this study. Because of the limited information available for some of the selected compound in these two groups, therefore, a brief description about selected groups in this study are described below.

2.7.1 Progestogens.

Many of the hormonally-active micropollutants known to date, and of most concern, are steroids, or steroid-like molecules. Some of these, in particular synthetic compounds such as ethynyl estradiol (EE2), used in the contraceptive pill, are relatively resistant to biodegradation, and cross the gills of fish very readily, where they interact with specific receptors and/or enzymes to disrupt the endocrinology of the fish. These chemicals can cause adverse effects even when present in the aquatic environment at extremely low (sub ng/l) concentrations. So, based on that knowledge, it is likely that there are other classes of hormonally-active synthetic steroids, besides EE2, in the aquatic environment that could cause adverse effects. One obvious group would be the synthetic progestogens, which are the other active ingredient of many contraceptives (at doses higher than EE2) and are also used in hormone replacement therapy (Bromley, de Vries & Farmer, 2004). Endogenous (natural) progesterones play important roles in reproduction in fish, controlling maturation of the gametes (sperm and oocytes) in both sexes. Synthetic progestogens target the progesterone receptor in women, and as fish also have this receptor, so it seems likely that synthetic progestogens will target these receptors. All of this information suggests that synthetic progestogens may have effects on fish reproduction and the key issue is really at what concentration do synthetic progestogens cause adverse effects, and how different is this concentration to those in the aquatic environment (Sumpter, 2008). There is some evidence that natural progesterone and synthetic progestogens are present in wastewaters in Europe (Vulliet et al., 2007) and that advanced treatment, such as ozonation will effectively remove progesterone (Snyder et al., 2006). However, there is no comparison of removal in such advanced treatment processes with what may be achieved in biological processes, and in particular, evidence that nitrifying bacteria enhance removal rates of other hormonally active compounds with similar structures, such as the steroid estrogens (Vader et al., 2000; Leusch et al., 2005; Ren et al., 2007).

Ten compounds were selected as a part of the compounds in this study to represent the progestogens group and the selection was based on the amounts prescribed in 2006 in the United Kingdom for clinical use. In total, 1,700kg of progestogens were used, while estrogens and androgens were only 500kg and 300kg respectively (Runnalls et al., 2010). These compounds are cyproterone acetate, drosprinone, dydrogesterone, medroxyprogesterone, medroxyprogesterone acetate, megestrel acetate, norethindrone, norgestrel, progesterone, and tibolone.

2.7.2 Benzotriazoles.

Benzotriazole (1H-benzotriazole) and tolytriazole (4 or 5-methyl-1H-benzotriazole) are commonly used domestic and industrial chemicals (Voutsa et al., 2006; Giger, Schaffner & Kohler, 2006). Despite this, surprisingly little is known about the environmental contamination of surface waters by these compounds or, indeed, about any chronic toxicity that may result from the exposure of aquatic organisms to this chemical. The compounds are anticorrosive agents, making up between 10-20% of formulated aircraft deicing and antifreeze fluids, of which 8,000,000 litres are used per year in Canada alone (Cancilla et al., 1997). For this reason, much of the attention on environmental concentrations has focused on runoff from airports, as well as on the water bodies and groundwater systems receiving such runoff (Cancilla, Martinez & Van Aggelen, 1998; Corsi et al., 2003). However, benzotriazole is also used in machine dishwashing detergents for silver protection (Ort et al., 2005). Within the UK, dishwasher ownership has risen from less than 5% in 1977 to over 33% in 2006 (Waterwise, 2011). Recent studies have demonstrated that benzotriazole exhibits antiestrogenic activity in *vitro*, although this was not observed in *vivo* (Harris et al., 2007), although a wider range of tests would be required to be certain that this was generally true. Nonetheless, benzotriazole has been described as ‘‘toxic to aquatic organisms and can cause long-term adverse effects in the aquatic environment’’ (Hem & Weideborg, 1999). These compounds are therefore considered to represent chemicals with a range of uses, likely to be present in relatively high concentrations which exhibit low removal and hence have been selected for study.

Chapter Three: Materials, Methods, and Field Study Description

Overview

To date, various methods have been developed and introduced to measure emerging contaminants. However, quantification of low concentrations is one of the main challenges in the analysis of these contaminants (Petrovic, Gonzalez & Barcelo, 2003). This chapter will describe the experimental methods used in this work including reagents, chemical standards, and analytical procedure. In addition, it will describe the field study sites including the STWs, River Erewash and catchment, and tap water survey area sampling procedure. Moreover, modelling the River Erewash and its catchment and Thames catchment area and the method to calculate the concentrations of benzotriazoles for wider area are also described in this chapter. Furthermore, the validation of the methods used to determine the benzotriazoles and progestogens is also described.

3.1 Experimental method.

3.1.1 Reagents and chemical

3.1.1.1 Analysed standards

The purity of all analyzed standards was greater than 98%. Cyproterone–acetate (CPA), megetsrel–acetate (MTA), medroxyprogesterone (MDP), and progesterone (PGT) were purchased from QMx, (Essex, UK). Norethindrone (NTD), drosprinone (DSP), dydrogesterone (DHG), norgestrel (NGL), tibolone (TBL), and medroxyprogesterone–acetate (MPA) were obtained from LGC (Exeter, UK). Benzotriazole (BT) and tolyltriazole (TT) were purchased from Sigma-Aldrich (Gillingham, UK). Table 3.1 shows the full description of each chemical.

3.1.1.2 Internal standards

Deuterated norethindrone and progesterone with a greater than 98% chemical purity were obtained from QMx and Aldrich respectively. These internal standards were used to quantify the progestogens group. The internal standard 5, 6-dimethylbenzotriazole was used to quantify the benzotriazoles group and was also purchased from Sigma-Aldrich (Gillingham, UK). Table 3.1 also summarizes the details of the internal standards.

3.1.1.3 Solvents and standard solutions

Organic solvents were of HPLC grade: methanol, dichloromethane and methyl tertiary butyl ether MTBE were purchased from Rathburn Chemicals (Walkerburn, UK). Nitric acid was purchased from Fisher (Loughborough, UK) and then diluted to 3%. Stock solutions were prepared in methanol. For solid phase extraction, Oasis HLB (500mg/6cm^3) cartridges were obtained from Waters Ltd. (Watford, UK). The other three types of cartridges which are C18CC, C18EC, and Easy were obtained from Thames Restek Ltd. (Saunderton, UK). Reagent grade MilliQ water (18.2MΩ) (Millipore, Watford, UK) was used for blanks, spiked and preparation of solutions. For HPLC analysis an Ascentis c18 (10cm x 2.1mm) 2.7µm column from (Sigma, Gillingham, UK) was used to separate progestogens, and a Synergi 4µ Hydro-RP80A column from (Phenomenex, Macclesfield, UK) was used for benzotriazole separation, and both were protected by using a C18 Guard column. The standards used in the method were prepared by further dilution of the stock standards with methanol/ water (50:50v/v).

3.1.2 Reference standards preparation.

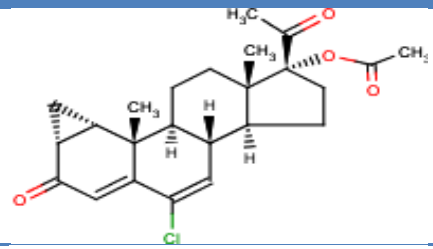
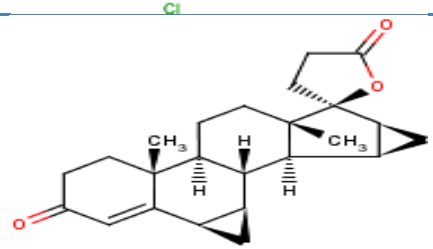
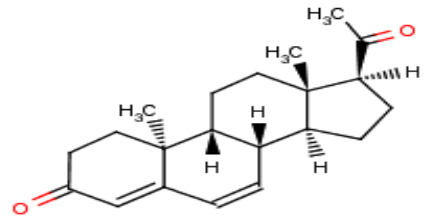
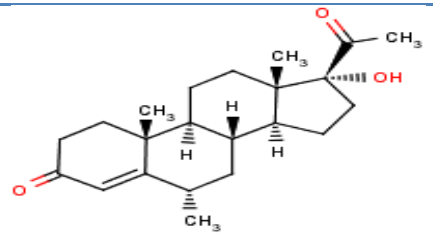
3.1.2.1 Progestogens

A standard stock solution of 0.1 g of each compound was dissolved into a 100 ml volumetric flask of HPLC grade methanol. Around 1000 µg/ml individual stock solution of each compound (deuterated and non-deuterated) were prepared in methanol. A series of mixed calibration standards containing all ten analytes in MeOH/H_2O (50/50), at a concentration range 0.5-500 ng/ml, and deuterated internal standards at 100 ng/ml were prepared from the stock solution.

3.1.2.2 Benzotriazoles

Standard solutions were prepared from individual stock solutions. About 0.1 g of each compound was dissolved into 100 ml of HPLC grade methanol in order to get 1,000 µg/ml. A series of mixed calibration standards containing analytes to produce six- point calibration at a concentration range 2.5 – 5,000 ng/ml and the internal standard (100 ng/ml) in methanol/water (50/50) were prepared, along with a solution of benzotriazole and tolyltriazole at 1000 ng/ml for use in spiking samples to evaluate method recovery and performance. A solution of internal standard was prepared in methanol at 10 µg/ml for addition to samples prior to extraction.

Table 3.1 Details of selected progestogens and benzotriazoles.

Compounds	abbr.	CAS	Formula	Log K_{ow} [a]	Systematic Name [b]	Structure [c]
Cyproterone acetate	CPA	427-51-0	$C_{24}H_{29}ClO_4$	3.87(±0.55)	H-Cyclopropa (1,2)pregna-1,4,6-triene-3,20-dione, 17-(acetyloxy)-6-chloro-1,2-dihydro-, (1beta,2beta)	
Drospirenone	DSP	67392-87-4	$C_{24}H_{30}O_3$	3.59(±0.68)	6-beta,7-beta;15-beta,16-beta-Dimethylene-3-oxo-17-alpha-pregn-4-ene-21,17-carbolactone	
Dydrogesterone	DHG	152-62-5	$C_{21}H_{28}O_2$	3.56(±0.31)	9-beta,10-alpha-Pregna-4,6-diene-3,20-dione	
Medroxyprogesterone	MDP	520-85-4	$C_{22}H_{32}O_3$	3.42(±0.13)	Pregn-4-ene-3,20-dione, 17-hydroxy-6-methyl-, (6-alpha)- (9CI)	

a: created from (ALOGPS 2.1)(VCCLAB, 2011) b, c: adapted from (U.S. NLM, 2010)

Table 3.1 Continued

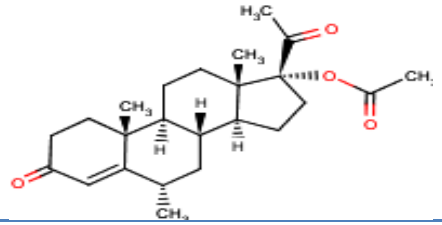
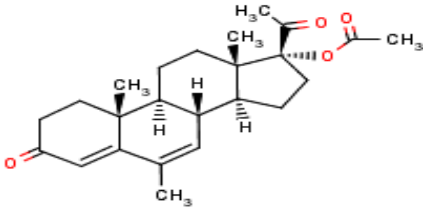
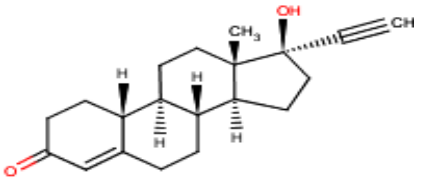
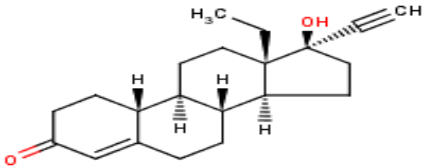
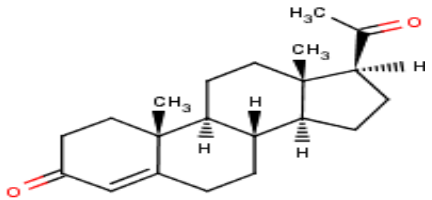
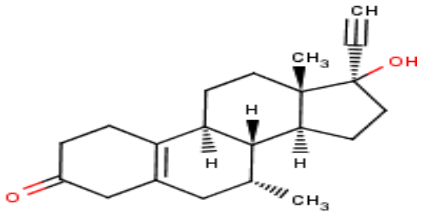
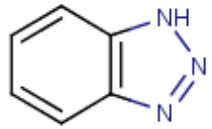
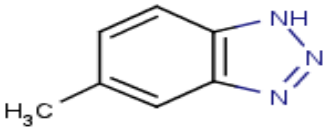
Compounds	abbr.	CAS	Formula	Log K_{ow} [a]	Systematic Name [b]	Structure [c]
Medroxyprogesterone acetate	MPA	71-58-9	$C_{24}H_{34}O_4$	3.63(±0.32)	Pregn-4-ene-3,20-dione, 17-hydroxy-6alpha-methyl-, acetate (8CI)	
Megestrel acetate	MTA	595-33-5	$C_{24}H_{32}O_4$	3.78(±0.37)	17-Hydroxy-6-methylpregna-4,6-diene-3,20-dione 17-acetate	
Norethindrone	NTD	68-22-4	$C_{20}H_{26}O_2$	3.29(±0.47)	19-Nor-17alpha-pregn-4-en-20-yn-3-one, 17-hydroxy- (8CI)	
*Norethindrone d6		52-78-8	$C_{20}H_{20}O_2D_6$		19-norethindrone-2,2,4,6,6,10-d6	
Norgestrel	NGL	6533-00-2	$C_{21}H_{28}O_2$	3.66(±0.44)	18,19-Dinor-17-alpha-pregn-4-en-20-yn-3-one, 13-ethyl-17-hydroxy-, (+-)-	

Table 3.1 Continued

Compounds	abbr.	CAS	Formula	Log K_{ow} [a]	Systematic Name [b]	Structure [c]
Progesterone	PGT	57-83-0	$C_{21}H_{30}O_2$	3.77(±0.19)	Pregn-4-ene-3,20-dione	
*Progesterone d9		15775-74-3	$C_{21}H_{21}O_2D_9$		progesterone-2,2,4,6,6,17α,21,21,21-d9	
Tibolone	TBL	5630-53-5	$C_{21}H_{28}O_2$	3.4(±0.62)	19-Norpregn-5(10)-en-20-yn-3-one, 17-hydroxy-7-methyl-, (7alpha,17alpha)-	
Benzotriazole	BT	95-14-7	$C_6H_5N_3$	1.24(±0.18)	1,2,3-Benzotriazole	
Tolyltriazole	TT	136-85-6	$C_7H_7N_3$	1.59(±0.23)	1H-Benzotriazole, 4(or 5)-methyl-	
* 5,6-dimethylbenzotriazole		4184-79-6	$C_8H_9N_3$		5,6-Dimethyl-1H-benzotriazole	

*: (internal standards) adapted from (ChemicalBook, 2008)

3.1.3 Instrumentation

Both LC/ESI$^+$ /MS/MS and LC/APCI$^+$/MS/MS system were used to perform the analyses. The system consisted of an HPLC (Hewlett Packard 1050) coupled to a Perkin Elmer Series 200 auto sampler and a PESciex API 365 triple quadruple mass spectrometer with atmospheric pressure chemical ionization (APCI) in positive mode in order to determine the concentration of progestogens. An electro spray ionization source (ESI) in positive mode was used to determine the concentrations of benzotriazoles. Acquisition and evaluation of data, in addition to instrument control, were carried out by Analyst software 1.4.2 (Applied Biosystems, Warrington, UK).

3.1.3.1 LC/MS/MS method

In order to get the maximum sensitivity for the chemicals of interest, the two modes ESI and APCI were selected and tuned by experiments. A 250µl syringe was installed on syringe pump. The flow rate for each individual standard that was infused by the syringe pump was 50 µl/min. The positive mode produced more parent ions than the negative mode and was then optimized in order to get higher sensitivity. The main parameters for the instrument to improve the signal intensity for chemicals were optimized as below:

- Temperature (T) = 300 °C.
- Nebulizer gas (NEB) = 6.
- Curtain gas (CUR) = 8.
- Collision gas pressure (CAD) = 4.
- Focussing potential (FP) = 115 V.
- Declustering potential (DP) = 15 V.
- Entrance potential (EP) = 4 V.

An HPLC method to separate the compounds was then developed. Table 3.2 shows the retention time and the masses for each compound.

Table 3.2 Retention time and masses for progestogens.

	Compound	Retention time (min.)	M1	M2
1	Cyproterone acetate	29.22	417.2	313.2
2	Drosprinone	27.24	367.2	97.1
3	Dydrogesterone	29.11	313.2	145.2
4	Medroxyprogesterone	29.34	345.1	123.1
5	Medroxyprogesterone acetate	29.84	387.1	123.1
6	Megestrel acetate	29.57	385.3	267.2
7	Norethindrone	26.70	299.2	109.2
8	Norethindrone d6	26.70	305.4	113.3
9	Norgestrel	28.27	313.4	109.2
10	Progesterone	30.05	315.1	97.1
11	Progesterone d9	30.05	324.3	100.2
12	Tibolone	28.25	313.2	145.2

For benzotriazoles, multiple reaction monitoring (MRM) was used with parent and fragment ions shown in Table 3.3. Two fragment ions were used for the determinants, one for quantification and the second for confirmation. Tolyltriazole (4(or 5)-methyl-1H-benzotriazole) consists of two isomers, the 4- and 5-methyl, however these were not separated by the chromatography, and are reported together as tolyltriazole

Table 3.3 Retention time and masses for benzotriazoles.

	Compound	Retention time (min)	[M+H+]	Quantification ion	Confirmation ion
1	**Benzotriazole**	2.03	120.1	65.1	92.2
2	**Tolyltriazole**	3.17	134.2	79.1	95.1
3	**5,6-dimethylbenzotriazole**	5.04	148.2	93.1	*

* Single ion used for the internal standard

3.1.3.2 Chromatography.

Progestogens

The concentration of analytes were determined using LC ($APCI^+$)/MS/MS at a flow rate of 0.2 ml/min. Analytes were separated using an Ascentis c18 (10cm x 2.1mm) 2.7µm column, (Sigma, Gillingham, UK). The total run time was 45 minutes, with data acquisition over a methanol/water (+0.4% formic acid) gradient of: 5% MeOH for 2 minutes, linear gradient to 80% MeOH over 25 minutes and held at 80% for 5 minutes, followed by a column wash for 1 minute and equilibration back to starting conditions for 13 minutes for a 46 minute cycle time as shown in Figure 3.1. The mass spectrometer was operated in a positive APCI mode using multiple reactions monitoring (MRM).

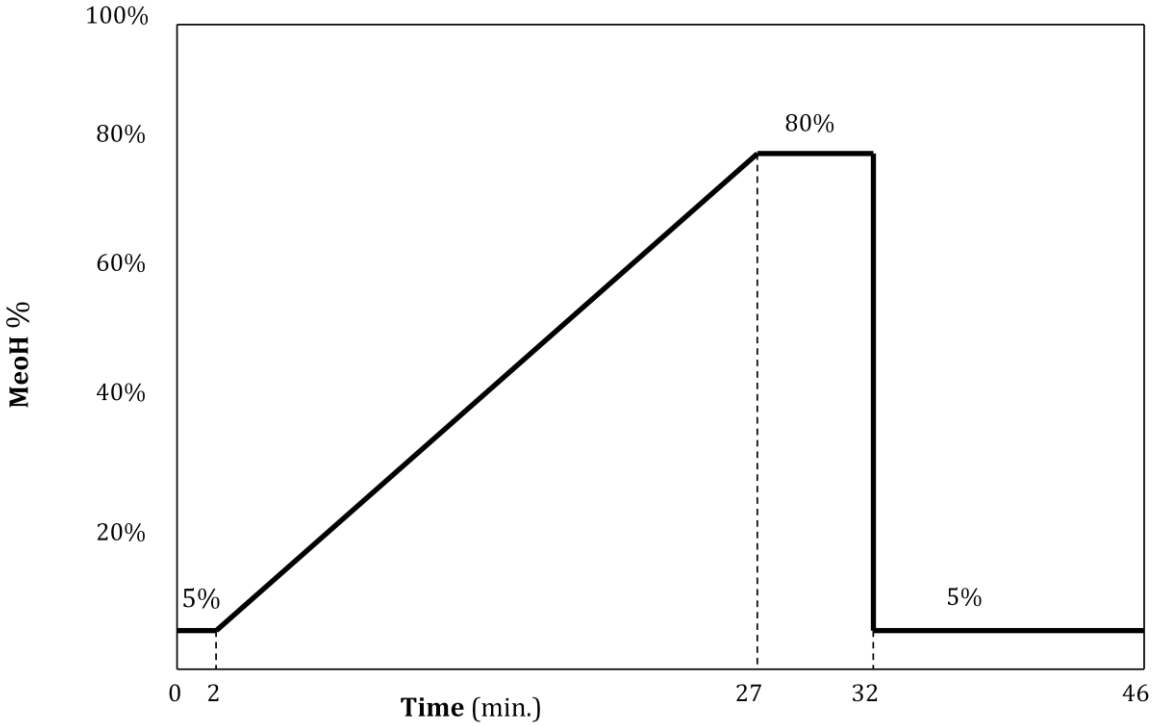


Figure 3.1 Gradient program of methanol for progestogens.

Benzotriazoles

Benzotriazole and tolyltriazole were quantified using a LC (ESI^+)/MS/MS system. Analytes were separated on a 7 cm x 2 mm i.d. Synergi 4µ Hydro-RP80A column (Phenomenex, Macclesfield, UK) at a flow rate of 0.2 ml/min with an initial mobile phase of 55% water and 45% methanol (both solvents with 0.4% formic acid). This was increased to 70% methanol / formic acid over four minutes, and held for a further four minutes before returning to initial conditions for the next sample as shown in the Figure

3.2. Instrument control, integration and quantification of samples were undertaken using the Analyst software.

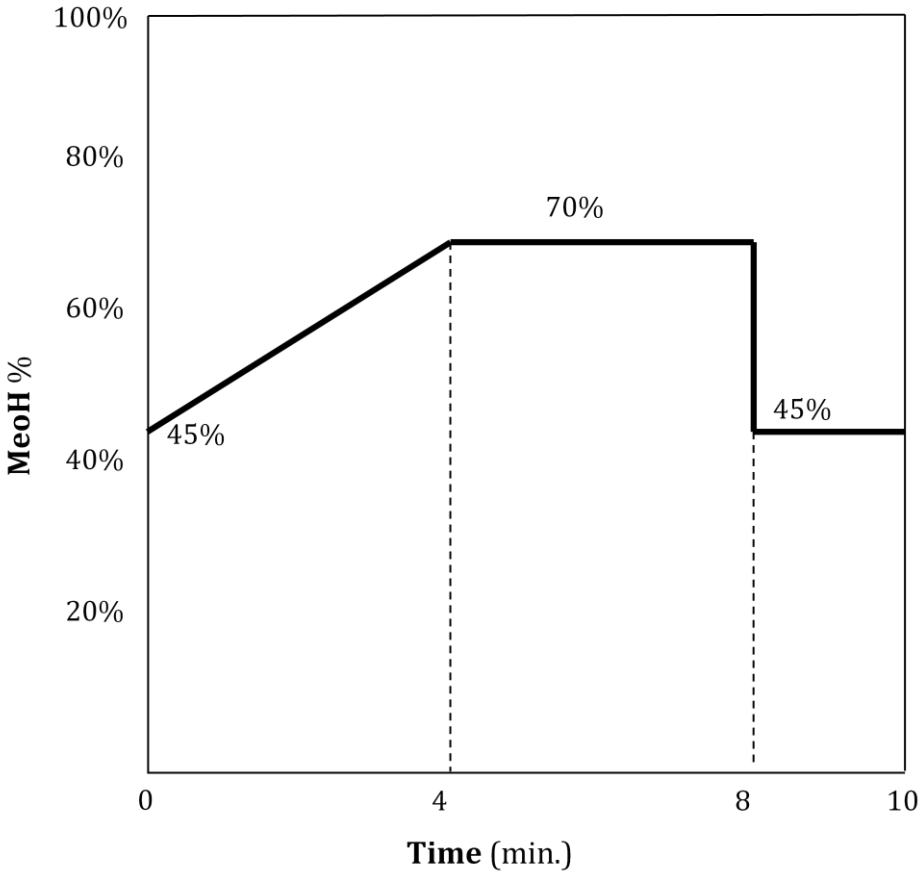


Figure 3.2 Gradient program of methanol for benzotriazoles.

3.1.4 Development of an analytical method for progestogens

One of the main important parts in the evaluating the occurrence of trace chemicals in the environment is the analytical method. However, much effort was involved in developing and validating these techniques. Examine different cartridges to obtain the best recovery was one of the attempts in order to develop the method. Trying to optimize the source parameters for the mass spectrometer was also one of the attempts. In addition to that, using gel permeation chromatography (GPC) cleanup was also one of the attempts to improve the analytical method by separating the big fragments from the fragments of the chemicals of interest. The purpose of all these efforts was to increase the accuracy for detection and determination the concentrations of these chemicals in the aquatic samples.

3.1.4.1 Cartridges types

During the period of developing the method, four different types of cartridges were examined. The purpose of this test was to assess the efficiency of these cartridges via finding the recovery for these compounds from these cartridges. Therefore, duplicate samples (each sample 1l of pure water 18 MΩ) were spiked with 100 ng, extracted and then analysed by LC/MS/MS for each type of cartridges. The results showed that C18cc provided the largest set of the mean recovery compared to the other cartridges, however HLB cartridges were chosen due to two reasons: firstly, there was no significant difference between C18cc and HLB mean recovery, and secondly Vulliet et al. (2007), recommended to use HLB cartridges in the analytical method for determination selected steroid sex hormones in wastewater. Table 3.4 shows the recovery percentage with each type of cartridges.

Table 3.4 Mean recovery percentage of progestogens from different cartridges.

	Cartridge type			
Compound	C18EC	HLB	C18CC	Easy
Drospirenone	53	72	78	35
Dydrogesterone	51	59	69	38
Medroxyprogesterone	54	79	78	51
Medroxyprogesterone acetate	41	62	61	33
Norethindrone	98	88	92	93
Norgestrel	53	87	81	49
Progesterone	42	62	64	38
Tibolone	60	80	83	51

3.1.4.2 Slough data and validation

Base line data

The first sampling was undertaken to detect these compounds from wastewater sample from Slough STW to the west of London was on 13 of September 2008. The purpose of this sampling was to investigate the presence or the background concentrations of the 12 emerging contaminants (10 progestogens, and two benzotriazoles) in wastewater sample associated with our study and also to inform us of how much we have to spike at appropriate levels for the next stage. Electrospray Ionization in positive (ESI^+) mode with LC/MS/MS was used to analyse these compounds. At that time, triplicate samples

(1 litre each) was taken from a bulk samples of 5 l collected from each crude and final effluent. The extraction of samples by SPE was within 5 hours of sampling. Concentrations of these compounds were in low ng/l for progestogens and two orders of magnitude higher for BT and TT as shown in Table 3.5.

Table 3.5 Concentrations of target compounds at Slough sewage treatment work in (ng/l) (1st sampling).

	Compounds											
Sample name	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL	BT	TT
Crude 1	39.6	13.2	18.8	22.2	11.0	7.2	15.5	27.4	30.2	38.8	2500	910
Crude 2	38.2	11.9	11.8	19.2	10.3	4.9	10.5	17.8	30.2	31.6	3180	1270
Crude 3	18.2	14.4	10.8	24.2	10.0	5.7	15.3	29.8	39.6	46.4	3330	1360
Mean	32.0	13.2	13.8	21.9	10.4	5.9	13.8	25.0	33.3	38.9	3000	1180
RSD %	37	9	32	11	5	20	21	25	16	19	15	20
FE 1	5.4	6.5	6.0	10.4	10.6	6.5	5.4	14.4	6.8	22.8	3450	2470
FE 2	5.6	6.6	9.0	6.0	8.5	1.4	4.1	12.6	5.6	23.0	3750	2530
FE 3	9.2	4.8	5.4	5.1	10.2	2.8	2.9	45.0	6.3	69.8	3360	2440
Mean	6.7	6.0	6.8	7.1	9.7	3.6	4.1	24.0	6.2	38.5	3530	2480
RSD %	32	17	28	40	11	73	30	76	10	70	6	2

All ten compounds in this study were found in the crude and final effluent samples. In addition, initial indications were that there was limited removal for some of these chemicals during the conventional process.

Method evaluation

The next sampling was on 23 of September from the STW that sampled previously. The purpose of this sampling was to validate the method by spiking at relevant concentration to detect these pollutants in wastewater sample. Electrospray ionization (ESI^+) in positive mode with LC/MS/MS was used to analyse these compounds.

For progestogens, samples were collected from crude and final effluent in 10 l and 20 l containers respectively. The crude samples were divided into ten samples, each sample was 0.5l, the first five samples were not spiked (no progestogens added), while the second five samples were spiked with 20 ng of progestogens to validate the method and to find out the recovery percent of the samples. For the final effluent, samples were categorised into three groups and each group consists of five samples with 1l each and

measured as unspike samples, spike samples with 5 ng, and spike with 10 ng of progestogens.

Benzotriazoles had the same procedure as progestogens except the volume of the sample was 200 ml and spiked with 500 ng of benzotriazoles in both crude and final effluent samples to determine the method validity and the recovery percentage of the samples.

From the results in Table 3.6, it is clear to see that all compounds were present also in the influent and final effluent samples of the wastewater; also the recovery for the high spike volume in the final effluent samples was more accurate than in low spike volume.

Table 3.6 Concentrations of target compounds at Slough sewage treatment work in (ng/l) (2nd sampling).

		Compounds											
		CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL	BT	TT
Crude sewage unspiked: sample volume is 0.5l for progestogens and 0.2l for benzotriazoles	Mean (ng/ sample Vol.)	14.4	5.2	12.6	5.3	4.5	4.5	4.1	7.7	10.8	11.9	553	667
	% RSD	40	23	20	32	10	12	24	26	8	20	1	2
Crude spiked at 20 ng of progestogens samples and 500ng of benzotriazoles samples	Mean (ng/ sample Vol.)	39.5	11.0	7.4	32.0	13.9	11.2	12.8	25.8	29.4	34.1	964	1092
	% RSD	36	60	51	62	31	28	8	65	14	43	4	6
	% Mean recovery	126	29	-26	129	47	33	42	112	93	82	84	86
Final effluent unspiked: sample volume is 1l for progestogens and 0.2l for benzotriazoles	Mean (ng/ sample Vol.)	8.1	4.7	8.3	6.8	4.3	4.0	1.7	5.8	4.2	9.7	314	487
	% RSD	68	44	28	119	96	90	71	90	106	78	4	6
Final effluent spiked at 5 ng for progestogens samples	Mean (ng/ sample Vol.)	7.9	6.0	5.9	13.2	9.4	9.8	5.7	12.4	7.6	18.1		
	% RSD	62	10	52	13	18	15	21	62	23	61		
	% Mean recovery	-4	27	-49	125	104	114	79	163	68	124		
Final effluent spiked at 10 ng for progestogens samples and 500 for benzotriazoles samples	Mean (ng/ sample Vol.)	10.0	11.6	12.6	18.0	11.3	7.7	10.4	13.8	11.1	23.4	921	1284
	% RSD	27	21	32	33	22	23	40	41	35	36	6	5
	% Mean recovery	20	69	43	109	71	36	86	100	69	101	101	118

3.1.4.3 Problem with and solution to progestogens analysis

Different limitations had occurred during development the method for analysing progestogens, especially with GPC, since the first sample set of data were very good compared to the later data set. Many problems had occurred with GPC such as the auto sampler did not communicate with the collector, therefore, that led to loss many samples. Changing the auto sampler to new one had made many problems specifically with the pressure. All these factors led to investigating a change from using the ESI^+ to $APCI^+$. Vanderford et al. (2003) used both ESI^+ and $APCI^+$ in analysing progesterone; therefore, the decision to investigate the method performance by using $APCI^+$ was made. To validate the method, a bulk samples of 15 litres was collected from Cranfield University STW in order to validate the method and check the method performance, therefore, the bulk sample was divided into three groups: the first group consisted of five samples each one was one litre as unspiked samples, five samples each one was 1l were spiked at 5ng represented the second group, and third group of five sample also each sample was one litre with 10ng spiked. Table 3.7 shows the method validation by using $APCI^+$ at two different levels of spiking. The mean recovery in both levels was very good (72%-129%), therefore, $APCI^+$ source was used to analyse the progestogens in this study, because it is less sensitive to suppression so it does not require cleanup.

Table 3.7 $APCI^+$ method validation for target compounds.

	Compound									
	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
Final effluent unspiked (n=5)										
Mean (ng/ l)	1.4	1.7	0.5	0.4	3.3	0.6	0.5	3.6	0.7	0.6
%RSD	34	29	55	67	50	43	53	20	13	70
Final effluent spiked at 5 ng/l (n=5)										
Mean (ng/ l)	6.1	8.1	4.8	5.6	6.9	6.9	4.5	8.0	4.6	5.4
% Mean recovery	95	129	84	100	72	125	78	110	78	72
%RSD	9	45	12	7	37	10	15	36	11	16
Final effluent spiked at 10 ng/l (n=5)										
Mean (ng/ l)	13.1	12.2	11.0	11.0	13.4	11.3	9.0	13.0	8.8	11.3
% Mean recovery	117	106	105	103	101	105	83	116	80	79
%RSD	11	12	34	7	36	9	4	26	9	27

3.1.5 Final analytical methods

3.1.5.1 Analytical method for progestogens

The final analytical methods of progestogens followed the method of Vanderford et al. (2003). Wastewater samples were filtered by GF/C (0.45µ) (Whatman, Maidstone, UK) filters. Internal standard (0.2 ml of 100 ng/ml) was then added to the sample. Cartridges were preconditioned with 5ml of methanol, followed by 5ml of reagent grade 18MΩ water before loading the sample with a flow rate between 5-10 ml/min using a vacuum manifold. After extraction, the cartridges were rinsed with 5ml of reagent water and dried with a stream of air for about 3hours. Cartridges were then eluted with 5 ml (90% of MTBE, 10% MeOH) followed by 5 ml of MeOH. These elutes were collected in 15 ml polypropylene tubes, and were subsequently evaporated on a miVac concentrator (Genevac, USA) at 35 °C on the [-OH] programme setting for 65 minutes and then evaporated to dryness with nitrogen. Samples were reconstituted with 0.2 ml of MeOH / H_2O (50:50 v/v) into 800µl small vials and transferred to auto an sampler prior to quantification by LC(APCI$^+$)/MS/MS. Figure 3.3 shows the systematic arrow diagram for the final analytical method for progestogens.

