Electronic Thesis and Dissertation Repository

Treatability of Micropollutants and Microplastics using Selected Wastewater Treatment Processes

Juan Li, *The University of Western Ontario*

Supervisor: Ray Madhumita B, *The University of Western Ontario*
Co-Supervisor: Dagnew Martha, *The University of Western Ontario*
A thesis submitted in partial fulfillment of the requirements for the Doctor of Philosophy degree in Chemical and Biochemical Engineering
© Juan Li 2021

Recommended Citation

Abstract

Micropollutants and microplastics are ubiquitously detected in environment, which are directly linked to human health and ecosystems safety. Conventional wastewater treatment plants (WWTPs) are regarded as a major source for discharging these contaminants into environment. Therefore, it is of great significance to study the behavior of these pollutants and their removal potential in WWTPs. This work investigated specific treatment processes to determine their efficiency in removing target micropollutants and microplastics.

Primary treatments in WWTPs are first step to removal solid particle and other floated materials from the water stream. Therefore, the process is important for microplastics particles. The behavior of microplastic, especially for microfiber from laundry water were investigated in coagulation process. Over 90% removal efficiency of microfibers in pure water and laundry wastewater occurred by ferric chloride and poly aluminum chloride. As 90% of microfibers transferred into primary sludge after coagulation, the effect of microfibers on anaerobic digestion was explored. Microfibers have showed positive effect on anerobic digestion with methane production increased 6% to 35%.

Micropollutants are frequently found at $\mu g/L$-ng/L in wastewater. Many hydrophobic organics tend to adsorb on primary and secondary sludge (biosolids), however show poor removal in anaerobic digestion. Thermal alkaline hydrolysis (TAH) as a pretreatment method for removal of several commonly found micropollutants in biosolids was investigated for improving the safety of biosolid reuse as fertilizer or other land applications. Optimum detection methods for simultaneous detection of five micropollutants from water and biosolids using LC-MS were established. The TAH was found as an effective process to remove micropollutants in biosolids with an average 40% removal efficiency for the target micropollutants.

Additionally, micropollutants also are frequently detected in secondary effluent. The effluent after reverse osmosis can drop to pH 5 to 5.5. Therefore, the pH could be a factor in UV photolysis efficiency of micropollutants for potable reuse of municipal wastewater. Direct UV photolysis was evaluated to remove target micropollutants at varying pH 5.0-8.0. Sulfa, fluroquinolones, and tetracycline group are sensitive to pH and sulfa group showed a high photodegradation rate.

Summary for Lay Audience

Micropollutants and microplastics, are two kinds of contaminants in environment, which come from many sources closely related to daily life. Micropollutants refer to industrial chemicals, pharmaceuticals, and personal care products such as antibiotics, hormones such as estrogen, synthetic musk, cleaners, and disinfectants. Microplastics are tiny plastic pieces with a diameter less than 5 mm from direct plastic fragments or breakdown of larger plastic pieces under environmental weathering activities. Both micropollutants and microplastics are regarded as global environmental problems due to their possible negative effects on human health and ecosystems. Generally, wastewater treatment plants (WWTPs) play an important role on protecting human and environment from contaminants as barriers. Therefore, the removal of several micropollutants and microplastics were studied in this work.

This research aimed at investigating various treatment processes in WWTPs related to the elimination of micropollutants and microplastics including coagulation, anaerobic digestion, post-treatment of anaerobic digestion, and UV disinfection. The results indicated coagulation as a process in primary treatment in WWTP, can remove over 90% microfibers by adding coagulants. Most of microfibers retained in sludge and enhanced methane production in anaerobic digestion. Furthermore, post-treatment with thermal-alkaline at pH 11.0 removed 40% of target micropollutant. This is beneficial for reuse of biosolids as fertilizer or other land applications.

UV disinfection is a promising process for potable reuse application. Selected micropollutants were removed efficiently by direct UV photolysis, in which pH affected the UV photodegradation performance. The results in this work are useful for understanding the treatment performance of coagulation, thermos-alkaline treatment, and UV photolysis for the removal of micropollutants and microplastics in WWTPs.

Keywords

Co-Authorship Statement

This This PhD thesis contains materials that are published, 'submitted' or in preparation for submission in peer reviewed journals as listed below.

Chapter 3: **Effect of coagulation on microfiber in laundry water**

Authors: Juan Li, Martha Dagnew, Madhumita B Ray

The primary author of this chapter was Juan Li under the supervision of Dr. Ray, Dr. Dagnew and. Juan conducted all the experimental work, data collection and analysis, interpretation, and drafting the manuscript with guidance from Dr. Ray, Dr. Dagnew.

Status: In preparation for submission to Science of the Total Environment.

Chapter 4: **Effect of microfiber and ozone pretreated microfiber on anaerobic digestion**

Authors: Juan Li, Martha Dagnew, Madhumita B Ray

The primary author of this chapter is Juan Li under the supervision of Dr. Ray, Dr. Dagnew and. Juan conducted all the experimental work, data collection and analysis, interpretation, and drafting the manuscript with guidance from Dr. Ray, Dr. Dagnew.

Status: In preparation for submission to Bioresource Technology.

Chapter 5: **Simultaneous quantification of five pharmaceuticals and personal care products in biosolids and their fate in thermo-alkaline treatment**

Authors: Juan Li, Lyne Sabourin, Justin Renaud, Samantha Halloran, Ajay Singh, Mark Sumarah, Martha Dagnew, Madhumita B Ray

The primary author of this chapter was Juan Li under the supervision of Dr. Ray and Dr. Dagnew. Juan Li, conducted all experiments, analyzed the data, and prepared the initial draft of the manuscript. Drs Dagnew and Ray have conceptualized the work, revised the manuscript, and provided funding and overall supervision of the work. Ms. Sabourin, Mr. Renaud, and Dr. Sumarah helped with the method development and LC-MS analysis at the AAFC-LRDC. Ms. Halloran and Dr. Ajay Singh provided the Lystek treated sludge and provided information about thermo-alkaline hydrolysis.

Status: Published

Reference: Li, J., Sabourin, L., Renaud, J., Halloran, S., Singh, A., Sumarah, M., ... & Ray, M. B. (2021). Simultaneous quantification of five pharmaceuticals and personal care products in biosolids and their fate in thermo-alkaline treatment. Journal of Environmental Management, 278, 111404.

https://doi.org/10.1016/j.jenvman.2020.111404

Chapter 6: **pH-dependence molar absorptivity of selected micropollutants and effect on UV photolysis**

Authors: Juan Li, Martha Dagnew, Madhumita B Ray

The primary author of this chapter is Juan Li under the supervision of Dr. Ray, Dr. Dagnew. Juan conducted all the experimental work, data collection and analysis, interpretation, and drafting the manuscript with guidance from Dr. Ray, Dr. Dagnew.

Status: In preparation for submission to Chemosphere.

Acknowledgments

It is my great pleasure to express my sincere gratitude to a great number of people who helped me in different ways throughout my PhD studies. First, I would like to convey my gratitude to my supervisor, Professor Madhumita Ray, for her support and guidance throughout my PhD research. Thank you for your patience, motivation, constant encouragement, and your immense knowledge. I would like also to thank my co- supervisor Dr. Martha Dagnew for her excellent encouragement, insightful comments, and brilliant support during my PhD. Thank you for taking me as your student to pursue my post- graduate studies in Canada.

I also offer my enduring gratitude to my colleagues and friends for your cooperation, assistance, and advice. In particular, thanks to Dr. Tulip Chakraborty, Sreejon Das, Siddharth Gupta, Dr. Ziming Zhao, from our research group; and Sun Lin, Wenjuan Mu, Shabir Razavi, Dr. Wudneh Ayele Shewa, Mohammad Noor Tamin from Dr. Dagnew's research group; Natasha Desrochers, Shawn Hoogstra, Megan Kelman, Dr. Justin Renaud, Jacob Walsh, Lyne Sabourin, Dr. Mark Sumarah from Agriculture and Agri-Food Canada. I really enjoyed working with you all.

To my family; I don't know how many words would convey my thanks to my parents, Liping Guo and Shunhe Li, who are sharing with my happiness, my achievement supporting and encouraging when I am crying, disappointed and frustrated. They are the ones who always supported me for pursuit my dream.

Finally, I want to thanks to myself, PhD is not an easy journey. During the past four years I went through wandering, doubting, hurting, hoping for some easy way out, but eventually, I insisted to shape myself a better person and better researcher. Drawing on a famous school motto, *I was built to be a mountain not a creek, rising to the high peaks with the small valleys at my feet. I was born to be great, not worthless, standing on the shoulders of the giants, the petty cowards beneath me.* Little girls, dream with ambition, lead with conviction, and see yourselves in a way that others may not, simply because they have never seen it before.

Table of Contents

List of Tables

List of Figures

List of abbreviations and symbols

AD   anaerobic digestion

ACN   acetonitrile

APCI   atmospheric pressure chemical ionization

BOD   biochemical oxygen demand

BPA   bisphenol A

BMP   biochemical methane potential

CH_4 methane

CIP   ciprofloxacin

CFX   ciprofloxacin

COD   chemical oxygen demand

CUPs   current-use pesticides

DC   doxycycline

DCM   dichloromethane

DI   deionized water

DFC   diclofenac

EMCs   endocrine-modulating chemicals

FLX   fluoxetine

FQs   fluoroquinolones

FTIR   fourier transform infrared spectroscopy

GC   gas chromatography

HCl   hydrochloric acid

HLB   hydrophilic-lipophilic balance

ILC   isotopically labelled compounds

K_d partition coefficient

K_{ow} octanol water coefficient

LC   liquid chromatography

LOD   limits of detection

LOQ   limits of quantification

MeOH   methanol

MIC   miconazole

MS   mass spectrometry

NaOH   sodium hydroxide

OFLX   ofloxacin

PA   polyamide

PAC   polyacrylate

PACl   polyaluminum chloride

PE   polyethylene

PES   polyester

PET   polyethylene terephthalate

pKa   acid dissociation constant

PP   polypropylene

PPCPs pharmaceuticals and personal care products

PS   polystyrene

PVC   polyvinyl chloride

QuEChERS quick, easy, cheap, effective, rugged and safe

RSD   relative standard deviation

sCOD   soluble chemical oxygen demand

SMX   sulfamethoxazole

sP   soluble phosphorous

SPE   solid phase extraction

SSE   signal suppression/enhancement

STZ   sulfathiazole

SXZ   sulfisoxazole

tCOD   total chemical oxygen demand

TAH   thermo-alkaline hydrolysis

TCC   triclocarban

TCS   triclosan

TP   total phosphorous

TWAS thickened waste activated sludge

TS   total solids

TSS   total suspended solids

VS   volatile solid

VSS   volatile suspended solids

UV   ultraviolet

WWTP wastewater treatment plant

List of Appendices

Chapter 1

1   Introduction

1.1   Rationale

Micropollutants and microplastics have become global concerns in recent years due to their potential negative effects on ecosystem and human health. Municipal wastewater treatment plants (WWTPs) are usually regarded as first barrier to protect water quality by removing these harmful contaminants before water can be discharged or reused. However, many micropollutants are found in WWTP effluents and aquatic systems at µg/L to ng/L level (Bollmann et al., 2019). Similarly, microplastics are also found at 0.28 to 3.14×10^4 particles/L and 0.01 to 2.97×10^2 particles/L in influent and effluent, respectively (Liu et al., 2021). The inadequate performance of WWTPs lead to major discharge of the micropollutants and microplastics into environment. Previous studies found that micropollutants can cause accumulation in food chain, hormone imbalance, long-term chronic effects, antibiotics resistance gene (Patel et al., 2019). Microplastics also can be vectors for micropollutants by absorbing them and releasing into the environment (Hüffer et al., 2019). Therefore, it is of great significance to investigate their behavior in WWTPs and efficacy of potential treatment processes for them.

As literature reported, primary treatment can remove most of microplastics compared to secondary and tertiary treatment. Coagulation is the main technology for microplastics removal in primary process, which have achieved 70% to 90% removal. Therefore, further investigation has been recommended focusing on the removal behavior of microplastic during coagulation process. After coagulation, over 90% microplastics were retained in sludge at an average concentration around 22.7×10^3 particles/kg dry sludge according to recent publications (Zhang et al., 2020). Municipal WWTPs are expected to satisfactorily the demands of cities. Sludge or biosolids can be recycled and used as fertilizer after treatment and stabilization for sustainable development. Those contaminants retained in biosolids have potential physical and chemical damages in environment such as: internal abrasions or disruption of the digestive system, reduced growth and reproduction (Mathalon et al., 2014). Anaerobic digestion is widely used to stabilize sludge and produce

renewable biosolid resources. The effect of microplastics on anaerobic digestion need to be clarified.

The occurrence and fate of micropollutants vary with compounds as well as WWTPs, mainly due to their physical and chemical characteristics, such as octanol-water coefficient (K_{ow}) and solubility (Dubey et al., 2021). Musk fragrances or hormones group of compounds with medium or high K_{ow} are more likely attached to sludge (Alvarino et al., 2018). The existence of micropollutants in biosolid have potential risk to environment and human. Thermal alkaline hydrolysis treatment is an innovative commercial technology to remove pathogens and provide high solid, low viscosity products. The good application prospect of removing micropollutants in biosolid should be investigated.

Hydrophilic micropollutants such as caffeine, sulfamethazine, and sulfamethoxazole are frequently detected in liquid stream/water (Quesada et al., 2019). UV advanced oxidation process (AOP), a proven technology for trace organic contaminants removal in drinking water treatment, there is a higher level of performance validation requirements given public health risks associated with potable reuse of wastewater. In general, the effluent from reverse osmosis treatment presented a lower pH range. The effect of pH on removal of UV photolysis require to be demonstrated, which can provide information for the application of UV for a pilot reuse process and improve energy efficiency.

Therefore, micropollutants and microplastics are significant concerns in reuse applications where robust and eco-efficient technologies are urgently needed. The technologies involved in this PhD research will be based on coagulation, anaerobic digestion and pre/post treatment, and direct UV photolysis process, which are common technologies in WWTPs. The development and investigation of these technologies for reuse application are very much needed. This research is directed towards addressing some of these issues as presented in the objectives below.

1.2 Objectives

The overall objective of this PhD thesis is to investigate the effect and removal of micropollutants and microplastics during specific process during wastewater treatment. The specific objectives are outlined below:

a) To investigate effect of coagulation on microfibers in laundry water, demonstrate the removal efficiency and mechanism of microfibers in laundry water during coagulation via ferric and polyaluminum chloride (PACl);

b) To evaluate impact of microfibers and ozone pretreated microfibers on anaerobic digestion and phosphorus removal;

c) To quantify pharmaceuticals and personal care products (PPCPs) in biosolids; determine partition of these compounds between water and various sludge, and investigation of the fate of model PPCPs in thermal and alkaline treatment;

d) To study and compare pH dependence molar absorptivity of model micropollutants and effect of pH on select micropollutants during direct UV photolysis. Although most processes run at natural pH of water around 7.0, the pH of permeate from reverse osmosis can be about 5.0.

1.3 Thesis organization

Chapter 1 presents a brief overview of the thesis and the rationale behind assessing the importance to investigate micropollutants and microplastics in WWTPs. It briefly summarizes the background, underlines the demand for this research and provides the specific research goals.

Chapter 2 provides a comprehensive literature review on micropollutants and microplastics including their definition and impact on environment and human, fate and occurrence in WWTPs, quantification of micropollutants and microplastics, and emerging and conventional processes for the treatment of microplastics and micropollutants. Additionally, the review also presents the research knowledge gaps and scope of further research.

Chapter 3 is a research article entitled "Effect of coagulation on microfibers in laundry water". The objective of this work was to understand the behavior of microfibers from practical laundry process during coagulation, where ferric chloride and PACl were used as coagulants. The removal efficiencies were 86%-96% and 30%-94% in control study and laundry water, respectively. Additionally, the presence of surfactant in detergent in laundry wastewater reduced the removal efficiency of microfibers by coagulation.

Chapter 4 is a research article entitled "Effect of microfiber and ozone pretreated microfiber on anaerobic digestion" In this study, the effect of various abundance of microfibers in anaerobic digestion was investigated. The methane production was enhanced by low concentration of microfibers, while inhibited at high concentration. Phosphorus was found to adsorb in microfibers in control study during coagulation, and also removed during anaerobic digestion due to the addition of microfiber. With increasing microfibers concentration, the removal efficiency of phosphorus was increased. Furthermore, after ozone pretreatment, the removal efficiency of P was also increased.

Chapter 5 is a published research article entitled "Simultaneous quantification of five pharmaceuticals and personal care products in biosolids and their fate in thermo-alkaline treatment". The aim of this study was to develop and optimize the simultaneous detection method for the selected PPCPs from complex biosolids matrix by LC-MS in ng/L range. The partition of these PPCPs between water and sludge was determined, which suggested 89%–98% sorption onto solid phase due to their high octanol-water coefficients. The compounds were detected in the range of 54 ± 3 to 6166 ± 532 ng/g in raw biosolids collected from a local WWTP. About 42% to 99% degradation of these compounds occurred after thermo-alkaline hydrolysis (TAH).

Chapter 6 is a research article entitled "pH dependence molar absorptivity of selected micropollutants and effect on UV photolysis." This work investigated the effect of pH on molar absorption of 12 micropollutants at 254 nm, and on direct UV photolysis by a UV collimated beam apparatus. Furthermore, the behavior of absorption scans from 200 nm to 800 nm at pH 5.0-8.0 were also studied, improving our understanding for the potential of photolysis using alternative sources of radiation.

Chapter 7 is the summary of major research findings along with some recommendations for future research.

1.4   Thesis format

This thesis has been prepared in the integrated-article format according to the specifications provided by the School of Graduate and Postdoctoral Studies located at the University of Western Ontario. Chapter 3 has been prepared for submission to Science of the Total Environment. Chapter 4 has been prepared for submission to Bioresource Technology. Chapter 5 of this thesis has been published in Journal of Environmental Management. Chapter 6 has been prepared for submission to Chemosphere.

1.5   Reference

Alvarino, T., Suarez, S., Lema, J., & Omil, F. (2018). Understanding the sorption and biotransformation of organic micropollutants in innovative biological wastewater treatment technologies. *Science of The Total Environment*, *615*, 297–306. https://doi.org/https://doi.org/10.1016/j.scitotenv.2017.09.278

Bollmann, U. E., Simon, M., Vollertsen, J., & Bester, K. (2019). Assessment of input of organic micropollutants and microplastics into the Baltic Sea by urban waters. *Marine Pollution Bulletin*, *148*, 149–155. https://doi.org/https://doi.org/10.1016/j.marpolbul.2019.07.014

Dubey, M., Mohapatra, S., Tyagi, V. K., Suthar, S., & Kazmi, A. A. (2021). Occurrence, fate, and persistence of emerging micropollutants in sewage sludge treatment. *Environmental Pollution*, *273*, 116515. https://doi.org/https://doi.org/10.1016/j.envpol.2021.116515

Hüffer, T., Wagner, S., Reemtsma, T., & Hofmann, T. (2019). Sorption of organic substances to tire wear materials: Similarities and differences with other types of microplastic. *TrAC Trends in Analytical Chemistry*, *113*, 392–401.

Liu, W., Zhang, J., Liu, H., Guo, X., Zhang, X., Yao, X., Cao, Z., & Zhang, T. (2021). A review of the removal of microplastics in global wastewater treatment plants: Characteristics and mechanisms. *Environment International*, *146*, 106277.

Mathalon, A., & Hill, P. (2014). Microplastic fibers in the intertidal ecosystem surrounding Halifax Harbor, Nova Scotia. *Marine Pollution Bulletin*, *81*(1), 69–79.

Patel, M., Kumar, R., Kishor, K., Mlsna, T., Pittman, C. U., & Mohan, D. (2019). Pharmaceuticals of Emerging Concern in Aquatic Systems: Chemistry, Occurrence, Effects, and Removal Methods. *Chemical Reviews*, *119*(6), 3510–3673. https://doi.org/10.1021/acs.chemrev.8b00299

Quesada, H. B., Baptista, A. T. A., Cusioli, L. F., Seibert, D., de Oliveira Bezerra, C., & Bergamasco, R. (2019). Surface water pollution by pharmaceuticals and an

alternative of removal by low-cost adsorbents: A review. *Chemosphere*, *222*, 766–780. https://doi.org/https://doi.org/10.1016/j.chemosphere.2019.02.009

Zhang, Z., & Chen, Y. (2020). Effects of microplastics on wastewater and sewage sludge treatment and their removal: A review. *Chemical Engineering Journal*, *382*, 122955. https://doi.org/https://doi.org/10.1016/j.cej.2019.122955

Chapter 2

2 Literature Review

2.1 Micropollutants and Microplastics

2.1.1 Micropollutants

Micropollutants, a wide range of organic chemicals, which cause significant concern due to their potential effect on environmental and human health. The substances may reach the environment by the discharge of partially or fully treated wastewater and sludge to agricultural lands. In the Canadian Environmental annual report (2017-2018), an inventory of approximately 23,000 substances is listed. There are various categories of micropollutants depending on the Chemical Abstracts Service (CAS), including pharmaceutical and personal care products (PPCPs), current-use pesticides (CUPs), and endocrine-modulating chemicals (EMCs). These categories are discussed below.

Pharmaceutical and personal care products (PPCPs)

The term pharmaceutical and personal care products (PPCPs) was first introduced by Christian G. Daughton in the 1999 issue of Environmental Health Perspectives. Most of the pharmaceuticals taken by humans and livestock are not metabolized in organisms but are discharged directly into the environment. In addition to antibiotics and steroids, many PPCPs have been detected in various environmental samples, animal tissues, and human blood. Typical examples include prescriptions and over-the-counter pharmaceutical preparations for the treatment of human or animal diseases (e.g. antibiotics, painkillers, antiepileptic and antihypertensive drugs, contraceptives, anti-inflammatory drugs, etc.) and various kinds of care products used in human daily life (e.g. soap, shampoo, cosmetics, hair gel, hair dye, etc.) (Boyd et al., 2004).

PPCPs includes a variety group of organic compounds and can be separated into two main types, pharmaceuticals, and personal care products (Figure 2.1). Among the pharmaceutical groups, many studies have been dedicated by research communities towards antibiotics for their extensive use in human medicine and agriculture, which might

affect human health and antibiotics resistance in the environment (Ben et al., 2019). For personal care products, antimicrobial agents, such as miconazole, triclosan and triclocarban are received significant due to their frequently detection in environment (Jia et al., 2020).
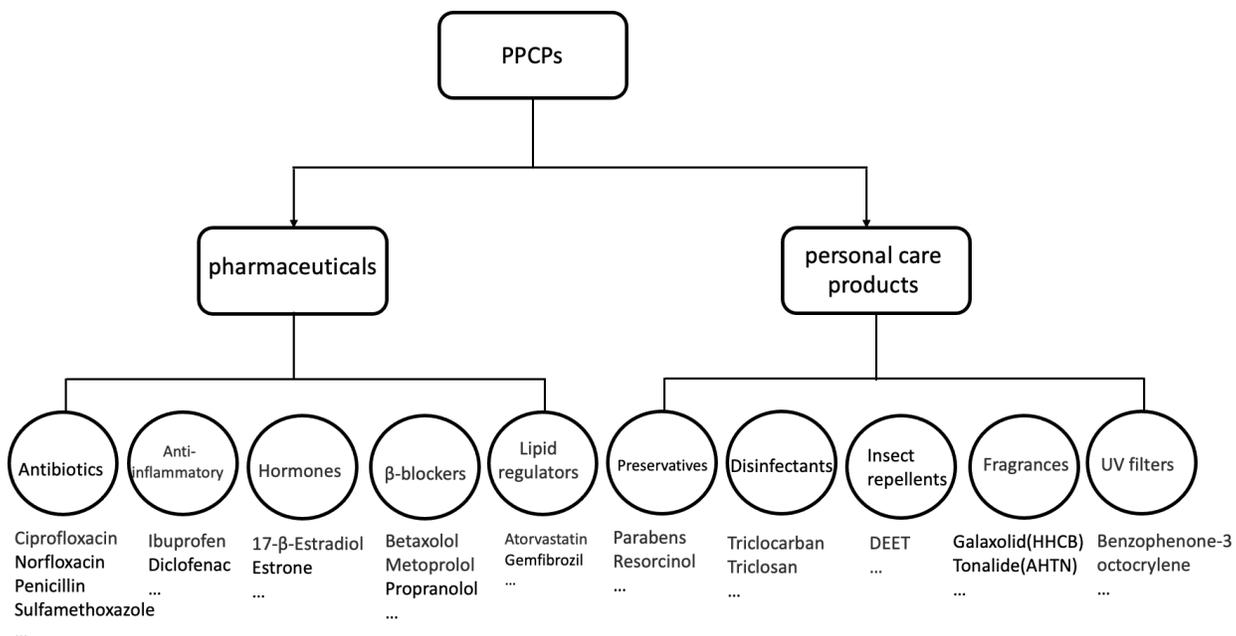
Figure 2.1: Typical classes of PPCPs and their representative chemicals in environment

In 2010, about 12.9 billions, 10 billions, and 6 billions antibiotic pills were consumed in India, China and the United State, respectively (Arun et al., 2017), and in 2012 the total consumption of antibiotics in European (outside hospitals) was 3400 t (Szymańska et al., 2019). More than 80 antibiotics have been detected in the waters of Austria, Germany, Britain, Italy, Spain, Switzerland, the Netherlands, the United States, and Japan. Residues of antibiotics were also detected in soil and sediment, sludge and even in fertilizer (Ternes et al., 2004). Globally, the use of antibiotics has been increased by 65% from 2000 and 2015 (Klein et al., 2018).

Current-use pesticides (CUPs)

CUPs include three major types: insecticides, herbicides and fungicides according to their functional application, such as glyphosate, atrazine and alachlor, which play a key role in

the agriculture to protect crops from insect, unwanted seed or fungal diseases (Lewis et al., 2016). Researchers have shown that CUPs adversely affect both aquatic and terrestrial ecosystems, which cause many health issues (Organization, 2018).

Due to the widely application of pesticides in agricultural land and their potential risks in environment, the occurrence and fate of pesticides are regularly investigated by research from many parts of the world. Four CUPs (trifluralin, chlorothalonil, chlorpyrifos, and dicofol) have been detected in surface seawater in China, ranging from 59.06 ± 126.94 pg·m-3 to 115.94 ± 123.16 pg·m^{-3}. These CUPs also end up in seawater due to precipitation, irrigation, or air-sea gas exchange (Hamza et al., 2016). Wang et al. (2018) investigated 15 CUPs in the Great Lakes basin and found total concentrations of 0.38–1760 pg/m^3 based on seasonal variations (S. Wang et al., 2018). The total amount of pesticides consumptions has been assessed to reach up to 3.5 million tons globally (A. Sharma et al., 2019), which also has been estimated to a further increased until 2027 due to continuous growth in agriculture industry (Fuhrimann et al., 2020).

Endocrine modulating compounds (EMCs)

EMCs are defined as substances that interfere with the normal function of endocrine or hormone systems. These are chemicals existing in the environment that can interfere with the human or animal endocrine system and may lead to abnormal effects through ingestion and accumulation over time, rather than from acute toxicity. Even if the environmental concentration is very low, they could still cause endocrine imbalance in organisms, resulting in a variety of abnormal phenomena such as reproductive disorders, larval death and even extinction. EMCs include some organic compounds like alkylphenol, alkylphenol polyoxyether, bisphenol A, phthalate, polychlorobiphenyl, etc.

2.1.2   Microplastics

Microplastics (MPs), refer to small synthetic polymer plastic particles smaller than 5 mm in diameter. With the development of human activities and industry, numerous MPs including synthetic fibers, microbeads, and fragments of irregular shape (Chubarenko et al., 2016) and other microplastics in cosmetics and personal care products have been given much attention for decades. Microplastics can be subdivided into two different types,

primary and secondary microplastics (Figure 2.2). Primary microplastics refer to manufactured plastic products with the micro size, including polyester, polystyrene, polypropylene, and polyethylene in personal care products. While secondary microplastics are consisting of the breakdown of large plastic products, such as facial scrubbers, fishing net, films under the environmental stressors.
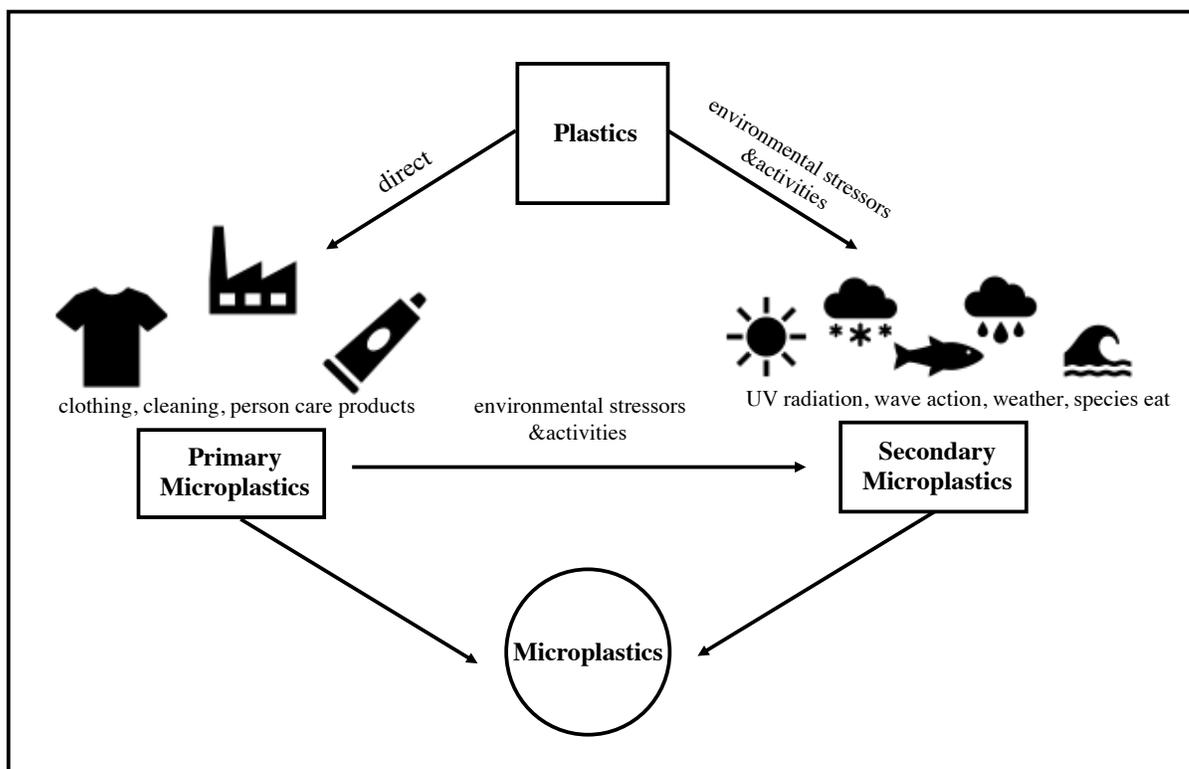
Figure 2.2: Primary and secondary microplastics in environment

2.1.3 Wastewater treatment plants (WWTPs) as a source of micropollutants and microplastics into environment

Generally, a typical wastewater treatment is not specifically designed to remove micropollutants or microplastics. Therefore, major sources of micropollutants as well as microplastics into the environment are the WWTPs (Murphy et al., 2016). Numerous micropollutants and microplastics originate from the treated effluent of conventional WWTPs, in which the microcontaminants are not completely removed. WWTPs act as

primary barriers to protect water bodies of contamination by micropollutants and microplastics. Therefore, it is of great significance to study about the effect and behavior of them in WWTPs.

A major part of most wastewater treatment systems including primary treatment, such as screening, grit removal, primary sedimentation, and primary clarification; secondary treatment which usually are biological processes and secondary settling, and tertiary treatment units also apply to advanced wastewater treatment systems such as membrane filtration process, gas stripping, ion exchange, advanced oxidation process and ultraviolet (UV) radiation disinfection.

1) Effect on primary treatment

Generally speaking, screening is the first unit in WWTPs, which of uniform size that is used to remove suspended solids from wastewater. The clear opening ranging from 6-150 mm (coarse screens), 6 mm (fine screens), and lower than 0.5 μm (microscreens). Considering the small size of microbeads, they may cause blockage to the fine screens and microscreens. For grit chambers, the horizontal-flow grit is generally designed for removal of 0.15 mm and 0.21 mm diameter particles. If MPs size is smaller than 0.15 mm, the process would not be efficient for removal of them and bigger size microplastics have potential effect grit removal process. Furthermore, microplastics can be an issue for coagulation and flocculation processes due to the interaction between their negative charge and alum sulfate, ferric chloride or other chemicals flocculating agents (Enfrin et al., 2019).

2) Effect on biological treatment

The secondary treatment processes are usually biochemical in nature. The effluent from primary treatment is treated in activated sludge plant using various aerobic-anaerobic modes. The sludge from secondary treatment goes for anaerobic digestion. Anaerobic digestion is one of most common treatment processes for sewage sludge stabilization. As mentioned before, 90% of microplastics are retained in sewage sludge, and the abundance of microplastics in sludge can be up to 170,900 particle/kg sludge (Talvitie et al., 2017). Thus, effects of microplastics during anaerobic digestion needs to be studied specifically.

Several studies showed that the presence of microplastics may inhibit methane production, which is one stage of the four steps in anaerobic digestion. PES (200 μm) resulted in an approximately 10% reduction in methane production at various microplastics abundances (Li et al., 2020). Wei et al. have indicated that PVC (1 mm) inhibited methane production by $75.8 \pm 0.2\%$ to $90.6 \pm 0.3\%$ compared to the control. It was reported that the high concentration of PE has inhibited methane production by 12.4%–27.5%, while low concentration of PE showed no significant influence on methane production (Wei et al., 2019). This indicates the effects of microplastics on methane production varied from the size and characteristics. Microplastics show slight negative correlation to hydrolysis rates, which was lower than the control (L. Li et al., 2020). Furthermore, the characteristics of sludge such as higher concentration of carbohydrate, $NH4^+$-N, NO_2-N, may lead to inhibition in anaerobic digestion process. The mechanistic study has found that the toxic components such as bisphenol A (BPA) leaching from PVC usually have more adverse effects on methane production and hydrolysis-acidification process (Wei et al., 2019). However, there is not enough information on the mechanism of inhibitions on anaerobic digestion due to microplastics and further studies are required.

3) Effect of tertiary treatment

Tertiary treatment usually including filtration and disinfection, varies in different treatment plants based on various treatment purposes. Mostly ultrafiltration and microfiltration processes are widely applied in WWTPs, unless tertiary treatment using reverse osmosis is required for potable water reuse application. In general, the abundance of microplastics is likely to cause membrane fouling by blockage, which could result in negative filtration performance(Li et al., 2020). Ma et al. have investigated the effect of PE particles on ultrafiltration membrane fouling. As shown in Figure 2.3, the membrane fouling was gradually aggravated in two stages (Abdelrasoul et al., 2013): first stage happens when size of microplastics particles larger than pores blocks the pores; second stage occurs owing to the accumulation of microplastics with size larger than pore size on the membrane surface forming filter cake. The membrane fouling results in increased transmembrane pressure, which affects pretreatment needs, cleaning requirements, operation time, performance, and increased energy consumptions. Understanding the effect of microplastics on filtration

performance is critical to avoid membrane fouling by microplastics. Thus, further research is needed towards to this purpose to keep filtration effective and stable.
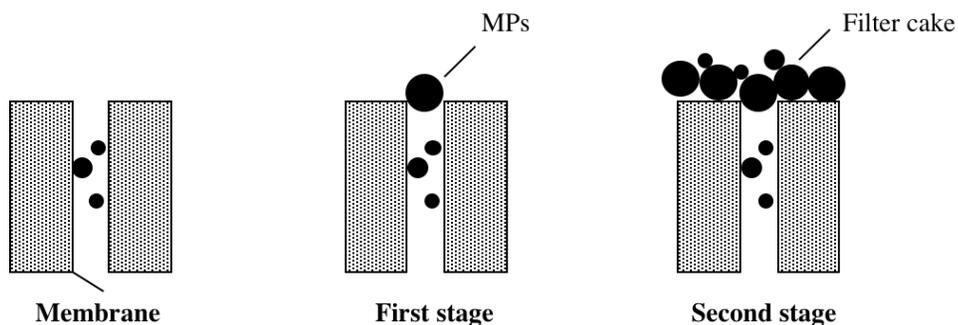
Figure 2.3: Membrane fouling mechanism caused by microplastics (MPs)

2.1.4   Impact of micropollutants and microplastics on environment

Thousands of industrial and natural chemicals related to anthropogenic activities have become one of the key worldwide concerns for human and environment. Although the concentration of micropollutants in the environment is usually detected at low level from ng/L to μg/L, they still have the potential risks to human health and ecological safety associated with a number of negative effects. EDCs are known to feminize males fish in river and lake via mimicking or interfering with natural hormone (Kidd et al., 2007). Bunzel et al. have investigated the effect of agricultural pesticides on macroinvertebrate communities; they indicated that pesticide can alter composition of species and change pesticide-resistance (Bunzel et al., 2013). The accumulated micropollutants in biosolids or soil have been found toxic to plants, affecting the growth of plant, equilibrium of soil ecosystems, and root elongation (Carvalho et al., 2014). The micropollutants on animals and plants may affect further down the food chain, increasing the risk to human health,

Many studies have reported that microplastics were ingested by various organisms and get accumulated in food chain, which led to possible impacts to human health and environmental safety (Murphy et al., 2016). The accumulation and effects of microplastics in aquatic systems have been exhaustively demonstrated in recent decades. In marine systems, microplastics have been frequently detected on both coastlines and deep-sea

locales of many countries (Van Cauwenberghe et al., 2013). In freshwater, microplastics also have been reported in recent publications such as lake Victoria in East Africa (Egessa et al., 2020), St. Lawrence River between Montreal and Quebec City (Crew et al., 2020), and Pearl River estuary in China (Yan et al., 2019). Furthermore, MPs were shown to have negative impacts on leading into other contaminants by adsorbing chemicals such as pharmaceutical and personal care products, current use pesticides, endocrine-modulating chemicals, and heavy metals. Fisner et al., observed adsorption of nonpolar micropollutants such as DDT, PCBs, and PAHs on plastic surface, and ingested by animals (Fisner et al., 2013).

2.2 Occurrence and fate of micropollutants and microplastics

2.2.1 Micropollutants in sediment and sludge

Micropollutants usually present in complex mixtures in WWTPs, in which partition onto solids occur due to their octanol-water coefficient (K_{ow}), partition coefficient (K_d) as well as acid dissociation constant (pKa). Micropollutants with high octanol-water partition coefficients (K_{ow}) are more likely to be partitioned into sludge or soil.

Micropollutants in soil or sediment mainly come from sludge from WWTPs and animal husbandry waste, which can then pollute waterways. When waste sludge or animal husbandry waste is used as fertilizer or soil modifier, the adsorbed micropollutants can be desorbed to groundwater or surface water through irrigation and other land application, and then pollute the water body (Chen et al., 2011). In addition, some micropollutants are adsorbed by plants (Golet et al., 2003), which become enriched in organisms and can enter the food chain. As shown in Figure 2.4, a large amount of micropollutants was detected in sludge. The content of micropollutants in the sludge of WWTPs is generally high, especially in Asia where it may reach mg/kg levels (Yang et al., 2015). The micropollutants can be partitioned to the sludge through adsorption due to their low solubility and high octanol-water coefficient (Venkatesan et al., 2012).
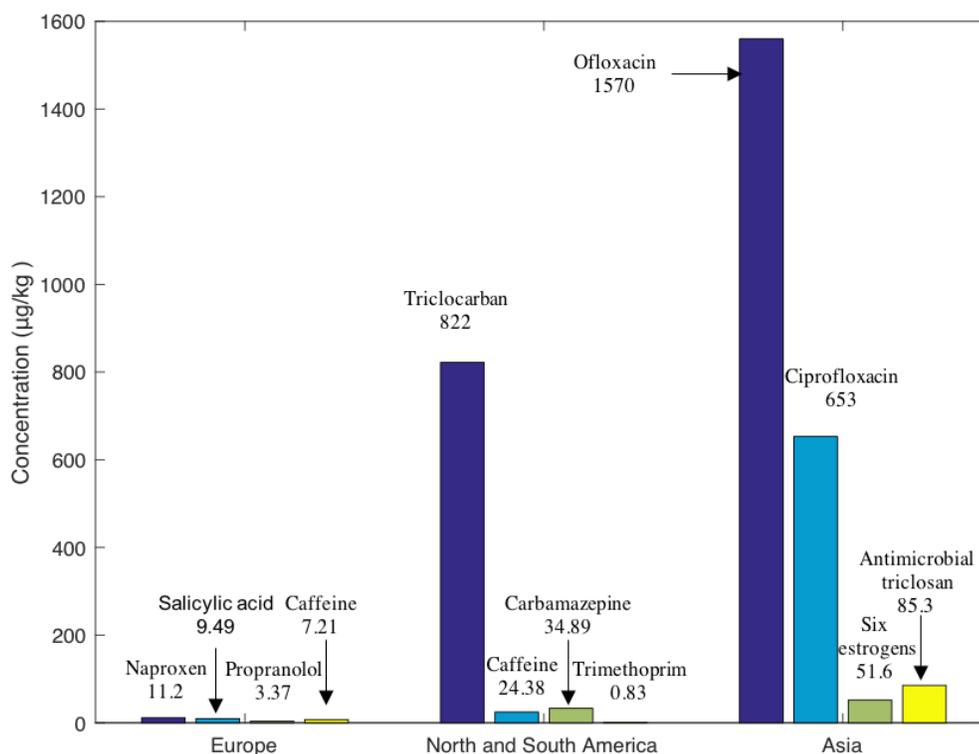
Figure 2.4: Micropollutants in sediment and sludge across difference continents

Sludge treatment and disposal are cost extensive processes in wastewater treatment. Firstly, sludge contains a large number of toxic and harmful substances, pathogenic microorganisms, bacteria, synthetic organic substances and heavy metal ions, which will have adverse effects on the surrounding environment. Secondly, with only 3-5% of solids, sludge volume is high with large amount of water.

2.2.2   Micropollutants in aquatic environment

Usually, a typical wastewater treatment plant (WWTP) is not specifically designed to remove micropollutants. The fate processes for micropollutants in a typical WWTP include primary treatment, secondary treatment, and tertiary treatment, while the removal processes include adsorption, coagulation and sedimentation, volatilization, biodegradation, and abiotic degradation (Das et al., 2017).

The primary treatment removes suspended solids from wastewater through sieve and sedimentation processes. In general, primary processes can remove 30% - 40% of biochemical oxygen demand (BOD), and 55% - 65% of suspended solids. However, micropollutants with high water solubility will not be removed during primary process. For example, sulfamethoxazole was found to only have 17% removal efficiency during the primary treatment due to its low sorption and hydrophilic nature (Yang et al., 2017).

The main task of the secondary treatment is to remove the settleable suspended matter and soluble biodegradable organic matter in wastewater by biological methods such as activated sludge, biofilm etc. The principle of biological treatment is the decomposition of organic matter and the synthesis of organisms through microbial process. Micropollutants removal efficiency significantly relies on the nature of micropollutants and reactor design. Chlortetracycline (>99%), doxycycline (64%), sulfamethoxazole (69%), and fluoxetine (66.7%) showed high removal efficiency in the secondary treatment (Yang et al., 2017). Table 2.1 shows the collected data of some micropollutants in an aquatic environment reported from different countries/regions, including Austria, China, France, Brazil, Italy, Vietnam, Spain, Mexico, Croatia, UK, and the US.

Table 2.1: Micropollutants in aquatic environment in different countries/regions

Selected compounds	Countries or regions	Sampling sites	Influent (μg/L)	Effluent / range (μg/L)	Removal Efficiency (100%)	Reference
Amoxicillin	Saronikos Gulf and the Elefsis Bay	WWTPs		0.005-0.128		(Alygizakis et al., 2016)
	Queensland, Australian	STPs	280	30	89.29	(Watkinson et al., 2007)
Antipyrine	US	Public source waters		<0.001		(Illinois, 2008)
Antipyrine	Vietnam	Groundwater, tap water and bottled water		0.04		(Kuroda et al., 2015)
chloramphenicol	North Italy	Hospitals		0.004-0.006		(Verlicchi et al., 2012)
chloramphenicol	Dalian, China	WWTPs	0-0.026	0-0.011	(-114)-100	(Xin Zhang et al., 2017)
Chlortetracycline	Vietnam	Surface water and wastewater		0.016		(Kuroda et al., 2015)
Chlortetracycline	North Italy	Hospitals		0.062-0.093		(Verlicchi et al., 2012)
Chlortetracycline	Eastern China	WWTFs	0.0001-0.03	0.0003-0.02	0-100	(Dong et al., 2016)
Ciprofloxacin	European countries,	WWTPs	0.413	0.0723	82.49	(Miege et al., 2009)

	Brazil and North America					
Ciprofloxacin	Seine, France	Rivers		<0.01		(Tamtam et al., 2008)
Ciprofloxacin	Guangzhou, China	Tap water		0.006-0.68		(Q.-J. Wang et al., 2010)
Diclofenac	European countries, Brazil and North America	WWTPs	1.34	0.68	49.25	(Miege et al., 2009)
Diclofenac	UK	STWs		0.6		(Ashton et al., 2004)
Diclofenac	Beiyun, China	Rivers		0.008-0.015		(Dai et al., 2015)
Doxycycline	North Italy	Hospitals		0.056-0.097		(Verlicchi et al., 2012)
Doxycycline	Dalian, China	WWTPs	0-0.162	0	0-100	(Xin Zhang et al., 2017)
Fluoxetine	US	STPs		0.012		(Schultz & Furlong, 2008)
Fluoxetine	US	Drinking water		<0.005		(Snyder, 2008)
Fluoxetine	Shanghai, China	WWTPs	0.0024	0.0016	33.33	(Wu et al., 2015)
Fluoxetine	Shanghai, China	Rivers		<0.001		(Wu et al., 2015)

Compound	Location	Matrix				Reference
Fluoxetine	Shanghai, China	Drinking water		0		(Wu et al., 2015)
Hydrochlorothiazide	US	WWTPs		<2.8		(Kostich et al., 2014)
Hydrochlorothiazide	Cuernavaca, Mexico	Surface water	0.015	0.0089	40.67	(Rivera-Jaimes et al., 2018)
Sulfamethoxazole	European countries, Brazil and North America	WWTPs	0.342	0.115	66.37	(Miege et al., 2009)
Sulfamethoxazole	Seine, France	Rivers		0.04-0.14		(Tamtam et al., 2008)
Sulfamethoxazole	UK	STWs		<0.05		(Ashton et al., 2004)
Sulfathiazole	Dalian, China	WWTPs	0-0.004	0-0.002	0-100	(Xin Zhang et al., 2017)
Sulfathiazole	Vietnam	Groundwater, tap water and bottled water		0.00075–0.001.5		(Kuroda et al., 2015)
Sulfathiazole	Seine, France	Nominal WWTPs	0.003-0.009	0-0.009	0-100	(Tamtam et al., 2008)
Sulfisoxazole	Eastern China	WWTFs	0-0.158	0-0.0003	0-100	(Dong et al., 2016)
Sulfisoxazole	Croatia	Surface water		<0.1		(Ivešić et al., 2017)

2.2.3　Microplastics in WWTPs systems

The microplastics entering WWTPs include fiber, films, flakes and spheres (Mahon et al., 2017; Talvitie et al., 2015). Chemically, they commonly are: polyethylene (PE), polypropylene (PP), polystyrene (PS), polyethylene terephthalate (PET), polyester (PES) polyamide (nylon, PA), polyvinyl chloride (PVC), synthetic rubber, polyacrylate (PAC), alkyds, and acrylic (Murphy et al., 2016). The MPs in WWTPs are hardly originate from any direct disposal of plastics items (plastic bags, bottles), manufactured pellets, and no storm water runoffs. Therefore, primary and secondary sources of microplastics in WWTPs are quite different from microplastics source in marine or freshwater system. WWTPs receive microplastic contaminants mainly derived from households and various municipal services.

Most frequently found emission of microplastics, fibers, are originated partly from laundry and textile handling activities (Hernandez et al., 2017), cosmetic/toothpastes (Carr et al., 2016) as well as personal care products such as shower gel, face cleanser and liquid hand soap (Gouveia et al., 2018). The presence of flake, film and fragment microplastics indicate the high impact of decomposition of industrial raw material production and breakdown or abrasion process of packaging (Xu et al., 2019). These studies indicate relativity between the microplastics source and human activities. Transport and removal patterns in WWTPs process are important to consider, as the distribution, effects, and degradation.

2.2.4　Microplastics in the environment

Major sources of microplastics in the environment are the wastewater treatment plants (WWTP) (Murphy et al., 2016). Many studies have reported MPs were ingested by various organisms and get accumulated in food chain, which led to possible impacts to human health and environmental safety (Murphy et al., 2016). Furthermore, MPs were shown to have negative impacts on leading into other contaminants by adsorbing chemicals such as pharmaceutical and personal care products, current use pesticides, endocrine-modulating chemicals, and heavy metals.

The knowledge on fate and transport of microplastics in WWTPs is much lower than that of aquatic environment. Until most recently, the studies of microplastics in WWTPs

recorded high concentrations of microplastics in varied WWTPs across courtiers and regions (Table 2.2). Microplastics have been found in: North America, in the 17 different facilities across United States (Mason et al., 2016), and a major urban WWTP in Vancouver (Gies et al., 2018); in Europe, Schmidt et al. provided an approximation of MPs annual discharge by WWTPs into 10 major river basins in Germany, a 14,000 population equivalents WWTP in Sweden has been demonstrated high abundance occurrence of microplastics (Magnusson et al., 2014). The Seyhan and Yüreğir WWTP in Turkey has detected millions of microplastics in both influent and effluent for 6 days in August (Gündoğdu et al., 2018). In Asia, the occurrence of microplastics in sewage sludge samples from 11 Chinese province have been investigated, which revealed tremendous abundance of microplastics up to hundred trillion (Li et al., 2018). After that, 650 million/day microplastics have been found in seven WWTPs of Xiamen as a typical coastal city in China (Long et al., 2019). Park et al., have investigated a nationwide survey of microplastics in 50 representative WWTPs providing current level of microplastics in Korea.

Table 2.2: Studies on detecting microplastics in WWTPs

Countries or regions	sampling	abundance	size classes (or mesh size)	reference
Vancouver, Canada	Influent	1.76(+/- 0.31) trillion/year	1μm	(Gies et al., 2018)
United States	Effluent	4 million/day (average)	125-355μm,>355 μm	(Mason et al., 2016)
Germany	Effluent	7 trillion /year (average)	10-5000 μm	(Schmidt et al., 2020)
Sweden	Influent	3.2 million/ hour	≥300 μm	(Magnusson & Norén, 2014)
Sweden	Effluent	1770/hour	≥300 μm	
Northern Italy	Effluent	160 million/day	5-1 mm, 1-0.5 mm, 0.5-0.1 mm, 0.1-0.01mm	(Magni et al., 2019)
Turkey	Influent	1 million–6.5 million/day	55 μm	(Gündoğdu et al., 2018)
Turkey	Effluent	220,000–1.5 million/day	55 μm	
China	Sewage sludge	156 trillion/year	37 μm-5 mm	(Li et al., 2018)
Xiamen, China	Effluent	650 million/day	355, 125, 63, and 43 μm	(Long et al., 2019)
Korea	Influent	10-470 /L	5 mm, 1 mm, 300 μm, and 100 μm	(Park et al., 2020)
Korea	Effluent	0.004-0.51 /L	5 mm, 1 mm, 300 μm, and 100 μm	(Park et al., 2020)

The concentration, size distribution and polymer type of microplastics in WWTPs varied with geographical location, analytical methods, population served, sampling season etc. Studies presented in Table 2.2 have demonstrated that WWTPs are a considerable source of contaminations of microplastics release to river, ocean or other waterway; meanwhile, high removal efficiency of MPs from influent to effluent was recorded in various WWTPs, while over 90 % of microplastics remained in WWTPs sludge; furthermore, fibers and fragments were most frequently detected microplastics, while most frequently observed polymer types were polyester and polyamide; in addition, tertiary treatment showed significant differences in removal of microplastics particles.

2.3   Quantifying micropollutants and microplastics

2.3.1   Extraction method of Micropollutants

An effective and valid detection method is a basis to attain micropollutant data in environment. Over the last few decades, studies on determination of micropollutants have been frequently documented. There are two main steps of an analytical method for micropollutants: extraction (usually coupled with clean-up) and analysis. Classical extraction techniques including Soxhlet and ultrasound-assisted extraction have been widely used in leaching organic micropollutants from sludge, due to their high efficiency and low solvent consumption, low energy use (Albero et al., 2019; Gago-Ferrero et al., 2015; Okuda et al., 2009). Novel extraction techniques are focused on ultrasound-assisted liquid extraction, pressurized liquid extraction, QuEChERS, microwave-assisted extraction, supercritical fluid extraction, and matrix solid-phase extraction (Kathi, 2017).

Soxhlet extraction commonly used in solid samples to analyze thermally stable compounds for over one century, is highly efficient for recovery. The conventional Soxhlet extraction operates as a batch system with a thimble-holder or between two plugs of glass wool and gradually extracted using an appropriate solvent. Afterwards, the analyte is dried, reconstituted and transferred into a compatible solvent (EPA, 1996). Modifications of the conventional extractor to shorten extraction time and automating the extraction include high pressure, automation, ultrasound- and microwave-assisted Soxhlet extraction Hirondart et al., 2020; Llompart et al., 2019).

Solid phase extraction (SPE) is widely used in environmental samples to concentrate the target chemicals for analysis. The procedures for SPE are generally as follows (USEPA, 2003): 1) activate extraction medium (disks or cartridges) using same solvents as used in the sample and necessary pH adjustment; 2) slow loading of sample; 3) clean up the cartridge by washing out unwanted components; 4) finally elute the targets analytes with strong solvent. The sample extracts are evaporated under a gentle air stream and then reconstituted in suitable solvent until analysis is performed (Figure 2.5).
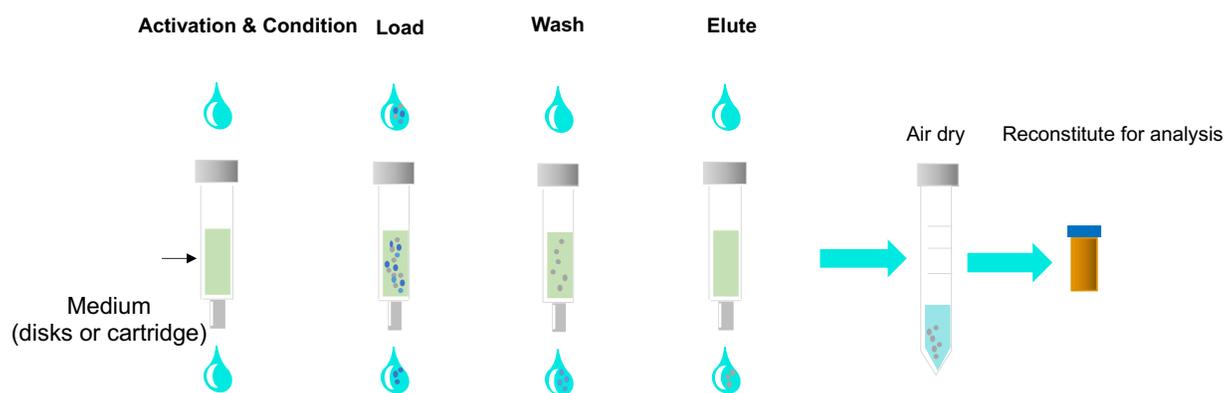
Figure 2.5: General procedures for solid phase extraction

QuEChERS extraction refers to quick, easy, cheap, effective, rugged, and safe extraction, which was developed by Anastassiades et al. (2003) to extract pesticide residues in fruits and vegetables. This method reduces the operating time, solvent volumes, expensive equipment, and processing for extraction. The main process of QuEChERS method consist of extraction, partition, and clean-up. Homogenized sample extracted with organic solvent initially is vortex mixed, followed by phase separation using salt solution (such as NaCl, $MgSO_4$). The solution is centrifuged to separate the solid particulates from the liquid extract. In the last step, the liquid phase is carefully removed and placed into a dispersive SPE (d SPE) containing 100 mg of sodium EDTA for cleanup. Extracts are evaporated and reconstituted for analysis.

2.3.2   Analytical methods for Micropollutants

Extraction method allows to remove interferences, and enrichment the analysts of interest until instrument analysis is performed. Gas chromatography (GC) or liquid chromatography (LC) separation coupled to mass spectrometry (MS) is the most common

analytical method employed for the determination and quantification of micropollutants in environment. GC can be applied for analysis of thermally labile compounds (Calza & Fabbri, 2014), while LC is usually used to separate non-volatile, thermally stable or polar chemicals.

GC-MS/MS GC-MS/MS methods are relatively inexpensive and highly efficient for the detection of volatile substances. The extracts generally introduced to GC system via injection port through an autosampler. Then, the target analytes pass through selected capillary column in GC oven at a programmed temperature. Helium is usually used as the carrier gas. Afterwards, the separated compounds are determined by MS, which can achieve high selectivity and sensitivity with different ionization source. Electron impact ionization (EI) is used routinely in GC-MS for the analysis of micropollutants. Peck et al (2006), reported five classes of micropollutants including triclosan, DEET, HHCB, AHTN etc. detection method using GC-EIMS in surface water and wastewater. EI ionization have the advantages of less matrix effect and large number of mass fragments (Pietrogrande et al., 2007). There are other detectors coupled to GC which can fit the requirements for analysis of micropollutants such as electron capture detector (Daso et al., 2012), triple quadrupole (Cristale et al., 2013), and quadrupole to time-of flight (Schoeman et al., 2017).

Recently, many studies of micropollutants using LC-MS/MS analysis are abundant in literature (Rivera-Jaimes et al., 2018). LC-MS approach has supplemented the scope of GC-MS with the advantages of higher versatility for less volatile and polar analytes. High performance liquid chromatography (HPLC) is most widely used in detection of micropollutant in environment sample using chromatography column to separate target analysis carried by a suitable mobile phase. C_{18} column is frequently applied for identification and quantification of environment samples with high efficiency (Ouyang et al., 2015). Appropriate organic solvents such as methanol (MeOH) and acetonitrile (ACN) are combined with formic acid or ammonium formate (0.01–0.5%). Similarly, HPLC is also coupled to MS with different ionization source. ESI source allows analytes remain un-fragmented and mild ionization, which matrix effect easily affects ESI source performance (Pérez-Lemus et al., 2019). ESI is by far the most commonly used ionization approach for polar analytes (Meng et al., 2021). Except for ESI, atmospheric pressure chemical

ionization (APCI) is a popular source with the advantages of more ionization options for low polar analytes and less background interferences (Silva et al., 2019). The selection of these parameters for LC-MS/MS is mainly dependent on the characteristics of samples and targets analytes. Recent publications about the detection and quantification of micropollutants at certain LC-MS/MS conditions (Table 2.3).

Table 2.3: Determination of micropollutants in environment samples by LC-MS/MS

Target micropollutants	Sample type	Column	Mobile phases	Ionization source	Reference
methadone, oxycodone, lorazepam, aripiprazole, cotinine	wastewater	Zorbax SB-Aq	0.1% formic acid: MeOH	ESI	(Pérez-Lemus et al., 2019)
29 pharmaceuticals, 6 metabolites	sludge form STP	Ultra-Biphenyl	0.1% formic acid: MeOH	ESI	(Subedi et al., 2017)
triclosan, triclocarban	biosolid matrix	C_{18}	ammonium acetate:MeOH	ESI	(Ashfaq et al., 2018)
78 compounds	aquatic matrix	C_{18}	0.1% formic acid: MeOH	ESI	(M. Hu et al., 2018)
32 compounds	river	C_{18}	0.01% formic acid: MeOH	ESI	(Xie et al., 2021)
ibuprofen, sulfamethoxazole, estrone, caffeine, carbamazepine,	groundwater	C_{18}	0.1% acetic acid:ACN	ESI	(Edwards et al., 2019)

trimethoprim					
acetaminophen, sulfamethoxazole, diclofenac, atenolol, metoprolol, diethyltoluamide, oxybenzone	STP influent and effluent	C_{18}	0.01% formic acid: MeOH	APCI	(Tan et al., 2015)
33 androgens, 14 estrogens, 12 progestins, 11 corticosteroids	aquatic matrix	C_{18}	0.1% formic acid: MeOH	APCI	(Huysman et al., 2017)
androsterone, cortisol, cortisone, epitestosterone, norethisterone	wastewater, surface water, drinking water	C_{18}	ACN	APCI	(Leusch et al., 2018)

2.3.3 Detection and analysis of MPs

Sampling and extraction

Despite an increasing number of studies have been reported presence of microplastics in freshwater environments, and marine geographic locations (Pivokonsky et al., 2018). The sampling techniques for detecting microplastics are limited due to the complex solid matrices. Yuan et al (2004) tested saturated sodium chloride (NaCl) solution as flotation agent, and 30% (v/v) H_2O_2 solution to obtain an extraction efficiency of 67 ~ 98% in Poyang Lake China.

In fact, comparing the detection methods in aquatic environments, solid system extraction has high similarities. The method requires: 1) the ability to remove background

contaminations; 2) have method that enable to detect low size range particle; 3) keep the assurance and precision of analysis; 4) ensure high recovery, efficiency and sufficient reproducibility. The steps of microplastics detection include homogenization, flotation, purification, sieving and filtration, and spectroscopy identification. Samples should be suitably preserved prior to analysis. Sludge samples from wastewater treatment plants are usually freeze-dried at -20 °C and ground to fine particles using a mortar and pestle or homogenized using a blender. In initial flotation of microplastics, density-based separation methods are usually considered for isolating low-density particles from higher-density sludge samples. Most common microplastics have densities in the range of 0.8-1.4 g/cm^3 (Yuan et al., 2019). For example, the density of PE and PP are usually less than 1.0 g/cm^3, while the density of PVC is more than 1.0 g/cm^3 up to 1.4 g/cm^3. Therefore, most microplastics in sample can be separated with a solution density greater than 1.0 g/cm^3. Frequently used salt solutions are saturated NaCl, $ZnCl_2$, NaI and $CaCl_2$, etc. The density of these salt solutions applied to isolate microplastics from environmental matrices are shown in Table 2.4.

Table 2.4: Summary of density of flotation solution

Solution	Density(g/cm^3)	Sample type	Typical type MPs	Reference
NaCl	1.2	surface waters and sediments	PP, PE	(Yuan et al., 2019)
NaCl	1.2	sediments	PP, PE, PS, PA	(Fries et al., 2013)
NaCl	1.2	ocean	PP, PE, nylon	(Pan et al., 2019)
$ZnCl_2$	1.7	river	PP, PS, fibers	(Horton et al., 2017)
$ZnCl_2$	1.5	sediments	fibers	(Liebezeit et al., 2012)
NaI	1.6	ocean		(Van Cauwenberghe et al., 2013)
NaI	1.6	sediments	PVC, fibers	(Claessens et al., 2013)
$CaCl_2$	1.3-1.35	seawater and sediments	fibers	(Stolte et al., 2015)

These salt solutions are suitable for flotation of microplastics, and the combination of two-flotation solution was more efficient than the single extraction, which was suitable for analysis of mud and water samples. The content of microplastics in tidal flat of coastal zone was tested using a combination of NaCl and $ZnCl_2$. Initial flotation was carried out with saturated NaCl solution, followed by secondary flotation with saturated $ZnCl_2$ solution. After the two processes, recovery rate was reached up to 97%. In addition, Claessens et al., (2013) investigated saturated NaCl and NaI solution to collect microplastics in sediments sample with a high recovery range from 94% to 98%. Salt solution combined with aeration could effectively improve recovery of microplastics containing polyethylene (PE) and polypropylene (PP) as the two main polymer types. Of all the salts used, NaCl is superior due to cost, extraction efficiency, and non-toxicity. However, high-density microplastics particles need to be extracted with high-density salt solutions (Li et al., 2019).