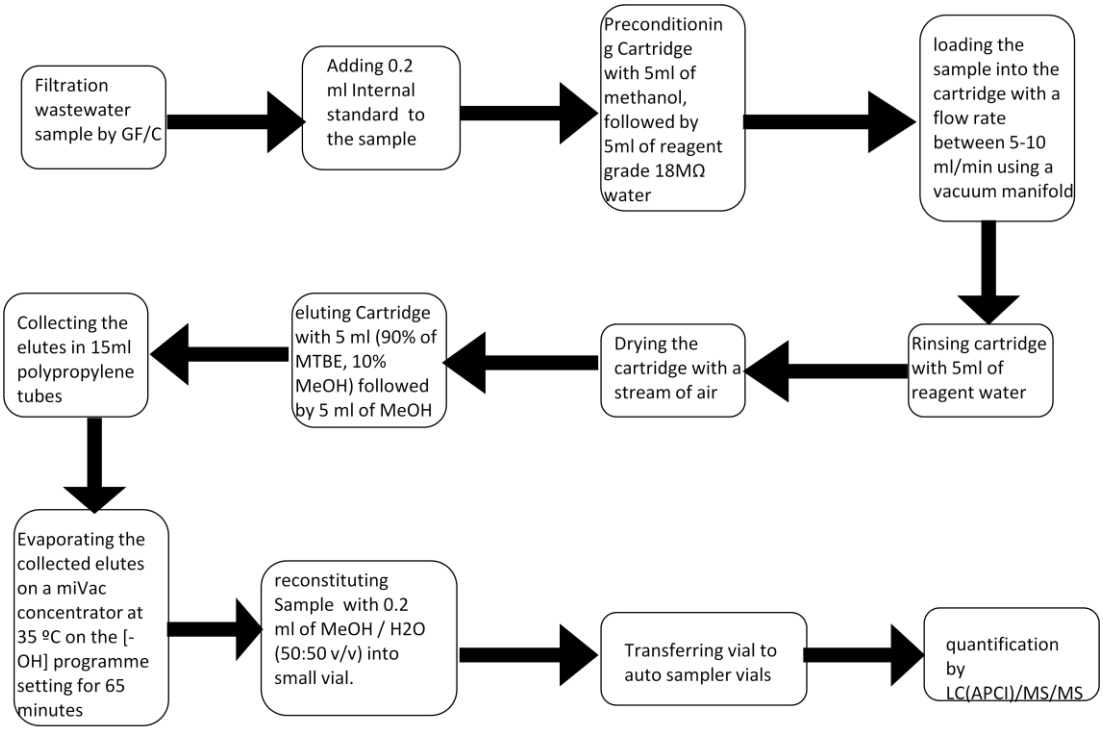


Figure 3.3 Flow chart for the final analytical method for progestogens.

The ion chromatography for the progestogens resulted from following this analytical method for one sample from final effluent is shown in Figure 3.4. The numbers in the graph refer to the each single compound as they listed in Table3.2.

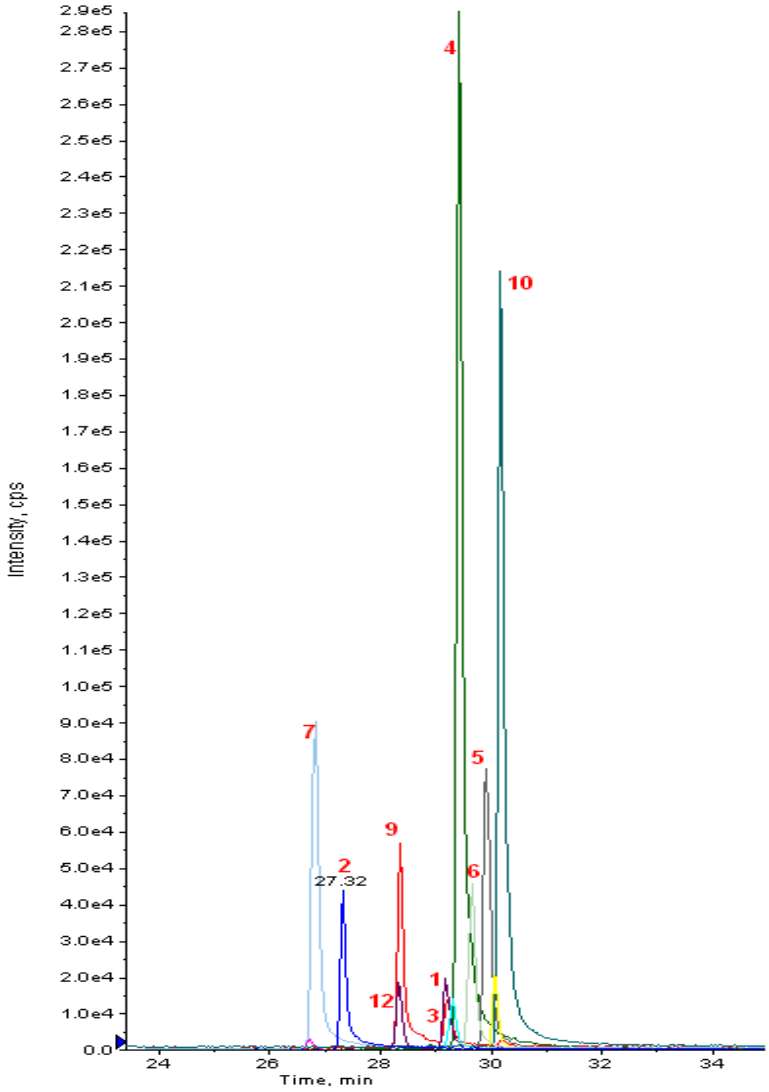


Figure 3.4 Ion chromatography for progestogens

3.1.5.2 Analytical methods for benzotriazoles

Samples of wastewaters, river waters and tap waters were enriched by SPE before quantification. Figure 3.5 shows the flow chart for the final analytical method for analysing benzotriazoles, which was based on that of Vousta et al. (2006). Before extraction, samples were filtered by GF/C filter paper, acidified to pH <3 with diluted nitric acid (3 %) (Fisher, Loughborough, UK) and internal standard (0.2 ml or 1 ml of 100 ng/ml in methanol, depending on the final volume that samples would be made up

to) was added. The Oasis cartridges were prepared by washing with 5 ml of methanol followed by 5 ml of reagent grade water. Samples were loaded onto the cartridges using a vacuum manifold at a flow rate of 5 to 10 ml/min. For wastewaters and river waters, a sample volume of 200 ml was used and 1000 ml for tap waters. After the extraction, the cartridges were rinsed with 5 ml of reagent water and dried with stream of air. Cartridges were then eluted with 5 ml of dichloromethane (DCM) with 3% methanol. These elutes were subsequently concentrated on a miVac concentrator (Genevac, Ipswich, UK) at 35 °C on the [-OH] programme setting for 45 minutes. They were then evaporated to dryness with a stream of nitrogen and re-dissolved in 0.2 ml or 1ml of methanol / water (50:50), depending on the required concentration factor needed, prior to quantification by LC (ESI^+)/MS/MS.

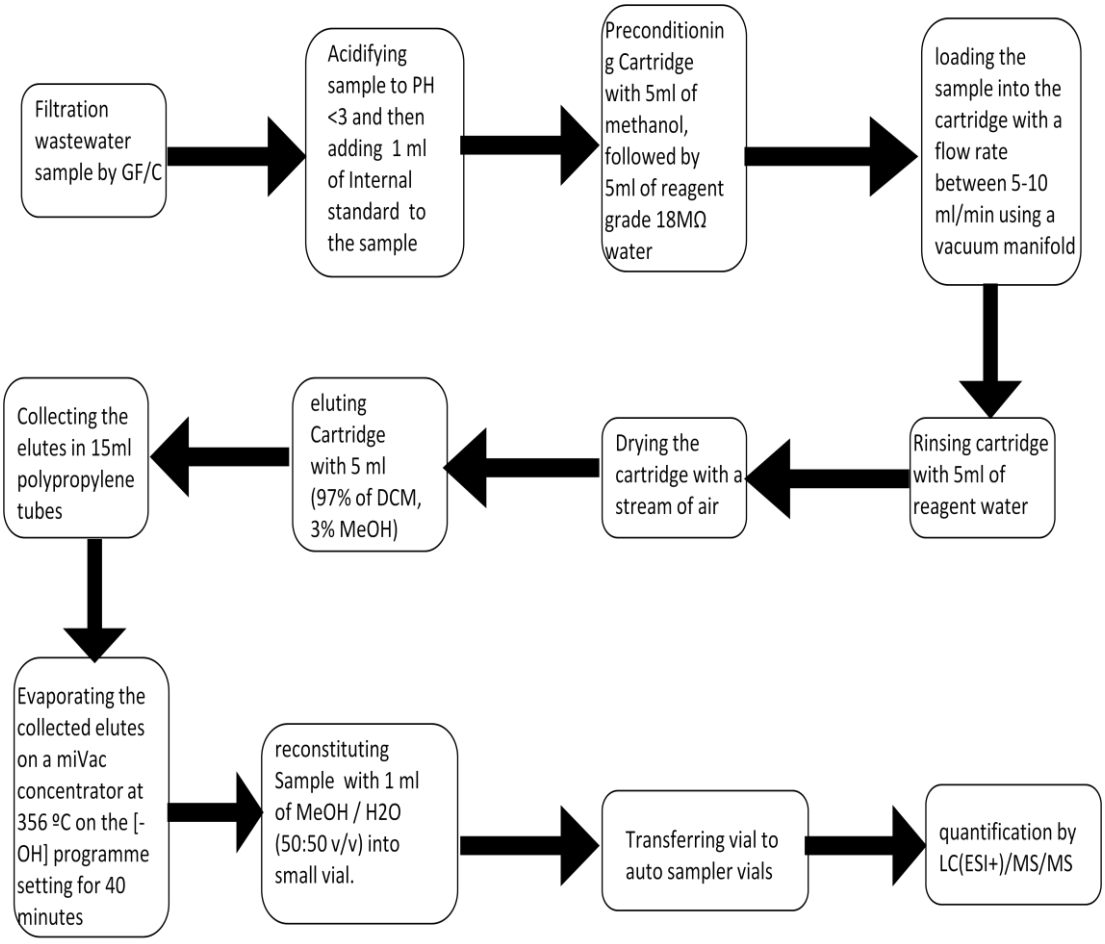


Figure 3.5 Flow chart for the final analytical method for benzotriazoles.

The total ion chromatogram (TIC) for the benzotriazoles resulted from following this analytical method for a typical sample is shown in Figure 3.6.

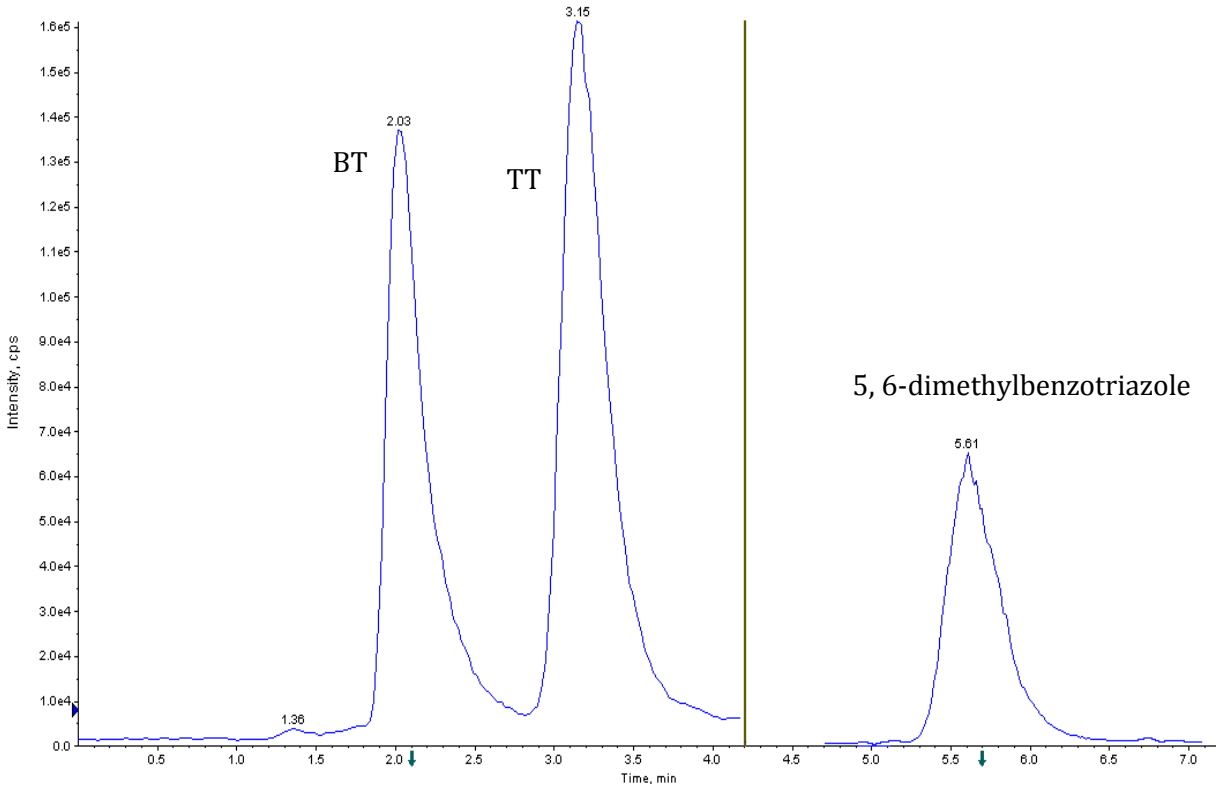


Figure 3.6 Ion chromatography for benzotriazoles.

3.1.6 Quality control

3.1.6.1 Limit of detection (LOD) and limit of quantification (LOQ)

The minimum concentration or amount of analyte that can be reliably distinguished is defined as the limit of detection (Taverniers, De Loose & Van Bockstaele, 2004). While the limits of quantification can be defined as the minimum concentration can be quantified in the sample which has the compound of interest. In this study the detection limit was calculated at least three times the signal to noise and limit of quantification was taken as 2 times the LOD according to (Taverniers, De Loose & Van Bockstaele, 2004). Table 3.8 shows the limit of quantification for the compounds of interest.

Table 3.8 Limit of quantification for each chemical in ng/l.

	Compounds									
Progestogens Sample	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
Final Effluent & Post Advanced treatment (1l)	0.2	1	1.6	0.4	0.4	0.4	1	2	0.4	1.6
Settled Sewage (0.5l)	0.4	2	3.2	0.8	0.8	0.8	2	4	0.8	3.2
Benzotriazoles Sample	**BT**	**TT**								
Tap Water (1l)	0.5	0.5								
Settled Sewage, Final Effluent & Rivers (0.2l)	2.5	2.5								
Post advanced treatment (0.1l)	5	5								

3.1.6.2 Blanks

Progestogens

To find out whether there was any contamination had occurred during sample preparation, extraction and analysis, at least one sample of 1l of non-spiked MilliQ water was analysed in each batch. Then, the value of the contamination was subtracted from the samples. For progestogens, Table 3.9 represents the data of the blank samples for the chemicals of interest and these samples were achieved during collecting and analysing the samples from STWs and during the degradation study. It is obvious to see that the blanks in the degradation study are better than the blanks that were done on site and that probably because of the quality of the MilliQ water in the both locations and also possibly more difficult to ensure a good washing for glassware on-site.

Benzotriazoles

Table 3.10 represents the blank data for benzotriazoles of all sample batches that achieved during the study which means from sewage treatment works (Slough, Hallam Fields and Newthorpe STWs), the Erewash River, the degradation study and the tap water in addition to some laboratory samples.

Table 3.9 Blanks data for progestogens.

Sample Name		CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
Slough STW	Blank 1	< 0.2	< 1	4.2	0.7	< 0.4	< 0.4	< 1	< 2	< 0.4	2.6
	Blank 2	< 0.2	< 1	4.3	0.9	< 0.4	0.3	< 1	< 2	< 0.4	< 1.6
Hallam Fields & Newthorpe STWs	Blank1	1.3	< 1	3.3	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
	Blank2	1.4	< 1	4.8	1.2	1.3	< 0.4	< 1	< 2	< 0.4	1.6
	Blank3	5.3	7.1	20.4	6.7	5.2	2.1	2.5	6.2	5.7	9.7
	Blank4	1.7	4.9	8.0	4.1	2.8	1.3	1.8	2.8	3.2	3.6
	Mean	2.4	3.1	9.1	3.0	2.3	0.9	1.2	2.5	2.3	3.8
	RSD %	80	110	85	98	94	109	94	108	119	108
Degradation Study	Blank1	2.3	4.3	1.8	0.6	5.7	< 0.4	< 1	3.3	< 0.4	< 1.6
	Blank2	2.2	4.1	3.8	2.8	2.0	2.9	1.8	8.0	0.8	6.2
	Blank3	1.0	4.0	1.8	0.4	2.8	0.3	< 1	1.6	0.5	2.1
	Blank4	< 0.2	< 1	< 1.6	1.3	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
	Mean	1.4	3.2	2.0	1.3	2.7	0.8	0.7	3.4	0.4	2.3
	RSD %	75	62	72	83	88	163	111	99	90	120

Table 3.10 Blanks data for benzotriazoles.

Sample Name		BT ng/l	TT ng/l
Slough STW	Blank (1)	< 0.5	< 0.5
	Blank (2)	< 0.5	< 0.5
Hallam Fields & Newthorpe STWs	Blank (1)	< 0.5	4.0
	Blank (2)	< 0.5	< 0.5
	Blank (3)	< 0.5	< 0.5
	Blank (4)	< 0.5	< 0.5
Erewash River	Blank (1)	< 0.5	< 0.5
	Blank (2)	1.0	2.0
Degradation Study	Blank (1)	< 0.5	< 0.5
	Blank (2)	2.4	3.0
Tap water	Blank (1)	1.5	0.8
	Blank (2)	0.9	0.6
	Blank (3)	< 0.5	< 0.5
	Blank (4)	1.1	< 0.5
	Blank (5)	< 0.5	< 0.5
	Blank (6)	1.6	< 0.5
	Blank (7)	1.7	0.6
	Blank (8)	1.8	0.7
Laboratory Test	Blank (1)	< 0.5	< 0.5
	Blank (2)	0.9	< 0.5
Mean		0.8	0.8

3.1.6.3 Recoveries.

Progestogens

One litre of MilliQ water and wastewater samples were spiked at different concentrations with each batch in order to determine the method performance. Recovery wastewater samples were selected from different stages through the whole processes, while MilliQ water samples was spiked. Blank samples were also subtracted from the spiked sample. Spiked samples were extracted at the same time of each batch. Table 3.11 shows the recovery percentage of these chemicals from spiked MilliQ water in the two different stages of the study. The mean recovery from the degradation study was better than that achieved on site, and the reasons for that is either the quality of the spiked water or the accuracy of the work in the field is less than that in the laboratory.

Another recovery test for progestogens was achieved with the final effluent in two occasions and in two different locations. A big container of 20l of final effluent samples were taken from Cranfield University sewage treatment work at the same day, each sample was 1l; five samples were unspiked, five samples were spiked at 5ng, and another five samples were spiked at 10ng in order to find the recovery percentage of the method. In addition, eight samples were collected from GAC effluent (final effluent) at Hallam Fields STW, each sample was 1l and four of these samples were spiked at 5ng and compared with the collected unspiked GAC samples in order to validate the method, the results are listed in the Table 3.12.

Table 3.11 Recovery data for progestogens from MilliQ water.

Sample Name		CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
Hallam Fields & Newthorpe STWs	Recovery (1)	126	151	108	162	88	117	94	130	92	96
	Recovery (2)	116	129	286	167	173	107	125	211	141	182
	Recovery (3)	75	-34	68	0	4	56	58	8	-22	-34
	Recovery (4)	125	120	95	79	54	107	57	111	7	66
	Mean	111	91	139	102	80	97	83	115	54	77
	RSD %	22	93	71	77	89	29	39	72	138	115
Degradation Study	Recovery (1)	104	105	71	102	107	102	77	107	71	84
	Recovery (2)	98	108	72	97	106	97	101	119	80	97
	Recovery (3)	101	102	83	98	100	99	104	109	92	84
	Recovery (4)	101	103	84	106	95	104	101	118	91	95
	Mean	101	104	78	101	102	100	96	114	83	90
	RSD %	2	3	9	4	5	3	13	5	12	8

Table 3.12 Recovery data for progestogens from final effluent and GAC effluent.

		CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
Cranfield University STW	**Final effluent (n=5) spiked at 5 ng/l**										
	% Mean recovery	95	129	84	100	72	125	78	110	78	72
	%RSD	9	45	12	7	37	10	15	36	11	16
	Final effluent (n=5) spiked at 10 ng /l										
	% Mean recovery	117	106	105	103	101	105	84	116	80	79
	%RSD	11	12	34	7	36	9	4	26	9	27
Hallam Fields STW	**GAC Effluent (n=4) spiked at 5 ng/ l**										
	% Mean recovery	95	96	110	70	95	100	78	72	49	57
	%RSD	17	21	30	30	56	21	18	34	41	58

Benzotriazoles

Table 3.13 represents the recovery data for benzotriazole of all sample batches that were undertaken during the study. Each sample was one litre of MilliQ water and spiked in different levels. Samples were taken from STWs (Hallam Fields and Newthorpe), Erewash River, tap water, and the degradation study in addition to some initial laboratory studies to validate methods. The recoveries of the samples were very good; the mean recovery was 95% and 101% for BT and TT respectively in the glass bottles. In plastic bottles, the recovery for TT was the same as in the glass bottles, but the recovery of BT was 91%.

Table 3.13 Recovery data for benzotriazoles.

Sample Name		Lab Recovery %	
		BT	TT
Hallam Fields & Newthorpe STWs	Lab Recovery (1)	134	155
	Lab Recovery (2)	102	120
	Lab Recovery (3)	113	111
	Lab Recovery (4)	121	122
Erewash River	Lab Recovery (1)	109	107
	Lab Recovery (2)	93	95
Lab Test	Lab Recovery (1)	91	94
	Lab Recovery (2)	79	74
	Lab Recovery (3)	81	86
	Lab Recovery (4)	79	85
	Lab Recovery (5)	63	87
	Lab Recovery (6)	85	82
	Lab Recovery (7)	82	90
Mean		95	101
RSD %		21	22
Plastic Bottles			
Tap water	Lab Recovery (1)	118	98
	Lab Recovery (2)	99	103
	Lab Recovery (3)	73	115
	Lab Recovery (4)	74	88
Mean		91	101
RSD %		24	11

3.2 Field studies.

This research involved a number of surveys involving sampling of water and effluents. These can be divided into three types, from the survey of STW (November 2008), the samples from The River Erewash (January and October 2009) and the tap water survey during May and June 2010. This section describes these events.

3.2.1 Sampling of wastewater at Hallam Fields and Newthorpe

Two STWs were selected for study the removal of progestogens and benzotriazoles during unit processes. Each of the STW discharged into the River Erewash. One (Newthorpe STW) was a trickling filter (TF) plant, with a rapid sand filter, and dry weather flow (DWF) of 10,282 m^3/day. Three sampling points were selected through

the processes which were: settled sewage, after trickling filter, and after the sand filter (final effluent) as shown in Figure 3.7.

The other site (Hallam Fields STW) was a nitrifying activated sludge (N/AS) works, also with a sand filter, treating a DWF of 10,022 m^3/day. Sampling points were at seven locations through the process which were four points in the conventional treatment represented by: crude, settled sewage, then after the biological treatment and finally after the sand filter (final effluent). For the advanced wastewater techniques, three types of advanced treatment were available and the sampling points were in three parallel streams which were: after the ozone tank, after the GAC tank, and after treatment with chlorine dioxide. These three systems were working in parallel. Each system treated between 200 to 1,000 m^3 of the effluent from the sand filter per day. Ozone was generated from liquid oxygen on site. The GAC treatment used coal-based activated carbon operated at a minimum 20 min empty bed contact time (EBCT). In order to produce ClO_2, hydrochloric acid, sodium hypochlorite solution and sodium chlorite solution were mixed on site. The dose into final effluent of ozone and ClO_2 was at a rate of 1mg/l. Figure 3.7 shows the sampling points at Hallam Fields STW.

Sampling was in November 2008 for four days from Monday to Thursday for both sites. The sampling was at two times for Hallam Fields STW, once at 9:00 am and the other batch of sampling was at about 2:00 pm. At Newthorpe STW, the sampling was once a day at 12:00 pm. Samples were collected in 2.5l amber glass bottles and filtered by (GF/C, whatman, UK) directly after collection and were then extracted onto SPE. Because of the concentrations of benzotriazoles collected from the first sampling campaign (November 2008) were out of the calibration curve for the samples from advanced treatment, one further day of sampling was undertaken on 16th of September 2009 from the Hallam Fields STW. Samples were collected by the staff at STW and sent to Brunel University, each sample was 100ml with 0.1ml of the internal standards. They were collected from the final effluent, post ozone, post GAC, and post ClO_2 at two times (one in the 9:00 am and the other was in 2:00 pm) to be within the calibration curve.

A crude and settled sewage sample of 500ml and 200ml for progestogens and benzotriazoles samples respectively. All sample for biological process, sand feed, final effluent and advanced treatment samples were 1000ml for progestogens. For Benzotriazole, all samples were 200ml except the advanced treatment samples were 100ml. Table 3.14 summarises the sampling volume of each sampling point in the STW.

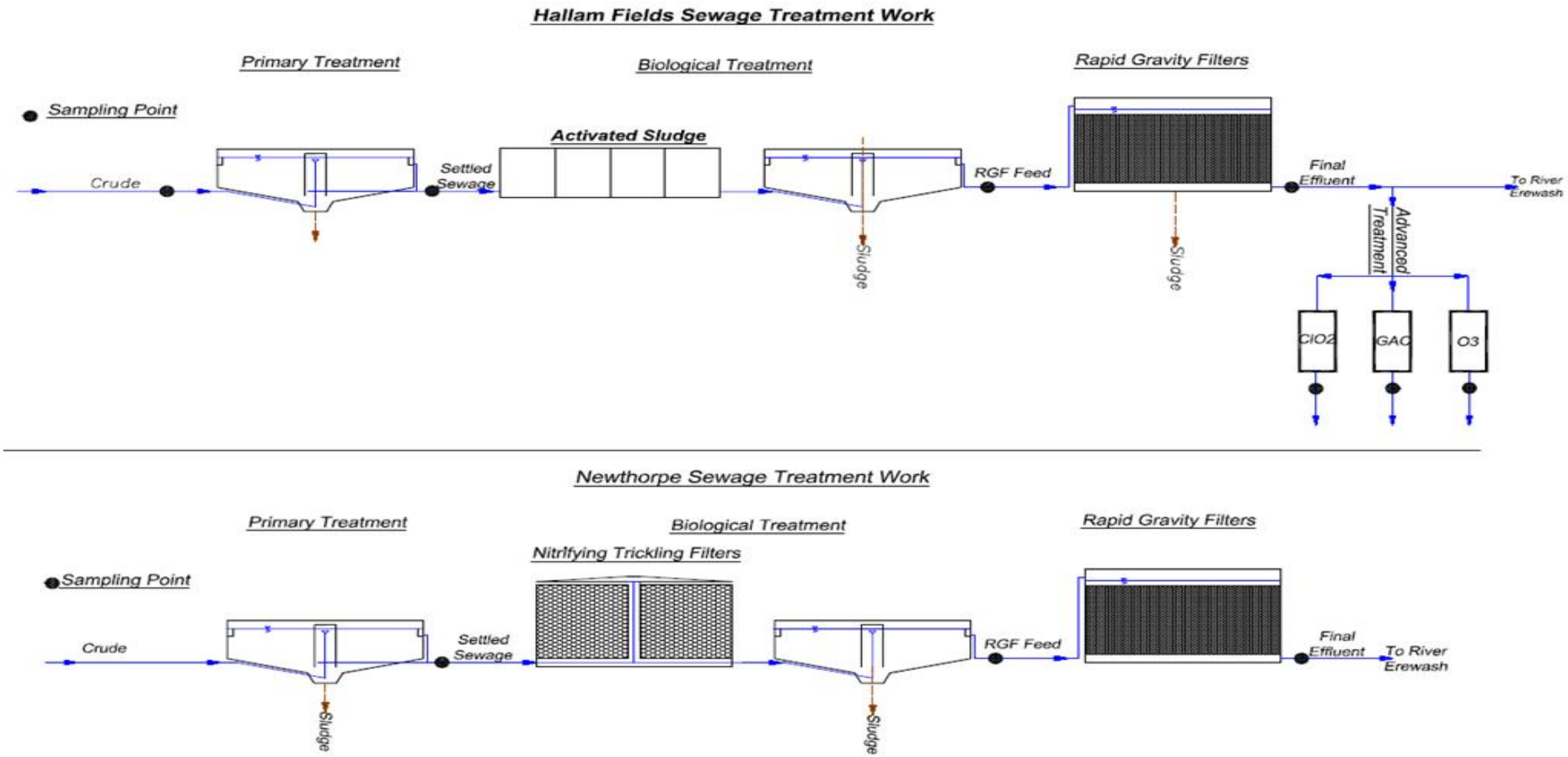


Figure 3.7 Flow diagram and sampling points for Hallam Fields (top) and Newthorpe STWs.

Table 3.14 Sampling volume of each sampling point in the STW.

Sampling Point	Sample Volume (l)	
	Progestogens	Benzotriazoles
Crude sewage	0.5	0.2
Settled sewage	0.5	0.2
RGF feed	1	0.2
Final effluent	1	0.2
Ozone output	1	0.1
GAC output	1	0.1
ClO_2 output	1	0.1

3.2.2 Sampling of the Erewash River

The River Erewash is a tributary of the River Trent and it rises in Kirkby-in-Ashfield, Nottinghamshire. The River Erewash drains an area of 211 km^2 above its confluence with the River Trent near Nottingham, UK. The mean flow of the River Erewash is 1.91 m^3/sec. Around 30% of the catchment is covered in urban development with the rest mainly arable and grazing (Marsh, Greenfield & Hannaford, 2005). There are eight sewage treatment works which discharge along the River Erewash.

Two sampling campaigns were conducted in order to determine the concentration of benzotriazoles and progestogens in the river and the STW existing in the Erewash River, the first campaign was in January 2009 and the second one was in October of the same year. There were 24 sampling points including eight wastewater treatment effluents existing on the river. The grid references of 11 sampling point along the River Erewash are in Table 3.15. The other five sampling points were on the brooks, tributaries, to the River Erewash. Samples were collected in 2.5l amber glass bottles with $CuSO_4.5H_2O$/nitric acid preservative. Sample volume was 1l for progestogens and 200 ml for benzotriazoles and they were filtered and extracted after less than 24 hours of collection. Figure 3.8 shows the sampling point on the River Erewash, on its brooks, and emission points of benzotriazoles from eight STWs.

Table 3.15 Grid references of the sampling point along the River Erewash.

Sample No.	Location on river system	Grid references
1	Erewash U/S of Kirkby STW	SK 485548
2	Kirkby STW FE	
3	Erewash D/S Kirkby STW (and U/S of Pinxton STW)	SK 464547
4	Pinxton STW FE	
5	Erewash D/S Pinxton STW (and U/S of Pye Bridge STW)	SK 443529
6	Pye Bridge STW FE	
7	Erewash D/S Pye Bridge STW	SK 440520
8	Bagthorpe brook	
9	Nethergreen brook	
10	Erewash U/S of Milnhay STW	SK 455466
11	Milnhay STW FE	
12	Erewash D/S of Milnhay STW	SK 463454
13	Gilt brook U/S of Newthorpe STW	
14	Newthorpe STW FE	
15	Gilt brook D/S of Newthorpe STW	
16	Erewash D/S of Gilt brook confluence	SK 475444
17	Erewash U/S of Hallam Fields STW	SK 478405
18	Hallam Fields STW FE	
19	Nut brook	
20	Erewash D/S of HF STW & Nut brook confluence (and U/S of Stapleford STW)	SK 484383
21	Stapleford STW FE	
22	Erewash D/S Stapleford STW (and U/S Toton STW)	SK 503341
23	Toton STW FE	
24	Erewash D/S of Toton STW (and just before confluence with R. Trent)	SK 510335

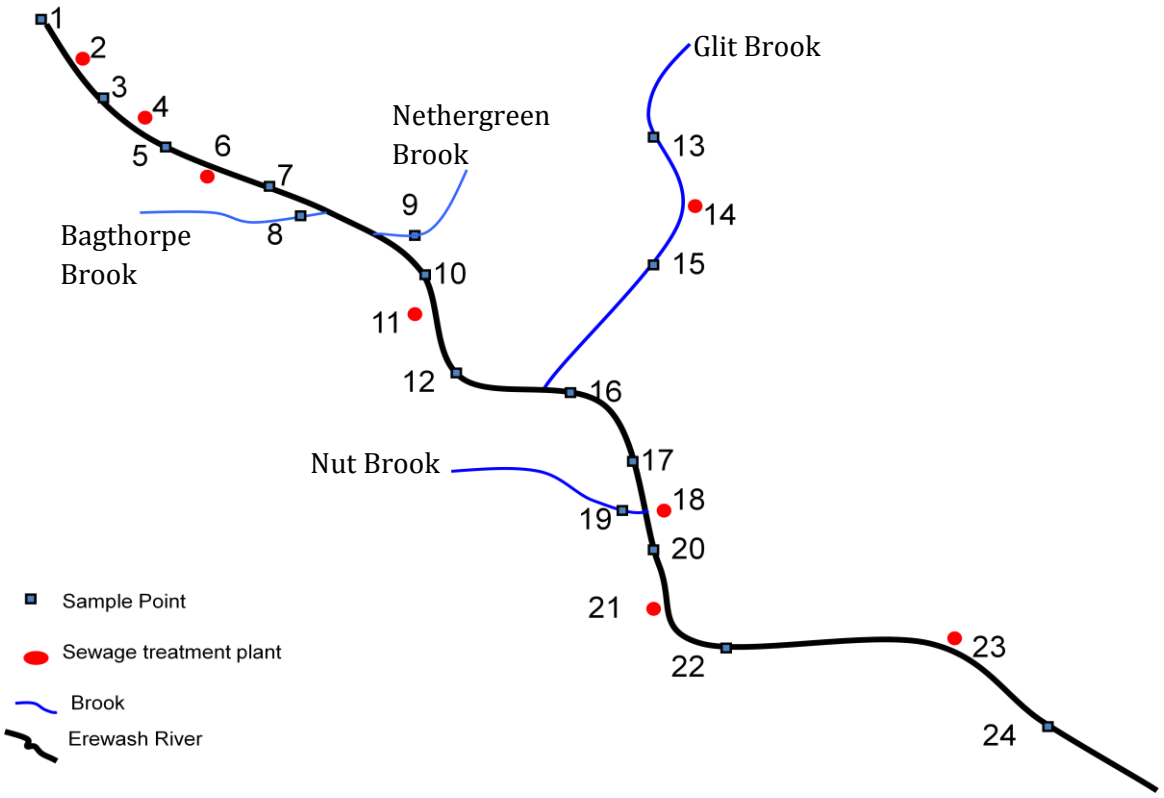


Figure 3.8 Schematic diagram of the sampling points on the River Erewash and its brooks. Numbers refer to locations identified in Table 3.15.