Giving that $ZnCl_2$ and NaI can be expensive and more hazardous, other emerging extraction techniques are also increasingly used to separate microplastics from sediments. For example, a density-independent method of extracting microplastics was established by pressurized fluid extraction technology (Fuller et al., 2016). The method could effectively identify microplastics particles less than 30 μm, and it could be applied to identify some common microplastics in soil and municipal waste. It was a promising alternative for determining concentration of microplastics in environmental samples.

The step of purification is to remove interfering impurities. The removal of organic matter is a key step for chemical identification of microplastics (Sun et al., 2019). Acidic, oxidative, alkaline, or enzymatic digestion methods are applied for purification. Commonly used reagents that can remove organic matter and their effects on microplastics are shown in Table 2.5.

Table 2.5: Effects of various reagents organic matter removal on MPs

Sample type	The reagent	Microplastic type	Reference
Soil and sludge	30% H_2O_2 solution, 70 °C	PE, PP, PA, PS, ABS and PET	(Li et al., 2019)
Lake water	30 % H_2O_2 solution, 25°C, overnight processing	PE, PP	(Yuan et al., 2019)
Wastewater	Fenton reagent (6.67 mg/ml), 10 min	PE, PP, PVC and nylon	(Tagg et al., 2017)
Sea water	Proteolytic enzyme-k, 50 °C	PS, PE, PVC, fiber	(Cole et al., 2014)
Seafood	10% KOH 60 °C for 24 h	PET	(Dehaut et al., 2016)
Fish tissues	HNO_3 12 mg/L, 70 °C, 2 h	microbeads, PP, fibers	(Munno, 2017)

H_2O_2 solution with volume fraction of 30% and Fenton reagent (a mixture of H_2O_2 and Fe^{2+}) are widely used in pretreatment of microplastics in seawater, fresh water and sediments (Hurley et al., 2018). Literature has reported most of the microplastics had no obvious damage, fracture, and degradation, as well as extraction efficiency improved significantly and the characteristic peaks in the spectrum were clear after oxidizing agents' treatment. Cole et al. (2014) have found proteolytic enzyme-k could separate microplastics quickly and removed 97% organic matter in biota-rich seawater samples and marine organisms. Furthermore, alkaline methods can produce a high recovery about 99% and have no significant effect on surface morphology, however, surface of some nylon particles was changed during treatment (Dehaut et al., 2016). In addition, acid method may also cause underestimations of microplastics in detection due to melting, yellowing or total destruction (Naidoo et al., 2017).

Generally, stainless steel sieves or glass fiber filters are used to extract microplastics from digestion solution. Sieve size is determined by size distribution of microplastics. Zhu et al.

(2018) used a stainless-steel sieve and separated microplastics into six size classes (<0.5 mm, 0.5–1 mm, 1–2 mm, 2–3 mm, 3–4 mm, 4–5 mm). Glass fiber filters are also commonly used for sample separation. For example, Zhang et al. (2018) used 0.7 μm pore size glass fiber filter to separate microbeads in the treated surface waters of the Bohai Sea. However, for accurate identification and quantification of microplastics further studies are required.

Identification of MPs

The identification of microplastics can be carried out by direct examination with naked eye or with aid of a microscope (Figure 2.6). Large and completely clean plastics can be sorted out directly, while smaller-sized ones need observation under stereoscopic microscope and scanning electron microscope (SEM). For microplastics particles with size of 1-5 mm or so, color, shape, and size can be directly judged by microscope, however, it is prone to human errors and misjudgment (Hidalgo-Ruz et al., 2012). Some particles or other substances similar to microplastics require further observation by SEM. By observing surface structure of microplastics in samples with SEM, possibility of visual misjudgment can be reduced and microplastics can be further separated accurately (Mahon et al., 2017).

Fourier transform infrared spectroscopy (FTIR) and its optimized technologies, such as micro FTIR, attenuated total reflectance (ATR) FTIR, and focal plane array detector based micro FTIR imaging, are used to identify chemical composition of microplastics. Chemical bonds and characteristic signals in samples can be identified by FTIR through the reference spectral library (Lo et al., 2018). Harrison et al. (2012) compared the performance of micro FTIR and ATR FTIR in analysis of PE, they have found both FTIR were successful in identifying polymer compositions (Harrison et al., 2012), while (ATR) FTIR was better in obtaining spectra of microplastics with irregular shapes it has limitation on particle size (larger than 500 μm) (Löder et al., 2015).

Raman spectroscopy is another commonly used analytical technique in microplastics chemical characteristics (Lenz et al., 2015). It can be used to analyze samples quite easily, quickly and without damage. Raman spectrum has greater spatial resolution, the thickness of sample has no influence on identification, and it is not disturbed by atmospheric water and CO_2 (Lares et al., 2019). However, detection results are susceptible to fluorescence

interference (Elert et al., 2017). Raman spectrum has advantages on examination ability of small particles down to 1 μm and have better performance on non-polar plastic functional groups than other detection methods (Lenz et al., 2015).
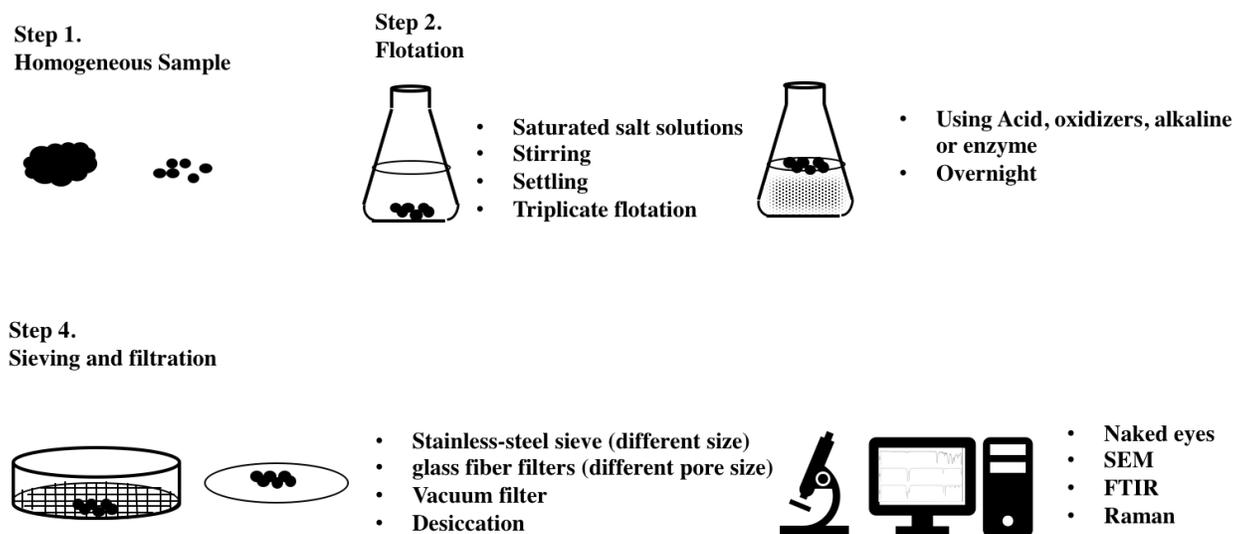
Figure 2.6: A schematic diagram of the detection procedure for MPs in the sewage sludge

2.4 Conventional and emerging processes for the treatment of micropollutants and microplastics

In WWTPs, primary treatment removes suspended solids by gravity, which is not the effective way to remove most micropollutants. The removal of micropollutants mainly occurs in biological treatment process and tertiary treatment. However, many micropollutants showed significantly varied removal behavior in different WWTPs due to their physical and chemical characteristics, which linked to specific treatment process.

2.4.1 Pretreatment and anaerobic digestion

Anaerobic digestion has been widely used in processing municipal wastewater sludge, agricultural wastes, industrial wastes, and municipal solid wastes. In general, the total solid concentration of anaerobic digestion is between 1.5%-5% in municipal wastewater

treatment plant. Based on the total solid content of anaerobic digestion substrate, anaerobic digestion can be divided into two categories: liquid and solid anaerobic digestion. When the total solid concentration is greater than 15%, it is solid anaerobic digestion (Shi et al., 2013). According to the temperature, anaerobic digestion can be divided into three types: low temperature anaerobic digestion (<25 ℃), moderate temperature anaerobic digestion (35 ℃), and high temperature anaerobic digestion (50 ℃). In order to improve anaerobic digestion rate, typically the sludge is pretreated to improve hydrolysis of complex organics and solubilize them facilitating the uptake of organics by the microorganism, subsequently shortening the time of anaerobic digestion, and increasing the biogas (methane) production.

At present, there are three main methods of sludge pretreatment: physical, chemical, and biological. Physical methods include hot water solution, mechanical pretreatment, ultra-sonication, microwaves, high pressure, and freeze-thaw methods. Chemical methods include acid and alkali treatment, oxidation, etc., and biological methods involve microorganism that can secrete extracellular enzymes to sludge, or directly adding lysozyme for hydrolysis of complex organics.

Vlyssides et al. (2004) studied the effects of thermal-alkali synergy on the anaerobic digestion of excess sludge. The temperature range of the experiment was between 50 ℃ - 90 ℃, and at pH 8 - 11. The correlations between the hydrolysis rate coefficient, pH, and temperature fitted kinetics polynomial model. After pretreatment of excess sludge at pH 11 and 90 ℃ for 10 hours, about 45% reduction in VSS occurred. The concentration of soluble COD was 70 mg/L, which was 3 times higher than that obtained at pH 11.0 and 50 ℃ for 10 hours. Methane production efficiency increased by 400% than that obtained at pH 8.0 and 50 ℃.

Kim et al. studied the effects of thermal-alkali synergy on anaerobic digestion. The ranges of NaOH concentration and temperature were 0-200 mg/L and 60-90 °C, respectively. The maximum solid disintegration (SD), which was determined by change in soluble COD was 77.8%, occurring at 0.16 mg/L NaOH and 90 ℃. The optimal methane production rate was 195.1 mL (73.9% increase over the control), occurring at 0.1 mg/L NaOH and 73.7 °C (J. Kim et al., 2013). The results presented that the addition of NaOH significantly affected

the community structure of anaerobic methane bacteria, but temperature had no effect on the community structure of methanogen bacteria.

Micropollutants are partially or totally removed during thermal hydrolysis pretreatment. 16 micropollutants were pretreated at 170 ℃ for 20 min prior to anaerobic digestion. Half of these compound showed high removal efficiency over 85%, while the rest of micropollutants had a removal percentage range from 30% to 60% (Taboada-Santos et al., 2019). However, the removal efficiency of micropollutants are significantly varies from compound. Fluoroquinolones (FQs) group show no obvious removal during pretreatment at 130 ℃ for 20 min due to their thermal stability (Zhang et al., 2018).

2.4.2   Adsorption on Activated Sludge

Activated sludge is a kind of flocs with porous structure and extracellular polymer, which has a large specific surface area. It has good surface adsorption capacity for organic matters and the suspended and colloidal organic matters in the sewage are easily condensed and adsorbed by activated sludge to be removed.

According to the physicochemical properties, micropollutants can be classified into three types: lipophilic, neutral, and acidic. A large part of micropollutants entering the wastewater treatment plant is adsorbed on the sludge. Xia et al., (2005) studies showed that the total mass concentrations of galoxolide and tonalid in the effluent of bioreactor were very high, 30 μg/L and 8.6 μg/L respectively, but the mass concentration in the water phase decreased to 2 μg/L and 0.5 μg/L respectively after filtration. This indicates that these substances were adsorbed to solid particles in large quantities. There are two main types of adsorptions of sludge: lipophilic adsorption and electrostatic attraction. Lipophilic adsorption refers to the process by which compounds containing aliphatic and aromatic groups enter into the lipophilic cell membrane of microorganisms and the lipids part of sludge, respectively. This process is mainly related to the lipophilicity of substances. For example, both galoxolide and tonalid musk belong to lipophilic organic compounds, and they are strongly hydrophobically adsorbed with a large amount of lipids in primary sludge.

Electrostatic attraction occurs between a compound with a positive group and a negatively charged cell surface. This process is mainly related to the ionic form of the compound in

aqueous solution. For example, the polarity of propylnorfloxacin is very strong, in neutral conditions will have positive charge, so it is easy to have electrostatic adsorption with negatively charged microorganism surface.

The transformation and microbial degradation are mainly through the following actions: 1) co-metabolism, microbial decomposition, or partial transformation but not as a carbon source; 2) mixed substrate growth, microbes use as a carbon source and energy, it can be completely mineralized.

The sludge age to a great extent affects the biodegradation efficiency. The removal rate generally increases with residence time of wastewater and the increase of sludge age. As the sludge age increases, the bacterial community may become more diversified, and the slow-growing microbes reach the appropriate number and degrade. Microbial metabolic activities become diversified to accommodate lower concentrations of sludge loading. In addition, different biological treatment processes will also have a certain impact on the biodegradation. For example, the traditional activated sludge process is limited by mass transfer, there are few compounds entering into the flocs, and most of the compounds are only converted in the external layer of flocs, so the reaction rate will be relatively low.

The degradation of these compounds by biological wastewater treatment mainly depends on the operating conditions. Compared with the conventional activated sludge process which only removes BOD, the removal of antibiotics by the A^2O (anaerobic/anoxic/oxic) process and the A/O (anaerobic/oxic) process is better.

The aerobic granular sludge technology has the advantages of using activated sludge (without carrier) to form a biofilm by self-immobilization and is mainly characterized in 1) the biomass concentration is high without settlement problem; 2) maintaining a higher biomass by fixing a low growth rate microorganism in the particle structure; 3) conducive to the growth of different functional organisms in small units, thereby enhancing the microbial structure. In addition, that bacteria in the biological membrane is more resistant to antibiotics than the free-floating bacteria due to the generation of extracellular polymeric material.

For example, comparing the removal efficiency of sulfamethoxazole from wastewater by aerobic granular sludge and conventional activated sludge; under the same conditions, the anoxic/anaerobic/aerobic SBRs was inoculated with suspended and particulate biomass for 90 days. When 2 μg/L SMX was added, the particle removal rate was 84%, which was significantly higher than that in suspended sludge removal rate (73%). The addition of SMX has little effect on the treatment efficiency of the reactor. The results showed that aerobic biodegradation was an effective removal pathway, but adsorption and anoxic/anaerobic treatment were not obvious. The biodegradation rate constant of the particles is obviously larger than the suspended biological mass (Xia et al., 2005).

2.4.3   Advanced oxidation process

Advanced oxidation processes (AOPs) are aimed at promoting the production of highly active hydroxyl radicals (·OH). Recent studies have shown that some advanced oxidation techniques have high degradation efficiency for PPCPs in (Shemer et al., 2006) .

The concept of AOPs was first proposed and applied to drinking water treatment by Glaze et al. in the 1980s. Since then, AOPs have been widely studied in wastewater treatment (Deng & Zhao, 2015). The redox potential of ·OH is 2.8 V (Table 2.6). The oxidation ability of ·OH is second only to the fluorine (3.03V). The ·OH oxidation rate constant of most organic compounds is $10^8 \sim 10^{10}$ $M^{-1} \cdot s^{-1}$ (Deng et al., 2015). The reaction of ·OH with organic compounds mainly goes through four steps: radical addition, dehydrogenation, electron transfer and free radical combination.

Ultraviolet radiation (UV) can be used as a source of external energy in photochemical advanced oxidation process, which is an important factor of ·OH production. In recent years, chemical oxidation and photochemical advanced oxidation processes have been widely studied for the treatment of PPCPs, including ultraviolet (UV), ultraviolet/hydrogen peroxide (UV/ H_2O_2), ozone/hydrogen peroxide(O_3/H_2O_2), ultraviolet/ozone gas (UV/O_3), Fenton oxidation, protophoton oxidation, UV/persulfate (UV/$S_2O_8^{2-}$), etc. (Esplugas et al., 2007; Giri et al., 2010; Zhang et al., 2009).

Table 2.6: Redox potential of some oxidizing agents

Oxidizing agent	F	HO-	O_3	$S_2O_8^{2-}$	H_2O_2	SO_4^{2-}	ClO_2	HClO	Br	I
E_0/V, 25°C	3.03	2.80	2.07	2.01	1.78	2.5~3.1	1.57	1.49	1.09	0.54

Direct UV:

Ultraviolet radiation is one of the essential factors in advanced photochemical oxidation process. The wavelength range of ultraviolet light is in the electromagnetic spectrum is 100 nm~400 nm. According to the different wavelength range, it can be divided into four regions: UVA (315～400 nm), UVB (280～315 nm), UVC (190～280 nm), VUV (<190 nm). In general, the application of ultraviolet light increases the reaction rate of the advanced oxidation process compared with the advanced oxidation process without light radiation.

The results show that the single ultraviolet radiation can also effectively remove some PPCPs (Table 2.7). At 254 nm wavelengths, ultraviolet photons are equivalent to 4.89 eV energy. After UV radiation, organic pollutants are excited by photons and decomposed by homolysis, heterolysis, and photochemical ionization. Under the condition of abundant oxygen, the superoxide radicals ($O_2 \cdot^-$) can be further formed. Although the oxidation ability of the $O_2 \cdot^-$(0.75eV) is not high but they can degrade the aromatic compounds. Because of the variety of PPCPs and the difference of physical and chemical properties, the degradation rate of PPCPs by single ultraviolet light is different.

Table 2.7: Research of PPCPs degradation by UV

System	Subject	Condition	Efficiency	Ref.
Distilled water	Clofibric acid(CA)	$\varrho0(CA)=1mg/L$, $T=(25\pm2)$ °C, $10W(\lambda=254nm)$	60min 100%, $k=1.5\times10^{-3}(s^{-1})$	(Giri et al., 2010)
Distilled water	Clarithromycin (CAM)	$\varrho0(CAM)=1mg/L$, $T=(25\pm2)$ °C, $10W(\lambda=254nm)$	30min 40%, $k=3.9\times10^{-4}(s^{-1})$	(Giri et al., 2010)
Distilled water	Diclofenac(DCF)	$\varrho0(DCF)=1mg/L$, $T=(25\pm2)$ °C, $10W(\lambda=254nm)$	20min 100%, $k=4.7\times10^{-3}(s^{-1})$	(Giri et al., 2010)
Milli-Q water	Clenbuterol	$\varrho0=5\mu g/L$, $T=20$°C, $8W(\lambda=185nm)$, $10W(\lambda=254nm)$	8W,10min 30%, $k=7.1\times10^{-4}(s^{-1})$; 10W,10min 80%, $k=3\times10^{-3}(s^{-1})$;	(I. Kim & Tanaka, 2009)
Milli-Q water	Oxytetracycline	$\varrho0=5\mu g/L$, $T=20$°C, $8W(\lambda=185nm)$, $10W(\lambda=254nm)$	8W,10min 65%, $k=1.6\times10^{-3}(s^{-1})$; 10W,10min 70%, $k=2.5\times10^{-3}(s^{-1})$;	(I. Kim & Tanaka, 2009)

UV/ H_2O_2 Photochemical oxidation:

Compared with single UV radiation, PPCPs can be degraded rapidly and efficiently by UV/ H_2O_2. Mainly because H_2O_2 can produce strong oxidant ·OH under UV irradiation ($\lambda <$ 280nm) (equation 1).

$$H_2O_2 + h\nu \rightarrow 2 \cdot OH \quad (1)$$

In the UV/ H_2O_2 system, there is not only the oxidation of ·OH to the target pollutant, but also the direct photolysis reaction of ultraviolet radiation and the oxidation of the target pollutant by H_2O_2, but mainly the effect of ·OH (Deng et al., 2015).

The oxidation ability of UV/ H_2O_2 process is very strong, which can degrade most of PPCPs quickly and effectively, while the reaction conditions are mild, and widely used. However, the utilization ratio of ultraviolet light source is low, the energy consumption is

relatively large, and the investment cost for large-scale wastewater treatment is high. The UV/ H_2O_2 degradation is shown in Table 2.8.

Table 2.8: Research of PPCPs degradation by UV/ H_2O_2

System	Subject	Condition	Efficiency	Ref
Distilled water	Naproxen (NPX)	$\varrho_0(NPX)=1mg/L, \varrho_0(H_2O_2)=11.03mmol/L$, $T=(25\pm2)$ °C, $10W(\lambda=254nm)$	30min 90%, $k=1.25\times10^{-3}(s^{-1})$	(Giri et al., 2010)
Milli-Q water	Tetracycline (TC)	$\varrho_0(TC)=20mg/L, \varrho_0(H_2O_2) = 0.02mmol/L$ $T=25, pH=2, 15W(\lambda=254nm)$	120min 100%, $k=4.8\times10^{-4}(s^{-1})$	(López-Peñalver et al., 2010)
Milli-Q water	Chlortetracycline (CTC)	$\varrho_0(CTC)=20mg/L, \varrho_0(H_2O_2) =$ $0.02mmol/L T=25, pH=2, 15W(\lambda=254nm)$	120min 100%, $k=8.3\times10^{-4}(s^{-1})$	(López-Peñalver et al., 2010)
Pure water	Carbamazepine (CBZ)	$\varrho_0(CBZ)=1mg/L, \varrho_0(H_2O_2) =20mmol/L$, $T=(25\pm2)$ °C, $300W(\lambda=254nm)$, $7.51\times10^4-8.46\times10^4Lux$	90min 97.1%, $k=1.42\times10^{-3}(s^{-1})$;	(N. Liu et al., 2015)
Pure water	Ciprofloxacin (CIP)	$\varrho_0(CBZ)=1mg/L, \varrho_0(H_2O_2) =20mmol/L$, $T=(25\pm2)$ °C, $300W(\lambda=254nm)$, $7.51\times10^4-8.46\times10^4Lux$	60min 100%, $k=1.11\times10^{-3}(s^{-1})$;	(H.-G. Guo et al., 2013)

O₃ oxidation:

The O_3 is a strong oxidizing agent, and the oxidation potential of ozone is 2.07 eV. Ozone can react slowly and selectively with organic compounds directly, and the rate constant of the reaction is generally 1~100 $M^{-1}\cdot s^{-1}$. When O_3 reaches saturation state in aqueous medium, especially when aqueous solution is alkaline (optimum pH is 9), there will be a rapid and non-selective oxidation reaction of producing ·OH. (equation 2) (Qi, 2010)

$$3O_3 + H_2O \rightarrow 2 \cdot OH + 4O_2 \qquad (2)$$

In general, the method of O_3 oxidation of PPCPs has the advantages of simple operation, mild reaction conditions, strong oxidation ability, fast reaction rate and no secondary pollution. However, the ozone generating equipment is complex, the utilization ratio of ozone is low, high cost, and the selectivity of oxidation reaction is high, and the PPCPs that can be effectively degraded are limited. The O_3 /H_2O_2 and UV/ O_3 process can not only increase the utilization rate of O_3, but also increase the oxidation rate of PPCPs and the effect of treatment. However, the energy consumption of ultraviolet lamp in UV/ O_3 process has to be considered.

Fenton method:

In the late 19th century, Fenton found that the mixed solution of Fe^{2+} and H_2O_2 could oxidize many organic compounds such as carboxylic acids, alcohols, and lipids. Although the Fenton reagent was found earlier, it was actually used to deal with toxic organic matter in 1960s. The essence of Fenton reaction is that Fe^{2+} catalyzes H_2O_2 to produce $\cdot$OH, which can be removed by other Fenton reagents or form $\cdot$OH$_2$ with weak oxidation ability, which results in the reduction of oxidation ability of Fenton reaction to pollutants.

In order to avoid this situation, the optimum molar ratio of Fe^{2+} and H_2O_2 is usually determined. In the Fenton reaction, the oxidation efficiency is also related to the pH of the solution, and the catalytic activity of Fe^{2+} is the highest at pH 2~4. The results show that the degradation efficiency of metronidazole by Fenton method is much higher than that of UV/ H_2O_2, while the dosage of H_2O_2 is only 1/25 UV/ H_2O_2 system (Shemer et al., 2006). The degradation efficiency of Fenton to adsorbable organic halides and COD was better than that of UV/ H_2O_2 and O_3/ H_2O_2. Therefore, it is generally used for pretreatment of municipal wastewater and drugs wastewater with more complex PPCPs composition.

To further improve the formation rate of $\cdot$OH and the reaction conditions, ultraviolet light was introduced as the optical Fenton reaction. The principle of the reaction is the same as that of the Fenton reaction, and the main oxidant is $\cdot$OH. The rate of $\cdot$OH production by optical Fenton method is faster than that by Fenton method, and the removal rate of refractory PPCPs is improved.

UV/$S_2O_8^{2-}$ method:

Persulfate $S_2O_8^{2-}$ is a strong oxidizing substance with a redox potential of 2.01 V, $S_2O_8^{2-}$ is activated to produce stronger oxidizing radical $\cdot SO^{4-}$ (E_0=2.5~3.1 eV) (Lou et al., 2016). Therefore, $\cdot SO^{4-}$ can oxidize most organic compounds. The reaction mechanism of persulfate to produce radical $\cdot SO^{4-}$ under UV radiation can be found in equation 3.

$$S_2O_8^{2-} + hv \rightarrow 2 \cdot SO^{4-} \qquad (3)$$

Except for activation of $S_2O_8^{2-}$ under UV irradiation to produce $\cdot SO^{4-}$, other methods such as the use of transition metal ions Fe^{2+}, Fe^{3+} can also catalyze persulfate to produce $\cdot SO^{4-}$. This method is simpler and more energy efficient than other methods and is considered to be the best method for $\cdot SO^{4-}$ production.

Compared with O_3 and H_2O_2, persulfate is easy to preserve, and no volatile gas is produced when reacting with organic pollutants, which indicated yield of $\cdot SO^{4-}$ is not affected during reacting. Secondly, $\cdot SO^{4-}$ can adapt to a wide range of pH, increasing the treatment range of wastewater.

At present, advanced oxidation processes are widely researched with many publications. However, the research on PPCPs by advanced oxidation process is mostly at the stage of laboratory, and the technical and cost problems still need to be solved for their application to large-scale water treatments. Secondly, the AOPs may produce more harmful substances than the parent pollutants while removing PPCPs, so it is necessary to further study the intermediate and final products of various kinds of PPCPs and the mechanism of its degradation and the changes of its toxicity. In the future, the research on PPCPs by advanced oxidation process should focus on the intermediate products of PPCPs degradation and its toxicity test, optimize the design of reactor, and develop highly efficient catalyst that can be recycled to reduce the cost. In addition, the degradation efficiency of advanced oxidation process for different types and properties of PPCPs is different.

2.4.4 Potential treatment process for microplastics

Generally speaking, WWTPs is not specially designed for removal microplastics. Thus, some treatment technologies were investigated to improve the quality of the final effluent by removing MPs, especially in tertiary phase. Initial field and laboratory studies have demonstrated that microplastics can be removed via advanced treatment processes. A

summary of various treatment technologies to remove microplastics from WWTPs is listed in Table 2.9.

Table 2.9: Summary of various treatment process/technologies

Treatment processes/technologies	Sample	Removal efficiency	Reference
	activated sludge	16.6%	(X. Liu et al., 2019)
Anaerobic/anoxic/aerobic (A^{2}O)	effluent	98–99% (> 106 μm)	(Lee & Kim, 2018)
	effluent	71.67 ± 11.58%	(L. Yang et al., 2019)
	effluent	47.1%-81.6%	(Hidayaturrahman & Lee, 2019)
Coagulation/flocculation	effluent	95% for 1 μm;	(Rajala et al., 2020)
		76% for 6.3 μm	
Disc-filter	effluent	89.7%	(Simon et al., 2019)
Disinfection	effluent	7.1%	(X. Liu et al., 2019)
Dissolved air flotation	effluent	95%	(Talvitie et al., 2017)
Electrocoagulation	artificial wastewater	> 90%	(Perren et al., 2018)
	effluent	99.9%	(Talvitie et al., 2017)
Membrane bioreactor (MBR)	effluent	98.3%	(Lares et al., 2018)
	effluent	79.01%	(Bayo et al., 2020)
Ozone	effluent	90%	(Hidayaturrahman & Lee, 2019)
Rapid sand filters (RSF)	effluent	> 90%	(Talvitie et al., 2017)
	effluent	75.49%	(Bayo et al., 2020)
Reverse osmosis (RO)	effluent	86%-90%	(Ziajahromi et al., 2017)

The efficiencies of various technologies for the microplastics pollutions of WWTPs have been also listed in Table 2.9. Anaerobic/anoxic/aerobic (A^2/O) process is a common secondary treatment process in WWTPs, which maintains a various microbial community for simultaneous removal of nitrogen and phosphorus also proved have high removal efficiency of MPs up to 99% in effluent. However, lower removal percentage about 16.6% occurred in active sludge. Membrane bioreactor (MBR) also showed high removal efficiency in a full-scale WWTP treating both municipal and industrial wastewater in Spain. Other treatment methods by different technologies such as coagulation, disc-filter, rapid sand filtration, and ozone, are tertiary treatment processes, and focus on the final effluent in WWTPs with a significant removal rate 75.49% to 99.9%. Despite different treatment methods showed positive performance on the removal of MPs, practical applications need to consider flexibility, energy consumption, operation time, maintenance, and cost. In order to obtain field application, more studies need to focus on overcoming the disadvantages of these methods.

2.5   Synopsis of literature

Both micropollutants and microplastics pose harmful effects on human and environment health because of their accumulation and recalcitrance (Geiger et al., 2016). Wastewater treatment plants are potential source and transport pathway of these contaminants in ecosystems. With the development of sophisticated analytical methods, multiple literature reviews have recently been published on the occurrence of micropollutants in WWTPs as well as microplastics (Hernandez et al., 2017; Yang et al., 2017). Based on literature, the concentrations of micropollutants in the WWTPs ranged from <1 ng/L to >1000 μg/L (Guo et al., 2017), and the abundance of microplastics in the WWTPs within the range of <0.01 particles/L – 2.4 ×10^5 particles/kg (Liu et al., 2021).

Conventional treatment facilities in WWTPs are inefficient to remove micropollutants and microplastics completely (Blair et al., 2019; Margot et al., 2015) due to their complex physicochemical properties such as partition coefficient, solubility, thermal stability and biodegradation. Therefore, suitable treatments in WWTPs are required for the removal of anthropogenic contaminants to reduce their risk before being discharged into environment (Lapointe et al., 2020). UV disinfection is one of common advance treatment technologies

to remove pharmaceuticals in secondary effluent (Lian et al., 2015). Also, coagulation/flocculation is widely used for removal of fine particles from a liquid stream, which is an indispensable process in WWTPs and will continue to be applied for carbon diversion and enhanced primary treatment (Shannon et al., 2008). Additionally, during liquid stream treatment, micropollutants and microplastics can absorb on to sludge (Murphy et al., 2016). Anaerobic digestion is considered as a promising and essential treatment process in WWTPs to remove biodegradable pollutants for stabilizing sludge.

2.6 Knowledge gap

Although the traditional WWTPs remove micropollutants and microplastics to some extent (Gündoğdu et al., 2018;Yang et al., 2017) further research is needed to enhance the performance of the commonly applied processes. To minimize the risk of pollutants discharged into natural ecosystem, both advanced and traditional wastewater treatment technologies, including coagulation and UV photolysis for effluent (water) treatment, thermal-alkaline pretreatment for anaerobic digestion are widely applied in WWTPs. However, due to the wide variety of micropollutants and microplastics, continuous research is needed for greater performance and process optimization for better removal of target pollutants. The challenges to understand the role of a certain treatment technology can be summarized as the following:

1) Coagulation process is widely applied in WWTPs as primary treatment approach to remove suspended and colloidal particles. However, information related to microplastics removal and mechanism is lacking, especially microfibers emanating from textile fabrics, requiring further research.

2) Over 90% of microplastics are remained in WWTPs sludge, which are recycled in land applications. Anaerobic digestion is most commonly used stabilization process of sludge. However, the effects of anaerobic digestion on microplastics/microfibers are still unknown.

3) UV disinfection has been considered as one of the most popular and ecofriendly process to remove micropollutants. UV photolysis destructs chemical bonds with different removal efficiency from ranging from 40%-100% (Wang et al., 2016). Therefore, photochemical

parameters of specific micropollutants should be investigated to increase the applicability of UV process. The photodegradation of many compounds is highly pH-dependent due to their acid-base speciation form which absorb UV-radiation differently (Lian et al., 2015). The effect of pH on molar absorptivity of selected micropollutants towards photolysis needs to be identified.

4) Many recent studies stated that high concentration of micropollutants are detected in sludge due to their high partition coefficient (K_d) and octanol-water partition coefficient (K_{ow}). The sludge or biosolids are recovered and reused after stabilization, in which a wide range of micropollutants can exist. Pre- and post-treatment processes such as thermal, chemical and combination of them are used to improve anaerobic digestion for greater stabilization of biosolids. To date, several studies have investigated the thermal-alkaline pretreatment to enhance biogas production and increased volatile suspended solids (VSS) degradation. However, limited data exist on the effect of thermal-alkaline process on micropollutants removal.

2.7 Reference

Albero, B., Tadeo, J. L., & Pérez, R. A. (2019). Ultrasound-assisted extraction of organic contaminants. *TrAC Trends in Analytical Chemistry*, *118*, 739–750.

Alygizakis, N. A., Gago-Ferrero, P., Borova, V. L., Pavlidou, A., Hatzianestis, I., & Thomaidis, N. S. (2016). Occurrence and spatial distribution of 158 pharmaceuticals, drugs of abuse and related metabolites in offshore seawater. *Science of the Total Environment*, *541*, 1097–1105.

Arun, S., Mukhopadhyay, M., & Chakraborty, P. (2017). A review on antibiotics consumption, physico-chemical properties and their sources in Asian soil. In *Antibiotics and Antibiotics Resistance Genes in Soils* (pp. 39–53). Springer.

Ashfaq, M., Li, Y., Wang, Y., Qin, D., Rehman, M. S. U., Rashid, A., Yu, C.-P., & Sun, Q. (2018). Monitoring and mass balance analysis of endocrine disrupting compounds and their transformation products in an anaerobic-anoxic-oxic wastewater treatment system in Xiamen, China. *Chemosphere*, *204*, 170–177. https://doi.org/https://doi.org/10.1016/j.chemosphere.2018.04.028

Ashton, D., Hilton, M., & Thomas, K. V. (2004). Investigating the environmental transport of human pharmaceuticals to streams in the United Kingdom. *Science of the Total Environment*, *333*(1–3), 167–184.

Bayo, J., López-Castellanos, J., & Olmos, S. (2020). Membrane bioreactor and rapid sand filtration for the removal of microplastics in an urban wastewater treatment plant. *Marine Pollution Bulletin*, *156*, 111211. https://doi.org/https://doi.org/10.1016/j.marpolbul.2020.111211

Ben, Y., Fu, C., Hu, M., Liu, L., Wong, M. H., & Zheng, C. (2019). Human health risk assessment of antibiotic resistance associated with antibiotic residues in the environment: A review. *Environmental Research*, *169*, 483–493. https://www.sciencedirect.com/science/article/pii/S0013935118304298

Blair, R. M., Waldron, S., & Gauchotte-Lindsay, C. (2019). Average daily flow of microplastics through a tertiary wastewater treatment plant over a ten-month period. *Water Research*, *163*, 114909.

Boyd, G. R., Palmeri, J. M., Zhang, S., & Grimm, D. A. (2004). Pharmaceuticals and personal care products (PPCPs) and endocrine disrupting chemicals (EDCs) in stormwater canals and Bayou St. John in New Orleans, Louisiana, USA. *Science of the Total Environment*, *333*(1–3), 137–148.

Bunzel, K., Kattwinkel, M., & Liess, M. (2013). Effects of organic pollutants from wastewater treatment plants on aquatic invertebrate communities. *Water Research*, *47*(2), 597–606. https://doi.org/https://doi.org/10.1016/j.watres.2012.10.031

Calza, P., & Fabbri, D. (2014). Advanced Mass Spectrometry-Based Techniques for the Identification and Structure Elucidation of Transformation Products of Emerging Contaminants. *Transformation Products of Emerging Contaminants in the Environment*, 325–350.

Carr, S. A., Liu, J., & Tesoro, A. G. (2016). Transport and fate of microplastic particles in wastewater treatment plants. *Water Research*, *91*, 174–182. https://doi.org/https://doi.org/10.1016/j.watres.2016.01.002

Carvalho, P. N., Basto, M. C. P., Almeida, C. M. R., & Brix, H. (2014). A review of plant–pharmaceutical interactions: from uptake and effects in crop plants to phytoremediation in constructed wetlands. *Environmental Science and Pollution Research*, *21*(20), 11729–11763.

Chen, F., Ying, G.-G., Kong, L.-X., Wang, L., Zhao, J.-L., Zhou, L.-J., & Zhang, L.-J. (2011). Distribution and accumulation of endocrine-disrupting chemicals and pharmaceuticals in wastewater irrigated soils in Hebei, China. *Environmental Pollution*, *159*(6), 1490–1498.

Chubarenko, I., Bagaev, A., Zobkov, M., & Esiukova, E. (2016). On some physical and dynamical properties of microplastic particles in marine environment. *Marine*

Pollution Bulletin, *108*(1–2), 105–112.

Claessens, M., Van Cauwenberghe, L., Vandegehuchte, M. B., & Janssen, C. R. (2013). New techniques for the detection of microplastics in sediments and field collected organisms. *Marine Pollution Bulletin*, *70*(1), 227–233. https://doi.org/https://doi.org/10.1016/j.marpolbul.2013.03.009

Crew, A., Gregory-Eaves, I., & Ricciardi, A. (2020). Distribution, abundance, and diversity of microplastics in the upper St. Lawrence River. *Environmental Pollution*, *260*, 113994. https://doi.org/https://doi.org/10.1016/j.envpol.2020.113994

Cristale, J., & Lacorte, S. (2013). Development and validation of a multiresidue method for the analysis of polybrominated diphenyl ethers, new brominated and organophosphorus flame retardants in sediment, sludge and dust. *Journal of Chromatography A*, *1305*, 267–275.

Dai, G., Wang, B., Huang, J., Dong, R., Deng, S., & Yu, G. (2015). Occurrence and source apportionment of pharmaceuticals and personal care products in the Beiyun River of Beijing, China. *Chemosphere*, *119*, 1033–1039.

Das, S., Ray, N. M., Wan, J., Khan, A., Chakraborty, T., & Ray, M. B. (2017). Micropollutants in wastewater: fate and removal processes. *Physico-Chemical Wastewater Treatment and Resource Recovery*, *3*, 75–117.

Daso, A. P., Fatoki, O. S., Odendaal, J. P., & Olujimi, O. O. (2012). Occurrence of selected polybrominated diphenyl ethers and 2, 2′, 4, 4′, 5, 5′-hexabromobiphenyl (BB-153) in sewage sludge and effluent samples of a wastewater-treatment plant in Cape Town, South Africa. *Archives of Environmental Contamination and Toxicology*, *62*(3), 391–402.

Dehaut, A., Cassone, A.-L., Frère, L., Hermabessiere, L., Himber, C., Rinnert, E., Rivière, G., Lambert, C., Soudant, P., Huvet, A., Duflos, G., & Paul-Pont, I. (2016). Microplastics in seafood: Benchmark protocol for their extraction and characterization. *Environmental Pollution*, *215*, 223–233.

https://doi.org/https://doi.org/10.1016/j.envpol.2016.05.018

Deng, Y., & Zhao, R. (2015). Advanced oxidation processes (AOPs) in wastewater treatment. *Current Pollution Reports*, *1*(3), 167–176.

Edwards, Q. A., Sultana, T., Kulikov, S. M., Garner-O'Neale, L. D., & Metcalfe, C. D. (2019). Micropollutants related to human activity in groundwater resources in Barbados, West Indies. *Science of The Total Environment*, *671*, 76–82. https://doi.org/https://doi.org/10.1016/j.scitotenv.2019.03.314

Egessa, R., Nankabirwa, A., Ocaya, H., & Pabire, W. G. (2020). Microplastic pollution in surface water of Lake Victoria. *Science of The Total Environment*, *741*, 140201. https://doi.org/https://doi.org/10.1016/j.scitotenv.2020.140201

Elert, A. M., Becker, R., Duemichen, E., Eisentraut, P., Falkenhagen, J., Sturm, H., & Braun, U. (2017). Comparison of different methods for MP detection: what can we learn from them, and why asking the right question before measurements matters? *Environmental Pollution*, *231*, 1256–1264.

Enfrin, M., Dumée, L. F., & Lee, J. (2019). Nano/microplastics in water and wastewater treatment processes – Origin, impact and potential solutions. *Water Research*, *161*, 621–638. https://doi.org/https://doi.org/10.1016/j.watres.2019.06.049

EPA, M. (1996). Method 3540C. Soxhlet extraction. *URL: Http://Www. Epa. Gov/Osw/Hazard/Testmethods/Sw846/Pdfs/3540c. Pdf*.

Esplugas, S., Bila, D. M., Krause, L. G. T., & Dezotti, M. (2007). Ozonation and advanced oxidation technologies to remove endocrine disrupting chemicals (EDCs) and pharmaceuticals and personal care products (PPCPs) in water effluents. *Journal of Hazardous Materials*, *149*(3), 631–642.

Fisner, M., Taniguchi, S., Moreira, F., Bícego, M. C., & Turra, A. (2013). Polycyclic aromatic hydrocarbons (PAHs) in plastic pellets: Variability in the concentration and composition at different sediment depths in a sandy beach. *Marine Pollution Bulletin*, *70*(1–2), 219–226.

Fries, E., Dekiff, J. H., Willmeyer, J., Nuelle, M. T., Ebert, M., & Remy, D. (2013). Identification of polymer types and additives in marine microplastic particles using pyrolysis-GC/MS and scanning electron microscopy. *Environmental Sciences: Processes and Impacts*, *15*(10), 1949–1956. https://doi.org/10.1039/c3em00214d

Fuhrimann, S., Klánová, J., Přibylová, P., Kohoutek, J., Dalvie, M. A., Röösli, M., & Degrendele, C. (2020). Qualitative assessment of 27 current-use pesticides in air at 20 sampling sites across Africa. *Chemosphere*, *258*, 127333. https://doi.org/https://doi.org/10.1016/j.chemosphere.2020.127333

Fuller, S., & Gautam, A. (2016). A Procedure for Measuring Microplastics using Pressurized Fluid Extraction. *Environmental Science and Technology*, *50*(11), 5774–5780. https://doi.org/10.1021/acs.est.6b00816

Geiger, E., Hornek-Gausterer, R., & Saçan, M. T. (2016). Single and mixture toxicity of pharmaceuticals and chlorophenols to freshwater algae Chlorella vulgaris. *Ecotoxicology and Environmental Safety*, *129*, 189–198.

Gies, E. A., LeNoble, J. L., Noël, M., Etemadifar, A., Bishay, F., Hall, E. R., & Ross, P. S. (2018). Retention of microplastics in a major secondary wastewater treatment plant in Vancouver, Canada. *Marine Pollution Bulletin*, *133*, 553–561. https://doi.org/https://doi.org/10.1016/j.marpolbul.2018.06.006

Giri, R. R., Ozaki, H., Ota, S., Takanami, R., & Taniguchi, S. (2010). Degradation of common pharmaceuticals and personal care products in mixed solutions by advanced oxidation techniques. *International Journal of Environmental Science & Technology*, *7*(2), 251–260.

Golet, E. M., Xifra, I., Siegrist, H., Alder, A. C., & Giger, W. (2003). Environmental exposure assessment of fluoroquinolone antibacterial agents from sewage to soil. *Environmental Science & Technology*, *37*(15), 3243–3249.

Gouveia, R., Antunes, J., Sobral, P., & Amaral, L. (2018). Microplastics from Wastewater Treatment Plants---Preliminary Data. In M. Cocca, E. Di Pace, M. E.

Errico, G. Gentile, A. Montarsolo, & R. Mossotti (Eds.), *Proceedings of the International Conference on Microplastic Pollution in the Mediterranean Sea* (pp. 53–57). Springer International Publishing.

Gündoğdu, S., Çevik, C., Güzel, E., & Kilercioğlu, S. (2018). Microplastics in municipal wastewater treatment plants in Turkey: a comparison of the influent and secondary effluent concentrations. *Environmental Monitoring and Assessment*, *190*(11). https://doi.org/10.1007/s10661-018-7010-y

Guo, H.-G., Gao, N.-Y., Chu, W.-H., Li, L., Zhang, Y.-J., Gu, J.-S., & Gu, Y.-L. (2013). Photochemical degradation of ciprofloxacin in UV and UV/H 2 O 2 process: kinetics, parameters, and products. *Environmental Science and Pollution Research*, *20*(5), 3202–3213.

Guo, X., Yan, Z., Zhang, Y., Kong, X., Kong, D., Shan, Z., & Wang, N. (2017). Removal mechanisms for extremely high-level fluoroquinolone antibiotics in pharmaceutical wastewater treatment plants. *Environmental Science and Pollution Research*, *24*(9), 8769–8777. https://doi.org/10.1007/s11356-017-8587-3

Hamza, R. A., Iorhemen, O. T., & Tay, J. H. (2016). Occurrence, impacts and removal of emerging substances of concern from wastewater. *Environmental Technology & Innovation*, *5*, 161–175.

Harrison, J. P., Ojeda, J. J., & Romero-González, M. E. (2012). The applicability of reflectance micro-Fourier-transform infrared spectroscopy for the detection of synthetic microplastics in marine sediments. *Science of the Total Environment*, *416*, 455–463.

Hernandez, E., Nowack, B., & Mitrano, D. M. (2017). Polyester Textiles as a Source of Microplastics from Households: A Mechanistic Study to Understand Microfiber Release during Washing. *Environmental Science and Technology*, *51*(12), 7036–7046. https://doi.org/10.1021/acs.est.7b01750

Hidalgo-Ruz, V., Gutow, L., Thompson, R. C., & Thiel, M. (2012). Microplastics in the

marine environment: a review of the methods used for identification and quantification. *Environmental Science & Technology*, *46*(6), 3060–3075.

Hidayaturrahman, H., & Lee, T.-G. (2019). A study on characteristics of microplastic in wastewater of South Korea: Identification, quantification, and fate of microplastics during treatment process. *Marine Pollution Bulletin*, *146*, 696–702. https://doi.org/https://doi.org/10.1016/j.marpolbul.2019.06.071

Hirondart, M., Rombaut, N., Fabiano-Tixier, A. S., Bily, A., & Chemat, F. (2020). Comparison between pressurized liquid extraction and conventional Soxhlet extraction for rosemary antioxidants, yield, composition, and environmental footprint. *Foods*, *9*(5), 584.

Horton, A. A., Svendsen, C., Williams, R. J., Spurgeon, D. J., & Lahive, E. (2017). Large microplastic particles in sediments of tributaries of the River Thames, UK – Abundance, sources and methods for effective quantification. *Marine Pollution Bulletin*, *114*(1), 218–226. https://doi.org/https://doi.org/10.1016/j.marpolbul.2016.09.004

Hu, M., Müller, E., Schymanski, E. L., Ruttkies, C., Schulze, T., Brack, W., & Krauss, M. (2018). Performance of combined fragmentation and retention prediction for the identification of organic micropollutants by LC-HRMS. *Analytical and Bioanalytical Chemistry*, *410*(7), 1931–1941. https://doi.org/10.1007/s00216-018-0857-5

Hurley, R. R., Lusher, A. L., Olsen, M., & Nizzetto, L. (2018). Validation of a Method for Extracting Microplastics from Complex, Organic-Rich, Environmental Matrices. *Environmental Science and Technology*, *52*(13), 7409–7417. https://doi.org/10.1021/acs.est.8b01517

Huysman, S., Van Meulebroek, L., Vanryckeghem, F., Van Langenhove, H., Demeestere, K., & Vanhaecke, L. (2017). Development and validation of an ultra-high performance liquid chromatographic high resolution Q-Orbitrap mass spectrometric method for the simultaneous determination of steroidal endocrine disrupting

compounds in aquatic matrices. *Analytica Chimica Acta*, *984*, 140–150. https://doi.org/https://doi.org/10.1016/j.aca.2017.07.001

Illinois, E. P. A. (2008). *Report on Pharmaceuticals and Personal Care Products in Illinois Drinking Water*. Springfield, Illinois: Illinois Environmental Protection Agency.

Ivešić, M., Krivohlavek, A., Žuntar, I., Tolić, S., Šikić, S., Musić, V., Pavlić, I., Bursik, A., & Galić, N. (2017). Monitoring of selected pharmaceuticals in surface waters of Croatia. *Environmental Science and Pollution Research*, *24*(29), 23389–23400.

Jia, Y.-W., Huang, Z., Hu, L.-X., Liu, S., Li, H.-X., Li, J.-L., Chen, C.-E., Xu, X.-R., Zhao, J.-L., & Ying, G.-G. (2020). Occurrence and mass loads of biocides in plastic debris from the Pearl River system, South China. *Chemosphere*, *246*, 125771. https://doi.org/https://doi.org/10.1016/j.chemosphere.2019.125771

Kathi, S. (2017). An overview of extraction, clean-up and instrumentation techniques for quantification of soil-bound xenobiotic compounds. *Xenobiotics in the Soil Environment*, 101–118.

Kidd, K. A., Blanchfield, P. J., Mills, K. H., Palace, V. P., Evans, R. E., Lazorchak, J. M., & Flick, R. W. (2007). Collapse of a fish population after exposure to a synthetic estrogen. *Proceedings of the National Academy of Sciences*, *104*(21), 8897 LP – 8901. https://doi.org/10.1073/pnas.0609568104

Kim, I., & Tanaka, H. (2009). Photodegradation characteristics of PPCPs in water with UV treatment. *Environment International*, *35*(5), 793–802.

Kim, J., Yu, Y., & Lee, C. (2013). Thermo-alkaline pretreatment of waste activated sludge at low-temperatures: Effects on sludge disintegration, methane production, and methanogen community structure. *Bioresource Technology*, *144*, 194–201. https://doi.org/https://doi.org/10.1016/j.biortech.2013.06.115

Klein, E. Y., Van Boeckel, T. P., Martinez, E. M., Pant, S., Gandra, S., Levin, S. A., Goossens, H., & Laxminarayan, R. (2018). Global increase and geographic

convergence in antibiotic consumption between 2000 and 2015. *Proceedings of the National Academy of Sciences*, *115*(15), E3463–E3470.

Kostich, M. S., Batt, A. L., & Lazorchak, J. M. (2014). Concentrations of prioritized pharmaceuticals in effluents from 50 large wastewater treatment plants in the US and implications for risk estimation. *Environmental Pollution*, *184*, 354–359.

Kuroda, K., Nakada, N., Hanamoto, S., Inaba, M., Katayama, H., Do, A. T., Nga, T. T. V., Oguma, K., Hayashi, T., & Takizawa, S. (2015). Pepper mild mottle virus as an indicator and a tracer of fecal pollution in water environments: comparative evaluation with wastewater-tracer pharmaceuticals in Hanoi, Vietnam. *Science of the Total Environment*, *506*, 287–298.

Lapointe, M., Farner, J. M., Hernandez, L. M., & Tufenkji, N. (2020). Understanding and Improving Microplastic Removal during Water Treatment: Impact of Coagulation and Flocculation. *Environmental Science & Technology*, *54*(14), 8719–8727.

Lares, M., Ncibi, M. C., Sillanpää, M., & Sillanpää, M. (2018). Occurrence, identification and removal of microplastic particles and fibers in conventional activated sludge process and advanced MBR technology. *Water Research*, *133*, 236–246. https://doi.org/https://doi.org/10.1016/j.watres.2018.01.049

Lares, M., Ncibi, M. C., Sillanpää, M., & Sillanpää, M. (2019). Intercomparison study on commonly used methods to determine microplastics in wastewater and sludge samples. *Environmental Science and Pollution Research*, *26*(12), 12109–12122.

Lee, H., & Kim, Y. (2018). Treatment characteristics of microplastics at biological sewage treatment facilities in Korea. *Marine Pollution Bulletin*, *137*, 1–8. https://doi.org/https://doi.org/10.1016/j.marpolbul.2018.09.050

Lenz, R., Enders, K., Stedmon, C. A., Mackenzie, D. M. A., & Nielsen, T. G. (2015). A critical assessment of visual identification of marine microplastic using Raman spectroscopy for analysis improvement. *Marine Pollution Bulletin*, *100*(1), 82–91.

Leusch, F. D. L., Neale, P. A., Arnal, C., Aneck-Hahn, N. H., Balaguer, P., Bruchet, A.,

Escher, B. I., Esperanza, M., Grimaldi, M., Leroy, G., Scheurer, M., Schlichting, R., Schriks, M., & Hebert, A. (2018). Analysis of endocrine activity in drinking water, surface water and treated wastewater from six countries. *Water Research*, *139*, 10–18. https://doi.org/https://doi.org/10.1016/j.watres.2018.03.056

Lewis, K. A., Tzilivakis, J., Warner, D. J., & Green, A. (2016). An international database for pesticide risk assessments and management. *Human and Ecological Risk Assessment: An International Journal*, *22*(4), 1050–1064.

Li, L., Geng, S., Li, Z., & Song, K. (2020). Effect of microplastic on anaerobic digestion of wasted activated sludge. *Chemosphere*, *247*, 125874. https://doi.org/https://doi.org/10.1016/j.chemosphere.2020.125874

Li, L., Liu, D., Song, K., & Zhou, Y. (2020). Performance evaluation of MBR in treating microplastics polyvinylchloride contaminated polluted surface water. *Marine Pollution Bulletin*, *150*, 110724. https://doi.org/https://doi.org/10.1016/j.marpolbul.2019.110724

Li, Q., Wu, J., Zhao, X., Gu, X., & Ji, R. (2019). Separation and identification of microplastics from soil and sewage sludge. *Environmental Pollution*, *254*, 113076. https://doi.org/https://doi.org/10.1016/j.envpol.2019.113076

Li, X., Chen, L., Mei, Q., Dong, B., Dai, X., Ding, G., & Zeng, E. Y. (2018). Microplastics in sewage sludge from the wastewater treatment plants in China. *Water Research*, *142*, 75–85. https://doi.org/https://doi.org/10.1016/j.watres.2018.05.034

Lian, J., Qiang, Z., Li, M., Bolton, J. R., & Qu, J. (2015). UV photolysis kinetics of sulfonamides in aqueous solution based on optimized fluence quantification. *Water Research*, *75*, 43–50.

Liebezeit, G., & Dubaish, F. (2012). Microplastics in Beaches of the East Frisian Islands Spiekeroog and Kachelotplate. *Bulletin of Environmental Contamination and Toxicology*, *89*(1), 213–217. https://doi.org/10.1007/s00128-012-0642-7

Liu, N., Zheng, M., Sijak, S., Tang, L., Xu, G., & Wu, M. (2015). Aquatic photolysis of carbamazepine by UV/H 2 O 2 and UV/Fe (II) processes. *Research on Chemical Intermediates*, *41*(10), 7015–7028.

Liu, W., Zhang, J., Liu, H., Guo, X., Zhang, X., Yao, X., Cao, Z., & Zhang, T. (2021). A review of the removal of microplastics in global wastewater treatment plants: Characteristics and mechanisms. *Environment International*, *146*, 106277.

Liu, X., Yuan, W., Di, M., Li, Z., & Wang, J. (2019). Transfer and fate of microplastics during the conventional activated sludge process in one wastewater treatment plant of China. *Chemical Engineering Journal*, *362*, 176–182. https://doi.org/https://doi.org/10.1016/j.cej.2019.01.033

Llompart, M., Celeiro, M., & Dagnac, T. (2019). Microwave-assisted extraction of pharmaceuticals, personal care products and industrial contaminants in the environment. *TrAC Trends in Analytical Chemistry*, *116*, 136–150.

Lo, H.-S., Xu, X., Wong, C.-Y., & Cheung, S.-G. (2018). Comparisons of microplastic pollution between mudflats and sandy beaches in Hong Kong. *Environmental Pollution*, *236*, 208–217.

Löder, M. G. J., & Gerdts, G. (2015). Methodology used for the detection and identification of microplastics—A critical appraisal. In *Marine anthropogenic litter* (pp. 201–227). Springer.

Long, Z., Pan, Z., Wang, W., Ren, J., Yu, X., Lin, L., Lin, H., Chen, H., & Jin, X. (2019). Microplastic abundance, characteristics, and removal in wastewater treatment plants in a coastal city of China. *Water Research*, *155*, 255–265. https://doi.org/https://doi.org/10.1016/j.watres.2019.02.028

López-Peñalver, J. J., Sánchez-Polo, M., Gómez-Pacheco, C. V, & Rivera-Utrilla, J. (2010). Photodegradation of tetracyclines in aqueous solution by using UV and UV/H2O2 oxidation processes. *Journal of Chemical Technology & Biotechnology*, *85*(10), 1325–1333.

Lou, X., Xiao, D., Fang, C., Wang, Z., Liu, J., Guo, Y., & Lu, S. (2016). Comparison of UV/hydrogen peroxide and UV/peroxydisulfate processes for the degradation of humic acid in the presence of halide ions. *Environmental Science and Pollution Research*, *23*(5), 4778–4785.

Magni, S., Binelli, A., Pittura, L., Avio, C. G., Della Torre, C., Parenti, C. C., Gorbi, S., & Regoli, F. (2019). The fate of microplastics in an Italian Wastewater Treatment Plant. *Science of The Total Environment*, *652*, 602–610. https://doi.org/https://doi.org/10.1016/j.scitotenv.2018.10.269

Magnusson, K., & Norén, F. (2014). Screening of microplastic particles in and down-stream a wastewater treatment plant. *IVL Swedish Environmental Research Institute*, *C 55*(C), 22. https://doi.org/naturvardsverket-2226

Mahon, A. M., O'Connell, B., Healy, M. G., O'Connor, I., Officer, R., Nash, R., & Morrison, L. (2017). Microplastics in sewage sludge: Effects of treatment. *Environmental Science and Technology*, *51*(2), 810–818. https://doi.org/10.1021/acs.est.6b04048

Margot, J., Rossi, L., Barry, D. A., & Holliger, C. (2015). A review of the fate of micropollutants in wastewater treatment plants. *Wiley Interdisciplinary Reviews: Water*, *2*(5), 457–487.

Mason, S. A., Garneau, D., Sutton, R., Chu, Y., Ehmann, K., Barnes, J., Fink, P., Papazissimos, D., & Rogers, D. L. (2016). Microplastic pollution is widely detected in US municipal wastewater treatment plant effluent. *Environmental Pollution*, *218*, 1045–1054. https://doi.org/https://doi.org/10.1016/j.envpol.2016.08.056

Meng, Y., Liu, W., Liu, X., Zhang, J., Peng, M., & Zhang, T. (2021). A review on analytical methods for pharmaceutical and personal care products and their transformation products. *Journal of Environmental Sciences*, *101*, 260–281. https://doi.org/https://doi.org/10.1016/j.jes.2020.08.025

Miege, C., Choubert, J. M., Ribeiro, L., Eusèbe, M., & Coquery, M. (2009). Fate of

pharmaceuticals and personal care products in wastewater treatment plants–
conception of a database and first results. *Environmental Pollution*, *157*(5), 1721–
1726.

Munno, K. E. (2017). *Microplastic retention by type in several species of fish from the Great Lakes*.

Murphy, F., Ewins, C., Carbonnier, F., & Quinn, B. (2016). Wastewater Treatment Works (WwTW) as a Source of Microplastics in the Aquatic Environment. *Environmental Science and Technology*, *50*(11), 5800–5808. https://doi.org/10.1021/acs.est.5b05416

Naidoo, T., Goordiyal, K., & Glassom, D. (2017). Are Nitric Acid (HNO 3) Digestions Efficient in Isolating Microplastics from Juvenile Fish? *Water, Air, & Soil Pollution*, *228*(12), 470.

Organization, W. H. (2018). *International code of conduct on pesticide management: Guidelines on highly hazardous pesticides*. Food & Agriculture Org.

Ouyang, X., Leonards, P., Legler, J., van der Oost, R., de Boer, J., & Lamoree, M. (2015). Comprehensive two-dimensional liquid chromatography coupled to high resolution time of flight mass spectrometry for chemical characterization of sewage treatment plant effluents. *Journal of Chromatography A*, *1380*, 139–145.

Pan, Z., Guo, H., Chen, H., Wang, S., Sun, X., Zou, Q., Zhang, Y., Lin, H., Cai, S., & Huang, J. (2019). Microplastics in the Northwestern Pacific: Abundance, distribution, and characteristics. *Science of The Total Environment*, *650*, 1913–1922. https://doi.org/https://doi.org/10.1016/j.scitotenv.2018.09.244

Park, H. J., Oh, M. J., Kim, P. G., Kim, G., Jeong, D. H., Ju, B. K., Lee, W. S., Chung, H. M., Kang, H. J., & Kwon, J. H. (2020). National Reconnaissance Survey of Microplastics in Municipal Wastewater Treatment Plants in Korea. *Environmental Science and Technology*, *54*(3), 1503–1512. https://doi.org/10.1021/acs.est.9b04929

Peck, A. M. (2006). Analytical methods for the determination of persistent ingredients of

personal care products in environmental matrices. *Analytical and Bioanalytical Chemistry*, *386*(4), 907–939. https://doi.org/10.1007/s00216-006-0728-3

Pei, J., Yao, H., Wang, H., Ren, J., & Yu, X. (2016). Comparison of ozone and thermal hydrolysis combined with anaerobic digestion for municipal and pharmaceutical waste sludge with tetracycline resistance genes. *Water Research*, *99*, 122–128. https://doi.org/https://doi.org/10.1016/j.watres.2016.04.058

Pérez-Lemus, N., López-Serna, R., Pérez-Elvira, S. I., & Barrado, E. (2019). Analytical methodologies for the determination of pharmaceuticals and personal care products (PPCPs) in sewage sludge: A critical review. *Analytica Chimica Acta*, *1083*, 19–40. https://doi.org/https://doi.org/10.1016/j.aca.2019.06.044

Perren, W., Wojtasik, A., & Cai, Q. (2018). Removal of microbeads from wastewater using electrocoagulation. *ACS Omega*, *3*(3), 3357–3364.

Pietrogrande, M. C., & Basaglia, G. (2007). GC-MS analytical methods for the determination of personal-care products in water matrices. *TrAC Trends in Analytical Chemistry*, *26*(11), 1086–1094. https://doi.org/https://doi.org/10.1016/j.trac.2007.09.013

Pivokonsky, M., Cermakova, L., Novotna, K., Peer, P., Cajthaml, T., & Janda, V. (2018). Occurrence of microplastics in raw and treated drinking water. *Science of The Total Environment*, *643*, 1644–1651. https://doi.org/https://doi.org/10.1016/j.scitotenv.2018.08.102

Qi, Y. (2010). *Ozonation of water and waste water: A practical guide to understanding ozone and its applications*. Taylor & Francis.

Rajala, K., Grönfors, O., Hesampour, M., & Mikola, A. (2020). Removal of microplastics from secondary wastewater treatment plant effluent by coagulation/flocculation with iron, aluminum and polyamine-based chemicals. *Water Research*, 116045. https://doi.org/https://doi.org/10.1016/j.watres.2020.116045

Rivera-Jaimes, J. A., Postigo, C., Melgoza-Alemán, R. M., Aceña, J., Barceló, D., & de

Alda, M. L. (2018). Study of pharmaceuticals in surface and wastewater from Cuernavaca, Morelos, Mexico: occurrence and environmental risk assessment. *Science of the Total Environment*, *613*, 1263–1274.