3.2.3 Sampling of tap water

To determine whether there were any benzotriazoles in the potable supply, we undertook sampling on four occasions for 20 tap water points during May and June 2010 for a total of eighty tap water samples, from locations in the south east of England, these sampling points were predominantly within a 15 mile radius of Uxbridge, in west London, although some were up to 30 miles to the west and one location was 80 miles to the north east. One litre of the each sample was collected in the morning and they were then extracted within less than five hours of collection.

3.2.4 Domestic dishwasher powders and tablets

During May and June 2010, consumer products were purchased from UK supermarkets and then analysed in order to quantify the benzotriazoles concentration in the dishwasher powder and tablet, approximately 20 g of each product collected from six brands of dishwasher powder and tablet were weighed and dissolved in 1l of MilliQ

water by using a magnetic stirrer. Duplicated samples were analysed after one hour of dissolution. Then, further dilutions (1:100) and some samples had more dilution in order to be with the range of calibration standards. An external standard was used to analyse the samples to quantify the benzotriazoles. Table 3.3 shows the retention time and the masses used to quantify benzotriazoles.

3.3 Degradation study

The goal of studying the degradation of progestogens and benzotriazoles in rivers was to determine the half life of these chemicals. Therefore, two plastic tanks each one about 100l in volume was filled with about 60 l of water taken from the Grand Union Canal side River in Maple Cross, Hertfordshire, about 0.5 km downstream of maple Lodge STW (Figure 3.9). Each tank had one air pump and this pump distributed the air via two bubble diffusers in the tank in order to maintain the dissolved oxygen levels. In addition to that, two agitators with two magnetic bars were installed in each tank in order to keep the water mixed. For two weeks, dissolved oxygen, pH and temperature were measured every day in the morning from each tank. Also, samples were taken from each tank in the morning for progestogens and benzotriazoles. The sample volume of progestogens was 1l, and for the benzotriazoles was 200ml. Duplicate samples for progestogens and benzotriazoles was collected in days 1, 4, 9, and 14 of the study. Four samples representing laboratory Recoveries were analysed during the study and the percentage of recovery of all chemicals were from 78% to 114% (Table 3.11). Four samples for progestogens and two samples for benzotriazoles each one was 1l of MilliQ water was used as blank sample, the mean concentration of all chemicals were between 0.4 and 3.4 ng/l (Table 3.9) (Table 3.10).

The half lives of these 12 compounds (progestogens and benzotriazoles) were calculated according to the equation (1):

Half life ($t_{1/2}$) = $\ln 2/k_{tot.}$ …….. (1)

Where $k_{tot.}$ represents the overall degradation rate of the compound (Sinkkonen & Paasivirta, 2000).

Figure 3.9 Location of the sampling point for the degradation test, about ≈ 850m downstream Maple Lodge STW (Google Earth).

3.4 Modelling

In collaboration with Richard Williams at the Centre of Ecology and Hydrology (CEH), this work involved visiting CEH in order to see the approach used for River Erewash modelling and its catchment area. The model was run by Richard Williams using the data provided from the first sampling campaign only of Hallam Fields and Newthorpe STWs for progestogens and benzotriazoles in the influent and final effluent. A single grab sample was collected from the River Colne (tributary of the River Thames) in order to investigate the occurrence of these compounds. Therefore, the Thames River catchment area was also modelled to evaluate their wider occurrence in the UK. The first approach was used by predicting the concentrations in the River Erewash by using the final effluent concentrations that measured at STWs and the second approach was depending on the per capita load. The catchments modelled were those of the River

Trent, where the Erewash is located, and the River Thames, which was the known source for some of the tap water samples, in particular from the area around Uxbridge, west London.

3.4.1 Overview of the model

Modelling was carried out using the LF2000-WQX model Williams et al. (2009), which is a system designed for making predictions of river concentrations of chemicals which are disposed of "down-the-drain" (i.e. chemicals whose main entry to the river system is through sewage treatment works discharges). It is based on the simple mass balance mixing equation which is applied in a Monte Carlo simulation using the method of combining distributions proposed by (Warn & Brew, 1980). Good estimates of river flow are important to correctly estimate in-stream concentrations resulting from point source discharges. In LF2000-WQX, river flow is calculated using the well established LowFlows 2000 hydrological model which estimates the flow duration curve (i.e. the statistical distribution of flows) at ungauged stations (Keller & Young, 2004; Young, Grew & Holmes, 2003). Point-source effluent emissions are combined with reach-specific flow statistics to calculate in-river concentrations after mixing at the point of discharge taking account of concentrations coming from upstream. Concentration changes along the river network due to dilution from tributaries not receiving discharges and degradation are taken into account. Accumulation in other environmental compartments (e.g. sediments) is not considered. The result is a statistical distribution of calculated in-river concentrations for each river reach, based on the variability of reach-specific river discharge, emissions from the STWs in the upstream catchment as well as in-stream degradation (Price et al., 2010).

3.4.2 Method of extrapolation to the Trent basin

To run the model at a catchment scale, to obtain values for sewage treatment works discharges that could be used more widely throughout the catchment, the measured concentrations in effluents were transformed into equivalent per capita output loads (equation 2).

$$PC_{L_i} = A(C_{eff,i} \times DWF_j)/Pop_j \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2)$$

Where PCLi (ng/person/day) is the per capita load for chemical i, C_{eff} (ng/l) is the measured effluent concentration, DWF (m^3/s) is the dry weather flow from sewage

treatment works j and Pop is the population served by the works and A is a unit conversion factor. To do that, these steps were followed to calculate the load per capita.

1. Calculate a per capita load in the effluent from the eight sewage treatment works on the Erewash based on the measured effluent concentrations of BT and TT. This is simply the measured concentration multiplied by the dry weather flow from the STW divided by the number of people served by the works. The values are given in Table 3.16.
2. Find an average per capita value checking if there was any difference between types of STW. If all STWs are the same then we can calculate one average value and a standard deviation.

   Ofwat, water services regulation authority, in 2008 classified STWs into seven different categories in order to discriminate between different treatment processes in terms of their pollutant removal efficiency (Ofwat, 2008). Some of these classifications relavant to the works on the Erewash are:

   SAS (Secondary activated sludge): As primary, plus STWs whose treatment methods include activated sludge (including diffused air aeration, coarse bubble aeration, mechanical aeration, oxygen injection, submerged filters) and other equivalent techniques including deep shaft process, extended aeration (single, double and triple ditches) and biological aerated filters as secondary treatment.

   TA2 (Tertiary A2): STWs with a secondary activated sludge process whose treatment methods include rapid-gravity sand filters, moving bed filters, pressure filters, nutrient control using physico-chemical and biological methods, disinfection, hard chemical oxygen demand (COD) and colour removal, where used as a tertiary treatment stage.

   TB2 (Tertiary B2): STWs with a secondary biological process whose treatment methods include rapid gravity sand filters, moving bed filters, pressure filters, nutrient control using physico-chemical and biological methods, disinfection, hard COD and colour removal, where used as a tertiary treatment stage (Ofwat, 2008).
3. A Welsh 2-way t-test showed that SAS type STWs had a significantly lower per capita effluent values than either TA2 or TB2 and so two values were calculated (Table 3.17). Actually this is rather surprising as it would be expected that tertiary treatments would remove more organic chemicals in general.

4. So now we can set up all STW input per capita loads in the River Trent to be the effluent per capita loads calculated above from the non-SAS type works. For these works a removal efficiency of 0% was applied (i.e. the input per capita load was the same as the effluent per capita load as we measured effluents). For SAS works removal efficiencies of 37.3% and 50.1% were set for BT and TT removal (253/678*100 and 728/1452*100) respectively.

Table 3.16 River Erewash sewage treatment works and calculated per capita effluent loads.

STW	Type	DWF m^3/s	DWF m^3/day	Concentration (ng/l)		Population	Mass (g/d)		PerCap (µg/day)	
				BT	TT		BTm	TTm	BTcap	TTcap
Kirkby	SAS	0.069	5970	840	2685	24538	5.0	16.0	204.4	653.3
Pinxton	TA2	0.049	4242	1335	4125	8166	5.7	17.5	693.5	2142.9
Pye Bridge	TB2	0.024	2048	2895	5450	7043	5.9	11.2	841.7	1584.5
Milnhay	TA2	0.082	7050	1655	3400	25957	11.7	24.0	449.5	923.5
Newthorpe	TB2	0.119	10282	2470	5500	43382	25.4	56.5	585.4	1303.5
Hallam Fields	TA2	0.116	10022	2090	5700	44895	20.9	57.1	466.6	1272.5
Stapleford	SAS	0.066	5737	1375	3665	26203	7.9	21.0	301.0	802.4
Toton	TB2	0.194	16762	3605	5200	58584	60.4	87.2	1031.4	1487.8

Table 3.17 Summary of per capita loads (µg/day) calculated from effluent data from STWs on the River Erewash.

	TA2 & TB2		SAS	
	Mean	STDEV	Mean	STDEV
BT	678	227	253	68
TT	1452	407	728	105

Chapter Four: Results of Conventional Treatment

Overview

Primary treatment, biological treatment and sometimes tertiary treatment which is usually designed to remove ammonia or suspended solids are the main components of conventional wastewater treatment. This chapter will report the occurrence of the compounds of interest in these three stages at Hallam Fields and Newthorpe STWs. Therefore, samples were taken after each unit process in order to observe the occurrence of these chemicals and to evaluate how effective each unit process was at removing them. In addition, some operation parameters and physiochemical parameters will also be described.

4.1 Physiochemical parameters and sampling conditions.

The two STWs were sampled during November 2008. The biological treatment at Hallam Fields (Figure 4.1) was N/AS served 44,895 population equivalent (PE). The second STW, Newthorpe, utilised trickling filters as a biological treatment and served 43,382 PE (Figure 4.1). The main sanitary parameters of these sewage treatment works are listed in Table 4.1 and represent the mean of eight and four samples from Hallam Fields and Newthorpe STWs respectively.

Figure 4.1 Photographs of the two sewage treatment works, Hallam Fields (left) and Newthorpe (right).

Table 4.1 Operating parameters for sewage treatment works.

Operating Parameters		Hallam Fields STW				Newthorpe STW			
Biological process		Activated Sludge Process				Trickling Filter			
PE		44,895				43,382			
Flow (DWF)		10,022 m^3/day				10,282 m^3/day			
Parameter	Unit	Settled Sewage	ASP Effluent	Final Effluent	Removal %	Settled Sewage	Humus Tank Effluent	Final Effluent	Removal %
pH		8.0	7.9	7.9		8.0	7.9	7.8	
Ammoniacal Nitrogen as N	mg/l	14.6	0.3	0.3	98	23.0	1.3	0.5	98
Nitrogen, Total Oxidized as N	mg/l	1.7	14.9	14.7		0.4	24.8	21.6	
Phosphorus , Total as P	mg/l	3.2	0.8	0.7	77	5.1	0.9	0.3	93
BOD + ATU (5 day)	mg/l	52.9	2.0	1.0	98	80.3	3.3	1.0	99
COD (Total)	mg/l	144.0	23.6	24.1	83	240.8	43.5	29.3	88

According to the results shown in Table 4.1, the removal efficiency of BOD_5 was 98% and 99% for Hallam Fields and Newthorpe STWs respectively. Ammoniacal nitrogen as N was also removed with 98% removal efficiency at both STWs. Although suspended growth as N/AS plant may give more removal, however TF STW has shown good removal efficiency for phosphorus compared to the N/AS STW represented by 93% and 77% removal efficiency respectively, this perhaps because of few numbers of samples and time of sampling might have biased the results. It is apparent from this table that there was no variation in pH in settled sewage and final effluent for both STWs. Despite of high concentration of COD in Newthorpe STW compared to Hallam Fields STW, the removal efficiency of COD at trickling filter process was higher than nitrifying activated process, which was not expected according to previous literature and that might be again due to sampling bias. During the sampling, there was no rain or wet weather; as a result the concentrations of the chemicals of interest in the influent would be expected to be at relatively high concentrations due to the lack of dilution, giving high loadings to the STWs.

Sampling bias

The daily volumes of wastewater streams were very large. In addition, the analytical costs of pharmaceuticals in aqueous samples are more expensive than other compounds. Therefore, samples must be collected in order to represent the concentration of selected chemicals in wastewater. Table 4.2 shows the sampling regime achieved through the study the occurrences and removal of progestogens and benzotriazoles at the two STWs. At Hallam Fields STW, two sampling times per day were achieved, the first sampling time was at 9:00 am and presumably if the retention time in the sewer system was 6 or 9 hours, in both cases that would mean that concentrations of these compounds were weak as inputs occurred at night. In the mean while, the second sampling time was at 2:00 pm and in this case the concentrations would be stronger in the effluent than at the influent. Thus this lag time may have had an effect on calculating the removal efficiency, making it to be lower than the true value.

Newthorpe STW had only one single sample per day and that was at 12:00pm, this means possibly strong concentrations in the influent and weak concentrations in the

effluent, consequently good removal efficiency will be determined. Generally, all these limitations, sampling time and sample numbers, might have led to some bias in the results and therefore there is associated uncertainty in the results.

Table 4.2 Sampling regime achieved at the two conventional STWs.

		time of discharge to sewer	
STW	**Sampling time**	time in sewer	
		6 hours	9 hours
Hallam Fields	09:00 (weak)	03:00 (weak)	00:00 (weak)
	14:00(strong)	8:00(strong)	5:00(strong)
Newthorpe	12:00(strong)	06:00(weak)	03:00 (weak)

4.2 Occurrence and removal of progestogens in conventional treatment works.

The concentrations of progestogens after each process at Hallam Fields and Newthorpe STWs are shown in Figure 4.2. Concentrations of the chemicals of interest in the settled sewage, rapid gravity filter (RGF) feed, and final effluent are described below:

4.2.1 Settled sewage

The concentrations of progestogens after the primary settling tank at Hallam Fields and Newthorpe STWs are shown in Figure 4.2A. The natural hormone, progesterone (PGT), was detected and quantified in all the samples at this stage at both STWs. It can be seen that there was an abundance of progesterone, where the mean concentration was 46.9 ng/l at Hallam Fields STW and was 41.6 ng/l at Newthorpe STW, no significant difference had occurred between both STW. Many compounds concentrations at both STW were similar, DHG and TBL mean concentrations in settled sewage at Hallam Fields STW was 35.4 and 29.4 ng/l, while at Newthorpe STW the mean concentrations of them were 35.2 and 15.9 ng/l. The cyproterone acetate (CPA) was quantified in seven out of eight samples with a mean concentration of 18.0 ng/l at Hallam Fields STW, and quantified in all samples with a mean of 13.5 ng/l at Newthorpe STW. MPA and DSP

were detected only in 25% of the samples of the settled sewage at Hallam Fields STW and 50% of the samples at

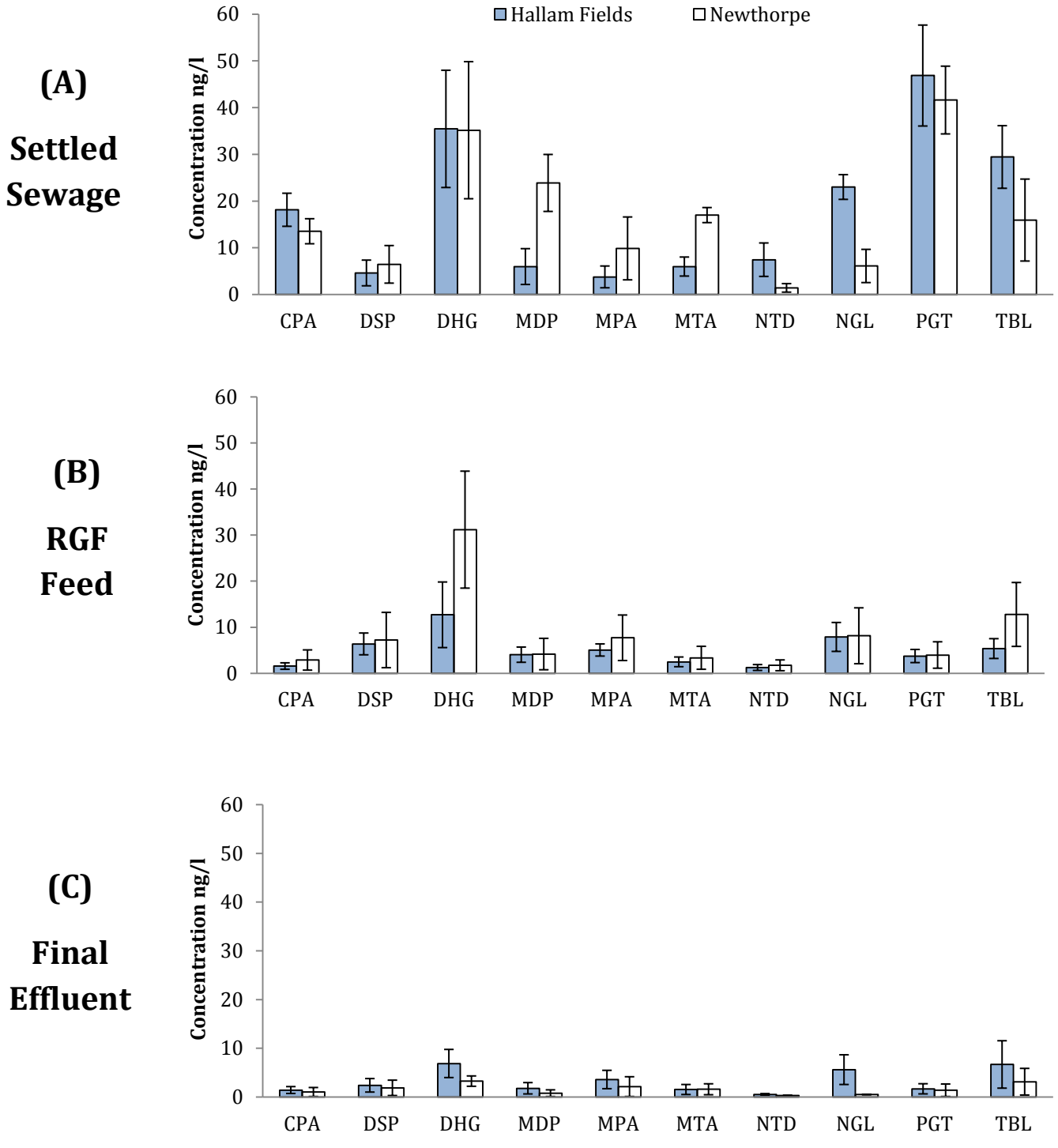


Figure 4.2 Concentrations of progestogens in ng/l (A. Settled Sewage. B. RGF Feed. C. Final Effluent) of two sewage treatment works.

Newthorpe STW, while NTD was quantified in half of the number of the samples and 75% of the sample at Hallam Fields and Newthorpe STWs. MPA, DSP, and NTD show again no significant difference in concentrations for each individual compounds at each STWs, and the mean concentration of each individual compound was below 10 ng/l in both STWs.

For a few compounds, significant differences (p=0.05) in concentrations were observed, Norgestrel in settled sewage at Hallam Fields STW 23 ng/l was a significantly above the concentration at Newthorpe STW of 6.1 ng/l. Conversely, MDP 6 ng/l and MTA 6 ng/l were significantly less at Hallam Fields STW than at Newthorpe STW of 23.9 ng/l and 17 ng/l respectively.

4.2.2 Rapid gravity filter (RGF) feed.

The concentrations of progestogens after the biological treatment (N/AS or TF) at Hallam Fields and Newthorpe STWs are shown in the Figure 4.2B. It can be seen that significant difference (p=0.05) for progesterone had occurred between the settled sewage and the RGF feed indicating a removal of 92% and 90% for Hallam Fields and Newthorpe STWs respectively. According to Figure 4.2B, it was apparent that CPA, NTD, NGL and TBL show significant removal had occurred during the N/AS process at Hallam Fields STW, while these chemicals apart from CPA show no removal had occurred during the TF process at Newthorpe STW. This indicates that the activated sludge process (ASP) may be more efficient to remove these compounds than the TF process.

Conversely, medroxyprogesterone (MDP) and MTA present a significant difference had occurred during TF as a biological process at Newthorpe STW, but there was no evidence for removal during N/AS and perhaps this because of these compounds had low concentrations entering the ASP at Hallam Fields STW. The original data for progestogens in the conventional treatment processes are found in appendix 1.

For a few compounds, drospirenone (DSP), DHG, and MPA show no significant removal was observed during the biological treatment at both STWs. It is possible that more removal could occurred , but not statistically observed in this study and that probably due to the number of samples limits statistical power. Generally, removal of compounds can be achieved by either adsorption or degradation and the range of Log K_{ow} (3.87 ±

0.68) was similar for all progestogens. This means similar removals were expected in terms of adsorption and the differences in the removal might be resulted from the degradation.

4.2.3 Final effluent

Figure 4.2C shows the concentrations of progestogens after the RGFs at Hallam Fields STW and Newthorpe STW. From Figure 4.2C, it can be seen that all compounds demonstrated removal through the STW. Although it appears that the performance of the RGF at Newthorpe STW was better than the performance of that at Hallam Fields STW, however there was no significant difference ($p=0.05$) had occurred across the RGF's for either STWs. Whereas the RGF at Newthorpe STW removal efficiency was between 53%-94%, and it was lower at Hallam Fields STW (0-62%). This difference in removal efficiencies may be real or might be biased because of the sampling strategy.

The effluent mean concentration of each individual compound at Hallam Fields STW was below 7 ng/l, while the concentrations of the final effluents at Newthorpe STW were below 3.5 ng/l. Most of the final effluent samples concentrations were below their limit of quantification in both STWs, half of LOD value was taken to calculate the mean concentration of each compound. For example, there were only two values at Hallam Fields STW were above the quantification limit and these values had possibly influenced the results for DSP, MDP, MTA, NTD, PGT, and TBL, therefore it is possible that the performance of the RGF at Hallam Fields STW looked less efficient than the RGF at Newthorpe STW.

The overall removal efficiency of progestogens during all the processes at each STW is illustrated in Figure 4.3. It is apparent that the natural hormone, progesterone, was the easiest compound which could be removed with 96% and 97% removal efficiency at Hallam Fields and Newthorpe STWs respectively. At Hallam Fields STW, the removal efficiency for most of the compounds were above 70% and only DSP and MPA were removed at 48% and 5% removal efficiencies, this is perhaps due to the low initial concentrations of these chemicals which were just above the limit of quantification. Newthorpe STW presented a very good removal for all compounds and the removal efficiencies were between 71% and 97%. In general, the ASP would be expected to give more removal than the TF process, however Hallam Fields STW looked less efficient to

remove these compounds than Newthorpe STW and as mentioned this may be due to sampling strategy. Therefore, it is possible that the removal efficiencies at Hallam Fields STW were more realistic than at Newthorpe STW.

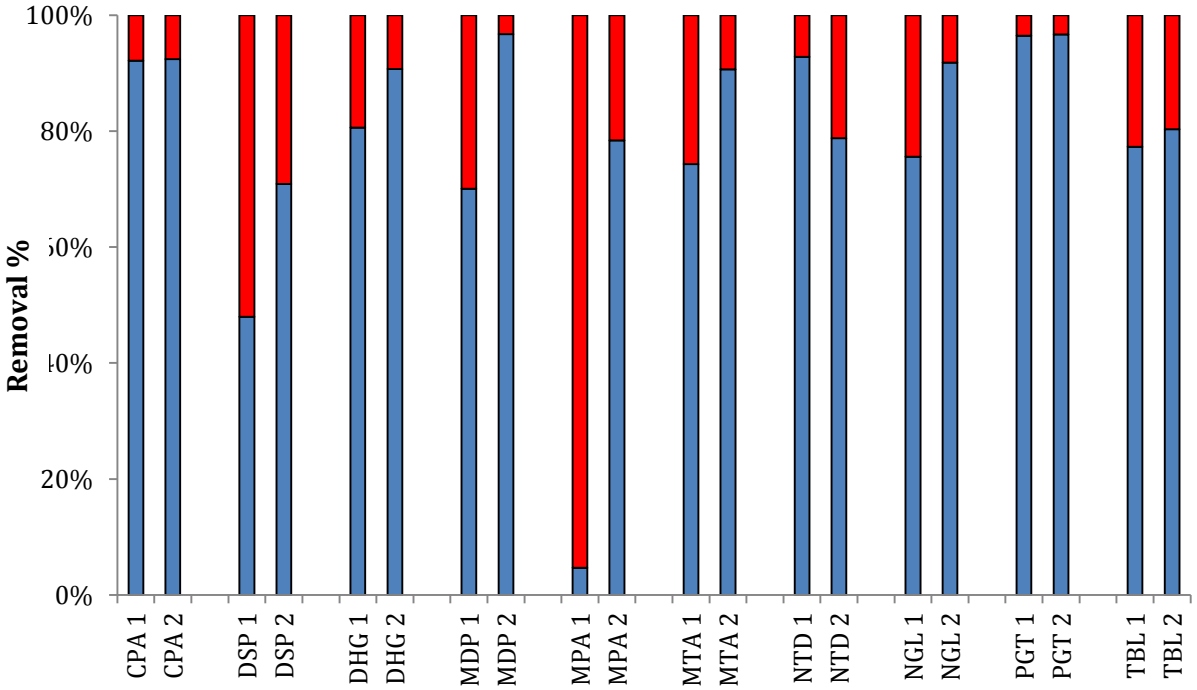


Figure 4.3 Percentage removal efficiencies of progestogens through the conventional treatment works. (1) Hallam Fields STW, (2) Newthorpe STW. ■ % removed during STW, ■ % Discharged to the surface water.

4.3 Occurrence and removal of benzotriazoles in conventional treatment works.

The results obtained from analyzing benzotriazoles at three different stages: settled sewage, biological treatment, and final effluent samples are illustrated in Figure 4.4. It is apparent that compared to progestogens there was an abundance of benzotriazoles that entered the STWs. Concentration of benzotriazoles were 2,000 to 3,500 ng/l compared to the tens of ng/l for progestogens. The proportions of BT to TT were the same at each STW. The results also showed that the concentrations of BT and TT in the settled sewage at each of the STW were the same, and although average concentrations of TT appeared to be above those of BT, there was no significant difference (p= 0.05).

According to Figure 4.4, there was some evidence that removal had occurred during the biological treatment for BT in both STWs, since the concentration of RGF feed was 1,778 and 1,438ng/l at Hallam Fields and Newthorpe STWs, but this difference was not significant and that possibly because of bias resulting from sample time/number. There was also a significant difference in the removal for TT at Hallam Fields STW during the N/AS process, this means that TT has probably degraded because of low K_{ow} log (1.24) meaning sorption is less likely. Although the data indicate removal occurred at Newthorpe STW, but that was not statistically significant. The original data for benzotriazoles in the conventional treatment processes are found in appendix 1.

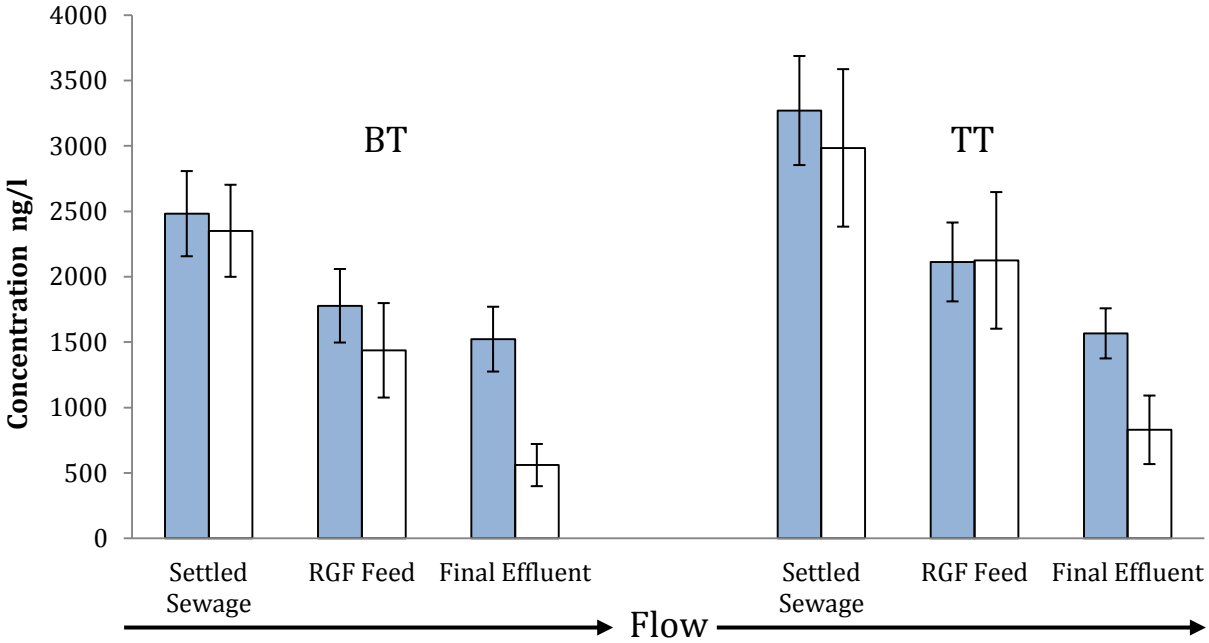


Figure 4.4 Concentrations of benzotriazole and tolyltriazole in (ng/l) ■ Hallam Fields STW, □Newthorpe STW.

Concentrations of these chemicals as they were discharged to the River Erewash in the final effluent were 1,523 and 1,567 ng/l for BT and TT at Hallam Fields STW and that would suggest a removal efficiency of 14% and 26% was achieved across the RGF, while the concentrations for BT and TT at Newthorpe STW were 560 and 830 respectively (Figure 4.4). No significant difference had occurred in the concentrations of benzotriazoles across the rapid gravity filters in both STWs. For BT or TT, a significant difference had occurred in the final effluent concentrations that discharged to the River Erewash between Hallam Fields STW and Newthorpe STW.

Although there is some evidence that removal had occurred through the conventional treatment works, but still there was a large amount of these chemicals were discharged to the surface water. Figure 4.5 shows the fluxes of benzotriazoles discharged from the conventional treatment works to the River Erewash which resulted from multiplying the flow by the concentration of these chemicals that discharge to the water body.

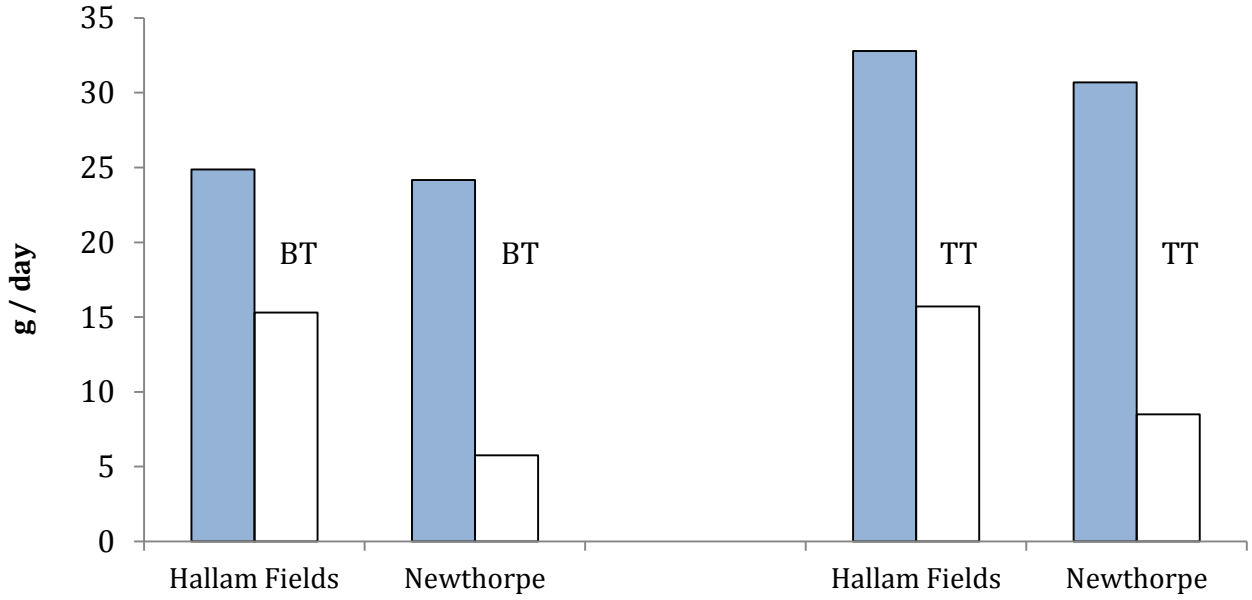


Figure 4.5 Fluxes of benzotriazoles through the conventional treatment works in g/d. ■ Settled sewage, □ Final effluent.

From Figure 4.5, it is apparent that there was a removal had occurred through all process with an overall removal efficiency of 39% and 76% for BT at Hallam Fields STW and at Newthorpe STW respectively. Moreover, there was a significant reduction in the concentration of TT had occurred during the process of the STW at STWs, 52% for Hallam Fields STW and 72% for Newthorpe STW. In general, the removal efficiency at Newthorpe STW appeared higher than Hallam Fields performance and that might be real or because of the bias in the sampling number and time.

Summary

This chapter illustrated the occurrences of progestogens and benzotriazoles during the conventional processes: primary sedimentation, biological treatment, and rapid sand filter at sewage treatment works. Because of these emerging contaminants were not removed completely, therefore further study to observe their occurrence in the advanced treatment will be undertaken in the next chapter.

Chapter Five: Results of Advanced Treatments

Overview

This chapter will report the effectiveness of the advanced treatment that was available at Hallam Fields STW. The effluents from three types of advanced treatment were sampled and analysed to determine the removal efficiencies of these techniques for the chemicals in this study. In total, eight samples were taken, one in the morning (9:00) and afternoon (14:00) for four days to monitor the removal of these chemicals via advanced technologies.

5.1 Operation parameters for ozone, granular activated carbon, and chlorine dioxide.

Part of the sand filter effluent was diverted to three pilot plants at Hallam Fields STW (O_3, GAC, and ClO_2) as shown in Figure 5.1. These three systems were working in parallel. Each system treated between 200 to 1000 m^3 of the effluent from the sand filter per day. A dose of 1mg/l of ozone was injected by injection system into the effluent with three minutes contact time; ozone was generated from liquid oxygen on site. The GAC treatment used coal-based activated carbon operated at a minimum 20 min empty bed contact time (EBCT). In order to produce ClO_2, hydrochloric acid, sodium hypochlorite solution and sodium chlorite solution were mixed on site; chlorine dioxide was injected into the effluent at a dose of 1 mg/l.