Schmidt, C., Kumar, R., Yang, S., & Büttner, O. (2020). Microplastic particle emission from wastewater treatment plant effluents into river networks in Germany: Loads, spatial patterns of concentrations and potential toxicity. *Science of The Total Environment*, 139544. https://doi.org/https://doi.org/10.1016/j.scitotenv.2020.139544

Schoeman, C., Dlamini, M., & Okonkwo, O. J. (2017). The impact of a wastewater treatment works in Southern Gauteng, South Africa on efavirenz and nevirapine discharges into the aquatic environment. *Emerging Contaminants*, *3*(2), 95–106.

Schultz, M. M., & Furlong, E. T. (2008). Trace analysis of antidepressant pharmaceuticals and their select degradates in aquatic matrixes by LC/ESI/MS/MS. *Analytical Chemistry*, *80*(5), 1756–1762.

Shannon, M. A., Bohn, P. W., Elimelech, M., Georgiadis, J. G., Mariñas, B. J., & Mayes, A. M. (2008). Science and technology for water purification in the coming decades. *Nature*, *452*(7185), 301–310. https://doi.org/10.1038/nature06599

Sharma, A., Kumar, V., Shahzad, B., Tanveer, M., Sidhu, G. P. S., Handa, N., Kohli, S. K., Yadav, P., Bali, A. S., Parihar, R. D., Dar, O. I., Singh, K., Jasrotia, S., Bakshi, P., Ramakrishnan, M., Kumar, S., Bhardwaj, R., & Thukral, A. K. (2019). Worldwide pesticide usage and its impacts on ecosystem. *SN Applied Sciences*, *1*(11), 1446. https://doi.org/10.1007/s42452-019-1485-1

Shemer, H., Kunukcu, Y. K., & Linden, K. G. (2006). Degradation of the pharmaceutical metronidazole via UV, Fenton and photo-Fenton processes. *Chemosphere*, *63*(2), 269–276.

Shi, J., Wang, Z., Stiverson, J. A., Yu, Z., & Li, Y. (2013). Reactor performance and microbial community dynamics during solid-state anaerobic digestion of corn stover

at mesophilic and thermophilic conditions. *Bioresource Technology*, *136*, 574–581.

Silva, R. D. O., De Menezes, M. G. G., De Castro, R. C., Nobre, C. D. A., Milhome, M. A. L., & Do Nascimento, R. F. (2019). Efficiency of ESI and APCI ionization sources in LC-MS/MS systems for analysis of 22 pesticide residues in food matrix. *Food Chemistry*, *297*, 124934.

Simon, M., Vianello, A., & Vollertsen, J. (2019). Removal of> 10 µm microplastic particles from treated wastewater by a disc filter. *Water*, *11*(9), 1935.

Snyder, S. A. (2008). Occurrence, treatment, and toxicological relevance of EDCs and pharmaceuticals in water. *Ozone: Science and Engineering*, *30*(1), 65–69.

Stolte, A., Forster, S., Gerdts, G., & Schubert, H. (2015). Microplastic concentrations in beach sediments along the German Baltic coast. *Marine Pollution Bulletin*, *99*(1), 216–229. https://doi.org/https://doi.org/10.1016/j.marpolbul.2015.07.022

Subedi, B., Balakrishna, K., Joshua, D. I., & Kannan, K. (2017). Mass loading and removal of pharmaceuticals and personal care products including psychoactives, antihypertensives, and antibiotics in two sewage treatment plants in southern India. *Chemosphere*, *167*, 429–437.

Sun, J., Dai, X., Wang, Q., van Loosdrecht, M. C. M., & Ni, B.-J. (2019). Microplastics in wastewater treatment plants: Detection, occurrence and removal. *Water Research*, *152*, 21–37. https://doi.org/https://doi.org/10.1016/j.watres.2018.12.050

Szymańska, U., Wiergowski, M., Sołtyszewski, I., Kuzemko, J., Wiergowska, G., & Woźniak, M. K. (2019). Presence of antibiotics in the aquatic environment in Europe and their analytical monitoring: recent trends and perspectives. *Microchemical Journal*, *147*, 729–740.

Taboada-Santos, A., Braz, G. H. R., Fernandez-Gonzalez, N., Carballa, M., & Lema, J. M. (2019). Thermal hydrolysis of sewage sludge partially removes organic micropollutants but does not enhance their anaerobic biotransformation. *Science of The Total Environment*, *690*, 534–542.

https://doi.org/https://doi.org/10.1016/j.scitotenv.2019.06.492

Tagg, A. S., Harrison, J. P., Ju-Nam, Y., Sapp, M., Bradley, E. L., Sinclair, C. J., & Ojeda, J. J. (2017). Fenton's reagent for the rapid and efficient isolation of microplastics from wastewater. *Chemical Communications*, *53*(2), 372–375. https://doi.org/10.1039/c6cc08798a

Talvitie, J., Heinonen, M., Pääkkönen, J. P., Vahtera, E., Mikola, A., Setälä, O., & Vahala, R. (2015). Do wastewater treatment plants act as a potential point source of microplastics? Preliminary study in the coastal Gulf of Finland, Baltic Sea. *Water Science and Technology*, *72*(9), 1495–1504. https://doi.org/10.2166/wst.2015.360

Talvitie, J., Mikola, A., Setälä, O., Heinonen, M., & Koistinen, A. (2017). How well is microlitter purified from wastewater? – A detailed study on the stepwise removal of microlitter in a tertiary level wastewater treatment plant. *Water Research*, *109*, 164–172. https://doi.org/https://doi.org/10.1016/j.watres.2016.11.046

Tamtam, F., Mercier, F., Le Bot, B., Eurin, J., Dinh, Q. T., Clément, M., & Chevreuil, M. (2008). Occurrence and fate of antibiotics in the Seine River in various hydrological conditions. *Science of the Total Environment*, *393*(1), 84–95.

Tan, E. S. S., Ho, Y. Bin, Zakaria, M. P., Latif, P. A., & Saari, N. (2015). Simultaneous extraction and determination of pharmaceuticals and personal care products (PPCPs) in river water and sewage by solid-phase extraction and liquid chromatography-tandem mass spectrometry. *International Journal of Environmental Analytical Chemistry*, *95*(9), 816–832. https://doi.org/10.1080/03067319.2015.1058929

Ternes, T. A., Joss, A., & Siegrist, H. (2004). Peer reviewed: scrutinizing pharmaceuticals and personal care products in wastewater treatment. *Environmental Science & Technology*, *38*(20), 392A-399A.

USEPA, U. S. (2003). Environmental Protection Agency. 2003. *Final NPDES General Permit for New Existing Sources and New Discharges in the Offshore Subcategory of the Oil and Gas Extraction Category for the Western Portion of the Outer*

Continental Shelf of the Gulf of Mexico, 1–117.

Van Cauwenberghe, L., Vanreusel, A., Mees, J., & Janssen, C. R. (2013). Microplastic pollution in deep-sea sediments. *Environmental Pollution*, *182*, 495–499. https://doi.org/https://doi.org/10.1016/j.envpol.2013.08.013

Venkatesan, A. K., Pycke, B. F. G., Barber, L. B., Lee, K. E., & Halden, R. U. (2012). Occurrence of triclosan, triclocarban, and its lesser chlorinated congeners in Minnesota freshwater sediments collected near wastewater treatment plants. *Journal of Hazardous Materials*, *229–230*, 29–35. https://doi.org/https://doi.org/10.1016/j.jhazmat.2012.05.049

Verlicchi, P., Al Aukidy, M., Galletti, A., Petrovic, M., & Barceló, D. (2012). Hospital effluent: investigation of the concentrations and distribution of pharmaceuticals and environmental risk assessment. *Science of the Total Environment*, *430*, 109–118.

Vlyssides, A. G., & Karlis, P. K. (2004). Thermal-alkaline solubilization of waste activated sludge as a pre-treatment stage for anaerobic digestion. *Bioresource Technology*, *91*(2), 201–206.

Wang, J., & Wang, S. (2016). Removal of pharmaceuticals and personal care products (PPCPs) from wastewater: A review. *Journal of Environmental Management*, *182*, 620–640. https://doi.org/https://doi.org/10.1016/j.jenvman.2016.07.049

Wang, Q.-J., Mo, C.-H., Li, Y.-W., Gao, P., Tai, Y.-P., Zhang, Y., Ruan, Z.-L., & Xu, J.-W. (2010). Determination of four fluoroquinolone antibiotics in tap water in Guangzhou and Macao. *Environmental Pollution*, *158*(7), 2350–2358.

Wang, S., Salamova, A., Hites, R. A., & Venier, M. (2018). Spatial and seasonal distributions of current use pesticides (CUPs) in the atmospheric particulate phase in the Great Lakes Region. *Environmental Science & Technology*, *52*(11), 6177–6186.

Watkinson, A. J., Murby, E. J., & Costanzo, S. D. (2007). Removal of antibiotics in conventional and advanced wastewater treatment: implications for environmental discharge and wastewater recycling. *Water Research*, *41*(18), 4164–4176.

Wei, W., Huang, Q. S., Sun, J., Dai, X., & Ni, B. J. (2019). Revealing the Mechanisms of Polyethylene Microplastics Affecting Anaerobic Digestion of Waste Activated Sludge. *Environmental Science and Technology*, *53*(16), 9604–9613. https://doi.org/10.1021/acs.est.9b02971

Wu, M., Xiang, J., Que, C., Chen, F., & Xu, G. (2015). Occurrence and fate of psychiatric pharmaceuticals in the urban water system of Shanghai, China. *Chemosphere*, *138*, 486–493.

Xia, K., Bhandari, A., Das, K., & Pillar, G. (2005). Occurrence and fate of pharmaceuticals and personal care products (PPCPs) in biosolids. *Journal of Environmental Quality*, *34*(1), 91–104.

Xie, L., Nakajima, F., Kasuga, I., & Kurisu, F. (2021). Simultaneous screening for chemically diverse micropollutants in public water bodies in Japan by high-performance liquid chromatography–Orbitrap mass spectrometry. *Chemosphere*, *273*, 128524. https://doi.org/https://doi.org/10.1016/j.chemosphere.2020.128524

Xu, X., Jian, Y., Xue, Y., Hou, Q., & Wang, L. (2019). Microplastics in the wastewater treatment plants (WWTPs): Occurrence and removal. *Chemosphere*, *235*, 1089–1096. https://doi.org/https://doi.org/10.1016/j.chemosphere.2019.06.197

Yan, M., Nie, H., Xu, K., He, Y., Hu, Y., Huang, Y., & Wang, J. (2019). Microplastic abundance, distribution and composition in the Pearl River along Guangzhou city and Pearl River estuary, China. *Chemosphere*, *217*, 879–886.

Yang, L., Li, K., Cui, S., Kang, Y., An, L., & Lei, K. (2019). Removal of microplastics in municipal sewage from China's largest water reclamation plant. *Water Research*, *155*, 175–181. https://doi.org/https://doi.org/10.1016/j.watres.2019.02.046

Yang, Y.-Y., Toor, G. S., & Williams, C. F. (2015). Pharmaceuticals and organochlorine pesticides in sediments of an urban river in Florida, USA. *Journal of Soils and Sediments*, *15*(4), 993–1004.

Yang, Y., Ok, Y. S., Kim, K.-H., Kwon, E. E., & Tsang, Y. F. (2017). Occurrences and

removal of pharmaceuticals and personal care products (PPCPs) in drinking water and water/sewage treatment plants: A review. *Science of the Total Environment*, *596*, 303–320.

Yuan, W., Liu, X., Wang, W., Di, M., & Wang, J. (2019). Microplastic abundance, distribution and composition in water, sediments, and wild fish from Poyang Lake, China. *Ecotoxicology and Environmental Safety*, *170*, 180–187. https://doi.org/https://doi.org/10.1016/j.ecoenv.2018.11.126

Zhang, Xiangyu, & Li, R. (2018). Variation of antibiotics in sludge pretreatment and anaerobic digestion processes: Degradation and solid-liquid distribution. *Bioresource Technology*, *255*, 266–272. https://doi.org/https://doi.org/10.1016/j.biortech.2018.01.100

Zhang, Xin, Zhao, H., Du, J., Qu, Y., Shen, C., Tan, F., Chen, J., & Quan, X. (2017). Occurrence, removal, and risk assessment of antibiotics in 12 wastewater treatment plants from Dalian, China. *Environmental Science and Pollution Research*, *24*(19), 16478–16487.

Zhang, Y., Liu, M., Qin, B., & Feng, S. (2009). Photochemical degradation of chromophoric-dissolved organic matter exposed to simulated UV-B and natural solar radiation. *Hydrobiologia*, *627*(1), 159–168.

Ziajahromi, S., Neale, P. A., Rintoul, L., & Leusch, F. D. L. (2017). Wastewater treatment plants as a pathway for microplastics: Development of a new approach to sample wastewater-based microplastics. *Water Research*, *112*, 93–99. https://doi.org/https://doi.org/10.1016/j.watres.2017.01.04.

Chapter 3

3   Effect of coagulation on microfiber in laundry water

3.1   Introduction

Microplastics pollution of the aquatic environment is one of the serious global environmental issues. Annual production of plastics and synthetic macromolecular polymers exceeds 320-335 million tonnes (Lapointe et al. 2020; Waring et al., 2018), and the global annual plastics discharge to the oceans are estimated to be 1.15 to 2.41 million tonnes per year (Lebreton et al. 2017). Due to human activities and environmental stressors, large plastic pieces gradually disintegrate to form small plastic particles, commonly called microplastics (MPs), with less than 5 mm diameter (Cooper et al., 2010; Jemec Kokalj et al. 2018). MPs are further categorized as primary and secondary particulates. Primary MPs usually are associated with cosmetic, clothing, cleaning and personal care products and enter the environment in microscopic size. Secondary MPs are the broken fragments from the degradable macro-scale plastic items due to mechanical and biological actions, sunlight and other environmental stressors (Hernandez et al., 2017; Lechner et al., 2015). Several environmental monitoring studies have indicated that MPs have been potentially stable in environmental matrices for a long time due to their chemical and physical stability (Chae et al., 2018; Cózar et al. 2014; Jiang et al. 2018). In recent years, research has already identified the physical and chemical damage such as internal abrasions or disruption of the digestive system, reduced growth and reproduction of aquatic or terrestrial organisms due to MPs (Galloway et al., 2016; Mathalon et al., 2014;). MPs also have been identified as a vector for contaminants by adsorbing persistent organic pollutants (POPs) and other toxic substances on their surface (Hüffer et al. 2019; Wang et al. 2018).

Microfibers resulting from the degradation of synthetic textile fabrics, such as polyester, acrylic, and polyamide during mechanical washing, are an essential source of microplastics in wastewater (Browne et al., 2011). An estimated 70 million tons of fibers are generated by the apparel industries every year, and significant microfibers pollution in the aquatic environment occurs due to laundry discharges (Carr et al., 2017). On average, washing a kilogram of cloth load releases 150,000 fiber particles (Tang et al., 2020), and

12.8×10^6 microfibers per cubic meter of water are released during the washing process (Pedrotti et al. 2021). Furthermore, fiber release experiments highlighted that washing a synthetic fabric (i.e. microfiber fabric) with only water can be a potential source of microplastics (Corami et al. 2020). Galvão et al. (2020) first time determined microfibers produced from residential washing of textiles in a household of 4 people; an estimated average discharge rate of 18,000,000 synthetic microfibers for a load of 6 kg of synthetic fibers was reported (Galvão et al. 2020). They also reported that only 7% of the synthetic fibers were larger than 500 μm in length, 40% were between 100 and 500 μm, and 53% were between 50 and 100 μm.

Due to the absence of accurate analytical methods to determine MPs and microfiber in complex wastewater metrics, the efficiency of MPs removal by various processes at municipal water treatment plants is uncertain. Coagulation and flocculation are typical primary treatment methods for removing suspended and colloidal particles in the wastewater. However, the extent of removal and mechanism of coagulation of MPs have been studied superficially using mostly pristine and monodisperse synthetic particles (Lapointe et al. 2020). Based on the best knowledge of authors, Larue et al. (2003) first studied the effect of coagulation/flocculation and electrocoagulation processes on latex particles (Larue et al. 2003). Later, several investigations have been reported by other researchers on other model MP particles, such as polyethylene (PE) microbeads and polystyrene (PS) microspheres, polyester (PEST) fibers as shown in Table 3.1.

Table 3.1: Previous studies on removal of MPs by coagulation

MPs type	Coagulants	Removal efficiency	Reference
Latex particles	ferric chloride, ferrous sulfate, or iron electrodes	> 90% at optimized condition	(Larue et al., 2003)
PE	aluminum chloride, ferric chloride, and polyacrylamide (PAM)	8%-20% (Al-based) 8%-12% (Fe-based) 5%-45% (PAM)	(Ma et al., 2019)
PE, PS, PEST	Al- based salts and cationic PAM	82%-99%	(Lapointe et al., 2020)
PE, rayon, PEST	Aluminum sulfate	86-99%	(Skaf et al., 2020)
PS	Ferric chloride, polyaluminum chloride, and polyamine.	95% for 1 μm MPs; 76% for 6.3 μm MPs	(Rajala et al., 2020)

Most of these studies reported high removal efficiency (>80%) of MPs during coagulation process in control treatment with inorganic and organic coagulants. One study had reported poor removal of pristine, less dense polyethylene microplastic particles (Ma et al., 2019) , of which mostly floated on water. To the best of our knowledge, no study has reported the removal of a model or authentic microfibers due to coagulation either in pure water or wastewater matrix. Microfibers comprise more than 85% of microplastics found on shorelines (Carr, 2017). Coagulation/Flocculation followed by sedimentation can reduce wastewater effluent's MPs burden, effectively transferring the significant load to primary sludge (Raju et al. 2018). However, the information related to microfiber coagulation emanating from textile fabrics is lacking. Therefore, the objectives of this work were to determine the coagulation performance of commonly used coagulant, ferric chloride (FeCl$_3$) and polyaluminum chloride (PACl) on microfibers of different sizes, isolated from residential laundry wastewater, both in pure water and laundry wash-water. Initially, coagulation tests were conducted using microfibers collected from the fabric mat on the lint screen of a household dryer and resuspending the microfibers in water. After that, the

effect of coagulation on real wash-water from a household laundry, both in the absence and presence of commercial detergent were investigated.

3.2   Materials and methods

3.2.1   Microfiber samples

Since it is difficult to isolate the fabric particles in wash-water from the washing machine, the fabric mat was collected from the lint screen of a dryer (GE, PCKS443EBWW). The fiber mat was dry ground using a coffee grinder (Black&Decker, Smartgrind). The relationship between mass and the number of microfiber particles of the fabric was determined following a simple gravimetric method described in the Supplementary Material, and the results are presented in Table S1. The results indicated that there were about 830 ± 75 particles/mg of fiber from the lint screen. The ground fiber was sieved using 90 μm & 125 μm sieve and was classified into three classes by particle size: < 90 μm, 90 - 125 μm, and > 125 μm. The sieved microfibers were stored in dry and capped bottles prior to the use in coagulation experiments. The types of microfibers were observed and identified using a microscope (Nikon Eclipse TS100) and a Fourier-transform Infrared Spectroscopy (FTIR, Perkin-Elmer, KBr disks), respectively.

3.2.2   Jar test

Coagulation experiments of microfibers were conducted using a jar test apparatus (Phipps&Bird, PB-700). Six 1 L beakers were filled with deionized water containing 30 mg/L microfiber samples of different sizes and stirred for 10 min to suspend the microfiber particles. The initial turbidity varied from 12-16 NTU, and the pH of the suspension was measured. $FeCl_3$ (0-30 mg/L) or PACl (0-5 mg/L) were added, and after one minute of rapid mixing at 145 rpm, the turbidity and pH were measured. Subsequently, turbidity was again determined after the flocculation step that involved slow mixing at 30 rpm for 30 minutes. After that, upon 120 min of settling (Ma et al., 2019), the final turbidity and pH of the solution were measured with pH meter (Thermo Scientific Orion Star A111) and Hach 2100AN turbidimeter, respectively. Control sedimentation experiments were conducted with microfiber particles in suspension but without chemical addition. The zeta potential of microfiber during coagulation was measured using Zetaplus (Brookhaven).

The coagulation study was conducted using (i) microfiber-DI water solution, (ii) microfiber-detergent-DI water solution, (iii) authentic wash-water without detergent, and (iv) authentic wash-water with detergent. The first experiment was conducted using 30 mg/L microfiber samples in DI water. In order to study the effect of surfactant on coagulation, 2 mg/L commercial liquid detergent (Tide, Procter & Gamble) was added into deionized water containing 30 mg/L microfiber samples. The chemical composition of the detergent used is given in Appendix A Table S2. Further, to compare the performance of the coagulation/flocculation process in microfiber DI water solution and authentic laundry wash-water, laundry wastewater samples were collected from a residential laundry machine (GE appliances) with 2 wash/spin combinations. The laundry processes with clothing only (no detergent) and clothing wash with 1 load (45 mL) detergent were investigated. Depending on the wash cycle and time, the initial turbidity of laundry wastewater varied from 20-302 NTU. All experiments were triplicated, and the standard deviation of all the tests were presented in error bar.

3.3 Results and Discussion

3.3.1 Removal of microfiber of different sizes

The typical dosage of $FeCl_3$ varies between 15- 40 mg/L for coagulation of raw or screened wastewater (Eddy et al. 2014). In this work, the dosage of $FeCl_3$ was varied from 0 - 30 mg/L. The overall removal efficiency of microfiber after 2 hours settling was 86%-89%, 90-92%, and 95%-96% for < 90 μm, 90 - 125 μm, and > 125 μm, respectively (Table 3.2). It can be seen that the microfiber particles all settled in 2 hours in the control test, and there is no clear advantage of $FeCl_3$ addition. As presented in Table 3.2, the removal efficiencies of larger size particles were slightly higher than smaller particles. Based on visual observations during the coagulation process, the large fibers formed clumps with higher settling velocities in water. Therefore, in a typical wastewater plant with a primary clarifier hydraulic retention time of 2-3 h, all microfibers of size between 90 - 125 μm should be settled in primary sludge. Similar results were found by Skaf et al. (2020) when model polyethylene microfiber particles of 5 μm diameter cut to 0.1 mm length, was removed well by coagulation.(Skaf et al. 2020) However, the authors did not report the removal of microfiber particles by sedimentation only.

Table 3.2: Turbidity reduction in coagulation with different size microfibers

Particle size (μm)	FeCl$_3$ (mg/L)	0	10	15	20	25	30
	Initial (NTU)	16.33±0.47	15.97±0.15	16.03±0.42	16.37±0.42	15.93±0.21	16.53±0.23
> 125	Final (NTU)	0.78±0.03	0.63±0.03	0.66±0.02	0.64±0.06	0.64±0.04	0.65±0.01
	Removal (%)	95%	96%	96%	96%	96%	96%
	Initial (NTU)	14.40±0.1	15.3±0.82	14.53±0.81	13.67±0.45	13.90±0.3	13.90±0.26
90-125	Final (NTU)	1.29±0.09	1.25±0.01	1.12±0.01	1.06±0.01	1.08±0.01	1.07±0.01
	Removal (%)	91%	92%	92%	92%	92%	92%
	Initial (NTU)	11.60±0.28	11.80±0.14	11.95±0.07	12.20±0.14	12.45±0.49	12.40±0.14
<90	Final (NTU)	1.61±0.02	1.54±0.02	1.57±0.02	1.34±0.02	1.34±0.02	1.34±0.03
	Removal (%)	86%	87%	87%	89%	89%	89%

However, for a shorter retention time, coagulant dosage did show some marginal benefit for particles smaller than < 90 μm, as shown in Figure 3.1, where removal increased with increasing dosage up to 15 mg/L. Previous studies reported a high dosage of 270-405 mg/L of aluminum chloride required for only 40% removal of MP particles below 500 μm, and 110-280 mg/L of ferric chloride for only <15% removal in drinking water treatment (Ma, et al., 2019).
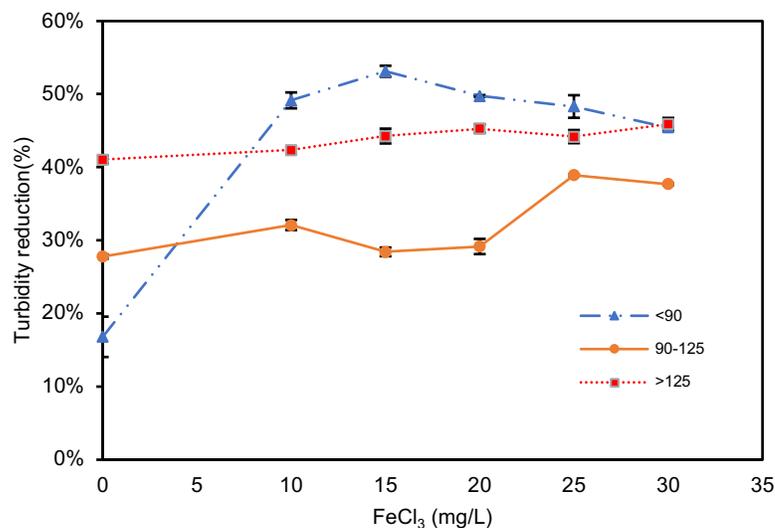
Figure 3.1: Coagulation of microfibers of different size

After coagulation, the microfibers and flocs were air-dried, and samples of microfiber particles were collected using a tweezer, placed on a glass slide with cover glass, and observed visually using a microscope. The images were taken at 20× magnification by the microscope. The images in Figure 3.2 show microfiber samples obtained from the lint screen of a dryer before and after coagulation. The microfibers are long, thick, and often occur in aggregates. Figure 3.2a shows microfiber clusters are primarily grey and black with some blue, green, and red color with a particle size ranging from 7.1 μm to 240 μm. These sizes are consistent with the microplastics size range of 20 to 1000 μm found in WWTP effluents (Enfrin et al., 2019). The coagulated fibers show the presence of ferric chloride, and microfibers looked bigger (Figure 3.2a vs. 3.2b); however, agglomeration of coagulated fibers could not be seen in Figure 3.2b.
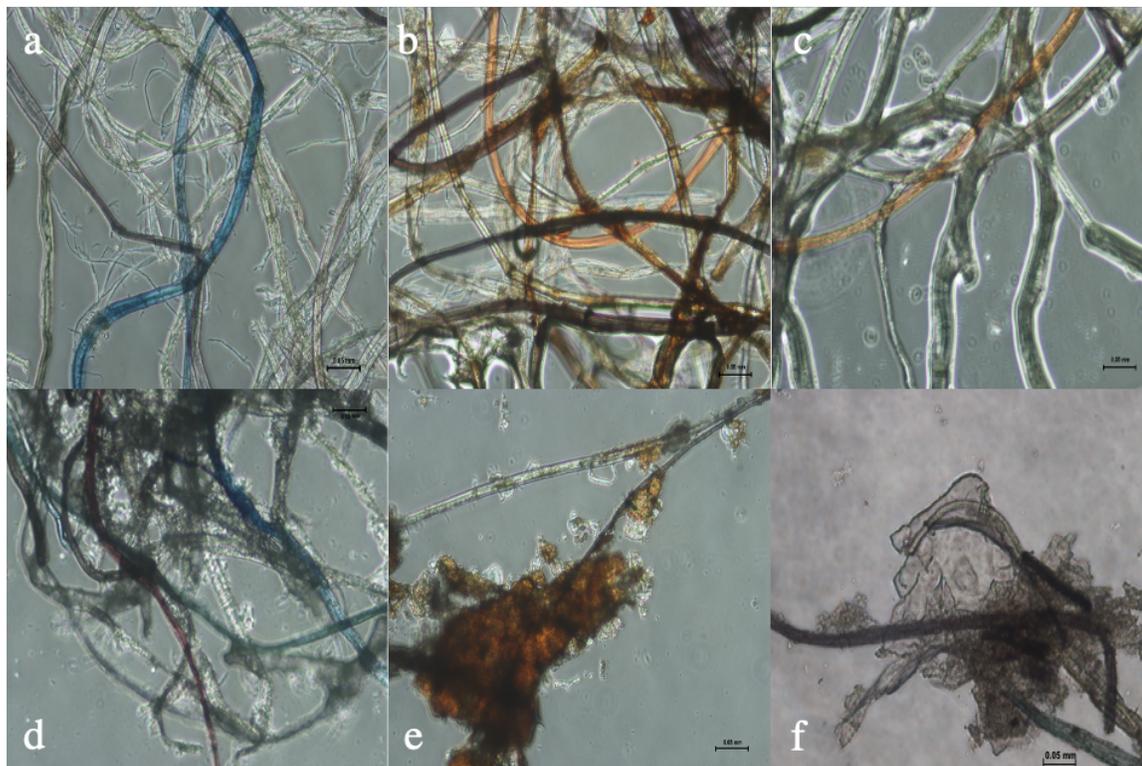
Figure 3.2: Morphology of microfiber before and after coagulation: (a) microfiber isolated from the dryer before coagulation; (b) microfiber flocs formed after coagulation with FeCl₃; (c) microfiber flocs formed by FeCl₃ with 2 mg/L surfactants, (d) microfibers in laundry wastewater; (e) microfiber flocs in laundry wastewater form after coagulation with FeCl₃, and (f) microfiber flocs formed by PACl in laundry wastewater with detergent

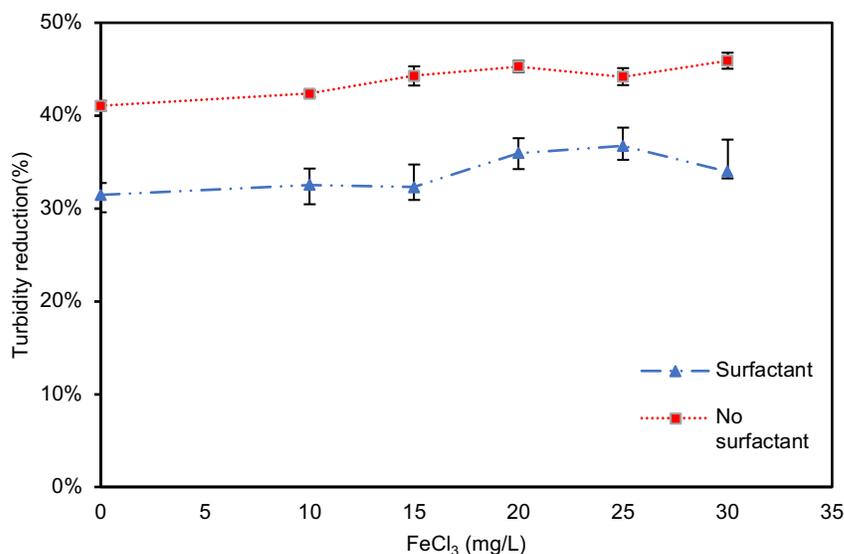## 3.3.2   Effect of surfactant on microfiber coagulation

The effect of surfactant on coagulation of microfiber (30 mg/L) was determined using 2 mg/L Tide detergent. The results show that the overall removal efficiency of microfiber > 150 μm and 90-125 μm after 2 hours settling was 86%-88% and 82% -89%, as shown in Table 3.3, which was slightly lower than the removal of microfiber without detergent.

Table 3.3: Turbidity reduction of microfiber coagulation with detergent

Particle size (μm)	FeCl₃ (mg/L)	0	10	15	20	25	30
> 125	Initial (NTU)	18.00±0.70	17.50±0.46	17.43±0.25	18.27±0.40	18.03±0.64	18.77±1.26
	Final (NTU)	2.44±0.34	2.51±0.43	2.73±0.21	2.49±0.36	2.56±0.31	2.65±0.08
	Removal (%)	86%	86%	84%	86%	86%	86%
90-125	Initial (NTU)	12.95±0.21	13.60±0.08	13.55±0.07	13.75±0.06	13.55±0.06	14.15±0.49
	Final (NTU)	2.36±0.14	1.48±0.08	1.74±0.03	2.20±0.06	2.44±0.06	2.49±0.04
	Removal (%)	82%	89%	87%	84%	82%	82%

As before, results for the flocculation period after 30 min are shown in Figure 3.3. The reduction in turbidity without surfactant ranged from 41%-46%, while in the presence of surfactant, the turbidity reduction was 31%-37%, and there were insignificant differences in turbidity reduction with increasing ferric chloride doses up to 30 mg/L. Surfactant facilitates stable suspension of microfibers, decreasing the removal by coagulation and settling. Both overall and 30 min removal efficiency was lower in the presence of detergent/surfactant. Similar behavior was found by Skaf et al., where higher residual turbidity was reported in the presence of laundry detergents during coagulation of synthetic microbeads. Microfiber clusters appear to contain fewer ferric particles in the presence of surfactants, as shown in Figure 3.2c.