O_3 system **GAC system** **ClO_2 system**

Figure 5.1 Pictures of the advanced technologies at Hallam Fields STW.

5.2 Occurrence and removal of progestogens in the advanced treatment processes.

In the following sections, performance of three processes in removing progestogens from the final effluent is presented. For final effluent, although 75% of values were below limit of quantification for six compounds (DSP, MDP, MTA, NTD, PGT, and TBL) and only 2 values were above LOQ; however the mean concentrations of progestogens ranged between 0.5 ng/l for norethindrone to 6.7 ng/l for tibolone. All the original data for progestogens are in appendix 2.

5.2.1 Removal by ozone

The Figure 5.2 shows the concentrations of the progestogens pre - and post ozone treatment. It is apparent that the concentrations post ozonation were similar to those pre ozonation, with the mean concentrations of the post ozonation for these compounds ranging from 0.7 ng/l for norethindrone to 5 ng/l for norgestrel. There was no significant difference in any concentration as a result of treatment with ozone, thus, ozone apparently did not remove the progestogens. Because the concentrations of progestogens were close to or below the limit of quantification, and there were relatively few samples, therefore, there is a high degree of uncertainty associated with these data, as shown by the error bars in Figure 5.2.

Five values out of eight sample concentrations for dydrogesterone (DHG), NTD, NGL, and TBL were less than the limit of quantification. These values were observed in the first two days of sampling (morning and afternoon). Half of the values of DSP, MDP, MTA, and PGT were below their quantification limit and that was in day 1 and 2 of sampling. The remaining compounds (CPA and MPA) had only two values under the limit of quantification and they were also observed in the afternoon of the first two days. Therefore, it is possible that there was analytical problem which occurred during the third and fourth days of the sampling (day three and four recoveries in Table 3.11) and that may have affected the average concentration of these compounds.

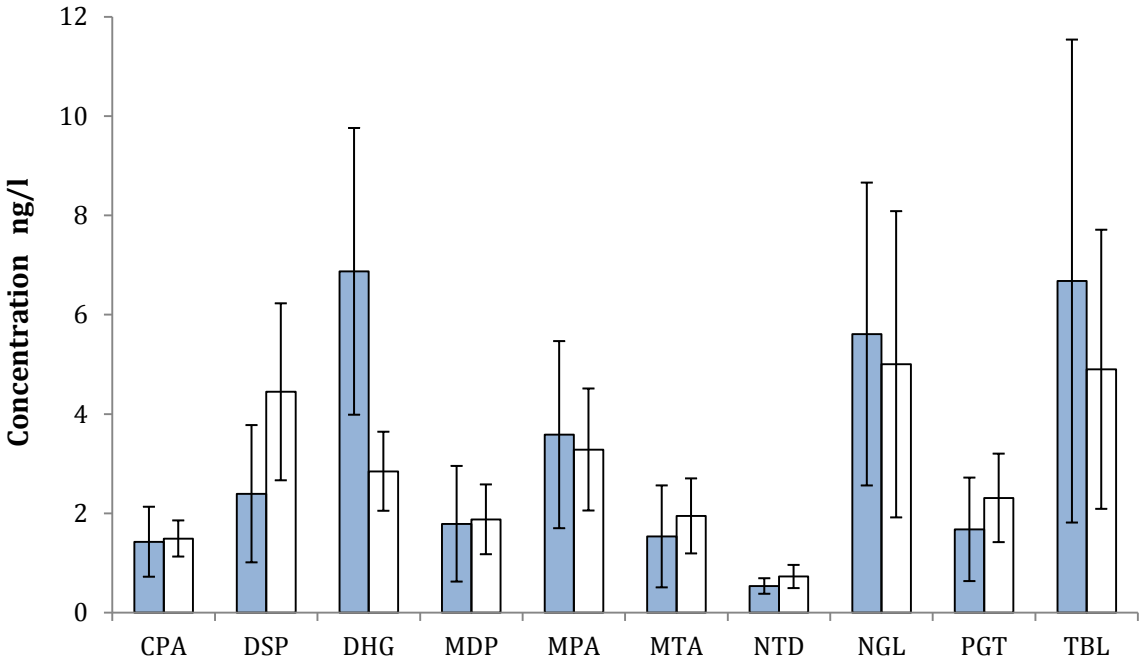


Figure 5.2 Removal of progestogens by ozone (■ Final Effluent (Pre-O_3), □ Post-O_3).

5.2.2 Removal by granular activated carbon (GAC).

The occurrence and removal of progestogens through the GAC treatment are shown in Figure 5.3. The average concentrations of progestogens ranged from 0.2 ng/l for cyproterone acetate to 5.7 ng/l for tibolone. There was more evidence for removal than was observed during ozonation, however it was not statistically significant with p=0.05. The average concentration of these compounds was calculated by assuming that each single value below the limit of quantification is equal to half of the limit of detection. Although no significant removal occurred, many compounds in the post GAC stage had 75% of values below LOQ and analytical uncertainty makes determining removal difficult. It was possible for some compounds like cyproterone acetate to show a significant difference between the final effluent (pre- GAC) and post GAC and that only achieved when p=0.1 and calculating the average based on 1/10 LOD for these concentrations below their limit of quantification.

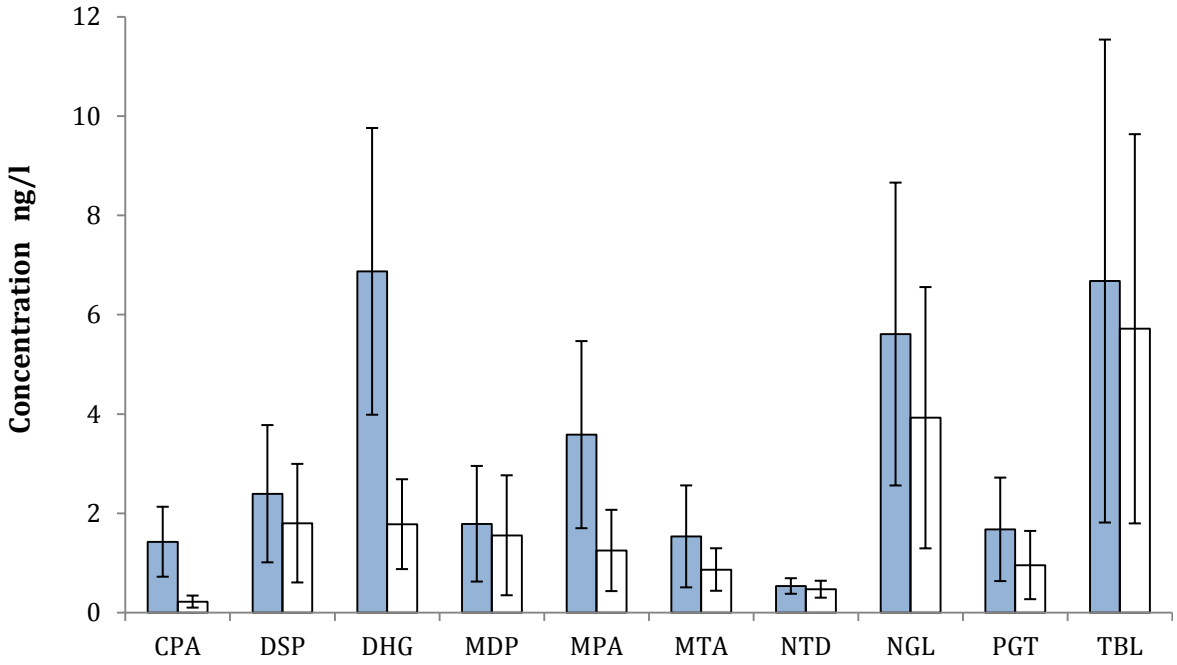


Figure 5.3 Removal of progestogens by GAC (■ Final Effluent (Pre-GAC), □ Post-GAC).

5.2.3 Removal by chlorine dioxide.

The Figure 5.4 shows the occurrence and removal of progestogens through the chlorine dioxide treatment. The ranges of the mean concentrations were from 0.6 ng/l for norethindrone to 2.3 ng/l for drospirenone for the post ClO_2 treatment. The mean for each compound was calculated based on ½ LOD for these values below LOQ. Although some removal had occurred in tibolone and dydrogesterone, however, there was no significant difference p=0.05 had occurred for these pharmaceuticals between pre and post ClO_2 in the chlorine dioxide system. Again analytical uncertainty or few numbers of samples led to make determination of the removal efficiency for this technology difficult. For example each compound like drospirenone or progesterone had five values below the limit of quantification and only three values were above that, thus removal is calculated using very few measured values.

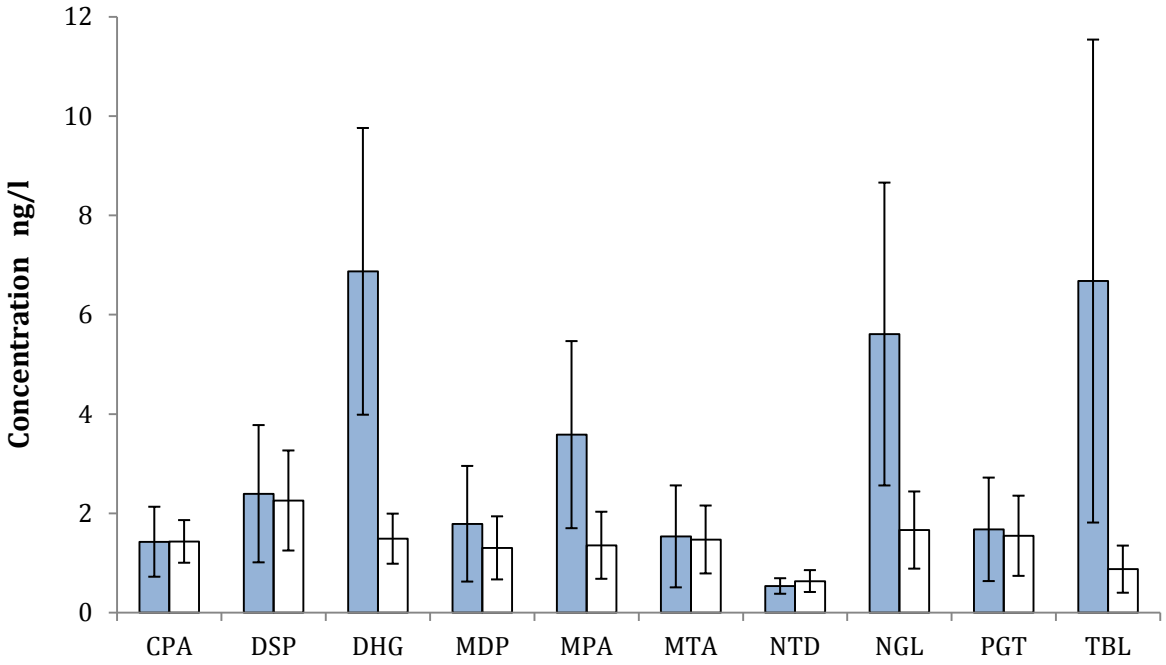


Figure 5.4 Removal of progestogens by chlorine dioxide (■ Final Effluent (Pre-ClO_2), □ Post- ClO_2).

5.3 Occurrence and removal of benzotriazoles in the advanced treatment process.

For benzotriazoles, during November 2008, one litre of each sample and 1ml of internal standards was added to the sample. After analysing and the running the samples, the concentration of each sample was out of the instrument (LC (ESI^+)/MS/MS) range of the calibration standards. The decision was made to sample 100ml (one in the morning and one in the afternoon) for one day from the Hallam Fields STW with 0.1ml of the internal standards to be within the calibration curve. Samples were collected by STW staff and sent to Brunel University on 16th of September 2009. All the original data for benzotriazoles are in appendix 2.

5.3.1 Removal by ozone.

The Figure 5.5 shows the concentrations of the benzotriazoles pre - and post ozone as an advanced treatment. Because a limited number of samples, it was difficult to examine if there was any significant difference had occur, however it was clear that there was a removal with a 74%, and the mean concentration of BT dropped from 517 ng/l to 136 ng/l. Correspondingly, there was some evidence for removal of TT during the ozone treatment, but again that was not significantly proved due to the limited number of sample, however the concentration of TT had reduced from 2,253 ng/l to 467 ng/l achieving 79% removal efficiency.

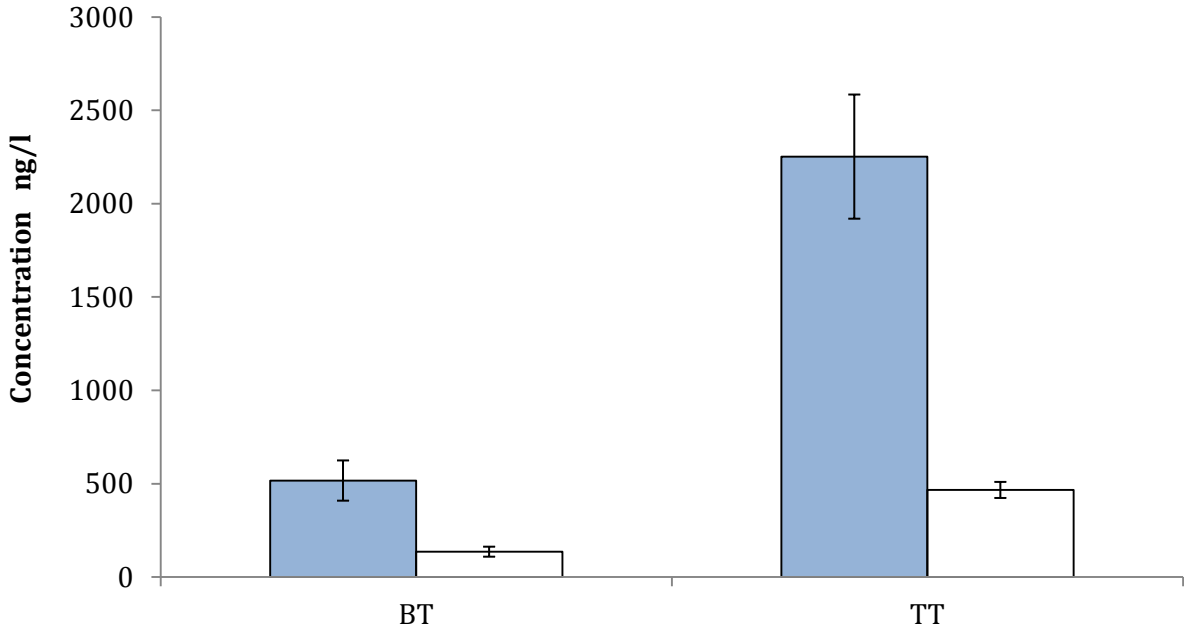


Figure 5.5 Removal of benzotriazoles by ozone (■ Final Effluent (Pre-O_3), □ Post-O_3)

5.3.2 Removal by granular activated carbon (GAC).

The Figure 5.6 shows the occurrence and removal of benzotriazoles for the pre- and post GAC treatment. It can be seen that a clear reduction was achieved by GAC, since the concentration of BT decreased from 517 ng/l to 50 ng/l presenting a removal efficiency of 90%. Interestingly, an excellent removal had occurred for TT with 98% removal efficiency and the concentration decreased from 2,253 ng/l to 51 ng/l. The removal of TT looked higher than BT and this is probably due to the properties of each compound, since TT is more hydrophobic than BT and therefore a high removal was achieved during GAC system.

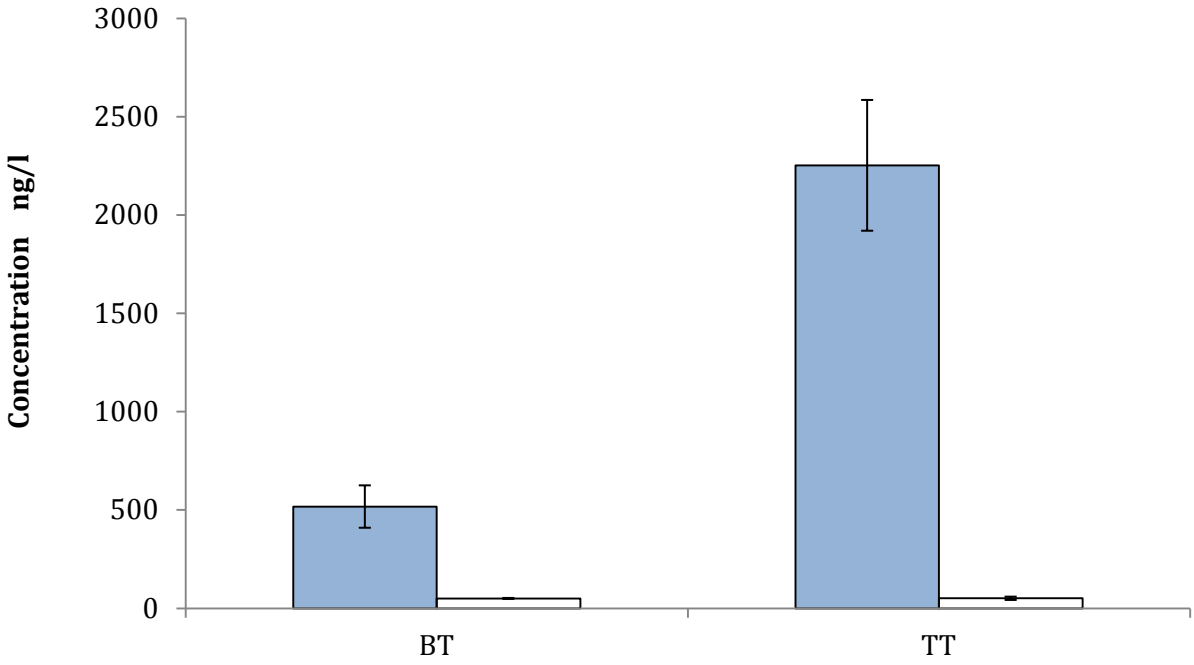


Figure 5.6 Removal of benzotriazoles by GAC (■ Final Effluent (Pre-GAC), □ Post-GAC).

5.3.3 Removal by chlorine dioxide.

The occurrence and removal of benzotriazoles through the chlorine dioxide treatment are shown in Figure 5.7. It is apparent that there was a decrease in the BT concentration from 517 ng/l to 40 ng/l achieving 73% removal efficiency, however it was difficult to determine whether there was a significant difference or not due to the limited number of samples. In contrast, there was some evidence for a significant difference had occurred through ClO_2 system for tolyltriazole. Therefore, it can be seen that a clear reduction to the pre chlorination concentrations from 2,253 ng/l to 469 ng/l as a post chlorination concentration.

Table 5.1 summarises the removal efficiencies for the three technologies used as an advanced treatment to remove BT and TT from the final effluent. It is apparent that GAC was the most efficient technology among these technologies to remove these compounds.

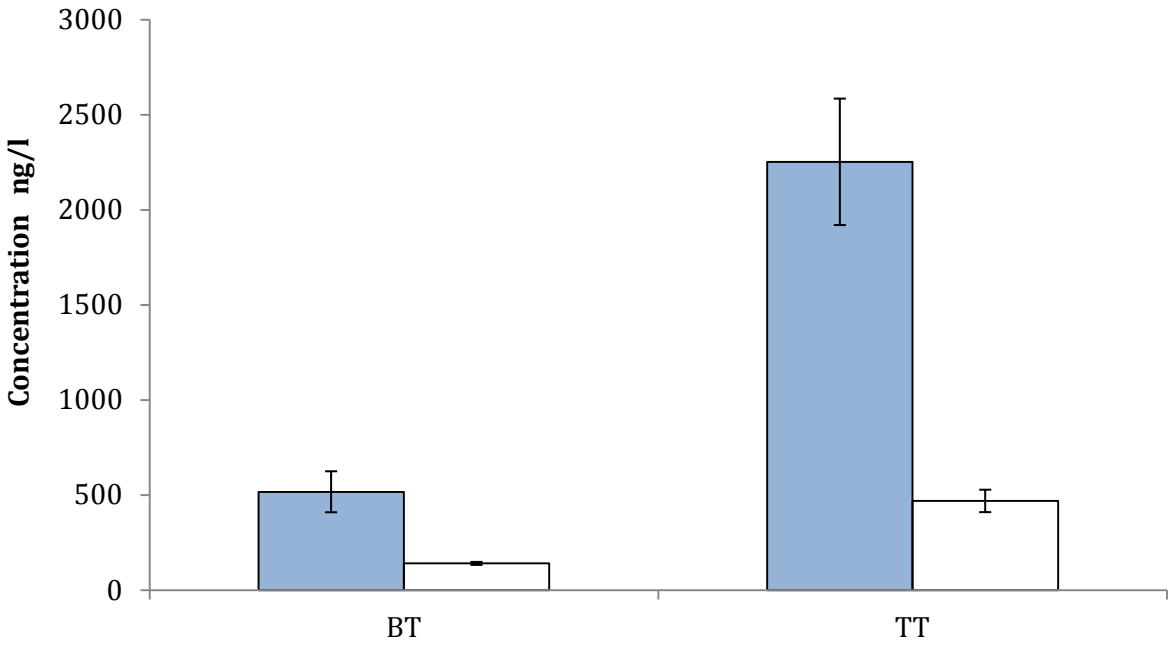


Figure 5.7 Removal of benzotriazoles by chlorine dioxide (■ Final Effluent (Pre-ClO_2), □ Post- ClO_2).

Table 5.1 Removal efficiencies for BT and TT through three different technologies.

	Removal Efficiency %	
Advanced Treatment	**BT**	**TT**
Ozone (O_3)	74%	79%
Granular activated carbon (GAC)	90%	98%
Chlorine dioxide (ClO_2)	73%	79%

Summary

This chapter illustrated the occurrences of progestogens and benzotriazoles during the advanced treatment process: ozone, granular activated carbon, and chlorine dioxide at Hallam Fields STW. In general, it is unlikely that progestogens would not be removed during the advanced treatment, while BT and TT would be removed. Although blanks of progestogens were subtracted from the values of each sample concentration, and because of high concentrations of progestogens in blank samples, it was possible that might lead to increase the uncertainty with the measured concentrations. Further study to observe the fate and behaviour of these compounds in the river water will be undertaken in the next chapter.

Chapter Six: The Fate of Progestogens and Benzotriazoles in the Laboratory Degradation Test

Overview

This chapter will illustrate the behaviour of progestogens and benzotriazoles during the laboratory degradation study. Two tanks were filled with river water and preliminary analysis had shown that BT and TT concentration were enough to study degradation. The original concentrations of progestogens were very close to the quantification limit, therefore tanks were spiked only with progestogens analysis in order to make the concentration of progestogens quantifiable and consequently be able to study the fate of them during the two weeks.

6.1 Basic parameters and initial concentrations.

The parameters measured during the study were temperature, pH, and dissolved oxygen (DO). The volume of river water in each tank was around 60l. All these parameters were measured once a day in each tank. Table 6.1 shows the volume of each tank and the mean of each parameter measured during two weeks. The initial temperature was 19 °C and the initial DO was 5 mg/l. Tanks were left for 24 hours to warm up and to stabilise the dissolved oxygen concentration and during that period temperature and DO were measured (after 8 hrs) and they were 21.2 °C and 7 mg/l respectively. The temperature was stable during the first ten days of the test and then there was gradually increasing in the temperature to reach 25 °C. The DO concentration varied from 7.0- 8.7 mg/l and this indicated that the tanks remained aerobic.

The measured concentration of progestogens in ng/l at each tank on Day=0 are listed in Table 6.2. The samples of these concentrations were taken after one hour of spiking the tanks with progestogens in order to quantify these compounds. There was no need to spike the tanks with benzotriazoles; where the initial concentrations of BT in tank 1 and 2 were 655 ng/l and 650 ng/l respectively, while the initial concentrations of TT were 2,370 ng/l in tank 1 and 2,365 ng/l in tank 2.

Table 6.1 Main measured parameters throughout the test (n=15).

Parameter	Units	Tank 1	Tank 2
Temperature (STDEV)	°C	22.5 (±1.9)	21.8 (±2.0)
pH (STDEV)	-	8.5 (±0.51)	8.5 (±0.53)
Dissolved Oxygen (STDEV)	mg/l	8.0 (±0.36)	8.0 (±0.24)

Table 6.2 Nominal concentrations for progestogens.

	Compounds (ng/l)									
	CPA	**DSP**	**DHG**	**MDP**	**MPA**	**MTA**	**NTD**	**NGL**	**PGT**	**TBL**
Tank 1	538	663	532	646	548	568	507	532	401	662
Tank 2	509	645	499	623	540	551	495	483	420	627

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6.2 Degradation of progestogens

The degradation of progestogens demonstrated differences between individual compounds. Some compounds were readily degraded and some were more recalcitrant. It is possible to split the ten compounds studied onto three groups according to their degradation rates, beginning with the natural hormone, progesterone, which was degraded rapidly, along with MDP and DHG. Figure 6.1 shows the concentration of progesterone over a period of 14 days and each point represent the average of concentrations in the two tanks. It can be seen that the initial measured concentration was 410 ng/l in the tank and after 24 hours of aeration and mixing (at 20 °C), the concentration had reduced to 6 ng/l. Further decrease in the concentration of PGT had occurred in the next 24 hours, where the concentration had reduced to 4 ng/l. During the remaining period of the test, the concentrations of PGT were fluctuated between below the limit of quantification or just above it, probably because of either analytical uncertainty or other compounds like medroxyprogesterone (MDP) might be metabolised to produce PGT and that will increase the concentration of PGT to be above the LOQ and then reduced again to be less than LOQ.

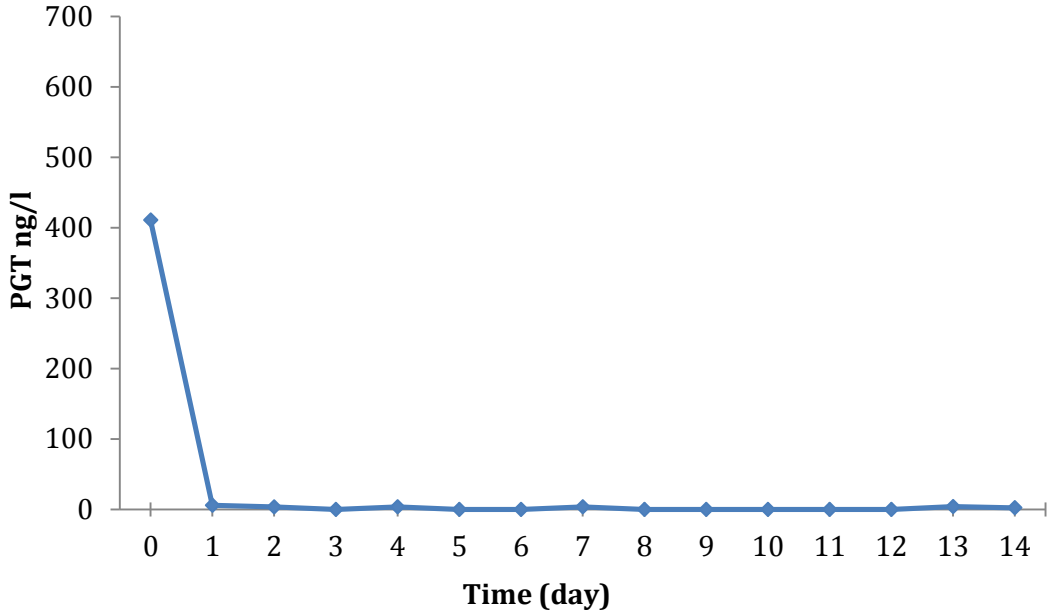

Figure 6.1 Concentration of progesterone throughout the 14 day test.

Medroxyprogesterone and DHG also showed degradation during the study, where the concentrations had reduced from 515 ng/l and 635 ng/l for MDP and DHG to 5 and 2 ng/l in 14 days achieving in that a degradation of 99.7% and 99.1% respectively. The concentration profiles of these compounds are shown in Appendix 3.

The second group of compounds represented by NTD, NGL, TBL, and MTA showed a slower degradation during the study. It can be seen from Figure 6.2 that the mean concentration from the two tanks of norgestrel (NGL) dropped from 508 ng/l to 19 ng/l throughout the 14 day of the test. The other graphs of the other compounds are illustrated in Appendix 3.

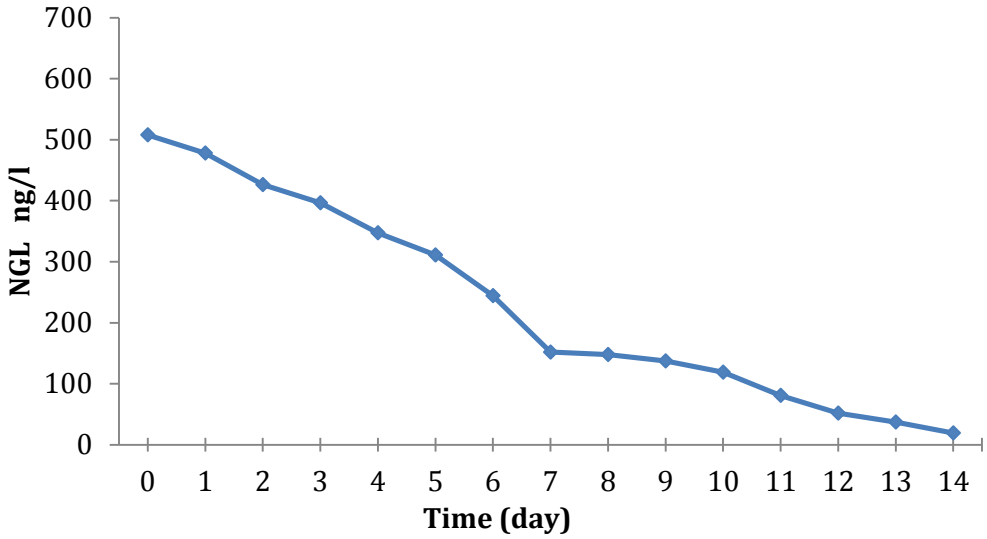

Figure 6.2 Concentration of norgestrel throughout the 14 day test.

Cyproterone acetate (CPA), MPA, and DSP as a third group showed least degradation during the study. For example, the initial concentration of CPA was 524 ng/l and after 14 days, it was 85 ng/l. Figure 6.3 demonstrates the behaviour of CPA for two weeks throughout the test. Each point in the graph represents the mean of the concentration from the two tanks. The other compounds figures of this group are illustrated in Appendix 3.

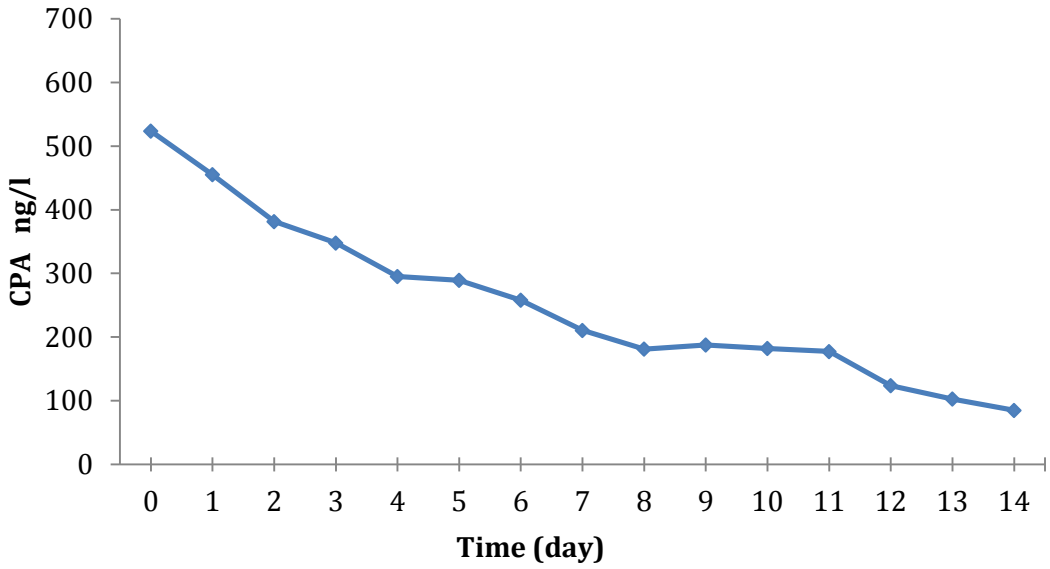

Figure 6.3 Concentration of cyproterone acetate throughout the 14 day test.

As previously explained in chapter three, the half life of each compound was calculated according to the equation:

Half Life ($t_{1/2}$) = ln (2)/$K_{tot.}$ …….. (1)

Table 6.3 shows the half lives of progestogens calculated according to equation (1) and also the degradation percentage of these compounds. Therefore, these compounds were divided into three groups according to their half lives. The first group which are PGT, MDP, and DHG, the half lives were less than 2 days. It can be seen also that the 99% of PGT was degraded during the first 24 hours of the test, therefore it was not possible to calculate an accurate half life for the PGT and that because of a single data point was measured. Also the results showed that more than 90% was degraded in the 2nd and 7th day of the study for MDP and DHG respectively.

The second group of compounds represented by NTD, TBL, NGL, and MTA, they demonstrate a half life between 2 to 4 days. It is apparent from Table 6.3 also that more than 90% of these compounds were degraded after 11 days for NTD and 12 days for the rest of this group. It also apparent from that the degradation rate according to the percentage degraded everyday was similar for most of these compounds in this group.

The third group represents by DSP, MPA, and CPA demonstrated least degradation and therefore the half life for each compound was more than four days (Table 6.3). It is also can be seen from the table that more than 90% of DSP was degraded after 13 days of the test, while less than 90% was degraded for MPA and CPA throughout the test.

Table 6.3 Degradation percentages and half lives for progestogens.

		Chemical Degradation %									
		PGT	MDP	DHG	NTD	TBL	NGL	MTA	DSP	MPA	CPA
Day	0	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**
	1	99	74	45	15	5	6	16	15	13	13
	2	99	93	69	23	13	16	27	29	27	27
	3	>99	97	73	31	24	22	45	50	42	34
	4	99	98	78	37	35	32	48	57	46	44
	5	>99	99	82	51	43	39	55	64	49	45
	6	>99	99	88	60	60	52	62	69	56	51
	7	99	99	90	70	70	70	69	73	65	60
	8	>99	>99	93	75	71	71	73	82	71	65
	9	>99	>99	97	80	77	73	76	82	74	64
	10	>99	>99	99	84	80	77	79	82	78	65
	11	>99	>99	99	92	85	84	85	84	81	66
	12	>99	>99	99	94	92	90	90	87	88	76
	13	99	99	99	96	94	93	91	89	89	80
	14	99	>99	99	99	97	96	94	92	89	84
Slope (K)		-2.52	-0.673	-0.378	-0.273	-0.233	-0.217	-0.189	-0.17	-0.163	-0.12
R Square		0.92	0.861	0.956	0.922	0.937	0.935	0.977	0.98	0.983	0.957
Half life(day)		<1	1.03	1.83	2.54	2.97	3.19	3.67	4.08	4.25	5.78

6.3 Degradation of benzotriazoles

For benzotriazoles, there was no need to spike the tank with BT and TT and that because of the preliminary analysis had shown that there was enough concentration of BT and TT to test their degradation. Therefore, each sample was 200 ml taken from each tank each day throughout the 14 day test and 1ml of internal standards was added to the sample. Figure 6.4 shows the fate of BT through two weeks of study. Each point represents the measured concentration of each day from each tank. From Figure 6.4, the results show that the initial concentration of BT was 655 ng/l and 650 ng/l for tank 1 and 2 respectively. During the two weeks, the concentrations of BT were around these values with RSD of 4.1% and at the end of the test, there was a slightly increase in the BT concentrations and that probably due to the increasing in the temperature that could have caused evaporation, although these tanks were covered with a lid, might increased the concentrations. Original data are available in Appendix 3.