Figure 3.3: Effect of surfactant on effectiveness of coagulation for microfibers

Generally, surfactants in detergent can lead to foam formation, which can affect the settling process. Other possible mechanisms for lower removal performance could be due to the surfactant stealth effect. The surfactant forms a protective film outside the surface of nano or microparticles, which can hide the adjacent suspended particulates.(Hu et al. 2018; Schöttler et al. 2016) The commercial detergent tide contains two kinds of surfactant, anionic surfactant sodium laureth sulfate (SLS) and nonionic surfactant lauramide monoethanolamin (MEA), shown in Appendix A Figure S1. The possible mechanism of anionic surfactant and nonionic surfactant during coagulation is shown in Figure 3.4. Anionic surfactant SLS has a negative charge due to the ionization of the sulfonic group, which could be adsorbed on the surface of microfiber, inducing stabilization of sol. Compared to anionic surfactants, the behavior of nonionic surfactant MEA is different. The formation of polyethylene glycol (PEG) film leads to a micelle-like structure, hindering microfiber particles from interacting with Fe-based coagulants. Both anionic and nonionic surfactants can potentially change the properties of the microfiber surface, affecting the agglomeration and precipitation of particles. Xia et al. (2020) suggested that the steric resistance of the PEG layer formed by tween 20 adsorbed on polystyrene microplastics

surface inhibited bentonite deposition and subsequent agglomeration and precipitation (Xia et al. 2020).
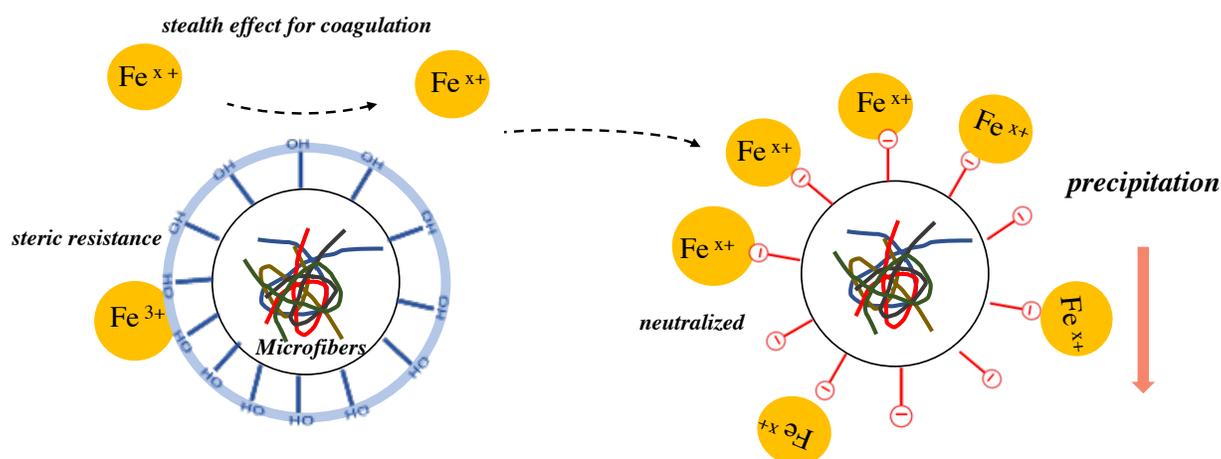
Figure 3.4: Mechanism of the surfactant effect on microfiber during coagulation process

3.3.3 Effect of Fe-based coagulation on microfiber in laundry wastewater

Further investigation was conducted using household laundry wastewater. The concentration level of surfactant in domestic or industrial laundry wastewater is much higher (Braga and Varesche 2014; Jardak, Drogui, and Daghrir 2016). Therefore, it is vital to explore the performance of coagulation in real laundry wastewater. The initial turbidity for different laundry cycles ranged from 20.87 to 302 NTU (Appendix A Table S4). The NTU values of regular laundry cycles with detergent are similar to numbers obtained in previous studies (Dimoglo et al. 2019; Nicolaidis and Vyrides 2014), whereas turbidity of wash-water in laundry cycles by washing clothing only (without detergent) was lower.
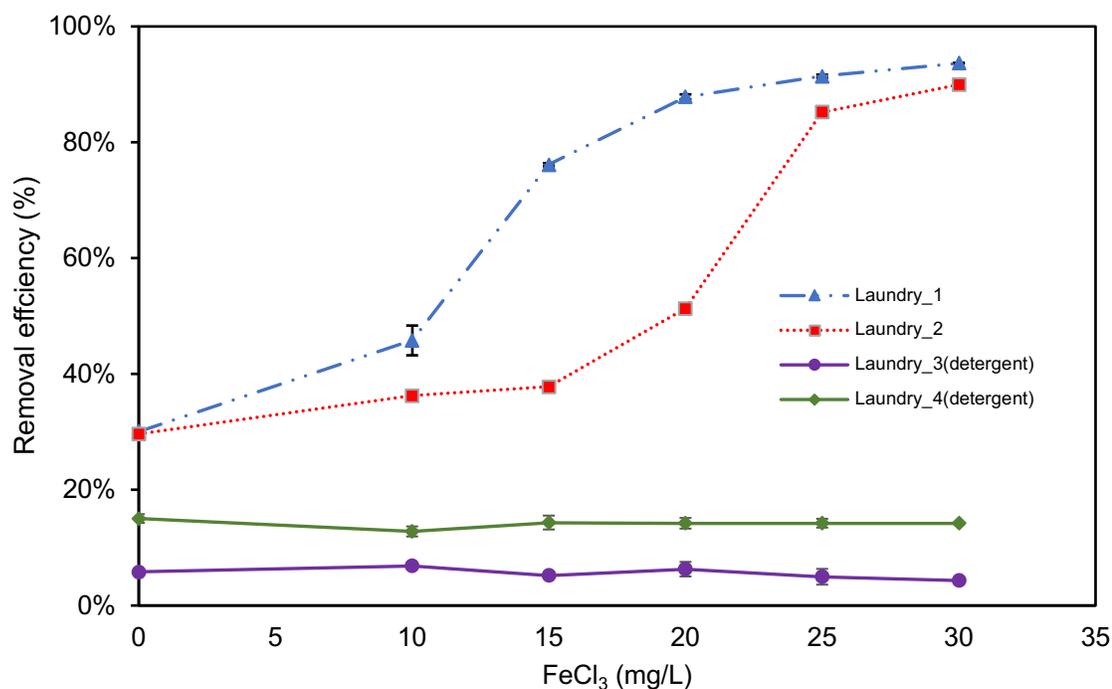
Figure 3.5: Removal efficiency of coagulation in laundry wastewater after 2 h settling (two trials for clothing washing only, two trials for clothing with 45 ml detergent)

The influence of coagulant concentration on the removal of microfiber in authentic laundry wastewater is quite significant, as can be seen in Figure 3.5, which was not observed in the case of microfiber isolated from the lint screen. Lint fibers were relatively larger in size and could be settled by themselves, and the effect of coagulation was not pronounced for microfiber size > 90 μm. Similar behaviors were observed for both trials for washing without detergent. Compared with the control condition, the Fe-based coagulants effectively removed microfibers from 46%-94% and 36%-90% for laundry cycle 1 and laundry cycle 2, respectively. However, in the presence of detergent, there was no coagulation of microfibers by $FeCl_3$, indicating that nano size microfibers will escape in the effluent without being trapped in the primary clarifier. Although the concentration of surfactant from detergent in laundry effluent is higher than what might be expected in a wastewater plant, there are many other sources of surfactant in wastewater, which will affect the coagulation performance of microplastics and microfiber.

The images using a microscope at 20× magnification show microfiber samples obtained from laundry wastewater before and after coagulation. Various colors of microfiber with similar size ranges from dozens to hundreds of micrometers were observed before coagulation (Figure 3.2d). As shown in Figure 3.2e, rusty colored larger cloudy flocs were formed after ferric chloride-based coagulation.

3.3.4 Effect of polyaluminum chloride (PACl) on microfiber in laundry wastewater

The removal efficiency of microfibers in laundry wastewater with detergent was less than 15% by coagulation with Fe-based salt. Therefore, other coagulants, such as polymer, were studied to achieve better removal performance. Polyaluminum chloride is widely used in water treatment plants (Zhou et al. 2021) at a lower concentration at 0 to 10 ppm (Almatin et al. 2019). The results of turbidity reduction are shown in Table 3.4. The overall removal efficiency of turbidity after 2 hours settling of laundry wastewater with and without detergent varied between 20%-98% and 22%-25%, respectively. Interestingly, in contrast to the results obtained with ferric chloride, the addition of PACl showed good turbidity removal performance in the presence of detergent. The removal efficiency initially increased with the increasing concentration of PACl up to 2 mg/L; after that, it remained constant when PACl concentration was increased up to 3 mg/L. The zeta potential of the laundry wastewater in the presence of PACl was measured to understand the observed contrasted $FeCl_3$ and PACl behavior. Typically, the colloidal suspension is unstable when zeta potential is around zero due to charge neutralization of the solution, and it is generally stable when the zeta potential is either negative or positive due to electrostatic repulsion between co-ions. The zeta potential of laundry wastewater crosses the zero point at a low dosage of PACl (below 0.5 mg/L) in the absence of detergent (Appendix A Figure S2a). Above a dosage higher than 0.5 mg/L, microfibers sol show positive zeta potential and hence low turbidity removal due to charge reversal and subsequent stabilization. In the presence of detergent, a slightly higher dosage of PACl is required to reach the zero zeta potential where charge neutralization occurs, leading to destabilization and removal of the microfibers (Appendix A Figure S2a). These results indicate that the primary mechanism of coagulation of microfibers using PACl is charge neutralization. In order to further

understand the effect of a low dosage of PACl (below 0.5 mg/L) in the absence of detergent, the coagulation experiment was conducted at a lower range of 0.1-0.5 mg/L of PACl. As shown in Table 3.5, the total removal efficiency increased from 22% to 98% with the addition of PACl from 0 to 0.5 mg/L. Similarly, charge reversal occurred at the lower dosage range around 0.4 to 0.5 mg/L PACl in laundry wastewater without detergent, as shown in Appendix A Figure S2c.

Table 3.4: Turbidity of PACl coagulation of laundry wastewater with and without detergent

| Laundry conditions	PACl (mg/L)	0	1	2	3	4	5
with detergent	Initial (NTU)	108±2.83	107±2.83	110.5±0.71	111.50±0.71	21.77±0.00	21.63±0.71
	Final (NTU)	94±0.42	87.00±0.28	2.31±0.72	6.84±0.59	10.03±0.24	24.20±0.99
	Removal (%)	13%	17%	97%	94%	91%	79%
without detergent	Initial (NTU)	42.80±0.28	42.55±0.21	43.25±0.35	42.75±0.78	42.60±0.14	42.30±0.42
	Final (NTU)	31.95±0.07	32.35±0.07	33.50±0.71	33.40±0.14	33.05±0.07	32.90±0.71
	Removal (%)	25%	24%	23%	22%	22%	22%

Table 3.5: Turbidity of low dosage of PACl coagulation of laundry wastewater in absence of detergent

Laundry conditions	PACl (mg/L)	0	0.1	0.2	0.3	0.4	0.5
without detergent	Initial (NTU)	83.25±0.21	87.95±0.78	88.00±0.14	87.65±0.49	85.25±0.49	87.85±0.21
	Final (NTU)	64.55±2.90	52.70±0.71	45.50±0.85	13.05±0.21	3.10±0.79	2.10±0.01
	Removal (%)	22%	40%	48%	85%	96%	98%

By comparing the removal performance in the same laundry wastewater, PACl proved much higher removal efficiency for the microfibers up to 99%, while $FeCl_3$ has lower than 20% removal efficiency (Appendix A Table S4). Both PACl and $FeCl_3$ are positively charged coagulants; the zeta potential was changed from -33.01 mv to 13.87 mv and -23.28 mv to -25.30 mv in PACl and $FeCl_3$ systems during coagulation (Appendix A Figure S3). Therefore, in addition to charge neutralization, adsorption-bridging on the polymer could play a significant role in microfibers destabilization (Zhang et al. 2021). Restabilization also occurs at a higher dosage, as shown in Figure S3, and dose optimization is required using a controlled study. Figure 3.2f illustrates an image of microfiber taken after coagulation and shows the corresponding changes in the relative presence of PACl. For coagulation using 0-5 mg/L PACl, the microfibers were clustered with a thin film of PACl precipitate.

Figure 3.6 depicts the microfibers' composition analyzed by Fourier transform infrared spectroscopy (FTIR in the range from 4000–400 cm^{-1}, the wavenumber precision at 0.01 cm^{-1}, and the resolution at 0.09 cm^{-1}). These fibers displayed the characteristic N-H stretching vibration or aromatic C-H stretching vibration located at 3400-3500 cm^{-1}, the alkyl C-H stretching vibration in the region immediately below 3000 cm^{-1}, and carbonyl

stretching peak at about 1600-1700 cm^{-1}. The peaks in the 1000-1300 cm^{-1} region corresponded to the C - H bending of aromatic carbons and the C-O-C asymmetric and symmetric stretching vibration of alkoxy ether, while the absorption at ~1000 cm^{-1} could be assigned to H-vibration attached to the aromatic ring (Cincinelli et al., 2017; González-Pleiter et al., 2020; Praveena et al., 2020). The test microfibers exhibited possible characteristic spectral features of polyester and/or polyamide by compared the similarity from polymer spectra library. The spectra of microfibers are similar irrespective of the experimental condition indicating no change in bond structure, which is expected as coagulation is merely a physical process. However, the spectra indicate the presence of microfibers in the flocs.
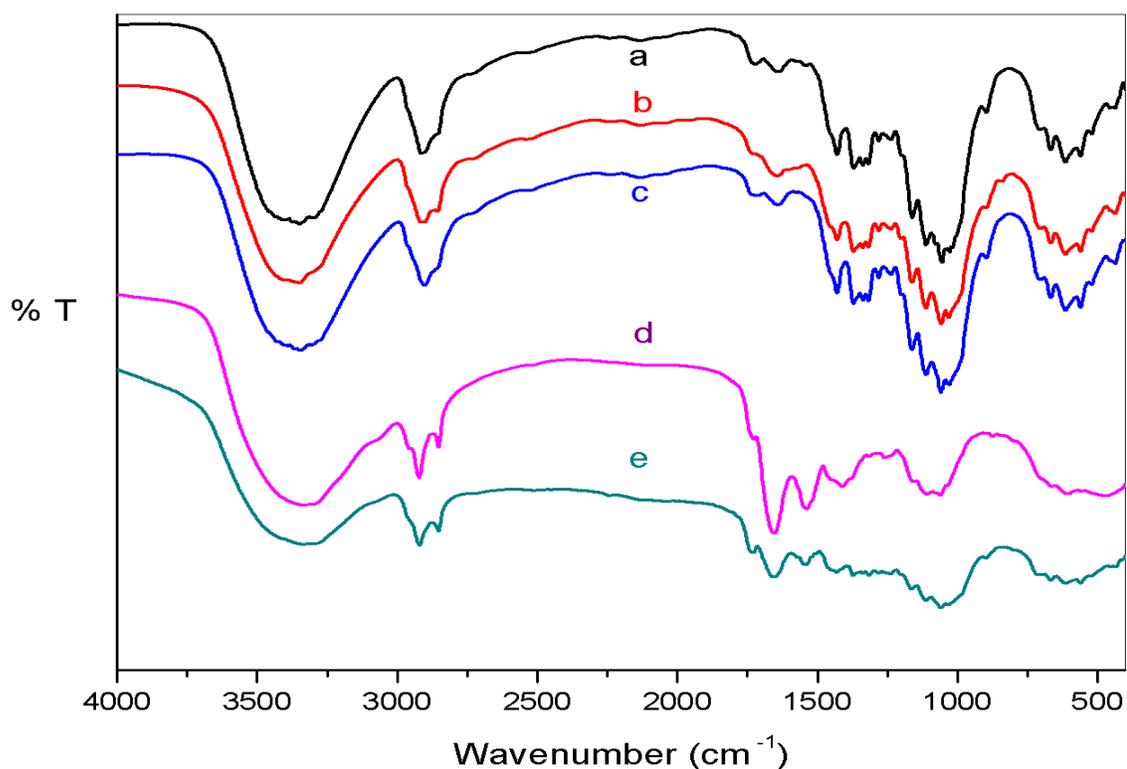
Figure 3.6: FT-IR spectra of microfibers before and after coagulation with FeCl3 (a) microfiber isolated from laundry dryer; (b) microfiber flocs formed by FeCl3; (c) microfiber flocs form by FeCl3 with 2 mg/L detergent; (d) microfiber in laundry wastewater; e) flocs of microfiber in laundry wastewater formed by FeCl₃

3.4 Conclusion

Microfiber is a large subgroup of microplastics. The presence of microfiber accounts for a large proportion of microplastics and becoming more common in municipal WWTPs influents. Ferric chloride, a commonly used coagulant in the primary treatment of wastewater, was used to treat suspension of microfiber collected from a lint screen in a dryer, as well as real wash-water from a household washer. Although lint microfibers of size greater than 90 μm settled by gravity within 2-3 hours without much effect of coagulant dose, more pronounced effect of Fe-based coagulation (0-30 mg/L) could be seen for smaller size and at lower settling time when initial turbidity was reduced by 86%-96%. In the absence of detergent, microfiber in wash-water from laundry could be effectively (90-94%) removed by coagulation. However, in the presence of surfactant in detergent, there was no benefit of ferric addition in authentic laundry effluent with initial turbidity of 84-300 NTU, where only 5-15% turbidity was removed after coagulation and flocculation. In comparison with $FeCl_3$, PACl removed 90% of particles in laundry wastewater, which provides a reference for the process improvement. Microfiber can be effectively removed by coagulation process in conventional WWTPs with a typical dosage of Fe-based coagulant or without ferric. Moreover, PACl could be a more desirable coagulant, especially in the laundry industry wastewater treatment. In addition, with 90% microfibers removed by settling/coagulation, fate of microfiber in the settled sludge and final removal need to be further investigated.

3.5 Reference:

Braga, J. K., & Varesche, M. B. A. (2014). Commercial laundry water characterisation. *American Journal of Analytical Chemistry*, *2014*.

Carr, S. A. (2017). Sources and dispersive modes of micro-fibers in the environment. *Integrated Environmental Assessment and Management*, *13*(3), 466–469.

Chae, Y., & An, Y.-J. (2018). Current research trends on plastic pollution and ecological impacts on the soil ecosystem: A review. *Environmental Pollution*, *240*, 387–395. https://doi.org/https://doi.org/10.1016/j.envpol.2018.05.008

Cincinelli, A., Scopetani, C., Chelazzi, D., Lombardini, E., Martellini, T., Katsoyiannis, A., Fossi, M. C., & Corsolini, S. (2017). Microplastic in the surface waters of the Ross Sea (Antarctica): Occurrence, distribution and characterization by FTIR. *Chemosphere*, *175*, 391–400. https://doi.org/https://doi.org/10.1016/j.chemosphere.2017.02.024

Corami, F., Rosso, B., Bravo, B., Gambaro, A., & Barbante, C. (2020). A novel method for purification, quantitative analysis and characterization of microplastic fibers using Micro-FTIR. *Chemosphere*, *238*, 124564. https://doi.org/https://doi.org/10.1016/j.chemosphere.2019.124564

Cózar, A., Echevarría, F., González-Gordillo, J. I., Irigoien, X., Úbeda, B., Hernández-León, S., Palma, Á. T., Navarro, S., García-de-Lomas, J., Ruiz, A., Fernández-de-Puelles, M. L., & Duarte, C. M. (2014). Plastic debris in the open ocean. *Proceedings of the National Academy of Sciences of the United States of America*, *111*(28), 10239–10244. https://doi.org/10.1073/pnas.1314705111

Dimoglo, A., Sevim-Elibol, P., Dinç, Ö., Gökmen, K., & Erdoğan, H. (2019). Electrocoagulation/electroflotation as a combined process for the laundry wastewater purification and reuse. *Journal of Water Process Engineering*, *31*, 100877. https://doi.org/https://doi.org/10.1016/j.jwpe.2019.100877

Eddy, M. &, Abu-Orf, M., Bowden, G., Burton, F. L., Pfrang, W., Stensel, H. D.,

Tchobanoglous, G., Tsuchihashi, R., & (Firm), A. (2014). *Wastewater engineering: treatment and resource recovery*. McGraw Hill Education.

Enfrin, M., Dumée, L. F., & Lee, J. (2019). Nano/microplastics in water and wastewater treatment processes – Origin, impact and potential solutions. *Water Research*, *161*, 621–638. https://doi.org/https://doi.org/10.1016/j.watres.2019.06.049

Galloway, T. S., & Lewis, C. N. (2016). Marine microplastics spell big problems for future generations. *Proceedings of the National Academy of Sciences*, *113*(9), 2331–2333.

Galvão, A., Aleixo, M., De Pablo, H., Lopes, C., & Raimundo, J. (2020). Microplastics in wastewater: microfiber emissions from common household laundry. *Environmental Science and Pollution Research*, *27*, 26643–26649.

González-Pleiter, M., Velázquez, D., Edo, C., Carretero, O., Gago, J., Barón-Sola, Á., Hernández, L. E., Yousef, I., Quesada, A., Leganés, F., Rosal, R., & Fernández-Piñas, F. (2020). Fibers spreading worldwide: Microplastics and other anthropogenic litter in an Arctic freshwater lake. *Science of The Total Environment*, *722*, 137904. https://doi.org/https://doi.org/10.1016/j.scitotenv.2020.137904

Hernandez, E., Nowack, B., & Mitrano, D. M. (2017). Polyester Textiles as a Source of Microplastics from Households: A Mechanistic Study to Understand Microfiber Release during Washing. *Environmental Science and Technology*, *51*(12), 7036–7046. https://doi.org/10.1021/acs.est.7b01750

Hu, J., Sheng, Y., Shi, J., Yu, B., Yu, Z., & Liao, G. (2018). Long circulating polymeric nanoparticles for gene/drug delivery. *Current Drug Metabolism*, *19*(9), 723–738.

Hüffer, T., Wagner, S., Reemtsma, T., & Hofmann, T. (2019). Sorption of organic substances to tire wear materials: Similarities and differences with other types of microplastic. *TrAC Trends in Analytical Chemistry*, *113*, 392–401.

Jardak, K., Drogui, P., & Daghrir, R. (2016). Surfactants in aquatic and terrestrial environment: occurrence, behavior, and treatment processes. *Environmental Science*

and Pollution Research, *23*(4), 3195–3216.

Jiang, C., Yin, L., Wen, X., Du, C., Wu, L., Long, Y., Liu, Y., Ma, Y., Yin, Q., Zhou, Z., & Pan, H. (2018). Microplastics in sediment and surface water of west dongting lake and south dongting lake: Abundance, source and composition. *International Journal of Environmental Research and Public Health*, *15*(10). https://doi.org/10.3390/ijerph15102164

Lapointe, M., Farner, J. M., Hernandez, L. M., & Tufenkji, N. (2020). Understanding and Improving Microplastic Removal during Water Treatment: Impact of Coagulation and Flocculation. *Environmental Science & Technology*, *54*(14), 8719–8727.

Larue, O., Vorobiev, E., Vu, C., & Durand, B. (2003). Electrocoagulation and coagulation by iron of latex particles in aqueous suspensions. *Separation and Purification Technology*, *31*(2), 177–192.

Lebreton, L. C. M., Van Der Zwet, J., Damsteeg, J.-W., Slat, B., Andrady, A., & Reisser, J. (2017). River plastic emissions to the world's oceans. *Nature Communications*, *8*(1), 1–10.

Lechner, A., & Ramler, D. (2015). The discharge of certain amounts of industrial microplastic from a production plant into the River Danube is permitted by the Austrian legislation. *Environmental Pollution*, *200*, 159–160. https://doi.org/https://doi.org/10.1016/j.envpol.2015.02.019

Ma, B., Xue, W., Ding, Y., Hu, C., Liu, H., & Qu, J. (2019). Removal characteristics of microplastics by Fe-based coagulants during drinking water treatment. *Journal of Environmental Sciences*, *78*, 267–275.

Ma, B., Xue, W., Hu, C., Liu, H., Qu, J., & Li, L. (2019). Characteristics of microplastic removal via coagulation and ultrafiltration during drinking water treatment. *Chemical Engineering Journal*, *359*, 159–167. https://doi.org/https://doi.org/10.1016/j.cej.2018.11.155

Mathalon, A., & Hill, P. (2014). Microplastic fibers in the intertidal ecosystem

surrounding Halifax Harbor, Nova Scotia. *Marine Pollution Bulletin*, *81*(1), 69–79.

Nicolaidis, C., & Vyrides, I. (2014). Closing the water cycle for industrial laundries: An operational performance and techno-economic evaluation of a full-scale membrane bioreactor system. *Resources, Conservation and Recycling*, *92*, 128–135.

Pedrotti, M. L., Petit, S., Eyheraguibel, B., Kerros, M. E., Elineau, A., Ghiglione, J. F., Loret, J. F., Rostan, A., & Gorsky, G. (2021). Pollution by anthropogenic microfibers in North-West Mediterranean Sea and efficiency of microfiber removal by a wastewater treatment plant. *Science of The Total Environment*, *758*, 144195. https://doi.org/https://doi.org/10.1016/j.scitotenv.2020.144195

Praveena, S. M., Syahira Asmawi, M., & Chyi, J. L. Y. (2020). Microplastic emissions from household washing machines: preliminary findings from Greater Kuala Lumpur (Malaysia). *Environmental Science and Pollution Research*. https://doi.org/10.1007/s11356-020-10795-z

Rajala, K., Grönfors, O., Hesampour, M., & Mikola, A. (2020). Removal of microplastics from secondary wastewater treatment plant effluent by coagulation/flocculation with iron, aluminum and polyamine-based chemicals. *Water Research*, 116045. https://doi.org/https://doi.org/10.1016/j.watres.2020.116045

Raju, Subash, Maddison Carbery, Aswin Kuttykattil, Kala Senathirajah, S. R. Subashchandrabose, Geoffrey Evans, and Palanisami Thavamani. 2018. "Transport and Fate of Microplastics in Wastewater Treatment Plants: Implications to Environmental Health." *Reviews in Environmental Science and Bio/Technology* 17(4):637–53.

Schöttler, S., Becker, G., Winzen, S., Steinbach, T., Mohr, K., Landfester, K., Mailänder, V., & Wurm, F. R. (2016). Protein adsorption is required for stealth effect of poly (ethylene glycol)-and poly (phosphoester)-coated nanocarriers. *Nature Nanotechnology*, *11*(4), 372–377.

Skaf, D. W., Punzi, V. L., Rolle, J. T., & Kleinberg, K. A. (2020). Removal of micron-

sized microplastic particles from simulated drinking water via alum coagulation. *Chemical Engineering Journal*, *386*, 123807.

Tang, N., Liu, X., & Xing, W. (2020). Microplastics in wastewater treatment plants of Wuhan, Central China: Abundance, removal, and potential source in household wastewater. *Science of The Total Environment*, *745*, 141026. https://doi.org/https://doi.org/10.1016/j.scitotenv.2020.141026

Wang, F., Wong, C. S., Chen, D., Lu, X., Wang, F., & Zeng, E. Y. (2018). Interaction of toxic chemicals with microplastics: A critical review. *Water Research*, *139*, 208–219. https://doi.org/https://doi.org/10.1016/j.watres.2018.04.003

Xia, Y., Xiang, X.-M., Dong, K.-Y., Gong, Y.-Y., & Li, Z.-J. (2020). Surfactant stealth effect of microplastics in traditional coagulation process observed via 3-D fluorescence imaging. *Science of The Total Environment*, *729*, 138783.

Zhang, Y., Zhou, G., Yue, J., Xing, X., Yang, Z., Wang, X., Wang, Q., & Zhang, J. (2021). Enhanced removal of polyethylene terephthalate microplastics through polyaluminum chloride coagulation with three typical coagulant aids. *Science of The Total Environment*, *800*, 149589. https://doi.org/https://doi.org/10.1016/j.scitotenv.2021.149589

Zhou, G., Wang, Q., Li, J., Li, Q., Xu, H., Ye, Q., Wang, Y., Shu, S., & Zhang, J. (2021). Removal of polystyrene and polyethylene microplastics using PAC and FeCl3 coagulation: Performance and mechanism. *Science of The Total Environment*, *752*, 141837. https://doi.org/https://doi.org/10.1016/j.scitotenv.2020.141837

Chapter 4

4 Effect of microfiber and ozone pretreated microfiber on anaerobic digestion

4.1 Introduction

The rapid industrialization and anthropogenic activities are causing many environmental problems. A significant problem is degradation of aquatic systems due to small-sized plastics ultimately causing harm to humans and the entire ecosystem. As reported, plastics, synthetic polymers with an annual production exceeding 320 million tonnes (Waring et al., 2018), are derived from various sources, including natural, organic resources such as resins, cellulose, coal and crude oil. In the last decade, small size plastics are commonly known as microplastics (MPs) with diameter less than 5 mm, which are gradually formed from large plastics due to mechanical actions, biological function, sunlight, and other environmental stressors (Jemec et al., 2018). Many environmental monitoring studies have shown that MPs are potentially stable in environmental matrices for a long time due to their chemical and physical stability (Cózar et al., 2014; Jiang et al., 2018). Significant concerns are raised on the effects of MPs on the ecosystem as reflected in the status of current research in this area. According to recent publications, the impact of MPs on environmental and health problems (Browne et al., 2013; Triebskorn et al., 2019) have been demonstrated, including physical harm, such as ingestion, internal abrasions and entanglement on both marine species and freshwater biota (Li et al., 2018). MPs are also regarded as potential carriers to adsorb and concentrate persistent organic chemicals such as polychlorinated biphenyls (PCBs), polycyclic aromatic hydrocarbons (PAHs), hexachlorocyclohexane isomers (HCHs), and dichlorodiphenyltrichloroethane family (DDTs) owing to their strong hydrophobicity and specific surface areas.

Wastewater treatment plants (WWTPs), not specifically designed to remove MPs, are considered as an important source of MPs to discharge into the environment (Carr et al., 2016). In North America, around 4 million MPs particles per day are originated from WWTPs effluents in 17 different facilities across the United States (Mason et al., 2016). While, WWTPs effluents are considerable source of MPs and their release to water bodies,

over 90% of MPs remained in WWTPs sludge due to their hydrophobic characteristics (Wei et al., 2019). An earlier study reported that the abundance of MPs has reached 156 trillion/year in sewage sludge samples and with an average concentration of $22.7 \pm 12.1 \times 10^3$ particles/kg of dry sludge in 11 provinces in China (Li et al., 2018). In Europe, the abundance of MPs in sewage sludge can be up to 170,900 particle/kg sludge (Lares et al., 2018). Sludge can be reused as fertilizer in agriculture and other land applications after treatment and stabilization (Nizzetto et al., 2016). The high amount of MPs in sludge transferred into biosolids could also increase the presence of MPs in agricultural soil (Corradini et al., 2019). Therefore, the effects of MPs during the sludge treatment processes need thorough investigation.

Anaerobic digestion is one of the most common treatment processes for sewage sludge stabilization in WWTPs. Recently, the behaviors of MPs during anaerobic digestion have captured attention in research communities. Multiple reviews have recently been published on the effect of MPs on anaerobic digestion. Wei et al. (2019) indicated that the presence of polyvinyl chloride (PVC) in anaerobic digestion inhibited methane production by $75.8 \pm 0.2\%$ to $90.6 \pm 0.3\%$ compared to control. Their other study reported that the high concentration of polyethylene (PE) had inhibited methane production by 12.4–27.5%, while the low concentration of PE showed no significant influence on methane production (Wei et al., 2019). Later studies by Li et al. (2020) reported that polyester (PES) has resulted in an approximately 10% reduction in methane production at various MPs abundances (Li et al., 2020). However, the evidence indicates that the effects of different MPs on methane production varied with their abundance and characteristics. For instance, the toxic component bisphenol A (BPA) leaching from PVC caused adverse effects on methane production and the hydrolysis-acidification process (Wei et al., 2019). In contrast, Polyamide 6 (PA6) enhanced methane production in anaerobic digestion by leaching caprolactam (Chen et al., 2021). Laundry wastewater is an important source of microfibers of diverse nature. Therefore, the effect of real MPs on anaerobic digestion needs to be understood in control studies.

To date, several investigations regarding anaerobic digestion have focused on the effect of synthetic model MPs. In the last decades, more than 60% of textiles are composed of

synthetic fibers, and about 12.8×10^6 particles/ L microfibers released from the domestic washing process discharged into WWTPs (Pedrotti et al., 2021). However, as far our knowledge goes, no research has focused on anaerobic digestion of real microfibers emanating from laundry wastewater. Pretreatment methods are generally used to improve anaerobic digestion of complex organics. In our earlier studies oxidative pretreatment methods such as ozonation, ferrate, ultrasonication etc. were tested for improved anaerobic digestion of different types of wastewater (Das et al., 2021; Elbeshbishy et al., 2011). Furthermore, ozonation is a typical pretreatment process prior to anaerobic digestion for enhancing digestibility. Based on the authors' knowledge, the effect of ozone pretreatment of microfiber has never been reported. In this context, this study investigates the potential impacts of microfibers collected from a fabric mat on the lint screen of a household dryer on carbon transformation and methane production during anaerobic digestion. Initially, pretreatment of microfiber spiked in deionized water (DI) was carried out at different ozone dosages. Then, the effect of various microfiber levels on methane production was conducted using biochemical methane potential tests (BMP). In addition, the phosphorus changes during anaerobic digestion were also monitored.

4.2   Material and method

4.2.1   Sources of sludge and microfiber

In this study, anaerobically digested sludge was obtained from Stratford WWTP (Stratford, ON). Primary sludge was collected from Greenway wastewater treatment plant (London, ON). The primary sludge was used as a substrate, and the anaerobic sludge was used as a seed. Table 4.1 lists the properties of the digested and primary sludge. Batch experiments were conducted with different microfiber abundances of 0, 20, 100, and 10,000 mg/L with and without ozone pretreatment, as shown in Table 4.2. The microfibers were collected from the fabric mat deposited on the lint screen of a dryer (GE, PCKS443EBWW). The corresponding number of microfibers was added into each reactor before adding sludge.

Table 4.1: Characteristics of the primary sludge and inoculum

Sample	Primary sludge (PS)	Anaerobically digested sludge
Total solids (TS) (mg/L)	29245±2496	32210±254.6
Volatile solids (VS) (mg/L)	21420±1881	17835±190.9
Total suspended solids(TSS) (mg/L)	29130±1867	31380±1372
Volatile suspended solids (VSS) (mg/L)	23425±318.2	20175±1096
Total chemical oxygen demand (TCOD) (mg/L)	43200±4666	26352±3.5
Soluble chemical oxygen demand (SCOD) (mg/L)	168.5±45.9	78.5±21

Table 4.2: Experimental design with different microfiber abundance and pretreatment conditions

Reactor	Experiment conditions
Blank	inoculum + buffer water
Control	inoculum + PS
R1	inoculum + PS with microfiber 20 mg/L
R2	inoculum + PS with microfiber 20 mg/L (ozone pretreatment)
R3	inoculum + PS with microfiber 100 mg/L
R4	inoculum + PS with microfiber 100 mg/L (ozone pretreatment)
R5	inoculum + PS with microfiber 1000 mg/L
R6	inoculum + PS with microfiber 1000 mg/L (ozone pretreatment)

4.2.2   Ozone pretreatment

The ozone was produced by an Ozonizer (model TG-40, Ozone solutions, Hull, Iowa, USA) and bubbled continuously (gas phase concentration = 900 ppm and 4L/min flow rate,

measured by ozone analyzer (model UV-100, Eco Sensors, Newark, California, USA). Assuming NTP, the concentration is calculated as: $C = 4$ L·min^{-1}/24.2 L·mol^{-1}×2900 ppm×10^{-6}×48 g ·mol^{-1}= 0.023 g·min^{-1}. The ozone concentration in the solution was determined by the indigo method, which was about 1 mg/L at pH 7. Ozone experiments were carried out in a 200 mL batch reactor with buffer water at different microfiber abundance (20 mg/L, 100 mg/L, 1000 mg/L). The ozone is continuously bubbled in the reactor through an 8.9 cm stainless steel tube with orifices. The ozone dosage was varied from 0 to 1.5 mg O^3 /g TS. The sCOD, tCOD, TS, TSS, VS and VSS of microfiber suspension were analyzed before and after the treatment. Based on preliminary tests of those parameters, specific dosage of 0.6 O_3 /g TS were selected.

4.2.3   Batch anaerobic digestion

Batch anaerobic digestion was conducted using an Automatic Methane Potential Test System (AMPTS II, BPC Sweden) equipped with 600 mL reactors. After adding microfibers with 0.6 O_3 /g TS ozone pretreated and without pretreatment into the reactor, the primary sludge and inoculum were added in the reactor with a food to microorganism (F/M) ratio of 0.5. The working volume of serum bottle reactors was 400 mL, and pH was at 7.0 using hydrochloric acid (HCl) or sodium hydroxide (NaOH). Then, the reactors were purged with N_2 for 15 mins to achieve anaerobic conditions. Finally, all reactors were closed with rubber stoppers and connected with a gas collection set, incubated at 37 °C at 150 rpm for 14 days digestion process. The batch tests were conducted in duplicates. The standard deviation of all the tests were presented in error bar.

4.2.4   Analytical method

The TS, VS, TSS, VS, TCOD, SCOD and soluble phosphorus (sP) concentration of the sludge were measured before and after batch-test according to standard methods (Adams, 2017). The soluble phosphorus (SP) was determined using a Discrete Analyzer (AQ300, Seal analytical). The dissolved samples were measured before filtering with a 0.45 μm filter. The dewatering efficiency was measured by the free water volume of sludge samples after centrifuging at 4000 rpm for 10 min. All the tests were conducted in duplicates. The standard deviation of all tests were presented in error bar. The microfibers were air-dried

and then were carefully collected using a tweezer, placed on a glass slide with cover glass, and observed visually using a microscope (Nikon Eclipse TS100).

4.3   Results and discussions

4.3.1   Effect of microfiber abundance on methane production during anaerobic digestion

The cumulative methane production from primary sludge in presence of different abundance of microfiber as shown in Figure 4.1. The blank and control represent buffer water and the primary sludge used for anaerobic digestion in the absence of microfiber. The methane production of all tests was recorded throughout the entire digestion period. After 14 days complete anaerobic digestion, the cumulative methane production remained nearly stable at 16.71 mL $\pm$ 0 CH$_4$/g COD, 17.75 $\pm$ 1.93 mL CH$_4$/g COD, 22.11 $\pm$ 1.62 mL CH$_4$/g COD, and 22.57 $\pm$ 0.08 mL CH$_4$/g COD for 0 mg/L, 20 mg/L, 100 mg/L, 1000 mg/L microfiber, respectively. In comparison to control, the methane production increased by 6%, 32%, and 35% in the presence of microfiber at different abundance of microfibers.
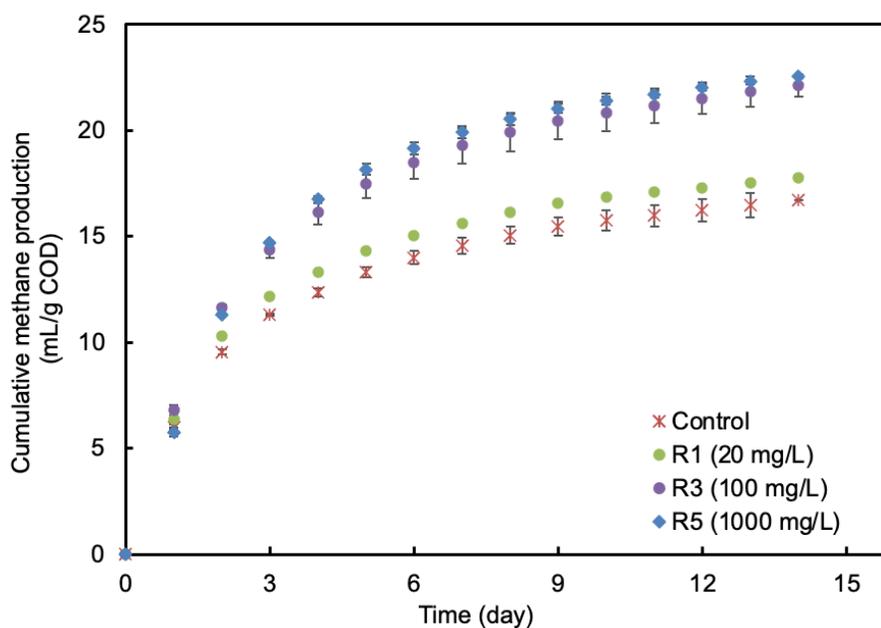

Figure 4.1: Cumulative methane production with and without ozone pretreatment at a different abundance of microfiber (Blank: buffer water only; Control: 0 mg/L; R1:20 mg/L; R3: 100 mg/L; R5: 1000 mg/L)

In this study, it was evident that higher concentration of microfiber enhanced methane production, which is in conflict with previous reports. Wei et al. (2019) reported that high levels of PVC (>20 particles/g) inhibited methane production by 9.4 % - 24.2 % due to the leaching of BPA on the hydrolysis-acidification process, and high levels of PE (100 and 200 particles/g) also decreased methane production by 27.5% (Wei, Huang, Sun, Dai, et al., 2019; Wei, Huang, Sun, Wang, et al., 2019). The positive results in this work are due to different nature of microfibers from various textiles. PES and PA are two main types of microfiber found in WWTPs (Magni et al., 2019). The behavior of these two microplastics seem to be considerably different from PVC and PE. The common mechanisms for microplastics that affect the anaerobic digestion process could be leaching of constituent that produce reactive oxygen species (ROS), affecting the microbial community, or incomplete digestion (Azizi et al., 2021). PES could cause a slight reduction of methane production due to incomplete digestion (L. Li et al., 2020), whereas PA enhanced methane production with all test dosages by improving the activities of key enzymes during anaerobic digestion (Chen et al., 2021). Our experiments indicate that microfibers in primary sludge may not inhibit anaerobic digestion.
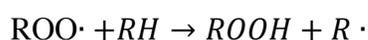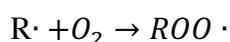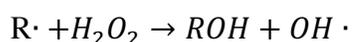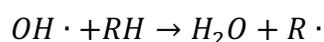
4.3.2 Effect of ozone pretreatment on microfiber during anaerobic digestion

In order to investigate possible changes in microfiber color, shape or surface after ozone treatments, the morphology was evaluated visually by a microscope, as shown in Figure. 4.2. The graph showed that microfibers have a rugged and crumpled surface, and the color of microfiber was faded after ozone pretreatment. Microscope images suggest that the oxidation treatments did physically alter microfiber.
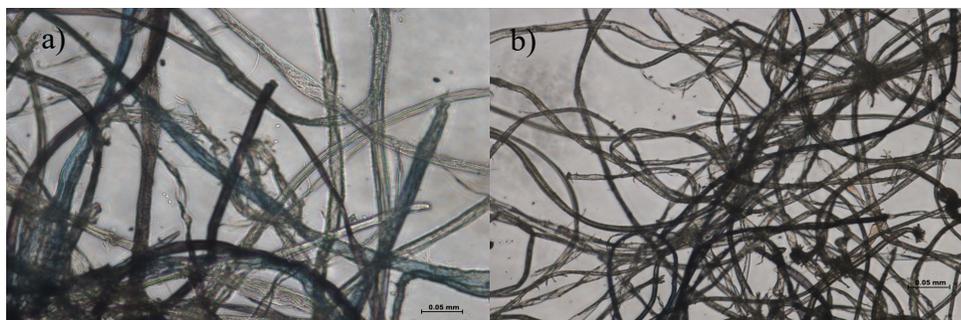
Figure 4.2: Microscope images for microfiber a) before and b) after ozone treatments

Various parameters of microfiber before and after oxidation treatments at a different dosage from 0-1.5 mg/g TS are presented in Table 4.3. TCOD, TS,VS,VSS remained more or less unchanged at 170-203 mg/L, 1267-1367 mg/L and 933-1033 mg/L, respectively. Turbidity of the solution decreased by around 29%, while sCOD has significantly increased 5 times after ozone treatments. The results indicated that the free radicals generated in the ozone system are likely to react with microfiber, release the soluble substances into solution. The common reaction of polymer and hydroxyl radicals were described by Carey (Carey, 1992) as follows:

$$OH \cdot + RH \rightarrow H_2O + R \cdot$$

$$R \cdot + H_2O_2 \rightarrow ROH + OH \cdot$$

$$R \cdot + O_2 \rightarrow ROO \cdot$$

$$ROO \cdot + RH \rightarrow ROOH + R \cdot$$

The FTIR spectra of microfibers revealed the direct chemical changes of microfibers during pretreatment and anaerobic digestion, especially on functional groups (Figure 4.3). The band around 3400-3300 cm^{-1} is related to the stretching of O-H groups (De Falco et al., 2019) or NH groups (Tang et al., 2021). It can be observed that the band around 3400-3300 cm^{-1} of ozone treatment microfiber was much wider and had a higher intensity

compared with the original microfiber, which demonstrated that microfiber contained more O-H or N-H groups after ozone treatment. For microfiber after anaerobic digestion, there was no obvious O-H/N-H stretching absorption peaks. The peaks shown at 2900 cm^{-1} to 2800 cm^{-1} were associated with C–H bond stretching vibration. The absorption peaks at 1700 cm^{-1} were related to C=O stretching, which was similarly broader and more intense ozone-treated microfiber than that of the original microfiber. Ozone-treated microfiber also has a more vigorous spectrum intensity on N-H bending (1650 cm^{-1} -1580 cm^{-1}) and O-H bending (1400 cm^{-1} -1300 cm^{-1}) compared with that before treatment. In addition, another change after ozonation was the significant increase in the intensity of the peak around 1200 cm^{-1} -1100 cm^{-1}, which was ascribed to C-O stretching. The results indicated certain chemical reaction were occurred during ozone pretreatment.
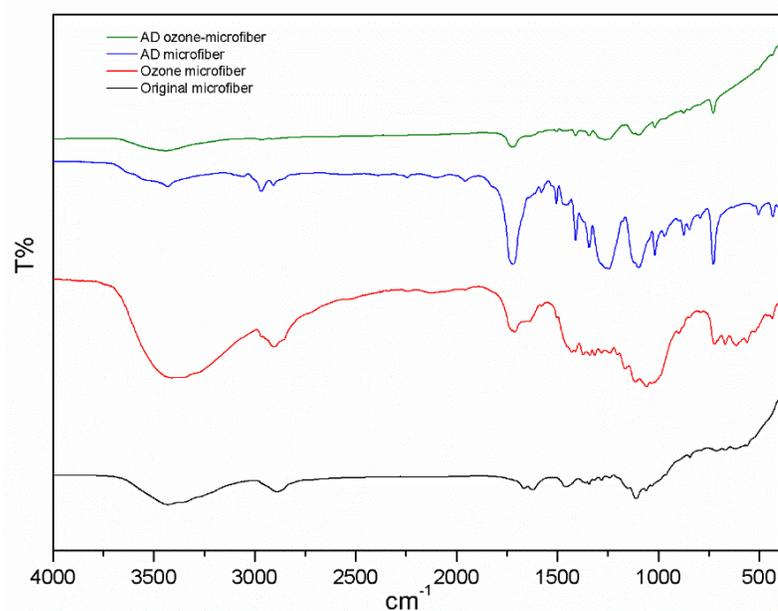

Figure 4.3: FTIR spectra of microfiber (black-original microfiber; red-ozone treated microfiber; blue-original microfiber after anaerobic digestion; green-ozone pretreatment microfiber after anaerobic digestion)

Table 4.3: Parameters of microfiber before and after ozone treatment

| Ozone dosage (mg/g TS)	Turbidity(NTU)	sCOD(mg/L)	tCOD(mg/L)	TS(mg/L)	VS (mg/L)
0	96.1	25	177	1367	967
0.1	64.7	125.5	170	1333	933
0.3	68.2	129.5	198	1400	1000
0.6	69.3	133	171	1300	1000
0.9	69.4	123.5	184	1300	1033
1.2	68.5	132.5	163	1333	1000
1.5	69.2	128	203	1267	1000

Earlier studies demonstrated that ozone react with polymer in main chains containing C=C bonds, aromatic rings or saturated hydrocarbons (He et al., 2015) to form carbonyl groups. For instance, polystyrene (PS) has aromatic rings; the CH_2 and CH_3 groups on aromatic rings are more likely to be chemically attacked by hydroxyl radicals. In contrast, polyethylene (PE) and polypropylene (PP) only have C-C single bond, which is more oxidation resistance (Gomes de Aragão Belé et al., 2021). Microfibers are complex matrices composed of polyester (PES), polyamide (PA), polyacrylonitrile (PAN), which also have similar carbonyl functional groups. The hydroxyl radicals could cleavage carboxylate and hydroxyl groups (Gabardo et al., 2021) or carbon-nitrogen single bond (Ren et al., 2019), as shown in Figure 4.4. In addition, the results indicated that ozone could oxidize microfibers, and increase soluble chemical oxygen demand.
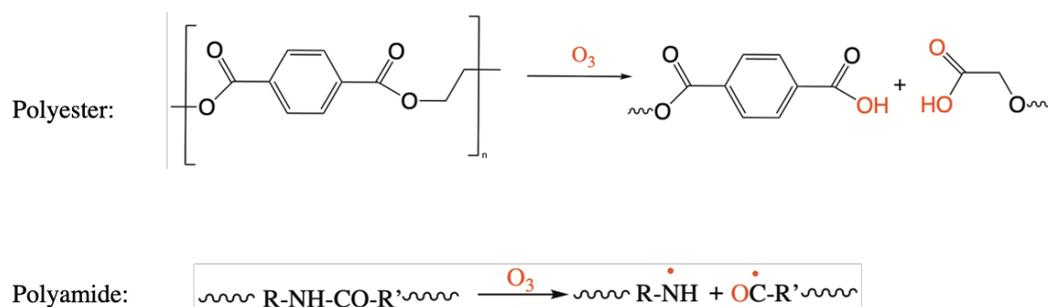
Figure 4.4: Possible reaction mechanisms of microfibers during ozone pretreatment

In this study, to investigate the impact of ozone pretreatment of microfibers on methane yield from primary sludge, batch anaerobic digestion was conducted and lasted for 14 days. Figures 4.5 a), b), and c) show the accumulated methane production in anaerobic digestion of PS spiked with 20 mg/L, 100 mg/L and 1000 mg/L microfibers with and without ozone pretreatment. During the 14 days digestion period, the methane production increased rapidly in the first 3 days, then rose slowly from day 4 to day 10, and achieved stationary in the last few days in each case. For low levels of microfiber (20 mg/L), the cumulative methane production during the entire digestion process was 17.75 ± 1.93 mL CH_4/g COD. In comparison, cumulative methane production with ozone pretreatment was 22.76 ± 1.13 mL CH_4/g COD, demonstrating a noteable increase of 28%. It is interesting to note that while the concentrations of microfibers increased, the positive effect of ozone pretreatment on methane production was significantly limited. The cumulative methane production was 22.11 ± 1.62 mL CH_4/g COD and 22.57 ± 0.08 mL CH_4/g COD for 100 mg/L and 1000 mg/L microfibers, respectively. After ozone pretreatment, the methane productions were 21.73 ± 0.1 mL CH_4/g COD, and 21.73 ± 1.59 mL CH_4/g COD, which showed on obvious changes in methane production for 100 mg/L microfibers and 1000 mg/L microfiber. The results indicated ozone pretreatment enhanced methane production at a low level of microfibers, while there is no noticeable effect on high concentration microfibers. The results indicated the mechanism for methane production increasing were not due to the biodegradation changes of ozone treated microfibers. One possible reason could be the ozone pretreatment release sub-micron scale microfiber and cracks the surface (Xu et al., 2021), which absorb toxicy substances during anaerobic digestion process in the presence

of low concentration microfiber. However, the specific mechanism of ozone pretreatment on microfiber needs to be further investigated.
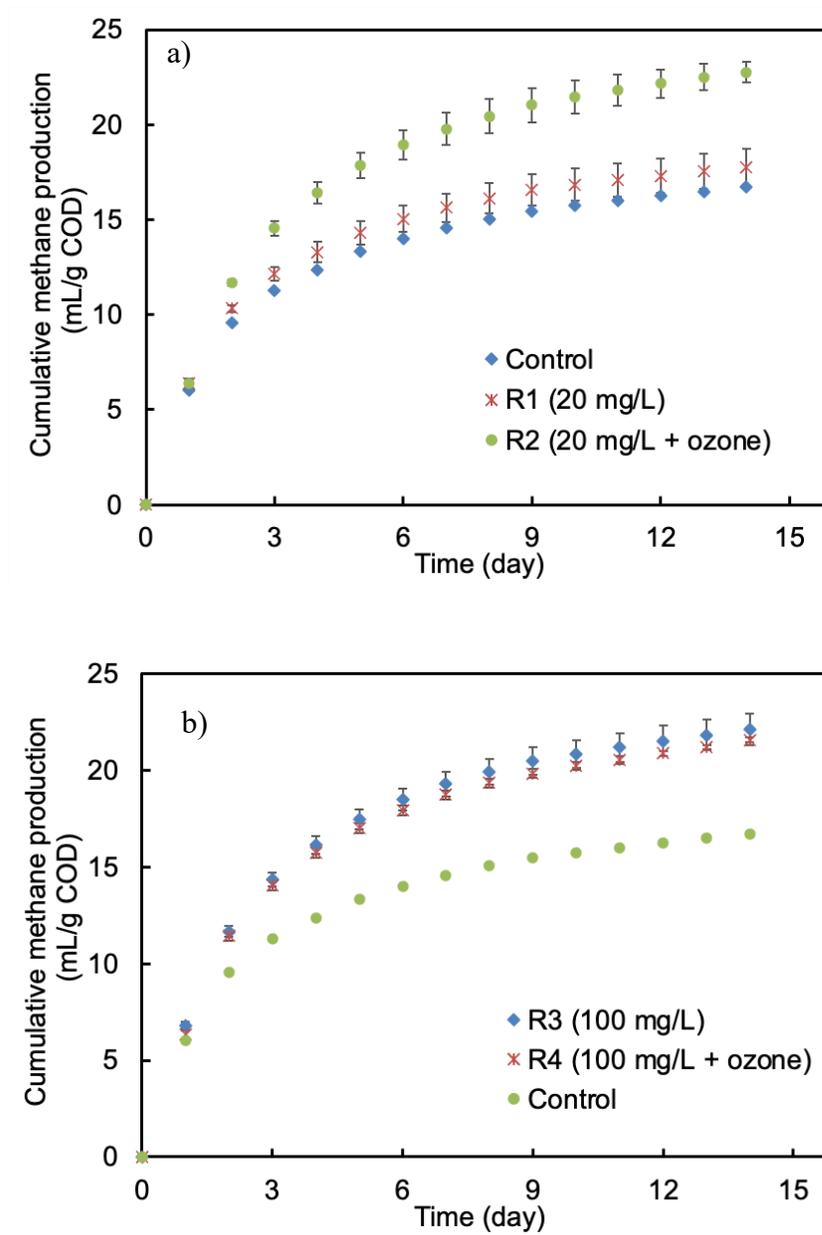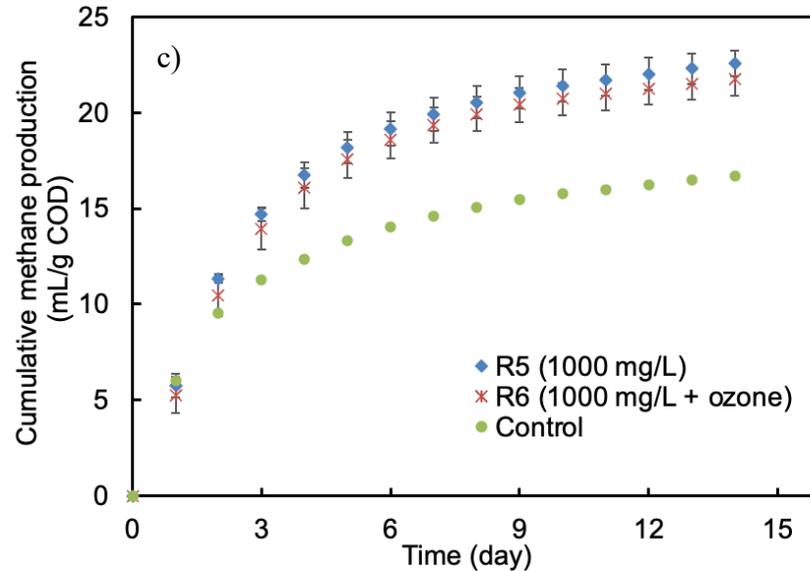
Figure 4.5: Cumulative methane production with and without ozone pretreatment (0.6 mg O3 /g TS) at different abundance of microfiber (a:20 mg/L; b: 100 mg/L; c:1000 mg/L)

4.3.3   Effect of microfibers on digested sludge characteristics

Pretreatment with an advanced oxidation process using ozonation has been used to improve the anaerobic digestion process (Uthirakrishnan et al., 2021). Ozone is a strong oxidizing agent, liberating extracellular and intracellular substances of microorganisms (Chacana et al., 2017). Previous results show ozone pretreatment only affected methane production at the lowest abundance of microfiber. However, the effect of ozonation on microfiber on the sludge characteristics is still unclear.

The average TS, VS, TSS, VSS, tCOD, sCOD, and sP concentrations of the digesters are shown in Table 4.4. Overall, the solids concentrations, especially VS decreased after ozonation. The VS reduction was 32%, 30%, 32%, 33%, 32%, 35%, and 40% in control reactor and R1 to R6, respectively. Ozone pretreatment does not affect the solids reduction in a short time digestion period, and the increasing abundance of microfibers has a slight enhancement, which was consistent with the results of methane production. The tendency in the tCOD and sCOD concentration changes agreed with the trend in solids concentrations. The tCOD and sCOD decreased from 28% to 46% and 37% to 45%,

respectively. Similar behaviors were observed with and without microfibers. Ozone pretreatment led up to 28% tCOD and 20% sCOD reduction in comparison with the control. Notably, soluble P decreased in the digester from 27% to 71% with microfiber addition, while the soluble P increased by 81% in the control reactor. It shows actually the microfiber is acting as a sink for the soluble P, reducing the content of phosphorus (Dong et al., 2021). The results indicated that to a certain extent microfiber and ozonated microfibers had influence on sludge characteristics. The corresponding removal of organic matters and nutrients contributed to complete anaerobic digestion, which was in agreement with the positive effects on methane production.

Table 4.4: The characteristics of sludge before and after anaerobic digestion

		Blank	Control	R1	R2	R3	R4	R5	R6
TSS (mg/L)	Before	27300	33325	33900	29500	33450	30200	32400	31900
	After	24300	26275	28050	24175	26725	24600	24425	24275
VSS (mg/L)	Before	15400	19400	20050	17275	19925	18000	19950	19575
	After	12300	13600	14450	12525	13775	12975	12875	12575
tCOD (mg/L)	Before	20350	27150	26975	23875	27325	26225	29800	31450
	After	16650	19080	19455	16365	19260	17160	19005	16905
sCOD (mg/L)	Before	329	567	596.5	593.5	599.5	607.5	625	620.5
	After	359	366.5	363	333	372	359	391.5	341.5
sP, PO_4^{3-} (mg/L)	Before	0.54	0.73	1.45	1.25	1.08	1.17	1.13	1.05
	After	0.71	1.32	0.56	0.36	0.48	0.46	0.82	0.55

The dewatering efficiency of sludge before and after anaerobic digestion under different microfibers abundance and pretreatment were also recorded in Figure 4.6. The dewatering efficiency before anaerobic digestion varied from 75.6 ± 1.13% to 81+ 1.98%. After

anaerobic digestion, the dewatering efficiency of all reactors was slightly higher than the control varying from 81.5 ± 0.35% to 85+ 2.12%. Similarly, Li et al., (2020) inspected the presence of commercial PES were found to promote the dewatering rate. The existence of microfiber have potential to enhance aggregation of sludge particles, which attributed to water release from sludge (Talvitie et al., 2017). Therefore, it can be surmised from the results that the presence of microfiber enhanced the dewatering ability in sludge during anaerobic digestion. This finding indicated that microfibers' presence might reduce sludge volume, reducing the cost of subsequent transportation and storage for sludge.
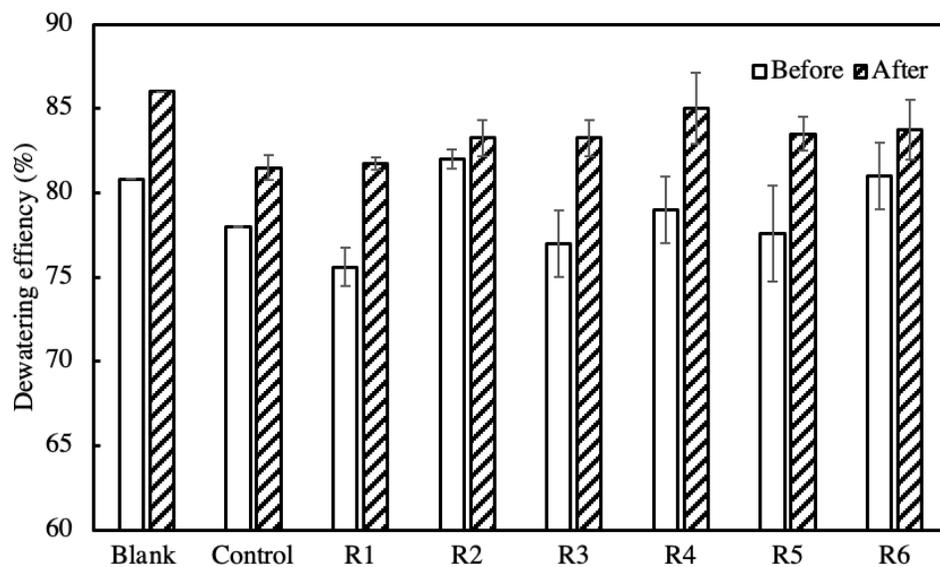
Figure 4.6: The dewatering efficiency before and after anaerobic digestion under various conditions (Blank: buffer water only; Control: 0 mg/L; R1:20 mg/L; R2: 20 mg/L+ ozone ; R3: 100 mg/L; R4: 100 mg/L+ ozone; R5: 1000 mg/L; R6: 1000 mg/L+ ozone.)

4.4 Conclusion

This work evaluated the effect of microfibers emanating from laundry wastewater and ozone pretreated microfibers on anaerobic digestion by monitoring cumulative methane production and sludge characteristics. The methane production of microfiber at a concentration range of 20 to 1000 mg/L was 16.71 mL $\pm$ 0 CH$_4$/g COD, 17.75 $\pm$ 1.93 mL CH$_4$/g COD, 22.11 $\pm$ 1.62 mL CH$_4$/g COD, and 22.57 $\pm$ 0.08 mL CH$_4$/g COD, respectively. High levels of microfibers at 100 mg/L and 1000 mg/L represented a significant increase of 32% and 35% compared to the control during the anaerobic digestion of primary sludge. Ozone pretreatment slightly increased methane production in low levels of microfibers (20 mg/L), while no apparent effects were observed in a high concentration of microfibers. The results showed that the microfibers and ozonation microfibers insignificantly affect the characteristics of sludge before and after anaerobic digestion. Especially for phosphorus removal, which were proved a weak correlation with microfiber. In addition, dewatering ability was slightly enhanced in the presence of microfibers and there was slight reduction in soluble phosphorus in presence of microfibers, probably due to adsorption. Overall, the occurrence of microfiber showed slight improvements in anaerobic digestion.