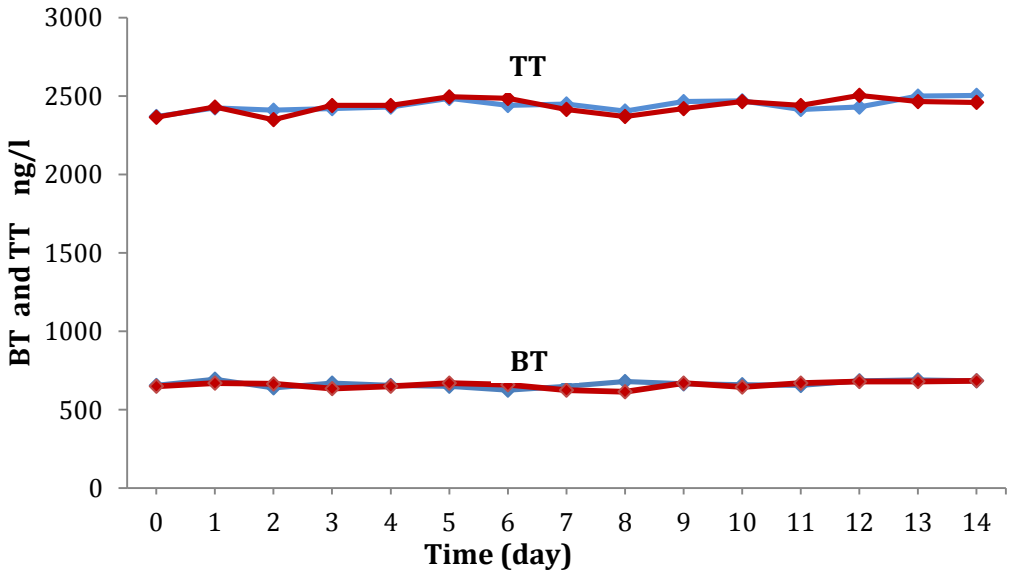

Figure 6.4 Concentrations of BT and TT in each tank throughout the 14 day test (tank 1 and tank 2).

In the mean while, the concentration of TT was 2,370 ng/l for tank 1 and 2,365 ng/l for tank 2. For the period of the test, TT concentrations were around the initial concentration with RSD of 9.85% except the last four days of the test when a slight increase in the TT concentration had occurred, probably because of the temperature. Also it is apparent that both tanks had the similar pattern during the test (Figure 6.4).

Figure 6.5 shows the mean concentrations of each compound that had been taken from each tank. It is apparent that the proportional between BT to TT was consistent. Benzotriazole and tolyltriazole showed no degradation had occurred throughout the test.

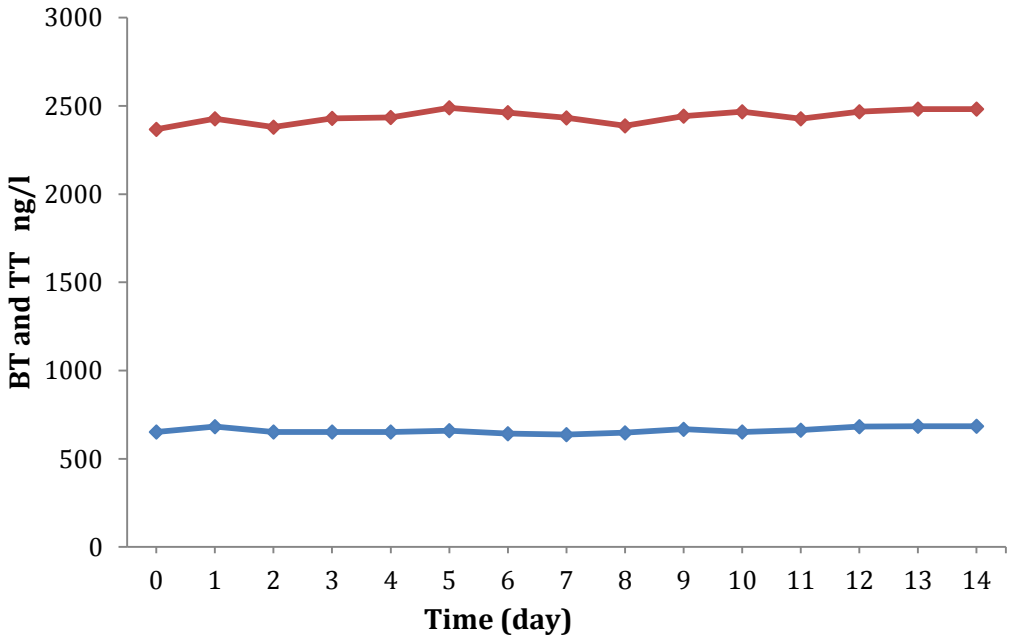

Figure 6.5 Fate of benzotriazole and tolyltriazole throughout the 14 day test (BT and TT).

Summary

This chapter illustrated the fate and behaviour of progestogens and benzotriazoles in the river water. The results show that progesterone was degraded rapidly, while the other compounds had a different degradation rates, and it was possible to categorise the progestogens into three groups according to their half life:

- Group 1: half life of PGT, MDP and DHG < 2 days.
- Group 2: half life of NTD, TBL, NGL and MTA between 2-4 days.
- Group 3: half life of DSP, MPA and CPA > 4 days.

For benzotriazoles, the results show no degradation had occurred throughout the test. These results for progestogens and benzotriazoles had led to investigate the occurrence of these compounds in the River Erewash, modelling the Trent catchment and Thames catchment to explore these compounds occurrences in wider area, and that what will be described in the next chapter.

Chapter Seven: Results of Monitoring and Modelling of Progestogens and Benzotriazoles in River Erewash, Trent and Thames Catchments

Overview

This chapter discusses the occurrence of progestogens and benzotriazoles in the River Erewash on the River Trent and its catchment, also the River Colne (tributary of the River Thames). Two sampling campaigns were achieved (each one represented by 24 sampling points including the effluent from eight STWs existing on the River) to observe the occurrences of progestogens and benzotriazoles in the River Erewash. A single grab sample was collected from the River Colne in order to investigate the wider occurrence of these compounds. In addition, the modelling of River Erewash and the River Trent catchment area which is where the Erewash located are described in this chapter. Thames River catchment area was also modelled to evaluate wider occurrence in the UK.

7.1 Details of selected sewage treatment works.

The types of the sewage treatment works and process details that were sampled during the survey are described in Table 7.1. During the sampling, the mean temperature river sample was 3.0 °C and 12.2 °C for January and October 2009 respectively. Average temperature of the final effluent samples was 7.8 °C in January and 15.3 °C in October.

Table 7.1 Types of treatments existed in the eight STWs on the River Erewash.

STWs	Population	DWF (m^3/d)	Type
Kirkby STW	24,538	5,970	(SAS) N/ASP + ferrous chloride dosing
Pinxton STW	8,166	4,242	(TA2) N/ASP + TFs + SFs + ferrous chloride dosing
Pye Bridge STW	7,043	2,048	(TB2) TFs + SFs + ferric sulphate dosing
Milnhay STW	25,957	7,050	(TA2) N/ASP + TFs + SFs + ferric sulphate dosing
Newthorpe STW	43,382	10,282	(TB2) TFs + SFs + ferric sulphate/PAC dosing
Hallam Fields STW	44,895	10,022	(TA2) N/ASP + SFs + ferrous chloride dosing
Stapleford STW	26,203	5,737	(SAS) N/ASP + ferrous chloride dosing
Toton STW	58,584	16,762	(TB2) N/ASP + TFs + SFs + ferric sulphate dosing

7.2 The occurrence of progestogens in the River Erewash.

Two sampling campaigns were conducted in order to observe the occurrence of progestogens in River Erewash. After collecting the samples from the first campaign which comprised of 16 samples along the river and its tributaries, with eight samples of the final effluent of the STWs discharging to the river. Because of the gel permeation clean up problems that occurred, all samples from the first survey were lost due to the low pressure in the auto sampler and that led to collect the wrong fraction. Consequently, there was no signal could be seen when the samples were run by LC (ESI^+)/MS/MS. The second sampling campaign was undertaken in October 2009. The same regime was followed as for sampling points and STW. There was high degree of uncertainty associated with the results and this uncertainty resulted from the values of the concentration of these compounds. There was no rain at that time which led us to expect progestogens concentrations to be relatively high in the river. Table 7.2 shows the concentrations of progestogens discharged from eight STWs existing on River Erewash and the concentrations of progestogens were fluctuated around the limit of their quantification, however Pye Bridge STW was the highest in terms of discharging the progestogens among the STWs. Table 7.3 shows the concentrations of progestogens along the River Erewash in October 2009. It can be seen that only drosprinone and norgestrel were above their limit of quantification, while the rest of these chemicals were close to or just above their limit of quantification and some of them like the natural hormone (PGT) was below the limit of quantification along the Erewash River. Because they were unlikely to be detected and quantified, it was decided not to model these chemicals in the catchments or to monitor in the tap water.

Table 7.2 Concentrations of progestogens (ng/l) discharged from eight STWs existing on Erewash River.

STWs	Effluent from STWs ng/l									
	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
Kirkby (N/ASP)	< 0.2	2.2	2.3	1.3	0.6	< 0.4	< 1	7.2	0.5	< 1.6
Pinxton (N/ASP)	< 0.2	5.7	2.5	< 0.4	1.1	< 0.4	< 1	2.9	< 0.4	12.1
Pye Bridge (TF)	3.3	7.0	39.2	10.4	3.6	3.0	7.0	16.5	6.2	24.4
Milnhay (N/ASP)	9.0	2.4	3.4	< 0.4	0.7	< 0.4	< 1	4.2	< 0.4	7.5
Newthorpe (TF)	< 0.2	2.1	2.9	0.5	1.3	< 0.4	< 1	4.5	< 0.4	9.8
Hallam Fields (N/ASP)	2.2	5.3	3.7	1.3	6.1	0.5	1.8	6.6	< 0.4	10.5
Stapleford (N/ASP)	< 0.2	6.0	2.2	0.8	2.9	< 0.4	< 1	5.4	< 0.4	7.0
Toton (N/ASP)	0.7	4.1	3.0	< 0.4	1.9	< 0.4	< 1	11.4	< 0.4	4.6

Table 7.3 Concentrations of progestogens (ng/l) in Erewash River.

Erewash River	Concentration in Erewash River ng/l									
	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
U/S of Kirkby STW	< 0.2	4.0	3.6	< 0.4	0.8	< 0.4	< 1	4.3	< 0.4	2.5
D/S Kirkby STW	< 0.2	4.4	< 1.6	< 0.4	1.3	< 0.4	1.1	4.3	< 0.4	4.6
D/S Pinxton STW	< 0.2	1.8	3.9	0.6	1.1	< 0.4	< 1	4.2	< 0.4	3.1
D/S Pye Bridge STW	< 0.2	2.0	2.0	0.6	1.1	< 0.4	< 1	3.2	< 0.4	2.1
U/S of Milnhay STW	< 0.2	2.6	2.0	0.6	< 0.4	< 0.4	< 1	3.4	< 0.4	< 1.6
D/S of Milnhay STW	3.1	4.5	2.0	0.5	< 0.4	< 0.4	< 1	4.7	< 0.4	4.1
D/S of Gilt brook confluence	2.2	6.0	4.1	< 0.4	3.5	0.5	< 1	5.7	< 0.4	4.1
U/S of Hallam Fields STW	2.8	2.5	2.1	< 0.4	0.6	< 0.4	< 1	1.7	< 0.4	3.8
D/S of HF STW & Nut brook	1.8	2.5	4.9	< 0.4	1.4	< 0.4	< 1	3.7	< 0.4	4.0
D/S Stapleford STW	1.8	9.6	2.3	0.5	2.3	< 0.4	< 1	5.7	< 0.4	1.7
D/S of Toton STW	2.0	5.0	< 1.6	< 0.4	< 0.4	< 0.4	< 1	4.4	< 0.4	7.2

7.3 Occurrence of benzotriazoles in River Erewash

7.3.1 The first survey in January 2009

The flow rate of the River Erewash on 6th of January was 0.846 m^3/sec and there had been no rain, indicating that the concentrations of benzotriazoles were also likely to be around their highest. The concentrations of benzotriazoles in the effluent samples from eight STWs existing on River Erewash are illustrated in Figure 7.1. It is apparent that the BT and TT were present in all the effluents of STWs. The treatment type, activated sludge or biological filters with or without tertiary treatment, had no clear relationship with the effluent concentrations. It can be seen that the concentration of BT was in the range from 840 ng/l at Kirkby STW to 3,605 ng/l at Toton STW with an average of 2,033 ng/l. In order to calculate the fluxes that were discharged from these STWs to the River Erewash, concentration of BT from each STW was multiplied by the discharge per day from each STW, and the results represent the total BT from each STW discharged to river. The summation of fluxes from eight STWs was 143 g/d. For tolyltriazole, the mean concentration of TT was 4,465 ng/l ranged from 2,685 ng/l at Kirkby STW to 5,700 ng/l at Hallam Fields STW, thus these STWs were discharging 291 g/d of TT everyday to the River Erewash. The original data are found in appendix 4.

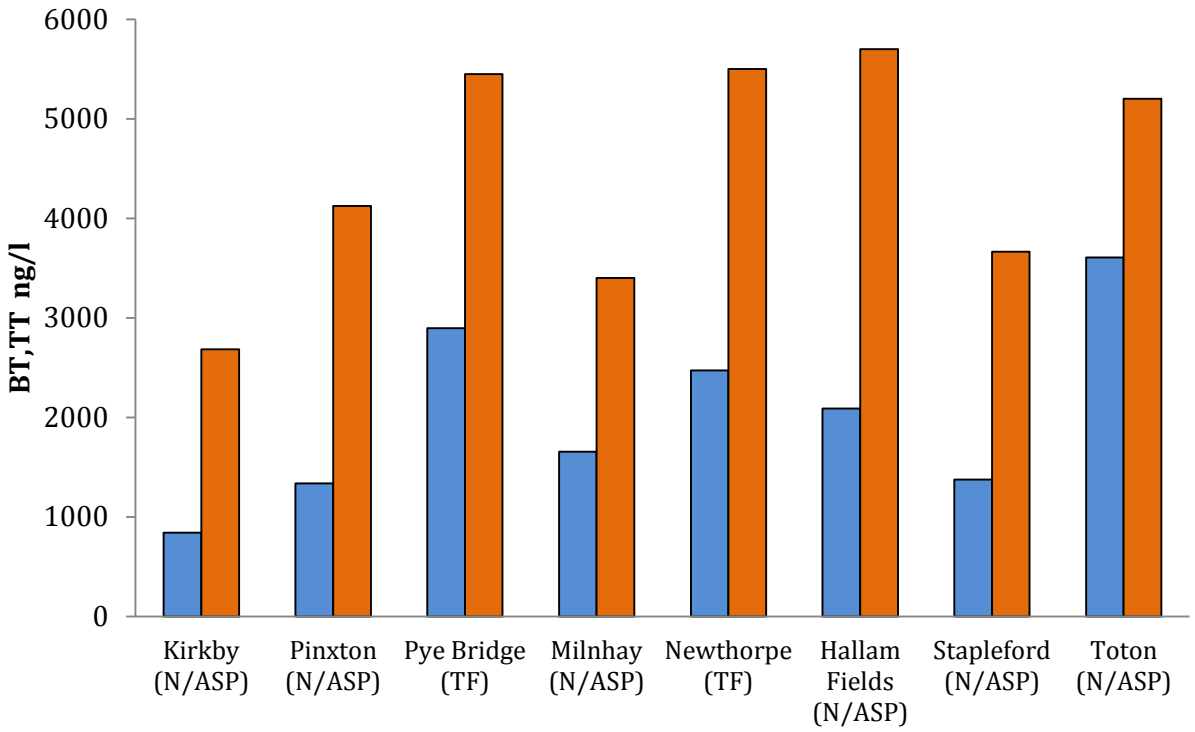

Figure 7.1 Final effluent concentrations of benzotriazoles from STWs, BT, TT, sites are shown as location downstream from the source of the river, left to right on x-axis in January 2009.

The graphical representation of River Erewash showing the trend of benzotriazoles concentrations is illustrated in the Figure 7.2. It can be seen that there was an increase in the concentration of both BT and TT along the River and that concentrations of TT were also higher than BT, which was the same as in effluent from STWs. From the Figure 7.2, it can be seen that there was initial concentrations of BT and TT in the upstream of the river. In addition, these two compounds were present in all samples which were taken from brooks, small tributaries and the main river samples. Upstream the STW on the Gilt Brook was the highest among the tributaries on the Erewash River with 118 ng/l of BT and this means that there is a possibility that some industries or some effluent that discharged their effluent directly to the brook . It is obvious to notice that there was an increase in concentrations after about 2.5 km from the first sampling point and that resulted from the inputs from the first STW, and then the increase was continuous due to the inputs from the other STWs. At the same time, there was a decrease in the concentrations of these

chemicals in the river water at some points and that probably resulted from the confluence of less contaminated tributaries, leading to dilution. Each black arrow represents the input from the STW to the River Erewash, while the blue arrows represent the brooks that existed on the river itself.

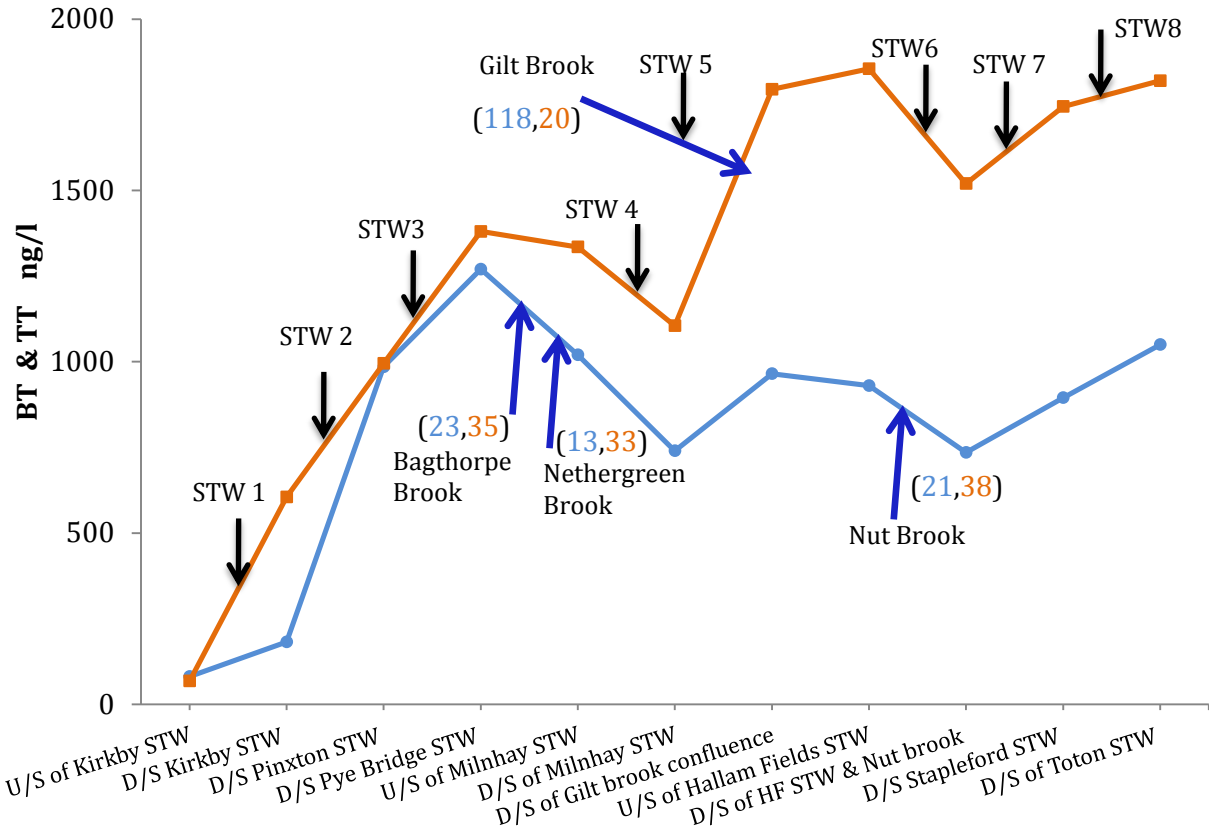

Figure 7.2 Graphical representation of the River Erewash showing concentrations in ng/l of BT and TT increasing downstream from the river source in January 2009.

7.3.2 The second survey in October 2009.

The second sampling survey was undertaken on the 29th of October 2009. There was also no rain on that day and the flow rate was 0.662 m^3/sec. The concentrations of final effluent samples of the STWs existing on River Erewash are illustrated in Figure 7.3. It is apparent that once again, BT and TT were present in all sewage effluent samples. The mean concentration of the effluents from these STWs was 652 ng/l with a range from 496 ng/l to

867 ng/l. While the average concentration of TT was 2,303 ng/l and varying between 2,040 ng/l to 2,730 ng/l. According to the effluent concentration and flow rate from each single STW, the total amount of BT was discharged to the River Erewash was 40.8 g/d and 140 g/d for TT. The original data are found in appendix 4.

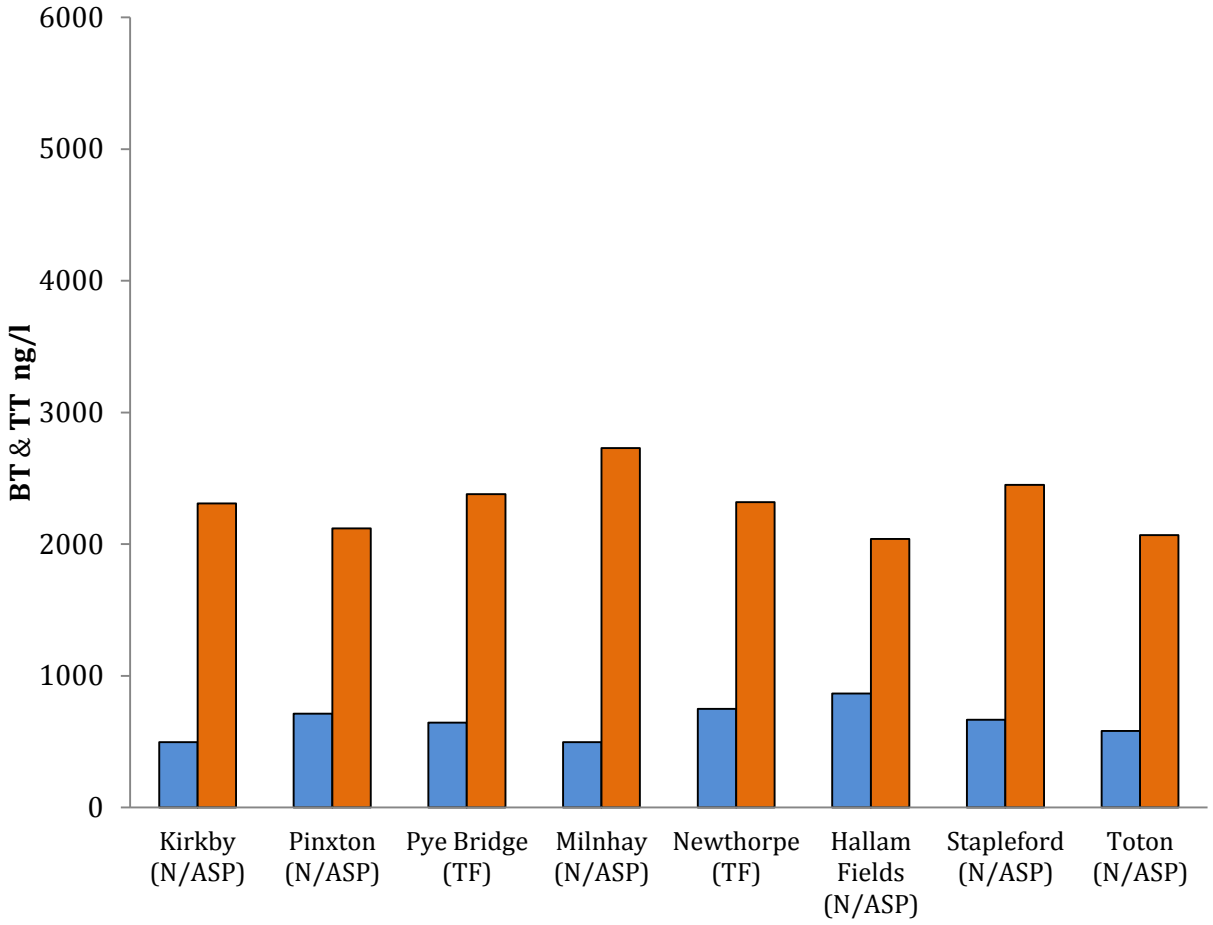

Figure 7.3 Final effluent concentration benzotriazoles from STWs, BT, TT, sites are shown as location downstream from the source of the river, left to right on x-axis in October 2009.

According to the final effluent concentration from these STWs, the geographical representation of BT and TT along the River Erewash is illustrated in Figure 7.4. It can be seen that the concentration of BT at the upstream of the River was 51 ng/l. Because of the inputs from the STWs existed on the river, there was an increasing in the concentrations of BT in the river, but this increasing was only after the first and second STWs, then there was

no wide variation in the concentrations although there was other STWs that discharge their effluent to the river and some tributaries with low concentration which might dilute the concentrations.

For TT, it is apparent that the concentrations of TT were varied from 84 ng/l at the upstream of the river to 1,570 ng/l at some point and that was resulted from the fluxes of the STWs to the River Erewash. In addition, it is obvious to see the impact of these STWs on the trend of the River by increasing the concentration in the river and also the tributaries had an opposite effect to the impact of STWs by diluting the concentrations.

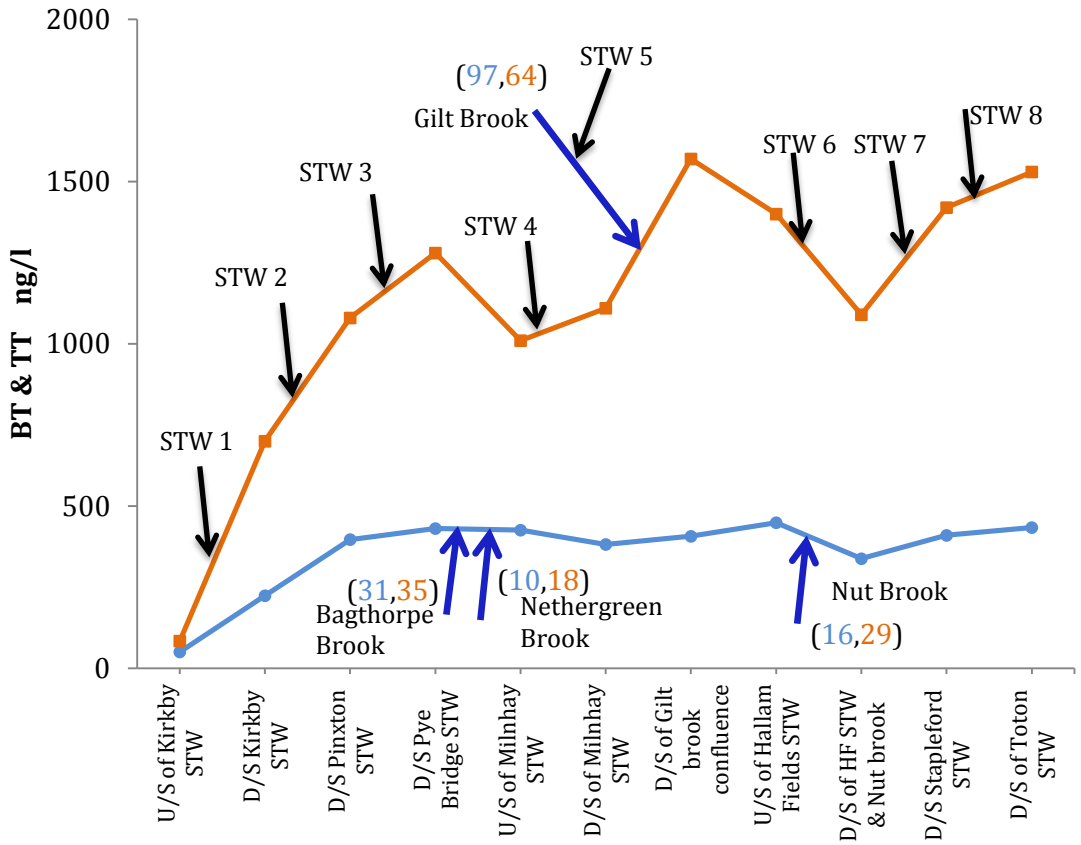

Figure 7.4 Graphical representation of the River Erewash showing concentrations in ng/l of BT and TT increasing downstream from the river source in October 2009.

In order to compare the occurrences of BT in the River Erewash in the two sampling campaigns (January and October), Figure 7.5 shows concentrations of BT along the River

Erewash in the two occasions. It can be seen that the concentrations of BT were similar only in the first two points in the river and that reflect that the background concentration in the upstream was consistent. The concentration of BT was less in October than in January, this could be due to change in use, such as in dishwashing at catering for school lunches, as it was half-term in October 2009. Although the effluent concentration of BT at Kirkby STW in January was higher than in October, however, the trend of the concentration was similar between the first two points. Downstream Pinxton STW, it can be seen clearly the effect of Pinxton STW and then Pye Bridge STW on the BT concentration along the river in January. Because of the dilution that had occurred due to the confluence between the River Erewash and some brooks, it can be seen the influence of these brooks on the BT concentration especially in January , while it is hard to find the effect of these tributaries in the BT concentrations on October.

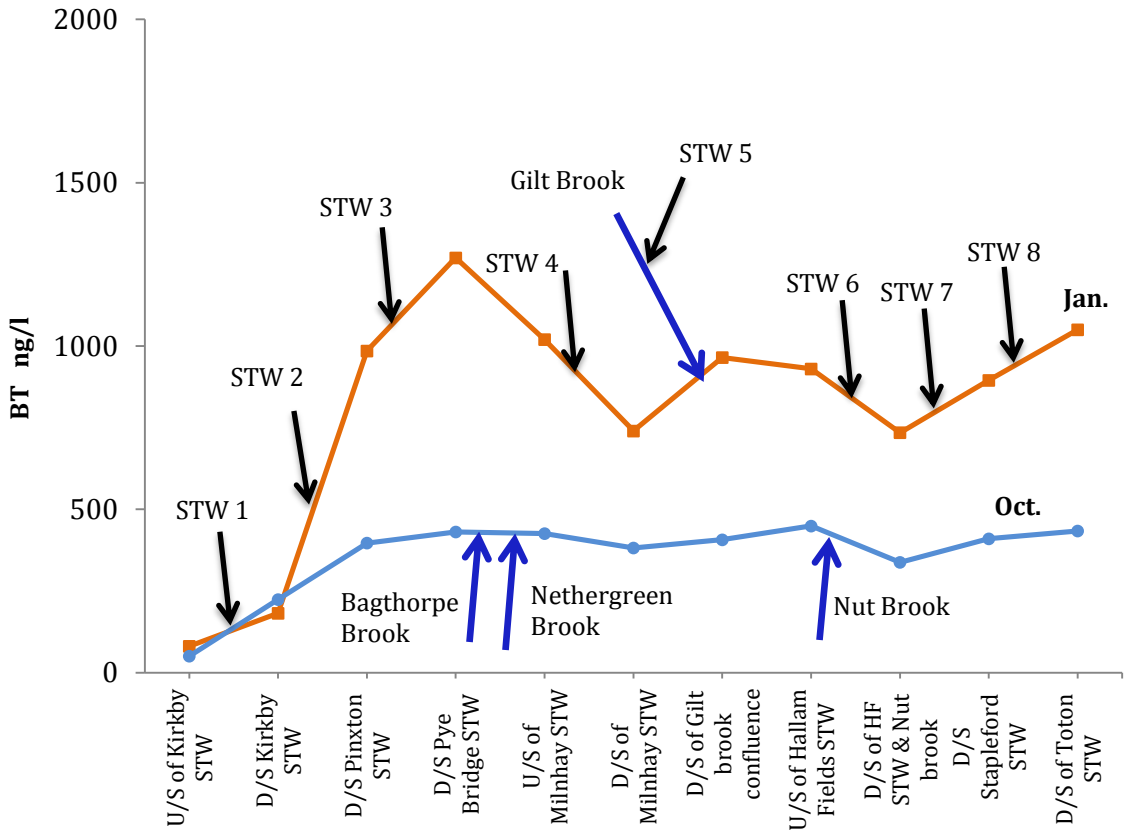

Figure 7.5 Concentrations of BT along the River Erewash in the two occasions, January ( ) and October ( ).

The concentrations of TT were also similar at the upstream of the river as shown in Figure 7.6. The graph also shows that until downstream of Minhay STW, TT concentrations had the same pattern, and then due to the effect of the Gilt Brook and Newthorpe STW discharge to the River Erewash, since the TT concentration in October at Newthorpe STW was half the concentration in January. Therefore as a result of this impact the differences between January and October in TT concentration were consistent between that sampling point and just before the confluence with the River Trent.

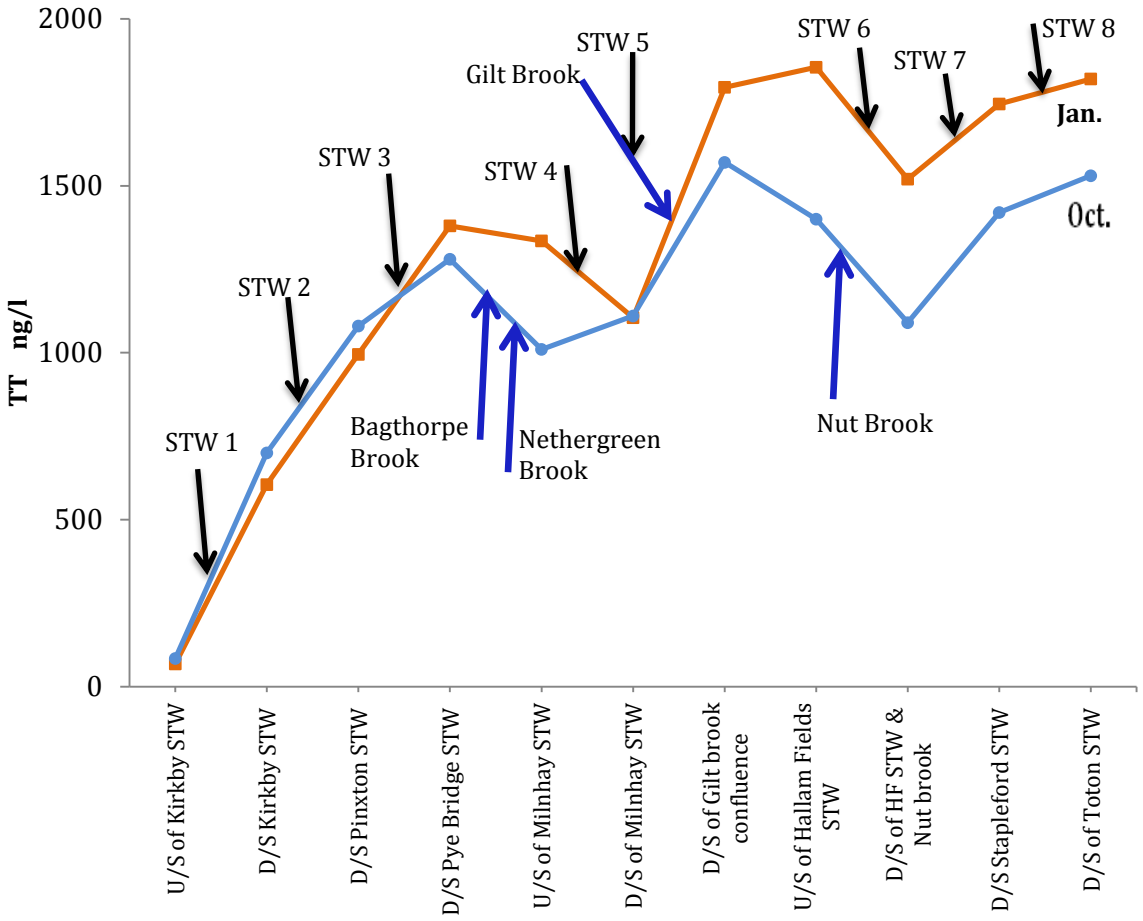

Figure 7.6 Concentrations of TT along the River Erewash in the two occasions, January (■) and October (●).

In general, although the flow rate in the River Erewash was in January higher than the flow rate in October, however the concentrations of BT and TT in the River Erewash looked higher in January than those in October. The reason for that is probably due the effluent

concentrations of these chemicals which was discharged from the STWs to the river was higher in January than those of October occasion, and as long as these chemicals had shown no degradation had occurred in the river from the previous chapter, therefore, it would be expected that BT and TT concentrations in the River Erewash would be higher than October concentrations.

7.4 Modelling of the River Erewash

In order to validate the model, the model was examined in terms of the final effluent concentrations of these chemicals which were discharged from STWs, therefore when using the measured sewage works effluent concentrations to drive the inputs to the LF2000-WQX model, the fit for BT with the observed values slightly underestimated measured concentrations (regression slope = 0.8; R^2 = 0.76; p<< 0.001) as shown in Figure 7.7. In contrast, it was able to reproduce the concentrations seen the river very well for TT, although there was a slight overestimate of the observed values (Figure 7.8). A regression of the observed against predicted TT values gave a line of slope = 1.2 and an R^2 value of 0.88 (p<< 0.001). This indicates that the municipal effluents were the predominant source of these compounds.

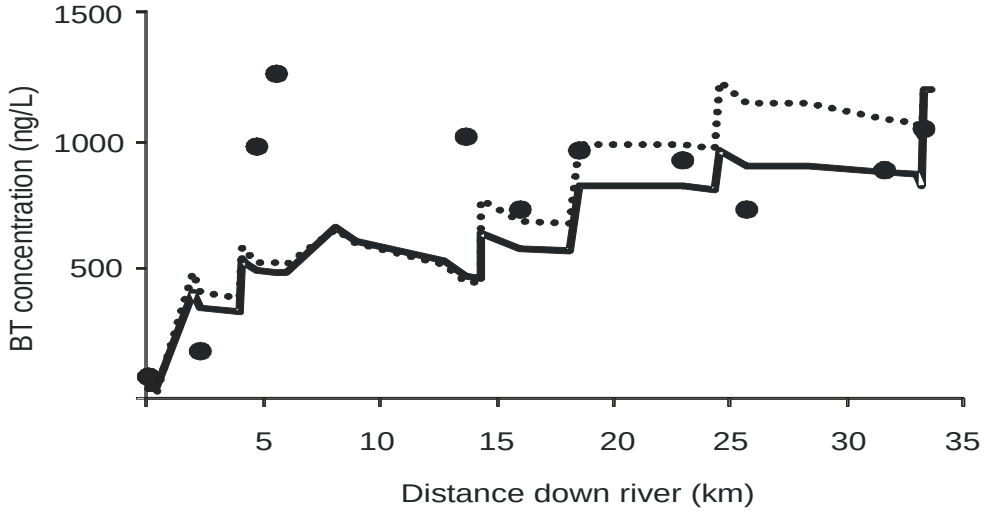

Figure 7.7 Model outputs, showing observed (●) and predicted concentrations of BT along the river. Modelled values were calculated using measured effluent concentrations (solid line) and per capita loads (dotted line).

The model was also tested in terms of per capita load in order to validate it , thus the predicted concentrations calculated based on per capita loads give very similar fits to the observed data as those using the measured data (Figures 7.7 and 7.8). The average underestimate of the observed values was unchanged for BT and consequently the regression statistics was similar. For TT, the regression statistics were almost the same as using the measured effluent data with a slope of 1.3 and an R^2 value of 0.87 ($p << 0.001$). It is clear that there were differences in the simulations although these were not large; the differences arise because per capita load is an estimation of the input from the sewage treatment works.

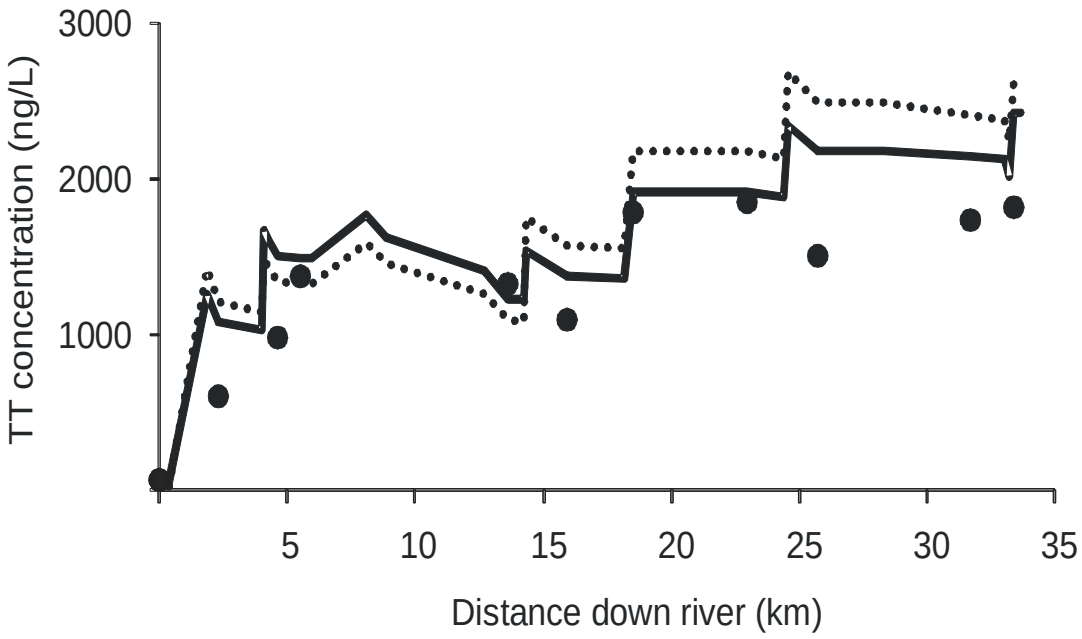

Figure 7.8 Model outputs, showing observed (●) and predicted concentrations of TT along the river. Modelled values were calculated using measured effluent concentrations (solid line) and per capita loads (dotted line).

For catchment modelling, it was not feasible to model the catchment in terms of using the concentrations of these chemicals from final effluent samples of the STWs, this is because these chemicals concentrations were not available from all STWs and for the whole catchment. In contrast, these results indicated that the calculated per capita loads in the

model for making predictions across the Trent catchment or Thames catchment would be a reasonable method for developing effluent concentrations at the unmeasured sites.

7.5 Catchment modelling

In order to save costs and resources of sampling and analysing many samples across the catchment, modelling was used to produce the concentration of benzotriazoles in the entire catchment by simulating the river Erewash to its catchment (Trent River catchment) based on per capita load. In addition and as long as there is no difference between the uses of these compounds in the area around River Trent catchment and the River Thames catchment, and also the operation types of the STWs was similar, therefore it was able to model the Thames catchment and the result was compatible with the real sample collected from one of the River Thames tributaries.

7.5.1 Trent River catchment modelling.

Across the River Trent catchment showed higher predicted mean concentrations of TT than of BT (Figure 7.9) because of its larger per capita input loads. Approximately 28% of river reaches showed BT predicted concentrations between 401 – 1,000 ng/l and 62% less than 400 ng/l for the upstream areas and less impacted area by STWs inputs including the main river Trent for most of its length as shown in Figure 7.10. For TT, 45% of concentrations were predicted to be between 401 – 1,000 ng/l with 90% less than 2,000 ng/l. The highest concentrations occurred just below sewage treatment works. The average concentrations of BT and TT across all the river reaches immediately downstream of a sewage treatment works discharge were 1,080 ng/L and 2,160 ng/l respectively (Figure7.9).

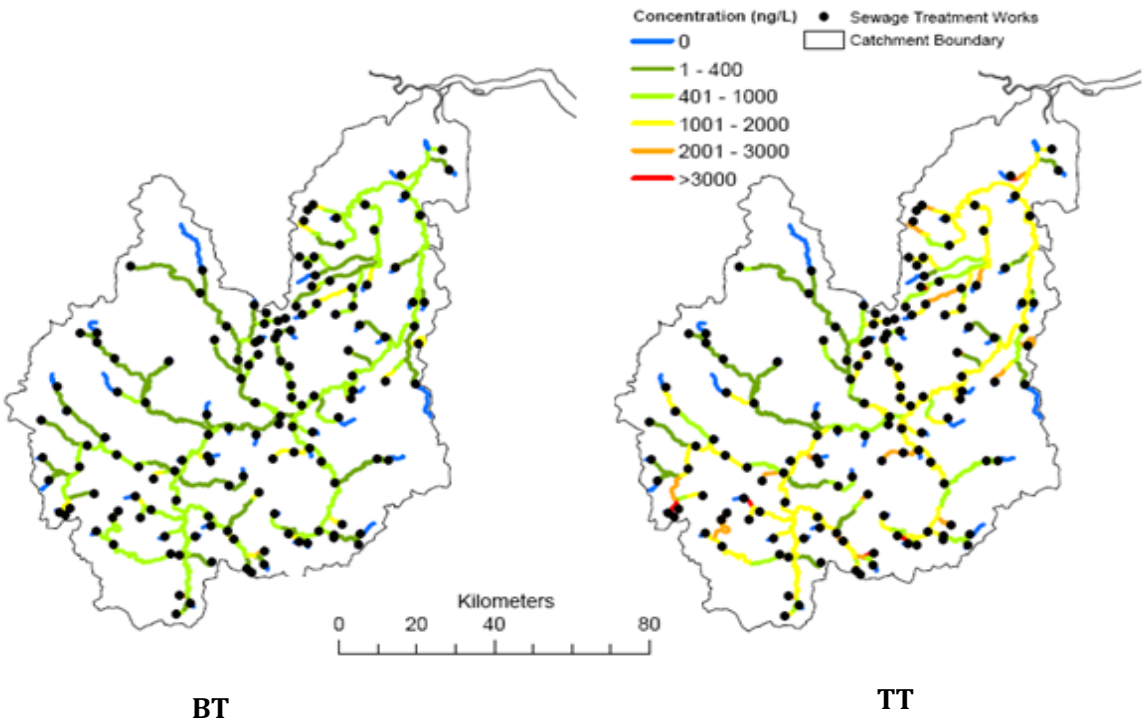

Figure 7.9 Model outputs for the River Trent catchment scale concentrations of benzotriazole and tolyltriazole (ng/l).

7.5.2 Thames River catchment modelling.

The purpose of modelling the Thames River catchment was to give an indication of the concentrations of BT and TT that might be found in the source water to water treatment works, in particular at Iver drinking water treatment which abstracts from the River Thames. Therefore a single grab sample taken from the River Colne, a tributary of the River Thames, in September 2010 to compare the average modelled values (BT 337 ng/l and TT 508 ng/l) and measured values of 224 and 453 ng/l respectively and the results were compatible. Across the catchment, concentrations of BT were predicted to range from 1 to 400 ng/l below the first STW inputs, with increasing values of 401 to 1,000 ng/l. While for TT, concentrations were predicted to range from 1 to 1,000 ng/l and frequently in the 1,000 to 2,000 ng/l range in the lower reaches as shown in Figure 7.10.

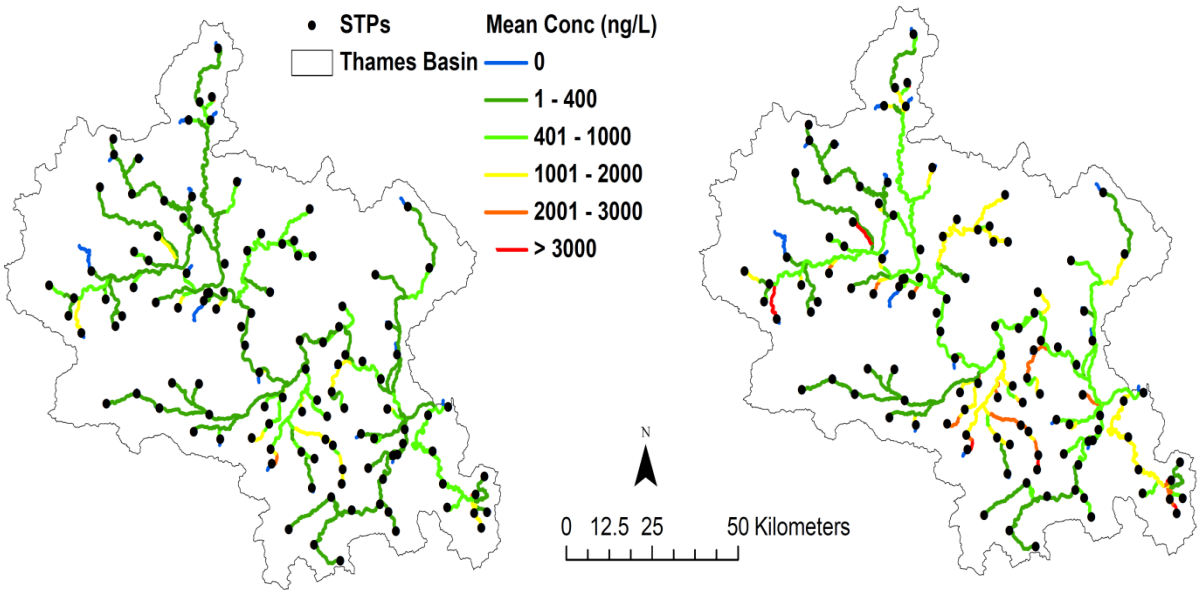

Figure 7.10 Model outputs for the River Thames catchment scale concentrations of benzotriazole and tolyltriazole (ng/l).

Summary

This chapter illustrated the concentrations of benzotriazoles in the river waters. The modelling shows that these compounds were present in wide area of the catchments (Trent and Thames). Because of benzotriazoles were not degraded in the river water (according to the results in chapter 6), and the waters in these rivers are used as a sources for drinking water treatment, therefore, investigations about the occurrence of benzotriazoles in the tap water will be undertaken in the next chapter.

Chapter Eight: Results of the Occurrence and Possible Source of Benzotriazole and Tolyltriazole in Potable Water

Overview

To date, around the world, many researchers reported some emerging contaminants in drinking water. Therefore this chapter will report the occurrences of benzotriazoles in the tap water around Uxbridge, West London. Although these chemicals are widely used in the producing of many products and then they might end up to the environment like break fluids, motor vehicle antifreeze and aircraft de-icing fluids. However, only one possible input (dishwasher detergents) that might contribute in the occurrences of these chemicals in the tap water will be described in this chapter.

8.1 Occurrence in drinking water

During the study to find the occurrence of benzotriazoles in the tap water, BT and / or TT were detected in all tap water samples analysed. Figure 8.1 shows Box plots of concentrations of BT and TT in tap water, median concentrations and the 25-75 percentile range are shown in the box with whiskers at 5 and 95 percentiles. From the Figure 8.1, it can be seen that the concentrations of BT were higher than the concentrations of TT in all samples, the opposite of that seen in the river survey. This is probably due to the fact that TT is more hydrophobic than BT and the granular activated carbon used in the drinking water treatment plant at Iver removed TT more effectively, therefore the TT concentrations were lower that BT concentrations. The mean concentration of BT was 30.9 ng/l ranging from 0.6 ng/l to 79.4 ng/l. While TT mean concentration was 15.1 ng/l and varied from 0.5 ng/l to 69.8 ng/l. The full data are shown in appendix 5.

Throughout the period of monitoring these chemicals in the drinking water, eight samples were collected from the MilliQ water system existed in the laboratory at Brunel University, which represented the blank samples which were also analysed. The MilliQ system at Brunel consists of carbon pre-cartridge followed by reverse osmosis (RO) and then ion exchange and all these three technologies classifies as types of advance water treatment. Therefore, the results show that mean concentrations of BT and TT were 1.2 and 0.5 ng/l

respectively. In addition, 25% and 50% of the values of the blank samples were below the limit of quantification (<0.5ng/l) for BT and TT respectively.

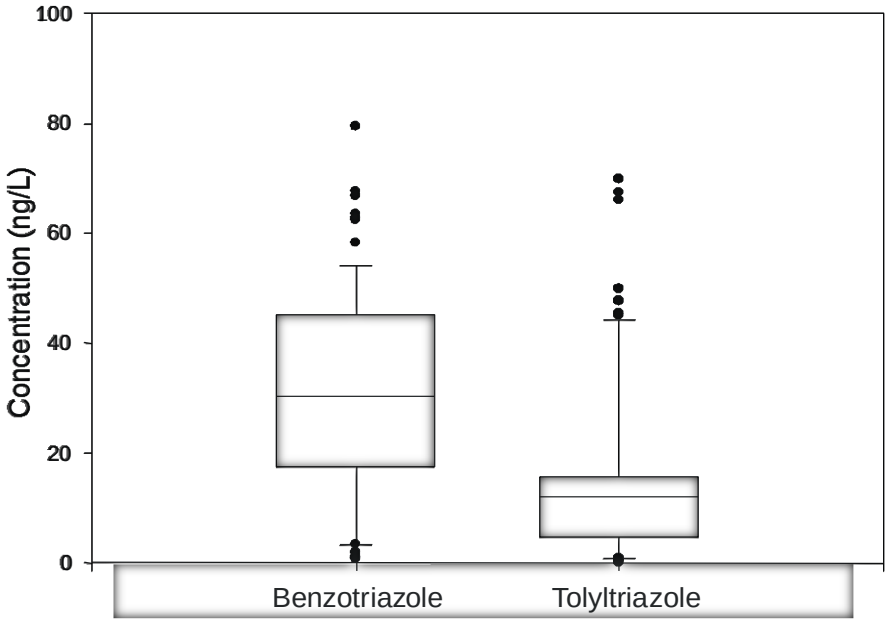

Figure 8.1 Box plots of concentrations of BT and TT in tap water. Median concentrations and the 25-75 percentile range are shown in the box with whiskers at 5 and 95 percentiles.

In terms of estimating the actual human exposure through the tap water, it is possible that taking average concentrations without considering to the spatial distribution might mislead. Therefore, Figure 8.2 shows the change in the concentrations of BT represented by 13 of the 20 locations over a 20 km distance from east to west around Uxbridge, west London. The results show that there was a significant difference between the concentration of BT in west of and Uxbridge, also there was some evidence of difference between the concentrations of BT at Uxbridge and east of it. In addition, the results show that the concentrations of BT in the area of 6-9 Km west of Uxbridge were <10 ng/l, while about 1.5 km around Uxbridge the BT concentrations were between 27.6-58.2 ng/l, and the BT concentrations were ranged from 40-80 ng/l at about 11-14 km to the east of Uxbridge. The other 7 samples excluded from Figure 8.2, four of them were to the north of Uxbridge, and

had between 17 and 29 ng/l. The other three samples were from between 40 to 100 km distant, with 5 to 25 ng/l BT.

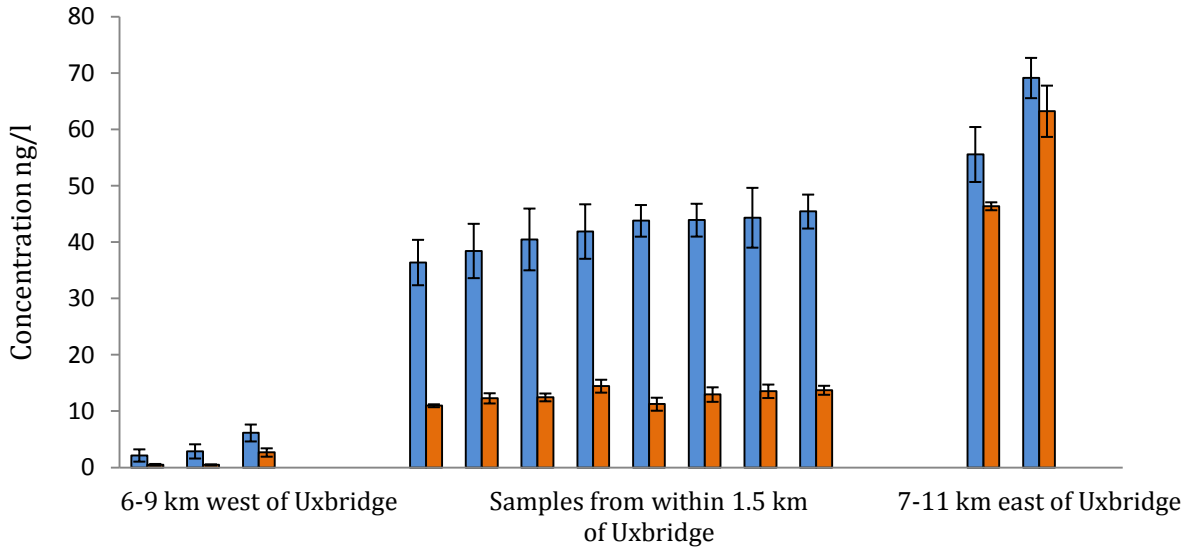

Figure 8.2 Change in concentration of BT ( ) and TT ( ) in tap water from 13 of 20 sampling points, error bars show maximum and minimum concentration.

For tolyltriazole, Figure 8.2 demonstrates also the spatial variation of the TT concentrations. Concentrations of <5 ng/l to the west, increased by an order of magnitude within 20 km east, where values ranged from 50 to 70 ng/l (Figure 8.2). Locations to the east and west of Uxbridge in Figure 8.2 are supplied by Thames Water, and differences in concentrations may be attributable to factors such as the source of drinking water supply (e.g. groundwater or surface water), treatment processes and mixing in distributions systems. Four of the 7 samples excluded from Figure 8.2 were to the north of Uxbridge, and had between 5 and 12 ng/l of TT. The other three samples were from between 40 to 100 km distant, with 1 to 28 ng/l TT.

The ratio of the BT concentration to TT concentration was different from west to east of Uxbridge. It can be seen the ratio of BT/TT within Uxbridge which is served by Veolia was different. This is probably because of the GAC system existed in the water treatment plant

that served Uxbridge. The GAC process adsorbed the TT because it is more hydrophobic and therefore, the removal of TT was higher than BT. It is possible that there were no, or limited, GAC systems in water treatment plants supplying areas to the east and west of Uxbridge and therefore the ratio of BT to TT was different.

8.2 Estimation of the inputs of BT and TT from dishwasher detergents

The analysis of dishwasher powders and tablets confirmed the presence of either BT or TT in all products (Table 8.1). The range of amount was between minimum of 0.5 mg per 20 g in one own brand tablet to 60 mg in a leading brand tablet product. In order to calculate the average load of BT and TT, market share information was used and use pattern also (Mintel International, 2009a; Mintel International, 2009b). To estimate how many washes were undertaken per day, we used the population equivalent of Hallam Fields, one of the eight the STWs on the River Erewash (44,895), assuming 2.36 people per household (The Office for National Statistics, 2003) to derive 19,023 households. With dishwasher ownership of 28% (Waterwise, 2008), this gave 5,326 households with dishwashers. One wash per day was assumed in each household. According to these information and assumption, the average load per wash was 1.45 mg of BT and 27.8 mg of TT as illustrated in Table 8.1. With an assumption of one use per household per day, this resulted in estimated inputs of 7.72 g of BT (5,326 x 1.45 mg) and 148 g of TT (5,326 x 27.8 mg) per day to this particular STW (Table 8.1). With a flow of 10 Ml/day, the estimate of concentrations in the influent at the STW was 772 ng/l BT and 14,800 ng/l TT.

The average concentrations in the effluent from the primary settling tanks (settled sewage) at Hallam Fields during four days in November 2008 were 2,482 ng/l of BT and 3,271 ng/l TT. It is apparent that the exercise underestimated the measured load of BT, and overestimated that of TT assuming minimal removal in primary treatment. As only one branded product contained TT, then the relative inputs of BT and TT from dishwasher detergents will depend very much on market share of that brand and on the proportion of people who use the tablet form of that brand. Given the assumptions made in calculating inputs, and possible limitations of the limited sampling regime in assessing true inputs, it

would appear that the use of these chemicals in dishwasher formulations may account for a significant proportion (at least 30%) of the inputs to the STW and subsequently the environment. To further improve this estimate, more accurate, catchment specific data on sales and use of dishwasher detergents and also looking at the use of detergents at restaurants and catering places such as hospitals and universities would be required, along with a more comprehensive sampling strategy at the STW. A full source apportionment exercise has not been undertaken; and other uses, such as in corrosion inhibitors for heating systems and motor vehicles could also contribute to the load to sewer, however the estimation has highlighted that the use of BT and TT in "down the drain" products may make a significant contribution to their concentrations in UK rivers. Appendix 5 contains all original date for benzotriazoles in drinking water samples and data analysis of dishwasher detergents.

Summary

Benzotriazole and TT were found in ng/l in drinking water samples, TT concentrations were lower than BT concentration due to its hydrophobicity. Dishwasher detergents as a one possible source of their occurrence in the aquatic environment was estimated and many factors need to be covered in order to improve this estimation.

Table 8.1 Amount of BT and TT present in dishwasher detergent products from UK supermarkets (mg per 20g of tablet or powder).

		Load in 20 g of product (mg)		Weighted load[a] (mg)		Market share (%)	Load per wash[b] (mg)	
	Type	BT	TT	BT	TT		BT	TT
Finish	tablet		60.0		51.4	54		27.8
	powder		17.0					
Fairy	tablet[c]	4.5		4.5		17	0.77	
Sainsbury's	tablet	0.9		3.9		5.4[d]	0.21	
	powder	16.0						
Morrison	tablet	0.5		3.8		4.0[d]	0.15	
	powder	17.0						
Asda	tablet	1.3		1.5		5.8[d]	0.09	
	powder	2.3						
Tesco	tablet[c]	2.2		2.2		10.8[d]	0.24	
Total market share						97.0		
Average load per wash[e]							1.45	27.8

[a] calculated based on 80% of people using the tablet formulation. = (mg in tablet x 0.8) + (mg in powder x 0.2)

[b] calculated by taking into account the market share of the product = weighted load/100*market share

[c] These products were only available in tablet form

[d] Market share of these products was assumed to be the same as the market share of food sold by the four major UK supermarkets (Mintel International, 2009b)

[e] Summing of load per wash gives an estimated input per wash based on the amount in each product and use of that product

Chapter Nine: Discussion

9.1 Occurrence and removal of emerging contaminants in conventional treatment

The research has shown that both the trickling filter and activated sludge process were effective at removing sanitary parameters (SS, BOD and ammonia). The data have also shown that there was little difference in the effectiveness of the two STWs, which may have been a consequence of the sampling regime that was used.

9.1.1 Progestogens.

Of the 10 progestogens covered in this study, only 8 appear to have been reported by other workers (CPA, DSP, MDP, MPA, MTA, NGL, NTD and PGT). Some compounds analysed in this present study, such as tibolone and dydrogesterone appear not to have been determined by other workers. Two of the compounds of interest have been reported in only single studies; in the case of cyproterone acetate by Sun et al. (2009), where CPA was not detected in the influent or effluent of a STW, and drosprinone was also reported as not detected (Vulliet et al., 2008). However the natural hormone, progesterone, has been observed by a number of workers. There are about 30 studies, since 2002, which have reported the occurrence and removal of progestogens in the aquatic environment. A review of these studies is listed in Table 9.1.

In this study, progesterone (PGT), the natural hormone, was the predominant compound and was detected and quantified in all samples of the influent with a concentration of 46.9 ± 30.6 ng/l and 41.6 ± 14.5 ng/l at both STWs. Similar findings were also reported by other studies that have shown that PGT was the most frequently detected compound but with surface waters not wastewaters (Vulliet et al., 2008; Chang, Wan & Hu, 2009). In addition, other studies showed that PGT was the predominant compound and the mean concentrations of PGT were compatible with the findings achieved in this study, 33.1 ± 8 ng/l (Fan et al., 2011) and 66 ± 36 ng/l (Chang et al., 2011). In contrast, other results have shown that norgestrel (NGL) was the predominant compound with 59.0 ± 28.3 ng/l (Liu et al., 2011), although PGT (4.1- 6.4 ng/l) was also observed in their study. This concentration of NGL was in agreement with our findings of 23 ± 7.5 ng/l, however, the high standard

deviations, and range of concentrations reported for these compounds (Table 9.1) highlight the uncertainty in reported values.

Progestogens are not the only steroids hormones studied in the waters and wastewaters. There are some studies which have demonstrated that androgens were the most predominant chemicals among the steroids hormones (Fan et al., 2011; Chang et al., 2011), followed either by the estrogens as the second contributor (Fan et al., 2011) or progestogens (Chang et al., 2011). Therefore more investigations are required to clarify the predominant chemicals in contrast to other hormones in particular as more progestogens are used in the hormonal medicine (factor of 3-100) than estrogens (Zeilinger et al., 2009).

The concentrations of progestogens in the final effluent of the STW measured by other studies are also listed in the Table 9.1. The overall removal efficiencies of progesterone in the two STWs studied in this study were 96% and 97% which is in agreement with other published work when they demonstrated that the removal efficiency achieved in two STW were 90% and 96% (Chang et al., 2008). In addition, synthetic progestogens, norgestrel (NGL) removed at a percentage of 89% (Qiao et al., 2009), while in our finding this was 76% and 92% at Hallam Fields and Newthorpe STWs respectively.

The present study has shown also that the synthetic progestogens were slower to be removed than the natural progestogen (PGT) in conventional biological treatment. Estrogens are an example of where the synthetic compound degrades more slowly when more than 98% of the natural hormones (E1+E2) were removed while 90% of EE2 was removed (Andersen et al., 2003). This finding is supported by the studies achieved by Joss et al. (2004), Shi et al. (2004) and Esperanza et al. (2007), to understand the behaviour of natural and synthetic estrogens when estrone (E1), 17β-estradiol (E2), and estradiol (E3) were more easily degraded than the synthetic estrogen 17α-ethinylestradiol (EE2), even though lower removal for (E1) had occurred due to the transformation of (E2) and (E3) to (E1). However, there is only one synthetic estrogen of significance (EE2) and there are many synthetic progestogens.

Removal of chemicals during biological treatment can be achieved either by degradation or sorption, and subsequent removal with the solids. In relation to degradation, given the nature of process, much focus is on the biological degradation, and chemical degradation is not usually considered as significant. Although it was not feasible to find out whether

progestogens undergo degradation or sorption during the biological treatment, however there is some evidence that the role of sorption process in removing steroids during the wastewater treatment is less significant than biodegradation even with the more hydrophobic compounds (Gomes et al., 2011), and other study also demonstrated that PGT does not partition to solids (Esperanza et al., 2007). Additionally, no significant removal had occurred when mercury chloride $HgCl_2$ was added to the sample in order to inhibit the biological activity (Chang et al., 2011). Furthermore, in our findings, there was no link between the log K_{ow} and removal of compounds during the biological treatment. This implies that most of PGT may have been removed via biodegradation and in our findings; N/AS and trickling filter were able to remove 92% and 90% of the PGT respectively. Therefore, it is likely that PGT degraded rapidly during the biological treatment and other studies support that observation (Esperanza et al., 2004; Labadie & Budzinski, 2005b). Conversely, Fan et al. (2011), demonstrated that no significant removal had occurred through the aerobic tank in their study at a nutrient removal plant (BNR) and that was probably due to the low initial concentration of PGT (4.5 ± 1.6 ng/l) at the influent of the aeration tank of their study as it had already degraded in the anoxic and anaerobic section of the nutrient removal plant.

Table 9.1 Occurrences of progestogens in the aquatic environment (ng/l)

Chemical	Surface water (ng/l)	Ground water (ng/l)	Drinking water (ng/l)	STW		References
				Influent (ng/l)	Effluent (ng/l)	
MDP	0.4 – 1	N.D			Max. 15	(Kolodziej, Harter & Sedlak, 2004; Kolodziej & Sedlak, 2007)
	N.D					(Zheng, Yates & Bradford, 2008)
	N.D	N.D				(Vulliet et al., 2008)
	N.D		N.D		N.D	(Sun et al., 2009)
				N.D	N.D	(Liu et al., 2011)
MPA	N.D			0.21- 4.42	0.03 – 0.42	(Chang et al., 2008)
	0.04 – 34				N.D	(Chang, Wan & Hu, 2009)
				1.08 ± 0.1	0.06 ± 0.03	(Fan et al., 2011)
	N.D – 1.4			18 -58	N.D – 1.1	(Chang et al., 2011)
MTA	N.D				0.35	(Chang et al., 2008)
	0.23 – 25				N.D	(Chang, Wan & Hu, 2009)
	N.D					(Sun et al., 2009)
				4.6 ± 4	0.2 ± 0.06	(Fan et al., 2011)
				1.9 – 9.3	N.D – 0.7	(Chang et al., 2011)
NTD				N.D – 8.9	N.D – 17.4	(Petrovic et al., 2002)
	< 872					(Kolpin et al., 2002)
	N.D				N.D	(Labadie & Budzinski, 2005a; Labadie & Budzinski, 2005b)

Table 9.1 Continued

Chemical	Surface water (ng/l)	Ground water (ng/l)	Drinking water (ng/l)	STW		References
				Influent (ng/l)	Effluent (ng/l)	
NTD					5.2 - 41	(Vulliet et al., 2007)
	N.D		N.D		N.D	(Kuster et al., 2008; Sun et al., 2009)
	2.7 & 2.8	4.2 – 5.6				(Vulliet et al., 2008)
	N.D			N.D	N.D	(Chang et al., 2008)
	N.D				N.D	(Kuster et al., 2009)
	N.D – 16				N.D	(Chang, Wan & Hu, 2009)
	N.D					(Velicu & Suri, 2009; Kuster et al., 2010)
				N.D	N.D	(Fan et al., 2011)
	N.D			5.3 – 12	N.D	(Chang et al., 2011)
	N.D			N.D	N.D	(Liu et al., 2011)
NGL				N.D - 16.1	N.D - 4	(Petrovic et al., 2002)
	N.D				N.D	(Labadie & Budzinski, 2005a; Labadie & Budzinski, 2005b)
					0.9 – 17.9	(Vulliet et al., 2007)
	N.D		N.D		N.D	(Kuster et al., 2008; Sun et al., 2009)
	5.3 & 7	7.4 – 11				(Vulliet et al., 2008)
	N.D			N.D	N.D	(Chang et al., 2008)
				5.6	1.1	(Pu et al., 2008)
	N.D				N.D	(Kuster et al., 2009)

Table 9.1 Continued

Chemical	Surface water (ng/l)	Ground water (ng/l)	Drinking water (ng/l)	STW		References
				Influent (ng/l)	Effluent (ng/l)	
NGL	6.2				N.D	(Chang, Wan & Hu, 2009)
	7.5		N.D	74.3	8.1	(Qiao et al., 2009)
	N.D					(Velicu & Suri, 2009; Kuster et al., 2010)
	N.D				N.D	(Fan et al., 2011)
	N.D			N.D	N.D	(Chang et al., 2011)
PGT	6		6			(Aherne, English & Marks, 1985)
				N.D – 1.9	N.D – 1.5	(Petrovic et al., 2002)
	199					(Kolpin et al., 2002)
	14 – 44					(Vanderford et al., 2003)
	N.D	N.D				(Kolodziej, Harter & Sedlak, 2004)
	N.D				N.D	(Labadie & Budzinski, 2005a; Labadie & Budzinski, 2005b)
					8 – 16.9	(Vulliet et al., 2007)
	1.4 – 4.2				2.1	(Kolok et al., 2007)
	Max. 27				N.D	(Kolodziej & Sedlak, 2007)
	0.32 – 1.39		0.93			(Kuster et al., 2008)
	N.D					(Zheng, Yates & Bradford, 2008)
				4.3 - >100	N.D – 3.2	(Pauwels et al., 2008)
	4.5 ±1.7				0.78 & 0.86	(Van der Linden et al., 2008)

Table 9.1 Continued

Chemical	Surface water (ng/l)	Ground water (ng/l)	Drinking water (ng/l)	STW		References
				Influent (ng/l)	Effluent (ng/l)	
PGT	1.7 & 3.5	2.5 – 4.1				(Vulliet et al., 2008)
	0.06 – 0.09			3.1 & 10	0.31 & 0.37	(Chang et al., 2008)
	0.72 – 6.5					(Standley et al., 2008)
	0.51 – 47.2					(Kuster et al., 2009)
	N.D – 26				0.48 – 1.4	(Chang, Wan & Hu, 2009)
	Max. 3.1		Max. 0.57			(Benotti & Brownawell, 2009)
	7.35 – 1.8					(Velicu & Suri, 2009)
	N.D		N.D		N.D	(Sun et al., 2009)
					N.D	(Mnif et al., 2010)
	N.D					(Kuster et al., 2010; Yoon et al., 2010; Singh et al., 2010)
			N.D			(Sodre, Locatelli & Jardim, 2010)
				6.62	0.99	(Fan et al., 2011)
	N.D – 1.7			35 - 108	0.8 – 2.3	(Chang et al., 2011)
	0.5-2.5			5.4 & 6.1	N.D	(Liu et al., 2011)

N.D: not detected

9.1.2 Benzotriazoles

Both benzotriazole and tolyltriazole have been reported to be ubiquitous in many European countries, and the concentrations of BT and TT reported in the aquatic environment are listed in Table 9.2. The concentrations of benzotriazoles in the influent STW were much higher than the progestogens group. This is because they are produced in high volume (HPV) and widely used as anti corrosives, in cooling and hydraulic fluids, in aircraft de-icing fluids, and in the formulation of dishwashing detergents (Hart et al., 2004). In our findings, the concentrations of BT (2.3-2.4 µg/l) in the influent was lower than reported by Weiss & Reemtsma (2005) and higher than of that Jover, Matamoros and Bayona (2009). A possible explanation for this might be that this difference is due to the difference in using these chemicals in formulations of products and patterns of use in different countries. In terms of TT, the concentrations of total TT were relatively similar to those reported in Germany (Weiss & Reemtsma, 2005).

In the biological treatment, BT and TT were partially removed. Other studies corroborated that when they observed that biological treatment was not able to remove these compounds completely (Voutsa et al., 2006; Reemtsma et al., 2006; Giger, Schaffner & Kohler, 2006). The removal efficiency of biological treatment was in the range of 28% - 39% for both chemicals at both STWs. This finding is in agreement with other studies, such as Weiss, Jakobs and Reemtsma (2006) when 37% of BT was removed. The physicochemical properties of this group has an important role in terms of their fate and occurrence in the wastewater, whereas low log K_{ow} (1.59 ± 0.23) for benzotriazoles, high degree of solubility and polarity in water and this means that BT and TT are difficult to get removed by sorption during conventional treatment. Furthermore, BT and TT are resistant to biodegradation (Voutsa et al., 2006; Giger, Schaffner & Kohler, 2006) and this is also a factor that affects their removal during conventional biological treatment.

Table 9.2 Occurrences of benzotriazoles in the aquatic environment (µg/l).

Chemical	Surface water	Ground water	STW		References
	(µg/l)	(µg/l)	Influent (µg/l)	Effluent (µg/l)	
BT					
	3.4	N.D	11.9 ± 1.2	9.6 ± 1.3	(Weiss & Reemtsma, 2005)
	1 (Max = 6.3)				(Giger, Schaffner & Kohler, 2006)
	0.175-1.2 (lake)				(Giger, Schaffner & Kohler, 2006)
	0.636 – 3.690		13 - 75	11 - 100	(Voutsa et al., 2006)
	2.7		12	7.7	(Weiss, Jakobs & Reemtsma, 2006)
	0.6			7.3	(Reemtsma et al., 2006)
				8	(van Leerdam et al., 2009)
	0.493				(Loos et al., 2009)
	0.213				(Loos, Locoro & Contini, 2010)
	N.D - 0.68			7.0-18	(Reemtsma et al., 2010)
	0.24		0.131	0.129	(Jover, Matamoros & Bayona, 2009)
	0.038 - 1.474				(Kiss & Fries, 2009)
		0.024 (Max=1.032)			(Loos et al., 2010)
TT					
	2.4				(Kolpin et al., 2002)
†	0.2	N.D	2.5 ± 2.6	2.2 ± 1.6	(Weiss & Reemtsma, 2005)
‡	0.1	N.D	2.0 ± 2 .0	2.1 ± 1.3	(Weiss & Reemtsma, 2005)

Table 9.2 Continued

Chemical	Surface water	Ground water	STW		References
	(µg/l)	(µg/l)	Influent (µg/l)	Effluent (µg/l)	
	0.2 (0.04 - 0.47)				(Giger, Schaffner & Kohler, 2006)
	0.03 - 0.23 (lake)				(Giger, Schaffner & Kohler, 2006)
	0.122 – 0.628		0.2 - 5.6	0.1 - 3.8	(Voutsa et al., 2006)
†	0.2		1.3	1.2	(Weiss, Jakobs & Reemtsma, 2006)
‡	1.2		2.1	2.1	(Weiss, Jakobs & Reemtsma, 2006)
	0.2			2.2	(Reemtsma et al., 2006)
				3	(van Leerdam et al., 2009)
	0.617				(Loos et al., 2009)
	0.081				(Loos, Locoro & Contini, 2010)
†	N.D - 0.13			0.8 - 1.2	(Reemtsma et al., 2010)
‡	0.2 - 0.46			1.0 - 5.0	(Reemtsma et al., 2010)
†	N.D - 2.16				(Corsi et al., 2003)
‡	N.D - 1.67				(Corsi et al., 2003)
†	0.925		N.D	N.D	(Jover, Matamoros & Bayona, 2009)
‡	1.561		N.D	N.D	(Jover, Matamoros & Bayona, 2009)
†	0.25 - 0.281				(Kiss & Fries, 2009)
		0.02 (Max= 0.516)			(Loos et al., 2010)

† For 5- isomer TT ‡ For 4- isomer TT

9.1.3 The mechanism of removal of emerging contaminants during wastewater treatment.

The main removal mechanism during the biological treatment has been reported to be biodegradation (Daughton & Ternes, 1999; Onesios, Yu & Bouwer, 2009). Although suspended growth process (activated sludge process) has demonstrated better removal than attached growth like trickling filter for many emerging contaminants (Svenson, Allard & Ek, 2003; Janex-Habibi et al., 2009; Limpiyakorn, Homklin & Ong, 2011). It also been noted that a nitrifying process is more able to remove ECs than other steps, since Anderson, Suarez and their co- workers have demonstrated that most of the natural estrogens (E1+E2) were largely biodegraded during the aerobic and anoxic stages, while the synthetic 17α-ethinylestradiol (EE2) was less biodegraded in the anoxic stage (Andersen et al., 2003; Suarez, Lema & Omil, 2010). However this present study did not clearly demonstrate that the ASP was better to remove them than the TF and this maybe a result of factors such as sampling/analytical issues related to the relatively small sample numbers and low concentrations, close to LOQ for the progestogens.

The removal efficiency for emerging contaminants varies from compound to compound. There is also broad variation in the removal efficiency for the same compound in different STWs. This implies that many parameters have influenced this removal rate and several studies have investigated these factors (Clara et al., 2005b; Radjenovic, Petrovic & Barcelo, 2007; Koh et al., 2008; Cirja et al., 2008). The physicochemical properties of the chemicals by means of hydrophobicity and hydrophilicity; operational conditions, such as sludge retention time (SRT), hydraulic retention time (HRT); and effluent characteristics like temperature, pH, and flow rate have been investigated and they have affected the removal efficiency of ECs during the conventional treatment. For example, longer SRT allow growing a more diverse bacterial population (including nitrifying bacteria) and that provides a wide spectrum of microorganisms that have the ability to metabolize the target compounds and consequently improve the removal efficiency (Johnson & Sumpter, 2001; Clara et al., 2005a; Koh et al., 2008;). Also increasing the HRT allows increased contact time and more time for biodegradation and adsorption leading to enhanced removal efficiency (Kirk et al., 2002; Svenson, Allard & Ek, 2003). Therefore high HRT and SRT are essential for high

removal rate (Miege et al., 2008). Temperature also has an influence on the removal of compounds, as noted when high concentrations of compounds were observed in effluent during winter compare to summer (Vieno, Tuhkanen & Kronberg, 2005).

It is important to understand how to remove these chemicals from wastewater as they may have an environmental impact, and although it can be considered that removal of 90% and above may be "good", this may not be sufficient to protect the environment. It is therefore worth considering at what concentrations they may be of concern.

9.1.4 The possible significance of these compounds in the environment

The effects of target compounds are not completely clear yet, although there are some studies which stated that at such levels of progesterone (maximum 1.39 ng/l) is considered not to pose a risk to human toxicity (Kuster et al., 2008). Moreover, medroxyprogesterone showed no or little effect (up to 5 mg/l) on aquatic organisms (*Ceriodaphnia dubia*) (Jukosky, Watzin & Leiter, 2008). However, there is some evidence of the possible impact of some of the chemicals of interest on wildlife and humans, for example, norethindrone showed a significant decrease in fecundity of fathead minnow at low level (1.2 ng/l) (Paulos et al., 2010). Additionally, there is some evidence of the impact of some of these, such as norgestrel and drosprinone have on the aquatic organisms; hence it was found that low ng/l range (3.3 ng/l for NGL, and 6.5 µg/l for DSP) exhibited an inhibition to the reproductive function in adult fathead minnows (Zeilinger et al., 2009). Although the mean effluent concentration of NGL at Hallam Fields STW was higher than 3.3 ng/l, however, the dilution rate of effluent in the river is an important factor to know whether this compound may have an impact on the aquatic organisms. Cyproterone acetate has been observed to exhibit an antiandrogenic activity (three times more than drosprinone) by preventing the androgen receptor from androgen binding (Fuhrmann et al., 1996).

The other group of interest, benzotriazoles, has been noted as toxic to aquatic organisms (Cornell, Pillard & Hernandez, 2000), and therefore may cause adverse effect in the aquatic

environment. Although benzotriazole (BT) is less toxic than TT (Pillard et al., 2001), however BT at 32 mg/l can interfere with the regulation of embryo development in protochordates such as *Ciona intestinalis* (Kadar, Dashfield & Hutchinson, 2010). Moreover, the Dutch Expert Committee for Occupational Standards concluded that there are some studies with rat and mice which demonstrated that BT may be carcinogenic (DECOS, 2000). In addition, Zhang et al. (2008) found that BT derivatives are able to induce growth inhibition in cancer cells and benzotriazole had an antiestrogenic activity in *vitro* but not in *vivo* (Harris et al., 2007). Limited chronic and acute ecological toxicity data indicate that BT is moderately toxic to *Lepomis macrochirus* (96-h LC50 =31 mg/l) and *Daphnia magna* (48-h LC50 = 74 mg/l) (Cornell, Pillard & Hernandez, 2000). It is therefore possible that such connections may exist between benzotriazoles and some impact on wildlife and aquatic organisms.

When discharged to the environment, there is frequently dilution of effluents in receiving waters which reduces the concentrations of the individual chemicals. However, groups of chemicals, such as the progestogens, may, if their effects are additive, still be of concern. Evidence for such effects was seen in a mixture of five similarly-acting chemicals: all were estrogens, even though one was a pharmaceutical, one a natural hormone, and three industrial chemicals was tested and they found that these chemicals acted in an additive manner, meaning that the effect of each chemical could be added together, and if the chemicals in a mixture all act in the same way, the effects will be additive (Brian et al., 2005). Additionally, their effects were not antagonistic or synergistic. In this study, the total concentrations of progestogens that discharged to the surface waters from the STWs were 32.1 ng/l and 16 ng/l for Hallam Fields and Newthorpe STWs respectively and the total concentrations of progestogens (natural and synthetic) along the River Erewash ranged from 10.9 ng/l to 27.0 ng/l. These progestogens were present in the same river sample and this would suggest that their effects would be additive assuming they are equally potent. Therefore to understand the threat, to wildlife or human health, a greater understanding of the risk posed by these very complex mixtures of chemicals is required.

In general, the concentrations of benzotriazoles were much higher in influent and effluent than progestogens. The activated sludge process and trickling filter considered in this study

have the ability to reduce the concentrations of emerging contaminants in wastewater but not to eliminate them completely. The concentrations of target compounds were lower in the effluent than those in the influent and that would suggest that biodegradation and sorption take place during STW. Conventional biological treatment, perhaps by chance, does remove amounts of a range of emerging contaminants. But although this can be more than 90% for some compounds, there are some other compounds poorly removed and this implies that there is a wide variation in their removal efficiency due to the differences in the physicochemical properties of each compound and also types of operation in addition to its operation parameters. According to these impacts, it seems that there is no emerging concern about BT and TT although there is no confirmed evidence about their action as a carcinogen (EPA, 2011). While for progestogens, there is some evidence that they may cause a problem with low ng/l. Thus, although conventional treatment removes "well", it may not be enough and therefore more treatment might be needed.

9.2 Occurrence and removal of emerging contaminants during advanced treatment

As previously explained that conventional treatment was designed to remove the conventional contaminants represented by the organic materials (BOD/COD) and suspended solids and in some of them were also designed to remove ammonia. Moreover, in this study and many other studies documented that conventional treatment only partially removes the emerging contaminants. In the results chapters, the sand filters used at Hallam Fields and Newthorpe STWs were included in the assessment of "conventional treatment". However, here they are considered as a tertiary treatment, because they are relatively low-energy that may enhance the removal of a range of chemicals.

Sand Filter

Although sand filter was not designed to remove emerging contaminants from wastewater, however, removal of progestogens and benzotriazoles did occur in this study with removal efficiency rate from 53 - 94% in trickling filter STW and from no removal to 62% at the activated sludge STW. In addition, the sand filter at Hallam Fields STW was able to remove 14% and 26% of BT and TT respectively and 61% was removed for BT and TT during the sand filter at Newthorpe STW. Therefore the performance of the sand filter at the TF plant appeared better than that one of ASP sewage work although there was no significant difference. There is very limited information available to demonstrate the performance of sand filter in terms of removing emerging contaminants from wastewaters. However, the results achieved by Gunnarsson et al. (2009) exhibited that estrone (E1) and bisphenol A (PBA) had reduced from 6.3 ng/l to 0.67 ng/l and from 780 ng/l to 420 ng/l respectively due to the fact that the sand filter demonstrated biological activity (nitrification).

Ozone (O_3)

The removal rates of progestogens (natural and synthetic) by using ozone in this study were relatively low ranging from (0 – 59%). Limited information is available about the removal of progestogens in the literature to compare with. There are many factors that might influence the removal rate of these chemicals such as the structure of target chemical, dose of ozone and contact time, initial concentration of the target compound and the quality of wastewater. Westerhoff et al. (2005) found that the steroids containing phenolic moieties such as estradiol, ethynylestradiol, or estrone are oxidised more efficiently than those without aromatic or phenolic moieties like progesterone and testosterone. This is supported by Broseus et al. (2009) when they found in drinking water (not wastewater) samples that many progestogens (natural and synthetic) were far slower to react with ozone compare to estrogens although progesterone was detected in one sample before ozonation and was not detected after. Another study confirmed that when similar conditions were applied for river water samples, the removal efficiencies of estrogens were higher than progesterone (Snyder et al., 2006).

Our study was not able to test the ozone dose, as it was arranged around other work at Hallam Fields. The applied dose of ozone used in present study during the treatment was

1mg O_3 /l with 3 minutes contact time which was close to another study by Snyder et al. (2006), when they found that with 2.4 mg O_3/l, the removal efficiency for progesterone differed at 2 and 6 minute contact time. Other studies suggest that 10mg O_3 /l with 40 minutes could achieve a high removal for PPCPs (Gabet-Giraud et al., 2010). In general, the applied dose at Hallam Fields was relatively lower than what other studies recommended, with O_3 doses ranging from 5 – 15 mg/l and contact time of about 15–30 minutes (Ternes et al., 2003; Verlicchi et al., 2010). Therefore, this factor probably affected the removal efficiency for progestogens, which is corroborated by reported estrogen removal at Hallam Fields which was relatively low (60-70%) when compared to other studies (Filby et al., 2010).

The initial concentration of the target compounds (pre - ozonation) plays an important role in measuring the removal efficiency, as when the initial concentration was very low or just above the limit of quantification, then it was difficult to find out how much was degraded or decomposed and consequently quantification of these chemicals resulted from an analytical variations rather than an actual concentration variation (Reungoat et al., 2010). Therefore to determine removal rates accurately at very low concentrations (< 5ng/l), sensitive and precise analytical methods are needed to reduce analytical uncertainty.

For benzotriazoles concentrations were high in relation to LOQ and the present findings seem to be consistent with other research which found that 70% of BT (Hollender et al., 2009) was removed with about 0.6 g O_3/ g DOC and at Hallam Fields 73% of BT was eliminated. Although Weiss, Jakobs and Reemtsma (2006) found that with 1 mg O_3 / mg DOC , more than 99% of benzotriazoles were removed, at Hallam Fields it was less with between 73 and 79% removal efficiency for BT and TT respectively with the same dose. The physicochemical properties of the compounds and the quality of the effluent have an influence on the oxidation process. Huber et al. (2005a), showed that ozone reacts readily with soluble compounds (such as BT and TT), which will be oxidized a more rapidly than compounds sorbed to particles due to limitation of reaction rates by diffusion.

Granular activated carbon (GAC)

The removal efficiency of GAC for progestogens appeared higher than using ozone however there was no significant difference in the post GAC and O_3 effluent apart from cyproterone

acetate (CPA). The effectiveness of GAC corroborates the observations of Rossner, Snyder and Knappe (2009), when they suggested that antimicrobial compounds, EDCs and other pharmaceuticals can be effectively removed via activated carbon adsorption processes. Although Fukuhara et al. (2006), found that adsorption of E1 onto activated carbon was higher than E2 due to differences in hydrophobicity, however, our study has been unable to demonstrate differences in removal of progestogens with different log k_{ow}. A possible explanation for this might be that due the low initial concentrations of progestogens and also the relatively similar log K_{ow} for the progestogens (3.29 - 3.87).

In terms of benzotriazoles, 90% and 98% of BT and TT were removed via GAC. The removal achieved via GAC for benzotriazoles was higher than for progestogens. However the high initial concentration of benzotriazoles (517 ng/l and 2,253 ng/l for BT and TT respectively) compared with low nanograms per litre range (< 7 ng/l) as a maximum initial concentration of progestogens could have affected the determination of removal rate. The main mechanism in removing these chemicals is sorption. The virgin GAC has been proved to be more efficient to remove hormones than the reactivated one (Rowsell et al., 2009). These workers conducted rapid, small-scale column tests and they found that 81% or more of estrogens were removed with the virgin GAC while around 65% were removed with reactivated carbon. However, the adsorption capacity is significantly reduced by the presence of natural organic matter (Fukuhara et al., 2006). When comparing O_3 and GAC, it is worth considering that ozone may result in degradation products while GAC removes the compounds by adsorption.

Chlorine dioxide (ClO_2)

The behaviour of ClO_2 against progestogens and benzotriazoles was similar to ozone treatment. Again in our findings, because of the low initial concentrations of progestogens, it was difficult to understand the performance of ClO_2 towards these chemicals. Although Filby et al. (2010), found that ClO_2 had the greatest removal effect of estrogens, however a study by Deborde et al. (2004), revealed that progesterone remained unchanged and did not react with chlorine while all other chemicals which have a phenolic group in their structure in their study were rapidly oxidized. Chlorination may also result in degradation products.

The total concentrations of progestogens discharged from the three advanced processes (O_3, GAC, and ClO_2) at Hallam Fields STW were 28.8 ng/l, 18.2 ng/l and 14.0 ng/l respectively. While for benzotriazoles these were 603, 101 and 609 ng/l respectively. Combinations of different types of advanced technologies may improve the effluent quality and that approach was recommended by Snyder et al. (2007) and Verlicchi et al. (2010) , when they found that reverse osmosis and advanced oxidation could remove many ECs and PPCPs. Additionally, the dilution rates of discharges within the receiving waters play an important role in the behaviour of these chemicals against the aquatic organisms. In addition, the degradation products produced during ozonation and chlorination of progestogens and benzotriazoles were not investigated in this study. In order to identify and understand their fate in advanced treatment and the toxicological impacts due to the formation of these degradation products, future studies and investigations would be required.

9.3 Is there a need to further remove emerging contaminants from effluents?

Progestogens and benzotriazoles are only partially removed during the STWs processes and this results in the presence of these compounds in the surface water. The dilution rate between the effluents and the river water and different half lives for target compounds, for example progesterone (PGT) and cyproterone acetate, this would suggest that different levels of concentrations in the river are expected. As mentioned before (section 9.1.1) there are some progestogens (cyproterone acetate, drosprinone, dydrogesterone, and tibolone) that have not been, or only on one occasion, studied or reported in the aquatic environment. However the other chemicals that were observed are in accordance with the concentrations observed in this study. The concentrations of medroxyprogesterone (MDP) in the Erewash River in this study (<0.4 – 0.6) ng/l was in agreement with Kolodziej, Harter and Sedlak (2004) and Kolodziej and Sedlak (2007) who found MDP in surface water in range of 0.4 – 1 ng/l. The concentrations of NGL reported in this study (1.7-5.7 ng/l) corroborates the findings of Vulliet et al. (2008), who found that NGL in two samples from urban dam of water receiving effluents from various sewage treatment plants, as well as direct industrial

effluents in France were 5.3 and 7.0 ng/l. Progesterone (PGT) concentrations in the Erewash River in the current study were lower with those of Kolpin et al. (2002) who found that PGT in the surface water was 199 ng/l, while in our results were below the limit of quantification (< 0.4 ng/l) in all samples along the river which is similar to Yoon et al. (2010) who reported < 0.5 ng/l and relatively consistent with Kuster et al (2008) (0.32-1.39 ng/l). It is likely that Kolpin et al. (2002) are incorrect because others have reported lower values (Table 9.1).

In terms of benzotriazoles concentration in the River Erewash, the findings of the current study do not support the previous research of Loos, Locoro and Contini (2010) who found that BT was the higher than TT in Danube River in Europe. However, it should be noted that it is not easy to compare small streams and big rivers because of many factors that play an important role like river flow, dilution rate, and loading of organic pollution in terms of sewage treatment works existing on the river. The concentrations of BT in the 1st campaign in January were in the range of 118 – 1,270 ng/l which is in consistent with other work (Giger, Schaffner & Kohler, 2006; Kiss & Fries, 2009). While the 2nd campaign, BT concentrations were less than the 1st survey however, these results supports the findings of others (Loos et al., 2009; Loos, Locoro & Contini, 2010). Regarding to tolyltriazole, the 4 and 5 isomers were not analyzed separately in this study; therefore the results are only compared with other studies which quantified the total tolyltriazole. The concentrations of TT along the River Erewash were quite similar in both campaigns ranging from 605 - 1855 ng/l. Although these results are higher than some published studies (Weiss & Reemtsma, 2005; Reemtsma et al., 2010), they are consistent with those of others (Weiss, Jakobs & Reemtsma, 2006; Jover, Matamoros & Bayona, 2009). Although the concentrations of BT and TT at the mouth of the River Erewash were higher than at the mouth of the Danube River, the mass fluxes of benzotriazoles discharged from the River Erewash toward the River Trent are very low when compared to large rivers such as Danube. The mass discharged from the Danube River to the Black Sea is 33.8 tonnes of BT per year and 17.0 t/year of TT (Loos, Locoro & Contini, 2010).

One of the main objectives of modelling the rivers and consequently its catchment is to understand the behaviour of target compounds along the river. The importance of

modelling is to inform us about the zones or sections in the river that have a concentration higher than the allowable limits or which may exceed the limit of concern. Consequently it will be easier to decide how to control the pollution points, which is normally the effluents from STWs, and also tell us where to invest in order to improve the quality of the effluents and then the quality of the river water rather than random investment of the STW which might lead to an enhancement to the quality of effluent but not to the river water.

The catchments of the Rivers Trent and Thames were modelled according to the principle of per capita load, due to the fact that the data about the concentrations of these chemicals from the discharging point (STWs and other points) were not available. The modelling of these two catchments showed that the highest concentrations for benzotriazoles were just downstream of the effluent discharges of the STWs existing on the streams and tributaries; this is not demonstrated in the main river (i.e. Trent and Thames). This is probably due to the flow rate in the main river compared to the other small rivers and also the dilution factor between the STW effluents to the receiving water body. Therefore it is worth investigating the load from specific STW that can contribute to the burden with high concentrations and flow rate in order to improve the overall quality by adding tertiary treatment instead of investing other STW that might not have a large impact at the catchment scale.

In order to achieve the modelling, there are many criteria that should be considered and one of these important criteria is the degradation rate or the half life of the chemicals of interest. Therefore in this study the degradation test, for both group of chemicals (progestogens and benzotriazoles) was conducted, as the main mechanism of progestogens removal is degradation (Labadie & Budzinski, 2005a; Fan et al., 2011).

The concentrations of progestogens were very low in samples taken for the degradation study, in agreement when progesterone was not detected Han River in South Korea and its tributaries (Yoon et al., 2010). In this study, and according to results, the degradation of progestogens was divided into three groups according to their half lives. The natural progestogen, progesterone (PGT), was degraded rapidly and the half life was less than one day. This is in accordance with Labadie and Budzinski (2005b) who found that the half-life

of progesterone was about 8 hours, although their samples were sewage effluent. However, in our study it was not planned to monitor the concentration of progesterone within the first day. Therefore it seems that the natural steroids are degraded quite rapidly in the aquatic environment, for example, both estrone and estriol, the natural estrogens, were found that losses had happened when they stored for a week (Baronti et al., 2000). The degradation of the synthetic progestogens was slower than that of the natural hormone under the same conditions and same findings were found by Chang et al. (2010).

The other group of interest, benzotriazoles (BT and TT), were resistant to degradation during the test. This result is corroborated by others who have stated that benzotriazole was not biodegraded (Hart et al., 2004; Weiss & Reemtsma, 2005; van Leerdam et al., 2009), however some studies have stated that the 5 - TT isomer is more easy to biodegrade than 4 - TT isomer (Weiss & Reemtsma, 2005; Weiss, Jakobs & Reemtsma, 2006; Reemtsma et al., 2010).

The results of this study indicate that there are some chemicals that may be of more concern because of their persistence. It was possible that high concentrations in water used for potable supply could result in their presence in tap waters. Benzotriazoles were the only chemicals studied and observed in the tap waters in this study and dishwasher detergents were identified as one possible source of these chemicals to the aquatic environment. Although the presence of chemicals in the environment and drinking water does not in itself pose a risk to health and the environment, there is concern that the possible effects of long term exposure to individual chemicals and / or mixtures of chemicals are not fully understood (Snyder et al., 2003; Jones, Lester & Voulvoulis, 2005). The implications of our findings depend to a large extent on the degree of (eco) toxicity of BT and TT. Chemicals are usually more toxic when administered chronically (long-term) than when exposure is acute (short-term). This difference can be expressed as the acute: chronic ratio. For many industrial chemicals, this ratio is 10 or less, meaning that the LOEC and NOEC derived from chronic toxicity tests are not appreciably lower than those derived from acute toxicity tests. However, if chemicals have specific modes of action (as, for example, pharmaceuticals do), then chronic toxicity tests often demonstrate that chemicals can be very much more toxic than anticipated based on the results of acute toxicity tests (Fent, 2008). For example, the

acute: chronic ratio of ethinyl estradiol is around 100,000 (Webb, 2004). This illustrates the importance of the current data gap with BT and TT; it is imperative to determine their chronic ecotoxicities (Harris et al., 2007) and data on carcinogenicity is conflicting (USEPA, 2011). Although there seems to be no particular reason to think that BT and/or TT will have a high acute: chronic ratio, and hence be of more concern than appears to be the case based on acute ecotoxicity data, caution should be exercised for the following reason. Many azoles are very active chemicals, with specific modes of action: many (imidazoles and triazoles) are fungicides used in agriculture, and others (e.g. fadrazole) are used for anti-estrogen treatment in diseases such as breast cancer (Trosken et al., 2004). Recent results have demonstrated that many commonly-used fungicides act as endocrine disrupters in vivo in both mammals (Taxvig et al., 2008) and fish (Rime et al., 2010). Structural alerts such as these can be useful in aiding the selection of appropriate chronic toxicity tests that should be helpful in determining whether or not BT and/or TT are significantly more toxic chronically than they are acutely.

Adding further uncertainty to the toxicity of BT and TT is the possibility that BT is a human carcinogen. A Dutch committee concluded that the weight of evidence indicated that BT may be a possible genotoxic carcinogen, although it was highlighted that the database was inconclusive (DECOS, 2000). Based on that assessment, and structural analogy, Australian drinking water quality guidelines suggest a maximum permissible concentration of TT of 7 ng/l (NRMMC-EPHC-NHMRC, 2008). Given this uncertainly, it could be strongly argued that the precautionary principle should be applied to both BT and TT, and exposure concentrations (to both aquatic wildlife and humans) minimised until appropriate chronic toxicity data become available on which to base any risk assessments.

It is therefore apparent that although conventional treatment may be seen as effective (with > 90% removal), this may not be good enough. We can use modelling to inform us about where in the catchment we might most effectively invest in any further treatment processes required to reduce concentrations in effluents. Investment in tertiary treatment may also consider a number of factors, such as the effectiveness at different sites, presence of degradation products and costs, which may be important both financially and through energy use (UKWIR, 2002).

Chapter Ten: Conclusions

The overall aim of this research was to study the effectiveness of conventional and advanced wastewater treatment in reducing discharges of emerging contaminants. This aim was achieved by studying the fate of progestogens (one natural and nine synthetic) and benzotriazoles (BT and TT) via determining the occurrence and removal of progestogens and benzotriazoles in wastewaters and in the aquatic environment in the UK.

The conclusions obtained throughout this research are:

1. Progestogens and benzotriazoles were present in the sewage system. In terms of predominance, the natural hormone, progesterone, was the most predominant progestogens. However, the concentrations of benzotriazoles were two magnitudes higher than progestogens and tolyltriazole predominated at both STWs.

2. Both the ASP and TF plant partially removed the compounds, and at both sites the sand filters contributed to improving overall removal.

3. In advanced treatment at Hallam Fields STW, results show that three techniques (O_3, GAC, and ClO_2) were not able to remove progestogens significantly. However, this may be due to the low initial concentration of these chemicals and probably due to low O_3 dose in terms of ozonation. However, a large reduction (73% - 90% for BT and 79% - 98% for TT) as observed for the benzotriazoles.

4. The adsorption method represented by GAC seemed more effective than the other two advanced (oxidation) treatments, O_3 and ClO_2, hence the removal efficiency ranged from 60 – 98 % for both group of compounds. However, the O_3 dose and contact time were low in this study and a range of factors would influence selection of tertiary processes.

5. In terms of degradation of progestogens in the river waters, the natural hormone was degraded rapidly (half-life < 24 hrs). Some synthetic progestogens had half-lives of between 2 to 4 days, and others were more recalcitrant and their half-lives were more than 4 days. While the other group of chemicals, benzotriazoles, were not degraded during the 14 days of the experiment.

6. Progestogens and benzotriazoles were present in the river waters. Concentrations of individual progestogens in the river water were relatively low, however the concentration of the sum of progestogens in the River Erewash was from 10.9 to 27.0 ng/l. For benzotriazoles, the concentrations of BT and TT were two magnitudes higher than the progestogens and the general trend along the river showed an increase in their concentration.

7. Two river catchments were modelled which indicated widespread occurrence of BT and TT, including in the waters used for abstraction for potable use.

8. In terms of tap water, benzotriazoles were quantified in all tap water samples around Uxbridge – London. Benzotriazole was present at concentration higher than TT in all samples measured. The mean concentrations of BT and TT were 30.9 ng/l to 15.1 ng/l respectively.

9. From the evaluation of possible inputs from dishwasher tablets and powders. It seems likely that they contribute significantly to concentrations of BT and TT present in the environment. Source control may be an effective way to improve the environmental quality.

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Appendix 1: Conventional Treatment

Concentrations of progestogens in the settled sewage in ng/l at Hallam Fields sewage treatment work.

Hallam Fields STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	18.1	20.1	64.5	26.7	13.7	5.7	9.0	27.2	26.3	36.4
AM	12.5	< 2	4.2	< 0.4	< 0.8	3.6	< 2	13.5	15.7	35.3
AM	10.5	< 2	29.5	< 0.4	< 0.8	< 0.8	< 2	15.8	40.9	36.7
AM	24.0	< 2	24.6	< 0.4	< 0.8	12.5	< 2	34.1	36.1	55.1
PM	23.1	13.8	< 3.2	19.8	15.2	8.6	5.8	24.6	32.9	38.2
PM	25.1	< 2	96.5	< 0.4	< 0.8	< 0.8	29.5	18.5	44.0	< 3.2
PM	< 0.4	< 2	62.6	< 0.4	< 0.8	1.5	< 2	31.4	65.2	< 3.2
PM	31.5	< 2	< 3.2	< 0.4	< 0.8	15.5	13.1	18.9	113.7	56.5

Concentrations of progestogens in the settled sewage in ng/l at Newthorpe sewage treatment work.

Newthorpe STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	9.0	7.3	< 3.2	27.7	10.3	12.9	4.2	16.1	25.2	29.1
AM	19.9	< 2	71.4	6.2	< 0.8	16.3	< 2	6.3	37.3	< 3.2
AM	16.0	< 2	40.8	27.3	< 0.8	18.4	< 2	< 4	44.0	< 3.2
AM	9.2	17.5	27.6	34.3	28.7	20.4	< 2	< 4	60.0	33.1

Concentrations of progestogens as a RGF feed in ng/l at Hallam Fields sewage treatment work.

Hallam Fields STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	0.3	< 1	< 1.6	< 0.4	5.0	< 0.2	< 1	< 2	< 0.4	< 1.6
AM	< 0.2	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	6.3
AM	1.9	13.3	27.9	9.4	3.9	5.2	4.7	17.8	7.4	2.3
AM	0.3	14.0	11.0	11.7	9.7	6.0	3.5	23.0	8.3	13.8
PM	0.7	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	3.8
PM	5.0	8.8	56.6	5.1	7.1	5.8	< 1	12.9	5.2	15.4
PM	4.3	< 1	4.4	< 0.4	9.7	< 0.4	< 1	2.1	< 0.4	< 1.6
PM	< 0.2	13.6	< 0.8	5.7	4.8	< 0.4	< 1	5.9	8.7	< 1.6

Concentrations of progestogens as a RGF feed in ng/l at Newthorpe sewage treatment work.

Newthorpe STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	1.1	3.1	1.7	2.2	3.8	2.6	1.1	5.6	2.0	3.6
AM	9.4	25.1	60.3	14.3	22.2	10.6	5.2	26.0	12.5	30.9
AM	0.9	< 1	41.7	< 0.4	< 0.4	< 0.4	< 1	< 2	1.3	< 1.6
AM	< 0.2	< 1	21.0	< 0.4	4.7	< 0.2	< 1	< 2	< 0.4	16.1

Concentrations of progestogens as a final effluent in ng/l at Hallam Fields sewage treatment work.

Hallam Fields STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	< 0.2	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	5.8	9.6	5.2	8.8	5.5	8.0	1.1	8.3	7.2	< 1.6
AM	0.3	< 1	19.2	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	< 0.2	< 1	5.8	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
PM	1.1	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	24.9	< 0.4	39.3
PM	1.2	7.7	20.1	4.9	3.1	3.6	1.4	9.2	5.6	11.7
PM	2.8	< 1	3.5	< 0.4	15.7	< 0.4	< 1	< 2	< 0.4	< 1.6
PM	< 0.2	< 1	< 1.6	< 0.4	4.1	< 0.4	< 1	< 2	< 0.4	< 1.6

Concentrations of progestogens as a final effluent in ng/l at Newthorpe sewage treatment work.

Newthorpe STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	< 0.2	< 1	< 0.8	< 0.4	< 0.4	0.6	< 1	< 2	< 0.4	< 1.6
AM	3.8	< 1	4.2	< 0.4	< 0.4	0.7	< 1	< 2	< 0.4	< 1.6
AM	< 0.2	< 1	5.4	< 0.4	< 0.4	< 0.2	< 1	< 2	< 0.4	< 1.6
AM	< 0.2	6.6	3.1	2.8	8.2	4.9	< 1	< 2	5.2	11.3

Concentrations of benzotriazoles in the settled sewage in ng/l at Hallam Fields sewage treatment work.

Hallam Fields STW	BT	TT
AM	1050	1435
AM	1400	4420
AM	2545	1870
AM	No Peak	No Peak
PM	3330	3530
PM	3115	3895
PM	2595	3365
PM	3340	4380

Concentrations of benzotriazoles in the settled sewage in ng/l at Newthorpe sewage treatment work.

Newthorpe STW	BT	TT
AM	1705	1840
AM	1860	2210
AM	3220	4490
AM	2620	3400

Concentrations of benzotriazoles as a RGF feed in ng/l at Hallam Fields sewage treatment work.

Hallam Fields STW	BT	TT
AM	800	1150
AM	2345	2240
AM	545	780
AM	2790	3315
PM	1310	1725
PM	2490	2720
PM	1655	2230
PM	2290	2745

Concentrations of benzotriazoles as a RGF feed in ng/l at Newthorpe sewage treatment work.

Newthorpe STW	BT	TT
AM	560	745
AM	1150	1895
AM	1890	3030
AM	2150	2830

Concentrations of benzotriazoles as a final effluent in ng/l at Hallam Fields sewage treatment work.

Hallam Fields STW	BT	TT
AM	670	855
AM	2395	1985
AM	745	1005
AM	2100	2070
PM	1425	1615
PM	NO IS	NO IS
PM	1175	1345
PM	2150	2095

Concentrations of benzotriazoles as a final effluent in ng/l at Newthorpe sewage treatment work.

Newthorpe STW	BT	TT
AM	469	695
AM	1035	1605
AM	320	511
AM	418	507

Appendix 2: Advanced Treatments

Concentrations of progestogens as a final effluent (pre-) in ng/l.

Hallam Fields STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	< 0.2	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	5.8	9.6	5.2	8.8	5.5	8.0	1.1	8.3	7.2	< 1.6
AM	0.3	< 1	19.2	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	< 0.2	< 1	5.8	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
PM	1.1	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	24.9	< 0.4	39.3
PM	1.2	7.7	20.1	4.9	3.1	3.6	1.4	9.2	5.6	11.7
PM	2.8	< 1	3.5	< 0.4	15.7	< 0.4	< 1	< 2	< 0.4	< 1.6
PM	< 0.2	< 1	< 1.6	< 0.4	4.1	< 0.4	< 1	< 2	< 0.4	< 1.6

Concentrations of progestogens after O_3 treatment in ng/l.

Hallam Field STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	2.1	< 1	< 1.6	< 0.4	0.5	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	2.6	< 1	< 1.6	< 0.4	2.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	1.1	5.6	3.0	3.5	5.5	2.7	1.1	4.9	4.0	4.4
AM	< 0.2	< 1	4.2	2.7	4.9	4.5	< 1	< 2	3.7	< 1.6
PM	< 0.2	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
PM	1.3	< 1	6.5	< 0.4	< 0.4	< 0.4	< 1	25.7	< 0.4	22.6
PM	2.0	13.6	4.1	4.9	10.2	5.2	2.1	6.9	6.5	10.2
PM	2.7	6.1	3.8	3.6	2.5	2.8	1.3	< 2	3.8	< 1.6

Concentrations of progestogens after GAC treatment in ng/l.

Hallam Field STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	< 0.2	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	< 0.2	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	1.1	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	< 0.2	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
PM	< 0.2	< 1	1.3	< 0.4	< 0.4	< 0.4	< 1	21.5	< 0.4	31.2
PM	< 0.2	9.9	7.6	9.9	6.4	3.0	1.7	6.9	5.6	12.1
PM	< 0.2	< 1	< 1.6	< 0.2	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
PM	< 0.2	2.7	3.3	2.0	3.0	2.5	< 1	< 2	1.4	< 1.6

Concentrations of progestogens after ClO_2 treatment in ng/l.

Hallam Field STW	CPA	DSP	DHG	MDP	MPA	MTA	NTD	NGL	PGT	TBL
AM	1.4	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	0.4	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	0.4	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
AM	1.8	7.4	4.1	3.3	3.1	4.1	< 1	6.0	6.2	4.2
PM	< 0.2	< 1	< 1.6	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
PM	1.7	5.1	3.1	4.6	5.1	4.3	1.5	4.3	3.7	< 1.6
PM	1.8	< 1	1.7	< 0.4	< 0.4	< 0.4	< 1	< 2	< 0.4	< 1.6
PM	3.9	4.1	1.8	2.0	2.1	2.9	1.7	< 2	2.0	< 1.6

Concentrations of benzotriazoles in the final effluent (pre-) and after the three technologies in ng/l.

		BT	TT
Final Effluent	**AM**	625	2585
	PM	410	1920
Ozone (O_3)	**AM**	109	424
	PM	163	510
Granular Activated Carbon (GAC)	**AM**	47	59
	PM	53	42
Chlorine dioxide (ClO_2)	**AM**	133	410
	PM	148	528

Appendix 3: Degradation Test

Main measured parameters throughout the test (n=15).

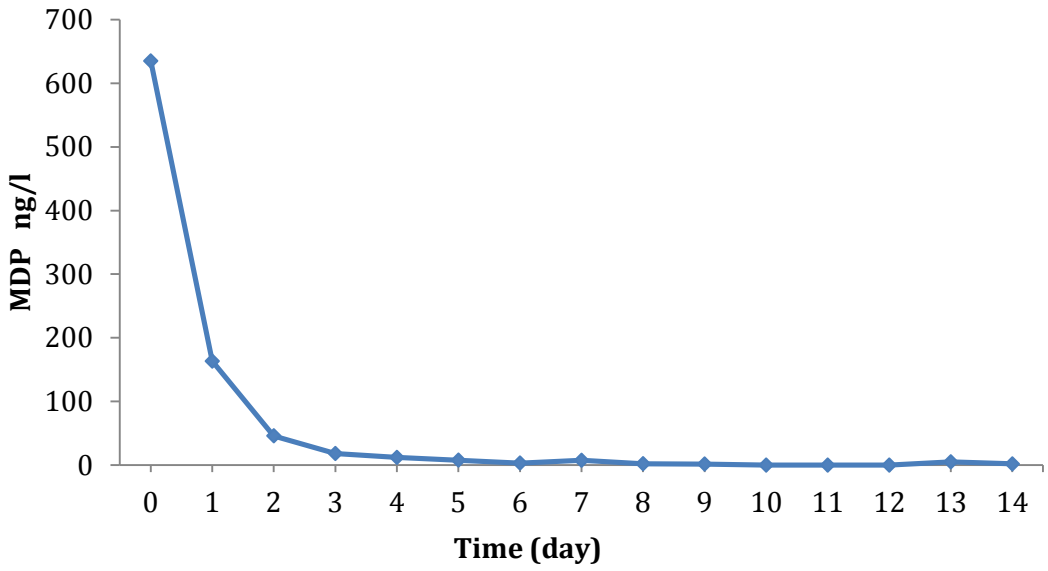
	PH		Dissolved Oxygen mg/l		Temperature C	
DAY	Tank 1	Tank 2	Tank 1	Tank 2	Tank 1	Tank 2
0	8.69	8.69	7.33	7.8	20.7	20.4
1	8.58	8.63	7.61	7.93	21	20.4
2	8.69	8.69	7.53	7.64	22.3	20.8
3	8.58	8.58	7.69	7.78	21.5	19.3
4	8.59	8.56	7.92	8	19.6	19.3
5	8.68	8.57	8.24	8.2	21.5	21
6	8.61	8.65	8.12	8.1	22.8	22.1
7	8.59	8.6	8.2	8.28	21.1	21
8	8.59	8.6	8.35	8.46	20.8	19.6
9	8.6	8.61	8.31	8.21	21.3	20.8
10	8.66	8.67	7.84	8.2	25.5	23.8
11	8.64	8.67	7.8	8	25	23.6
12	8.58	8.66	7.8	7.8	25	24.7
13	8.66	8.67	7.9	7.7	24.7	24.9
14	6.67	6.6	8.7	8.23	24.3	24.7

Concentration of benzotriazoles throughout the 14 day test.

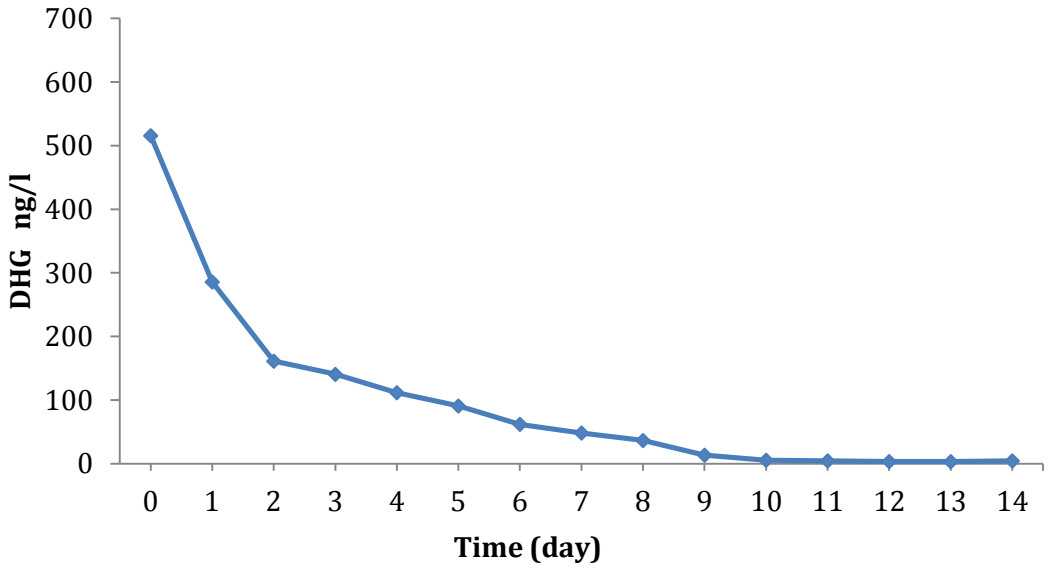
	BT		TT	
Day	Tank 1	Tank 2	Tank 1	Tank 2
0	655	650	2,370	2,365
1	695	670	2,425	2,430
2	640	665	2,410	2,350
3	670	635	2,420	2,440
4	655	650	2,430	2,440
5	650	670	2,485	2,495
6	625	660	2,440	2,485
7	650	625	2,450	2,415
8	680	615	2,405	2,370
9	665	670	2,465	2,420
10	660	645	2,470	2,465
11	655	670	2,415	2,440
12	685	680	2,430	2,505
13	690	680	2,500	2,465
14	685	685	2,505	2,460

Concentration of progestogens throughout the 14 day test.

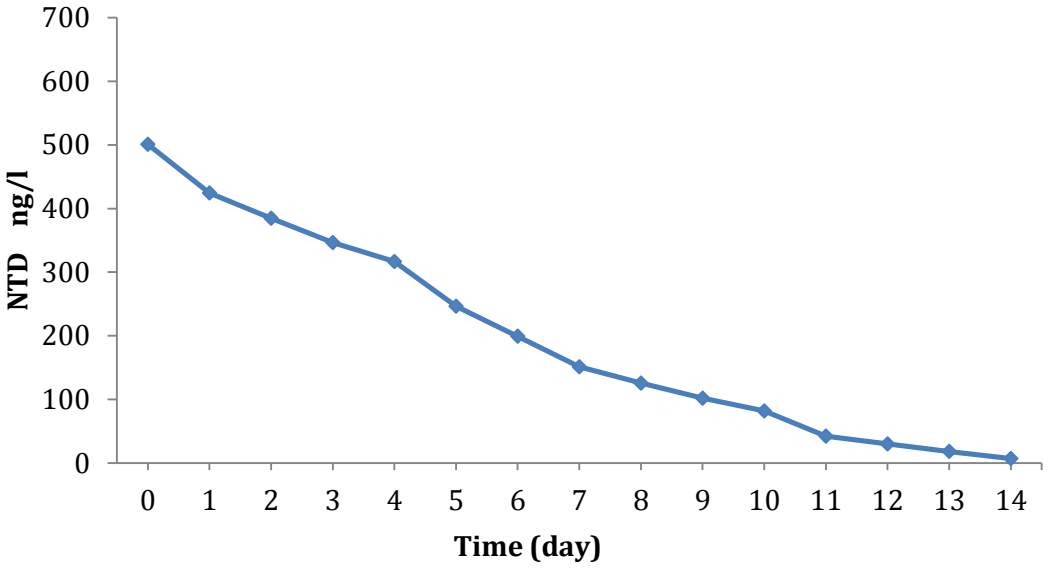
	CPA		DSP		DHG		MDP		MPA		MTA		NTD		NGL		PGT		TBL	
DAY	Tank 1	Tank 2	Tank 1	Tank 2	Tank 1	Tank 2	Tank 1	Tank 2	Tank 1	Tank 2	Tank 1	Tank 2	Tank 1	Tank 2	Tank 1	Tank 2	Tank 1	Tank 2	Tank 1	Tank 2
0	538	509	663	645	532	499	646	623	548	540	568	551	507	495	532	483	401	420	662	627
1	481	429	579	534	295	276	152	175	488	460	494	448	448	401	491	465	6	6	623	603
2	392	371	476	458	148	174	42	50	411	385	422	392	388	381	432	421	5	2	540	585
3	333	363	359	301	132	150	19	17	333	302	314	298	334	359	424	369	< 0.4	< 0.4	515	462
4	304	286	300	262	101	123	6	18	294	296	295	282	318	315	349	346	2	5	423	413
5	296	282	237	236	94	88	9	7	276	273	264	234	255	238	332	290	< 0.4	< 0.4	381	358
6	251	265	182	218	55	69	4	3	228	251	209	214	218	181	224	265	< 0.4	< 0.4	239	276
7	226	195	182	170	50	47	10	5	198	184	190	154	171	132	171	132	8	< 0.4	219	173
8	197	165	118	119	38	35	4	< 0.4	167	146	156	147	138	113	154	141	< 0.4	< 0.4	210	167
9	206	169	118	119	13	15	2	2	152	133	151	116	109	95	148	126	< 0.4	< 0.4	153	138
10	195	169	117	113	7	5	< 0.4	< 0.4	132	103	119	112	97	66	133	104	< 0.4	< 0.4	151	111
11	193	161	102	107	9	<0.8	< 0.4	< 0.4	117	94	98	75	47	38	88	74	< 0.4	< 0.4	89	101
12	130	118	91	77	3	4	< 0.4	< 0.4	75	61	61	51	28	32	46	58	< 0.4	< 0.4	47	50
13	110	96	72	75	3	4	9	1	64	59	58	46	19	17	39	36	8	< 0.4	34	40
14	90	80	49	53	4	5	4	< 0.4	62	54	38	28	6	8	19	19	3	2	13	25



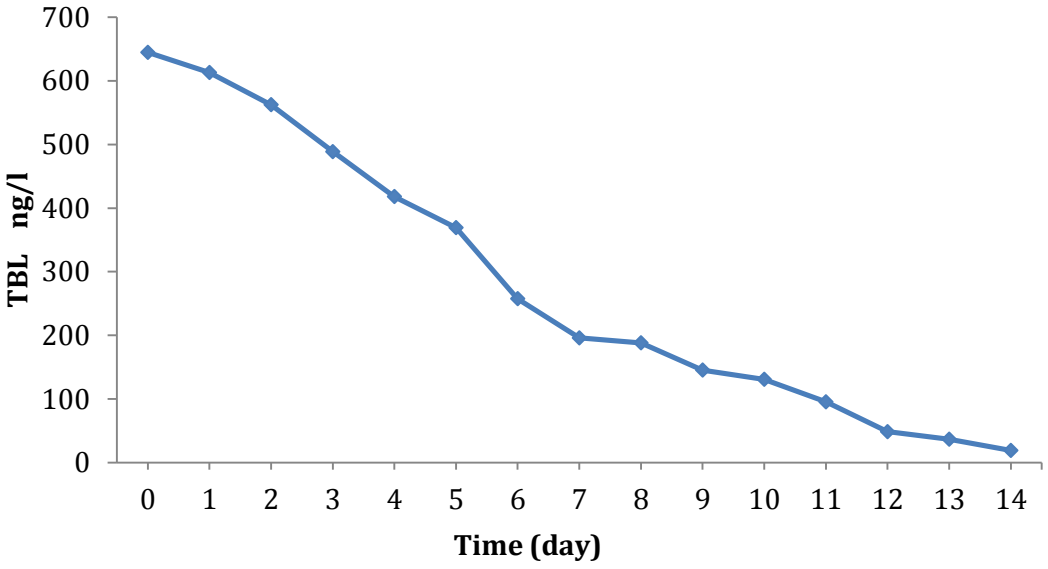
Concentration of medroxyprogesterone throughout the 14 day test



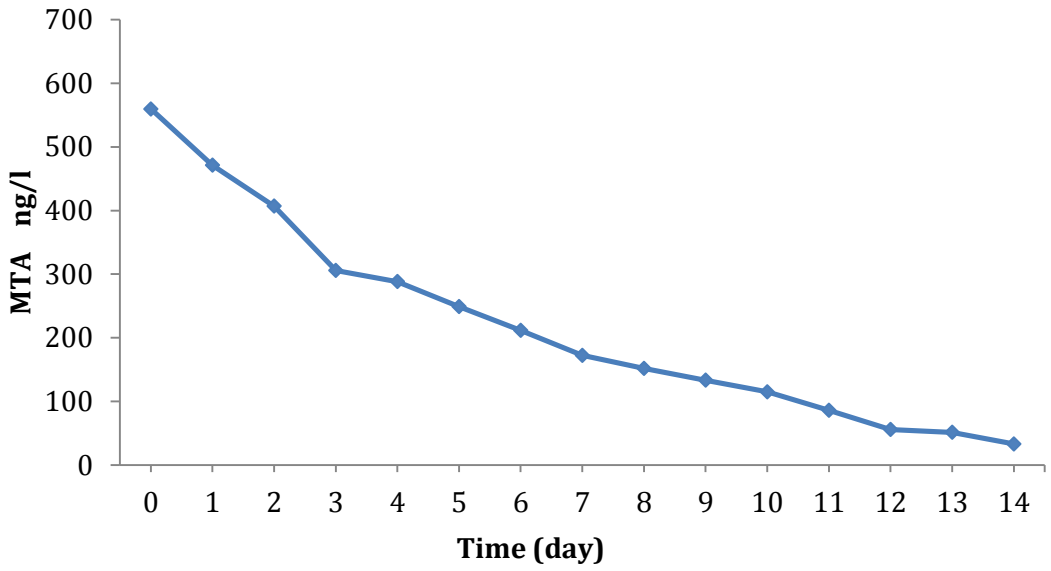
Concentration of dydrogesterone throughout the 14 day test.



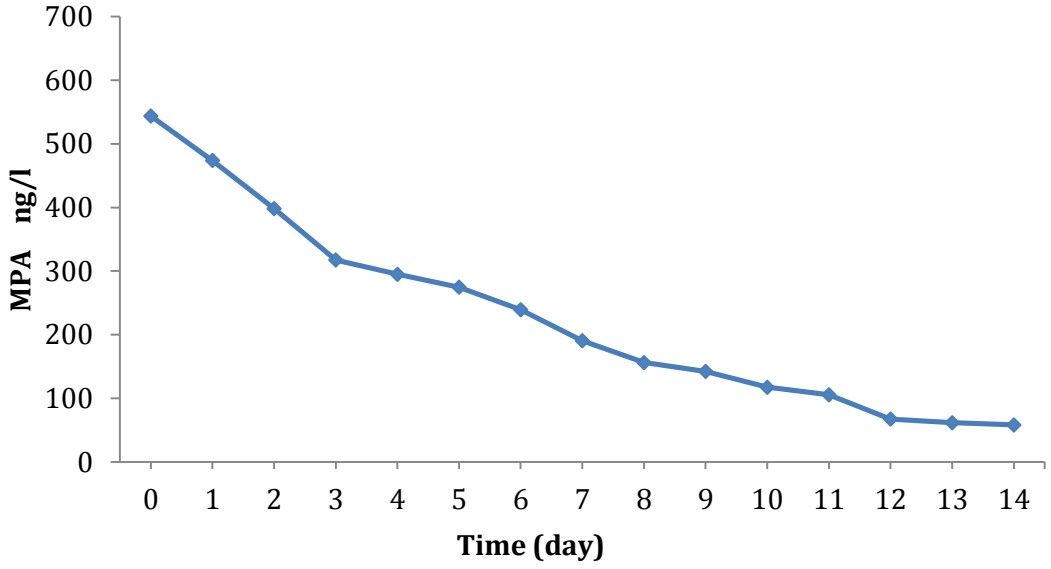
Concentration of norethindrone throughout the 14 day test.



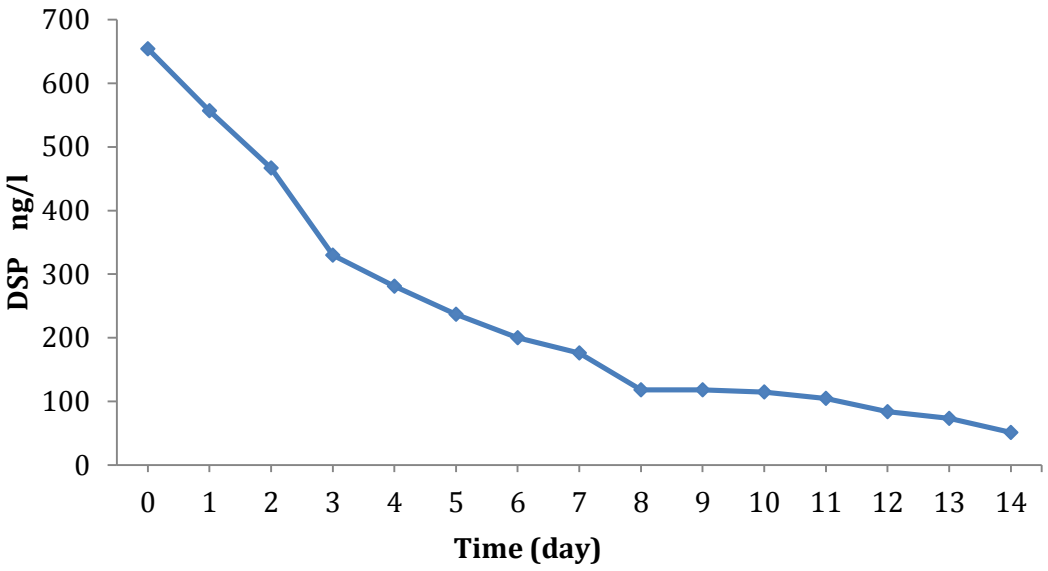
Concentration of tibolone throughout the 14 day test.



Concentration of megestrel-acetate throughout the 14 day test.



Concentration of medroxyprogesterone- acetate throughout the 14 day test.



Concentration of drosprinone throughout the 14 day test.

Appendix 4: Modelling

Fluxes of BT and TT in g/day discharged each day from STW to the River Erewash (6th January 2009).

STW	BT(ng/l)	TT (ng/l)	Q m^3/d	BT g/day	TT g/day
Kirkby FE (N/ASP)	840	2,685	5,970	5.0	16.0
Pinxton FE (N/ASP)	1,335	4,125	4,242	5.7	17.5
Pye Bridge FE (TF)	2,895	5,450	2,048	5.9	11.2
Milnhay FE (N/ASP)	1,655	3,400	7,050	11.7	24.0
Newthorpe FE (TF)	2,470	5,500	10,282	25.4	56.5
Hallam Fields FE (N/ASP)	2,090	5,700	10,022	20.9	57.1
Stapleford FE (N/ASP)	1,375	3,665	5,737	7.9	21.0
Toton FE (N/ASP)	3,605	5,200	16,762	60.4	87.2

Concentrations of BT and TT in ng/l along the River Erewash (6th January 2009).

Sampling point	BT (ng/l)	TT (ng/l)
U/S of Kirkby STW	81	68
D/S Kirkby STW	182	605
D/S Pinxton STW	985	995
D/S Pye Bridge STW	1,270	1,380
U/S of Milnhay STW	1,020	1,335
D/S of Milnhay STW	740	1,105
D/S of Gilt brook confluence	965	1,795
U/S of Hallam Fields STW	930	1,855
D/S of Hallam Fields STW & Nut brook	735	1,520
D/S Stapleford STW	895	1,745
D/S of Toton STW	1,050	1,820

Concentrations of BT and TT in ng/l from brooks as tributary to the River Erewash (6th January 2009).

Sample Name	BT (ng/l)	TT (ng/l)
Bagthorpe brook	23	35
Nethergreen brook	13	33
Gilt brook U/S of Newthorpe STW	118	20
Nut brook	21	38

Fluxes of BT and TT in g/day discharged each day from STW to the River Erewash (29th October 2009).

STW	BT(ng/l)	TT(ng/l)	Q m^3/d	BT g/day	TT g/day
Kirkby FE (N/ASP)	497	2,310	5,970	3.0	13.8
Pinxton FE (N/ASP)	714	2,120	4,242	3.0	9.0
Pye Bridge FE (TF)	646	2,380	2,048	1.3	4.9
Milnhay FE (N/ASP)	496	2,730	7,050	3.5	19.2
Newthorpe FE (TF)	749	2,320	10,282	7.7	23.9
Hallam Fields FE (N/ASP)	867	2,040	1,0022	8.7	20.4
Stapleford FE (N/ASP)	666	2,450	5,737	3.8	14.1
Toton FE (N/ASP)	581	2,070	16,762	9.7	34.7

Concentrations of BT and TT in ng/l along the River Erewash (29th October 2009).

Sampling point	BT (ng/l)	TT (ng/l)
U/S of Kirkby STW	50.8	84.3
D/S Kirkby STW	224	700
D/S Pinxton STW	397	1,080
D/S Pye Bridge STW	431	1,280
U/S of Milnhay STW	426	1,010
D/S of Milnhay STW	382	1,110
D/S of Gilt brook confluence	407	1,570
U/S of Hallam Fields STW	449	1,400
D/S of Hallam Fields STW & Nut brook	338	1,090
D/S Stapleford STW	410	1,420
D/S of Toton STW	434	1,530

Concentrations of BT and TT in ng/l from brooks as tributary to the River Erewash (29th October 2009).

Sample Name	BT (ng/l)	TT (ng/l)
Bagthorpe brook	31	35
Nethergreen brook	10	18
Gilt brook U/S of Newthorpe STW	97	64
Nut brook	16	29

Appendix 5: Drinking Water and Detergents

Concentrations of BT and TT in ng/l from 80 tap water samples around Uxbridge.

Sample Name	BT	TT	Sample Name	BT	TT	Sample Name	BT	TT	Sample Name	BT	TT
Sample_1	50.2	12.8	**Sample_21**	48.8	15.8	**Sample_41**	36.8	14.1	**Sample_61**	46	12.2
Sample_2	51.8	12.6	**Sample_22**	27.6	10.7	**Sample_42**	35.6	12.5	**Sample_62**	47	14
Sample_3	18.5	4.2	**Sample_23**	21.4	4.4	**Sample_43**	15.3	5.6	**Sample_63**	13.6	4.4
Sample_4	26.4	18.9	**Sample_24**	18.9	20.2	**Sample_44**	16.6	17.6	**Sample_64**	17.7	18.7
Sample_5	34.6	14.5	**Sample_25**	13.5	10.2	**Sample_45**	29.4	11.2	**Sample_65**	39.8	13.5
Sample_6	28.6	19.7	**Sample_26**	24.4	34.4	**Sample_46**	30	37.4	**Sample_66**	17.8	20.2
Sample_7	58.2	15.5	**Sample_27**	44.4	12.8	**Sample_47**	32.4	10.5	**Sample_67**	42.4	15.4
Sample_8	44.6	10.5	**Sample_28**	49.4	15.7	**Sample_48**	37.8	12.7	**Sample_68**	42.7	22.8
Sample_9	1.7	0.8	**Sample_29**	0.7	0.7	**Sample_49**	12.6	1.4	**Sample_69**	17.7	1.7
Sample_10	45.2	13.1	**Sample_30**	54	15.5	**Sample_50**	35.8	12.1	**Sample_70**	32.6	17.1
Sample_11	47.4	9.5	**Sample_31**	48	10.5	**Sample_51**	35.8	10.4	**Sample_71**	44	14.6
Sample_12	62.8	49.8	**Sample_32**	79.4	69.8	**Sample_52**	66.8	66	**Sample_72**	67.6	67.4
Sample_13	63.5	44.9	**Sample_33**	62.4	47.6	**Sample_53**	42.4	45.4	**Sample_73**	54	47.6
Sample_14	33	11.5	**Sample_34**	26.2	12.8	**Sample_54**	30.6	10.9	**Sample_74**	45.6	10.5
Sample_15	5.3	0.8	**Sample_35**	0.6	0.5	**Sample_55**	0.8	0.4	**Sample_75**	1.9	0.6
Sample_16	17.7	3.4	**Sample_36**	20.4	7.3	**Sample_56**	28	15.5	**Sample_76**	17.5	10
Sample_17	6.3	0.8	**Sample_37**	0.8	0.5	**Sample_57**	1.1	0.5	**Sample_77**	3.3	0.5
Sample_18	49.4	11.8	**Sample_38**	29.2	11.8	**Sample_58**	31.6	10.7	**Sample_78**	43.6	14.9
Sample_19	21.2	14.4	**Sample_39**	22.5	7.1	**Sample_59**	22.3	11.5	**Sample_79**	42.4	10.1
Sample_20	17.1	2	**Sample_40**	5	1.5	**Sample_60**	9.6	4.4	**Sample_80**	3.9	2.2

Concentrations of BT and TT in ng/l of dishwasher detergents from UK markets.

		mg per 20g		mg per 20g			
		Batch (1)		Batch (2)			
Sample Name	**Type**	**BT**	**TT**	**BT**	**TT**	**Average BT**	**Average TT**
Finish	tablet		57.3		61.7		60
Finish	powder		12.0		21.9		17
fairy	tablet	5.0		4.0		4.5	
Sainsbury's	tablet	1.0		0.8		0.9	
Sainsbury's	powder	17.8		15.0		16.4	
Morrison	tablet	0.6		0.5		0.5	
Morrison	powder	21.2		12.6		16.9	
ASDA	tablet	1.3		---		1.3	
ASDA	powder	2.5		2.1		2.3	
TESCO	tablet	2.9		1.5		2.2	