4.5 Reference

Adams, V. D. (2017). *Water and wastewater examination manual*. Routledge.

Azizi, S. M. M., Hai, F. I., Lu, W., Al-Mamun, A., & Dhar, B. R. (2021). A review of

mechanisms underlying the impacts of (nano) microplastics on anaerobic digestion.

Bioresource Technology, 124894.

Browne, M. A., Niven, S. J., Galloway, T. S., Rowland, S. J., & Thompson, R. C. (2013).

Microplastic Moves Pollutants and Additives to Worms, Reducing Functions Linked

to Health and Biodiversity. *Current Biology*, *23*(23), 2388–2392.

https://doi.org/https://doi.org/10.1016/j.cub.2013.10.012

Carey, J. H. (1992). An introduction to advanced oxidation processes (AOP) for

destruction of organics in wastewater. *Water Quality Research Journal*, *27*(1), 1–22.

Carr, S. A., Liu, J., & Tesoro, A. G. (2016). Transport and fate of microplastic particles

in wastewater treatment plants. *Water Research*, *91*, 174–182.

https://doi.org/https://doi.org/10.1016/j.watres.2016.01.002

Chacana, J., Alizadeh, S., Labelle, M.-A., Laporte, A., Hawari, J., Barbeau, B., &

Comeau, Y. (2017). Effect of ozonation on anaerobic digestion sludge activity and

viability. *Chemosphere*, *176*, 405–411.

https://doi.org/https://doi.org/10.1016/j.chemosphere.2017.02.108

Chen, H., Tang, M., Yang, X., Tsang, Y. F., Wu, Y., Wang, D., & Zhou, Y. (2021).

Polyamide 6 microplastics facilitate methane production during anaerobic digestion

of waste activated sludge. *Chemical Engineering Journal*, *408*, 127251.

https://doi.org/https://doi.org/10.1016/j.cej.2020.127251

Corradini, F., Meza, P., Eguiluz, R., Casado, F., Huerta-Lwanga, E., & Geissen, V.

(2019). Evidence of microplastic accumulation in agricultural soils from sewage

sludge disposal. *Science of The Total Environment*, *671*, 411–420.

https://doi.org/https://doi.org/10.1016/j.scitotenv.2019.03.368

Cózar, A., Echevarría, F., González-Gordillo, J. I., Irigoien, X., Úbeda, B., Hernández-León, S., Palma, Á. T., Navarro, S., García-de-Lomas, J., Ruiz, A., Fernández-de-Puelles, M. L., & Duarte, C. M. (2014). Plastic debris in the open ocean. *Proceedings of the National Academy of Sciences of the United States of America*, *111*(28), 10239–10244. https://doi.org/10.1073/pnas.1314705111

Das, S., Dagnew, M., & Ray, M. B. (2021). Anaerobic digestibility of resin acids in primary sludge: Effect of ozone pretreatment. *Chemosphere*, *262*, 128316. https://doi.org/https://doi.org/10.1016/j.chemosphere.2020.128316

Elbeshbishy, E., Aldin, S., Hafez, H., Nakhla, G., & Ray, M. (2011). Impact of ultrasonication of hog manure on anaerobic digestability. *Ultrasonics Sonochemistry*, *18*(1), 164–171. https://doi.org/https://doi.org/10.1016/j.ultsonch.2010.04.011

Gabardo, R. S., de Carvalho Cotre, D. S., Lis Arias, M. J., Moisés, M. P., Martins Ferreira, B. T., Samulewski, R. B., Hinestroza, J. P., & Bezerra, F. M. (2021). Surface Modification of Polyester Fabrics by Ozone and Its Effect on Coloration Using Disperse Dyes. In *Materials* (Vol. 14, Issue 13). https://doi.org/10.3390/ma14133492

Gomes de Aragão Belé, T., F. Neves, T., Cristale, J., Prediger, P., Constapel, M., & F. Dantas, R. (2021). Oxidation of microplastics by O3 and O3/H2O2: Surface modification and adsorption capacity. *Journal of Water Process Engineering*, *41*, 102072. https://doi.org/https://doi.org/10.1016/j.jwpe.2021.102072

He, G.-J., Zheng, T.-T., Ke, D.-M., Cao, X.-W., Yin, X.-C., & Xu, B.-P. (2015). Impact of rapid ozone degradation on the structure and properties of polypropylene using a reactive extrusion process. *RSC Advances*, *5*(55), 44115–44120.

Jemec Kokalj, A., Horvat, P., Skalar, T., & Kržan, A. (2018). Plastic bag and facial cleanser derived microplastic do not affect feeding behaviour and energy reserves of terrestrial isopods. *Science of The Total Environment*, *615*, 761–766. https://doi.org/https://doi.org/10.1016/j.scitotenv.2017.10.020

Jiang, C., Yin, L., Wen, X., Du, C., Wu, L., Long, Y., Liu, Y., Ma, Y., Yin, Q., Zhou, Z., & Pan, H. (2018). Microplastics in sediment and surface water of west dongting lake and south dongting lake: Abundance, source and composition. *International Journal of Environmental Research and Public Health*, *15*(10). https://doi.org/10.3390/ijerph15102164

Lares, M., Ncibi, M. C., Sillanpää, M., & Sillanpää, M. (2018). Occurrence, identification and removal of microplastic particles and fibers in conventional activated sludge process and advanced MBR technology. *Water Research*, *133*, 236–246. https://doi.org/https://doi.org/10.1016/j.watres.2018.01.049

Li, J., Liu, H., & Paul Chen, J. (2018). Microplastics in freshwater systems: A review on occurrence, environmental effects, and methods for microplastics detection. *Water Research*, *137*, 362–374. https://doi.org/https://doi.org/10.1016/j.watres.2017.12.056

Li, L., Geng, S., Li, Z., & Song, K. (2020). Effect of microplastic on anaerobic digestion of wasted activated sludge. *Chemosphere*, *247*, 125874. https://doi.org/https://doi.org/10.1016/j.chemosphere.2020.125874

Li, X., Chen, L., Mei, Q., Dong, B., Dai, X., Ding, G., & Zeng, E. Y. (2018).

Microplastics in sewage sludge from the wastewater treatment plants in China. *Water Research*, *142*, 75–85. https://doi.org/https://doi.org/10.1016/j.watres.2018.05.034

Liu, G., Zhu, Z., Yang, Y., Sun, Y., Yu, F., & Ma, J. (2019). Sorption behavior and mechanism of hydrophilic organic chemicals to virgin and aged microplastics in freshwater and seawater. *Environmental Pollution*, *246*, 26–33. https://doi.org/https://doi.org/10.1016/j.envpol.2018.11.100

Magni, S., Binelli, A., Pittura, L., Avio, C. G., Della Torre, C., Parenti, C. C., Gorbi, S., & Regoli, F. (2019). The fate of microplastics in an Italian Wastewater Treatment Plant. *Science of The Total Environment*, *652*, 602–610. https://doi.org/https://doi.org/10.1016/j.scitotenv.2018.10.269

Mason, S. A., Garneau, D., Sutton, R., Chu, Y., Ehmann, K., Barnes, J., Fink, P., Papazissimos, D., & Rogers, D. L. (2016). Microplastic pollution is widely detected in US municipal wastewater treatment plant effluent. *Environmental Pollution*, *218*, 1045–1054. https://doi.org/https://doi.org/10.1016/j.envpol.2016.08.056

Nizzetto, L., Futter, M., & Langaas, S. (2016). Are Agricultural Soils Dumps for Microplastics of Urban Origin? *Environmental Science and Technology*, *50*(20), 10777–10779. https://doi.org/10.1021/acs.est.6b04140

Pedrotti, M. L., Petit, S., Eyheraguibel, B., Kerros, M. E., Elineau, A., Ghiglione, J. F., Loret, J. F., Rostan, A., & Gorsky, G. (2021). Pollution by anthropogenic microfibers in North-West Mediterranean Sea and efficiency of microfiber removal by a wastewater treatment plant. *Science of The Total Environment*, *758*, 144195. https://doi.org/https://doi.org/10.1016/j.scitotenv.2020.144195

Ren, H., He, Z., Xu, J., Yan, X., & Liu, P. (2019). Evaluation of the Abilities of Ozone

    Resistance for Polyamide Fibers: Structures, Properties, and Aging Mechanism.

    Industrial & Engineering Chemistry Research, *58*(38), 17814–17823.

    https://doi.org/10.1021/acs.iecr.9b03284

Triebskorn, R., Braunbeck, T., Grummt, T., Hanslik, L., Huppertsberg, S., Jekel, M.,

    Knepper, T. P., Krais, S., Müller, Y. K., Pittroff, M., Ruhl, A. S., Schmieg, H.,

    Schür, C., Strobel, C., Wagner, M., Zumbülte, N., & Köhler, H.-R. (2019).

    Relevance of nano- and microplastics for freshwater ecosystems: A critical review.

    TrAC Trends in Analytical Chemistry, *110*, 375–392.

    https://doi.org/https://doi.org/10.1016/j.trac.2018.11.023

Uthirakrishnan, U., Godvin Sharmila, V., Merrylin, J., Adish Kumar, S., Dharmadhas, J.

    S., Varjani, S., & Rajesh Banu, J. (2021). Current advances and future outlook on

    pretreatment techniques to enhance biosolids disintegration and anaerobic digestion:

    A critical review. *Chemosphere*, 132553.

    https://doi.org/https://doi.org/10.1016/j.chemosphere.2021.132553

Wang, F., Wang, F., & Zeng, E. Y. (2018). *Chapter 7 - Sorption of Toxic Chemicals on

    Microplastics* (E. Y. B. T.-M. C. in A. E. Zeng (ed.); pp. 225–247). Elsevier.

    https://doi.org/https://doi.org/10.1016/B978-0-12-813747-5.00007-2

Wang, F., Wong, C. S., Chen, D., Lu, X., Wang, F., & Zeng, E. Y. (2018). Interaction of

    toxic chemicals with microplastics: A critical review. *Water Research*, *139*, 208–

    219. https://doi.org/https://doi.org/10.1016/j.watres.2018.04.003

Waring, R. H., Harris, R. M., & Mitchell, S. C. (2018). Plastic contamination of the food

    chain: A threat to human health? *Maturitas*, *115*, 64–68.

https://doi.org/https://doi.org/10.1016/j.maturitas.2018.06.010

Wei, W., Huang, Q.-S., Sun, J., Wang, J.-Y., Wu, S.-L., & Ni, B.-J. (2019). Polyvinyl Chloride Microplastics Affect Methane Production from the Anaerobic Digestion of Waste Activated Sludge through Leaching Toxic Bisphenol-A. *Environmental Science & Technology*, *53*(5), 2509–2517. https://doi.org/10.1021/acs.est.8b07069

Wei, W., Huang, Q. S., Sun, J., Dai, X., & Ni, B. J. (2019). Revealing the Mechanisms of Polyethylene Microplastics Affecting Anaerobic Digestion of Waste Activated Sludge. *Environmental Science and Technology*, *53*(16), 9604–9613. https://doi.org/10.1021/acs.est.9b02971

Chapter 5

5 Simultaneous quantification of five pharmaceuticals and personal care products in biosolids and their fate in thermo-alkaline treatment

5.1 Introduction

With the development of sensitive analytical methods and increasing awareness of environmental challenges, significant attention and research have been directed towards the detection and fate of pharmaceuticals and personal care products (PPCPs) in the environment (Li et al., 2017; Tranet al., 2018). These compounds are typically present in natural systems in trace concentrations and hence also termed as micropollutants. No discharge limit for them in environment has been established (Kosma et al., 2010). Many pharmaceuticals ingested by humans are not completely metabolized; some of the bioactive compounds remain un-metabolized or as insoluble components and are excreted through feces and urine into the wastewater treatment plants (WWTPs). Other possibilities of introduction into the environment include improper disposal or application of pharmaceuticals, leading to the release of active pharmaceuticals.

Conventional WWTPs processes are not designed for the removal of PPCPs; therefore, effluents discharged directly into aquatic systems may contain significant amount of PPCPs (Das et al., 2017). Compounds with high octanol-water partition coefficients (K_{ow}) are more likely to be partitioned into sludge and biosolids, causing potential problems of contaminating soil and groundwater due to agricultural amendments (Sherburne et al., 2016; Kodešová et al., 2019).

An estimated 780,000 dry tons of sludge/biosolids were generated in 2015 from municipal WWTP in Canada, of which 53.4% was used for land application as biosolids (Tessier et al., 2017). Land application of biosolids is a useful approach that recycles organic matter and nutrients, improves the physical, chemical, and biological properties of soils, re-establishes vegetation and restore degraded ecosystems (Torri et al., 2017). The biosolids program in Ontario, Canada, saves farmers approximately $5 million annually in fertilizer

costs (WEAO Residuals & Biosolids Committee, 2010). However, the presence of PPCPs in the sludge is a major concern for land application of treated biosolids (Xia et al., 2005).
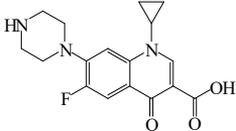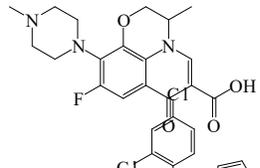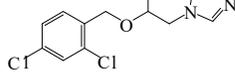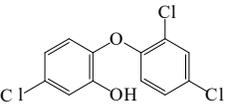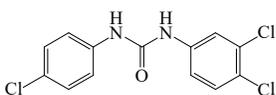
Establishing a valid extraction and detection method is an extremely important step to monitor the PPCPs in environmental samples at trace concentrations. Many recent studies were carried out indicating the importance of monitoring PPCPs in biosolids, where PPCPs were detected at higher concentrations in the range of mg/kg levels (Yang et al., 2015;). For example, triclosan, an antibacterial and antifungal agent present in many consumer products, was detected at high level (22700 ng/g) in anaerobically digested primary solids (Guerra et al., 2019). Azole antifungals were detected in the range of 3.7-11.1 ng/g (dry weight) in soils using ultrasound-assisted extraction method with recoveries in the range of 80.2–110.6% (Huang et al., 2018). Ciprofloxacin and ofloxacin, belonging to fluoroquinolones family, were detected in the primary sludge at 4.21 and 2.92 ng/g, respectively, using a pressurized liquid extraction (PLE) method and cleaning up by strong cation exchange (SCX) cartridges (Khadra et al., 2019). Due to the widely varying physical and chemical properties of the micropollutants, extraction methods need customization for simultaneous extraction and maximum recovery of several analytes present in water, soil and biosolids.

A variety of pre-treatment methods such as thermal, chemical, biological, and combination of them have been used to improve the anaerobic digestion of sludge for enhanced biogas production and greater inactivation of microbial pathogens (Takashima et al., 2014; Li et al., 2013; Tian et al., 2016). Similarly, post-treatment methods are used for greater stabilization of biosolids and odor removal. Among the many pre- and post-treatment methods tested at the lab scale, only a few mechanical, thermal, and thermochemical methods have been successfully applied at full scale. Based on a simple sustainability assessment, thermochemical treatment (at low temperatures $\leq 110\ °C$) of sludge and/or low temperature post treatment of biosolids was found to be an economical full-scale commercial method (Carrere et al., 2016). One such process is thermo-alkaline hydrolysis (TAH) conducted at pH 9.5 and 75°C, which is typically used as a post treatment for greater stabilization of biosolids after anaerobic digestion of sludge (Elbeshbishy et al., 2014). Amongst other benefits, the TAH allows odor removal and micropollutants removal

(Ahmad et al., 2019). Several studies have investigated the effect of TAH on enhanced biogas production and increased volatile suspended solids reduction efficiency, however, limited data exist on the effect of these processes on PPCPs in biosolids (Li et al., 2013).

Due to its relevance to the agriculture sector, it is important that biosolids treatment methods including TAH be evaluated in a more holistic way that includes their effects on PPCPs present. Towards this objective, five selected PPCPs (structures and the physio-chemical properties are shown in Table 5.1) including the fluoroquinolone antibiotics ciprofloxacin (CIP) and ofloxacin (OFLX), and three commonly used antimicrobial agents: miconazole (MIC), triclosan (TCS), and triclocarban (TCC), were detected and quantified in municipal biosolids before and following anaerobic digestion (AD) and TAH. These five PPCPs were chosen based on their high concentrations found in various biosolids in an internal work conducted by our group. In this study, it was hypothesized that thermo-alkaline hydrolysis has potential to reduce PPCPs in biosolids and producing more safety fertilizer to agriculture or other land application. Therefore, the major objectives of this chapter were: (1) to optimized simultaneous extraction and detection methods for the trace PPCPs which could applied to complex solid samples, (2) to determine partition of these compounds between water and various sludges depending on their solubility and octanol-water coefficient, (3) to explain the effect of TAH on the selected PPCPs in deionized water and biosolids collected from a local WWTP.

Table 5.1: The structure and important physiochemical characteristics of the selected PPCPs

Compound	Structure	pKa	log K_{ow}
Ciprofloxacin (CIP)		pKa_1: 6.18; pKa_2: 8.66	0.28
Ofloxacin (OFLX)		pKa_1: 5.97; pKa_2: 9.28	-0.39
Miconazole (MIC)		pKa_1: 6.77	6.1
Triclosan (TCS)		pKa_1: 12.7	4.76
Triclocarban (TCC)		pKa_1: 7.9	4.9

5.2   Materials and methods

5.2.1   Standards and chemicals

CIP ($\geq$ 98.0%), OFLX (98.0%) and TCC (98.0%) were purchased from Sigma-Aldrich, Canada. MIC (98.0%) and TCS (99.0%) were obtained from VWR, Canada. Isotopically labelled compounds (ILC) of ciprofloxacin-d8, ofloxacin-d8, miconazole-d5, triclocarban-13C6, and triclosan-d3 were purchased from Toronto Research Chemicals, Canada. LC-MS grade water, acetonitrile, methanol, and HPLC grade acetone, dichloromethane and hexane were purchased from Fisher Scientific, Canada. Solid-phase extraction (SPE) was performed using Oasis hydrophilic-lipophilic balance (HLB) cartridges, 200 mg/ 6 mL, Waters, Canada.

5.2.2   Sludge and biosolids samples

Sludge samples were collected from two municipal wastewater treatment plants, Guelph and London (Ontario, Canada), and the characteristics of the sludge are presented in Table 5.2. All sludge samples were stored at 4 °C. Primary sludge (PS) was filtered through 1 μm glass fiber filters (GF/C, Whatman) to separate the liquid and solid phases. Subsequently, solid samples were air-dried, ground to fine particles using a mortar and pestle, and stored in 1-L amber glass bottles at 4 °C until sample extraction.

Table 5.2: Characteristics of the sludge and biosolids samples

Sample	Location	pH	Total solid (TS%)
Primary sludge (PS)	London, ON	6.40	2.10%
PS-Waste activated sludge (PS-WAS) *75% PS and 25% thickened waste activated sludge (TWAS)*	Guelph, ON	7.66	4.51%
Biosolids	Guelph, ON	7.68	21.99%

5.2.3   Partition of the PPCPs in sludge

Since, the main objective of this work was to determine the effect of TAH on PPCPs present in biosolids as post-treatment, the subobjective was important to determine the amount of PPCPs partitioned on solids in different sludges which need to be treated before safe disposal and application. In this work, partition of the selected PPCPs on primary sludge (PS), and the combination of biosolids and PS-WAS with two ratios 15% : 85% (biosolids : PS-WAS, v/v) and 30% : 70% (biosolids : PS-WAS, v/v) was studied. These ratios of biosolids and PS-WAS sludge were used based on an actual practice in Guelph, ON, Canada, Wastewater Treatment Plant.

About 150 mL of primary sludge collected from the London Wastewater treatment plant (Greenway, London, ON) was taken in a 250 mL Erlenmeyer flask and agitated at 250 rpm for one hour. The PS was then spiked with the ILC as surrogate standards of the five PPCPs to obtain an end concentration of 250 ng/mL with constant stirring. About 12.5 mL of the spiked sludge was collected, and solid-liquid separation was achieved by centrifuging the sample at 3750 rpm for 10 minutes. The samples were analyzed using the methods described later in the analytical section. In order to eliminate the matrix effect of PS, about 12.5 mL of the un-spiked PS after mixing was taken and centrifuged at 3750 rpm for 10 minutes to separate the solid and liquid phases. Isotopically labelled PPCPs ($\approx$3.125 µg of each compound) as surrogate standards were then added to both the separated liquid and solid phases and allowed ample mixing time (several hours). The experimental method described above was used to determine the partition of PPCPs on the mixture of biosolids and PS-WAS.

The partition coefficient (K_d) in various biosolids samples was calculated using Equation 1.

$$K_d \left(\frac{L}{kg}\right) = \frac{\frac{C_S}{TSS}}{C_w} \times 10^6 \; (Ternes, 2007) \qquad (1)$$

Where C_s is the concentration of the PPCPs in solid phase (ng/L), TSS is total suspended solids (mg/L), and C_w is the concentration in liquid phase (ng/L).

5.2.4   TAH of PPCPs in deionized water (DI) and biosolids

To achieving the objective, the effect of TAH on the selected PPCPs in municipal biosolids, the TAH also was investigated on DI water as comparison. DI water and biosolids were spiked with the selected PPCPs. The samples were subsequently treated in a lab-scale thermo-alkaline treatment unit provided by Lystek International (Cambridge, ON, Canada) which consisted of the main reactor, steam line, biosolids and alkali injection port and a high-speed shearing blade.

Separately, 2.7 L of DI water and biosolids with 22% (solid content) was spiked with the ILC as surrogate standards to a final concentration of 0.3 µg/mL, and 0.01 M KOH was

added to make the final pH 9.5. Both alkalinized DI water and biosolids samples were spiked with the PPCPs and treated in the preheated TAH unit for 45 minutes and at 75 °C.

High-speed shearing (1800 rpm) was performed at the beginning to help mixing of the biosolids and DI water samples. All equipment were washed with deionized water (DI), followed by methanol and acetone before a new experiment was started in order to minimize the contamination from the previous experiment.

5.2.5   Analysis of PPCPs

Detection and analysis of a trace amount of PPCPs in complex matrix was also an important subobjective of this work. Sludge samples were separated into the liquid and solid phases. Subsequently, liquid samples and solids samples were extracted using acetonitrile and water at pH 2.0 (using HCl) in the ratio of 60:40 and cleaned using HLB cartridge, the schematic of the process is presented in Figure 5.1.
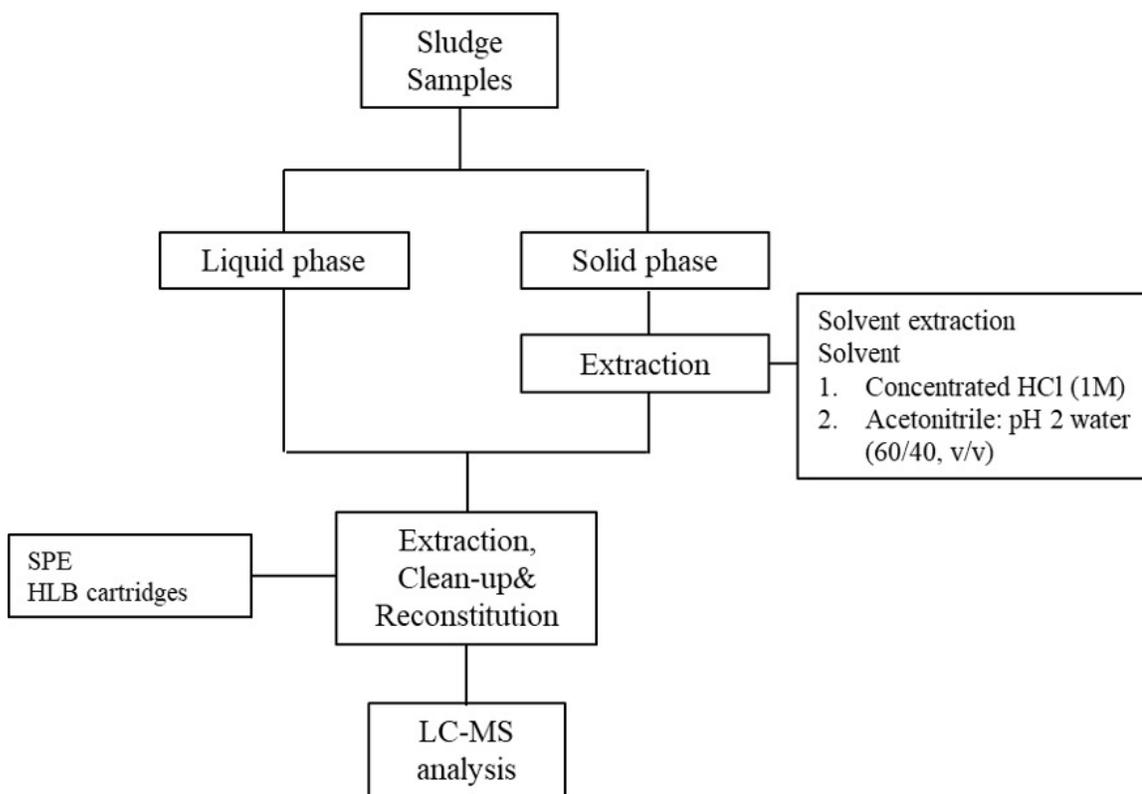
Figure 5.1: Analytical approach for simultaneous extraction of five selected PPCPs in sludge samples

Extraction of PPCPs from liquid phase

Liquid samples were extracted using the solid-phase extraction (SPE) method. The liquid fraction of the sludge samples (12.5 mL) was diluted by adding 20 mL of acidified water (HCl, pH 2) in a 50 mL polypropylene tube followed by centrifugation for 30 s. Hydrophilic- lipophilic balance (HLB) SPE cartridges were activated with 6 mL methanol, 6 mL acetone, and 6 mL methanol successively, followed by conditioning twice with 6 mL of acidic water (pH 2). The diluted samples were passed through the HLB cartridges at a rate of 1-3 mL/s. Subsequently, the HLB cartridges were washed with 3 mL of hexane and air-dried for 5 min. The retained PPCPs were then eluted from the HLB cartridges into polypropylene tubes (15 mL) with 3 additions of 1 mL methanol/acetone (50:50). The sample extracts were evaporated under a gentle stream of air and reconstituted in 1 mL methanol. Extracts were filtered through a 0.22 µm PTFE filter into 2 mL amber vials and stored at -4 °C for LC-MS analysis.

Extraction of PPCPs from the solid phase

The solid fraction of biosolids was first air-dried at room temperature and ground to fine particles using a mortar and pestle, and a mechanical laboratory blender. About 250 mg of the solid was transferred into 15 mL polypropylene conical tubes, followed by the addition of 5 mL of 60:40 (v/v) acetonitrile/water (pH 2). The final pH of the mixed solution was adjusted to 2.0 by adding 500 µL 1M HCl. The resulting solution was vortexed and shaken for 30 minutes and centrifuged at 3750 rpm for 10 min and the supernatant was decanted into a clean polypropylene tube (50 mL). A second extraction was conducted with the remaining solid fraction adding 5 mL of 50:50(v/v) acetonitrile/acetone, followed by vortexing and shaking for 3750 rpm for 10 min. The liquid extracts from successive extractions were brought up to a final volume of 20 mL with acidic water (pH 2), and then air dried for 30 minutes to a volume of 12 mL for centrifugation. The liquid extracts were cleaned up using HLB SPE cartridges prior to the LC-MS analysis.

5.2.6   LC-MS/MS Analyses

The target compounds were analyzed using a Thermo Q-Exactive Orbitrap mass spectrometer coupled to an Agilent 1290 HPLC. Chromatographic separation was performed using an Agilent Zorbax EclipsePlus RRHD C18 column with an injection volume of 5 µL for each sample at 35 °C. The mobile phase consisted of 0.1% formic acid in water solution (A) and 0.1% formic acid in acetonitrile (B) using a flow rate at 0.3 mL min^{-1}. The ion trap mass spectrometer was used with a heated electro-spray ionization source (HESI), with capillary temperature of 400 °C; sheath gas, 17 arbitrary units; auxiliary gas, 8 units; probe heater temperature, 450 °C; S-Lens rf level, 45%; and capillary voltage, 3.9 kV. All experiments and analysis were conducted triplicated, and the standard deviation were shown in error bar.

5.3   Results and discussion

### 5.3.1   Establish and verification of the extraction method

Recovery of the liquid phase and solid-phase extraction

As can be seen in Table 3.1, mostly basic in nature, the PPCPs studied are of different physico-chemical characteristics with wide range of K_{ow}, multiple pK_a values, and different functional groups. Hence, significant combinations of solvents with different compositions were tried to establish optimal extraction of all the five selected PPCPs simultaneously. As pH affects the ionization form of the antibiotics, both acidic and alkaline conditions were investigated for maximum recovery of the analytes (Semreen et al., 2019). As shown in Figure 4.3, it can be seen that extraction recovery (Re %) at pH 2 is much better than at pH 10 for all selected micropollutants except TCS, while TCC and TCS couldn't be extracted efficiently by considering the polarity of solvent, with relatively lower recovery at 8% and 57%, respectively. Therefore, pH 2 is considered for further extractions as most compounds could be extracted at this pH and various extraction solvents also are taken into account. Both TCC and TCS are relatively non-polar with high octanol-water coefficients and show strong affinity to HLB cartridges. Thereafter, hexane or hexane and dichloromethane (DCM) (sequentially) were used to improve recovery of TCC and TCS before eluting with methanol and acetone (50:50), and the results are presented in Figure 5.2. Cleaning up with

hexane produced better recovery of TCC and TCS than DCM, followed by hexane. Furthermore, additional clean-up is necessary to decrease the matrix effect for LC-MS analysis and increase the reproducibility of measurements.
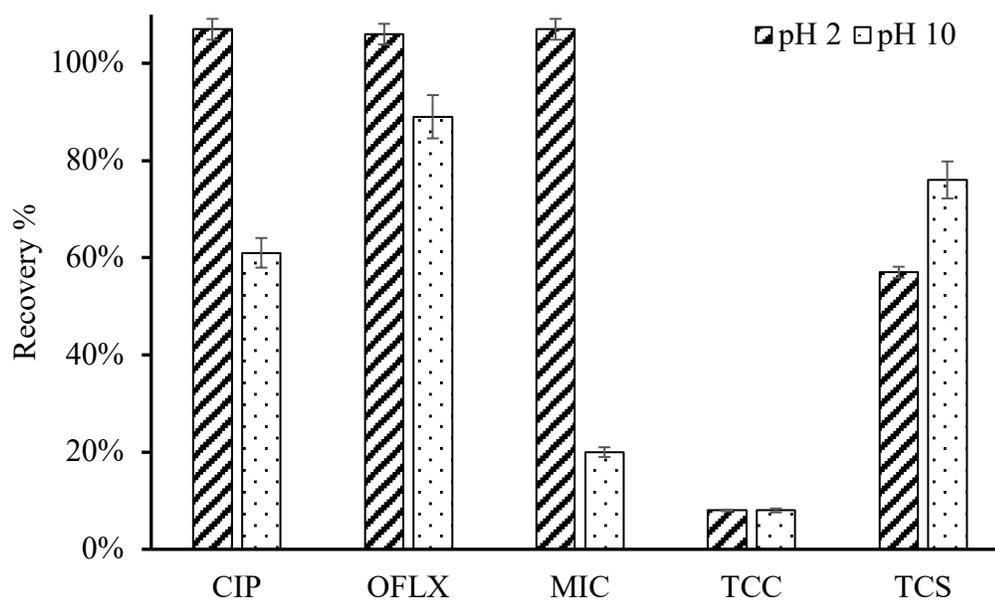
Figure 5.2: Effect of pH on five selected micropollutants in the liquid phase extraction
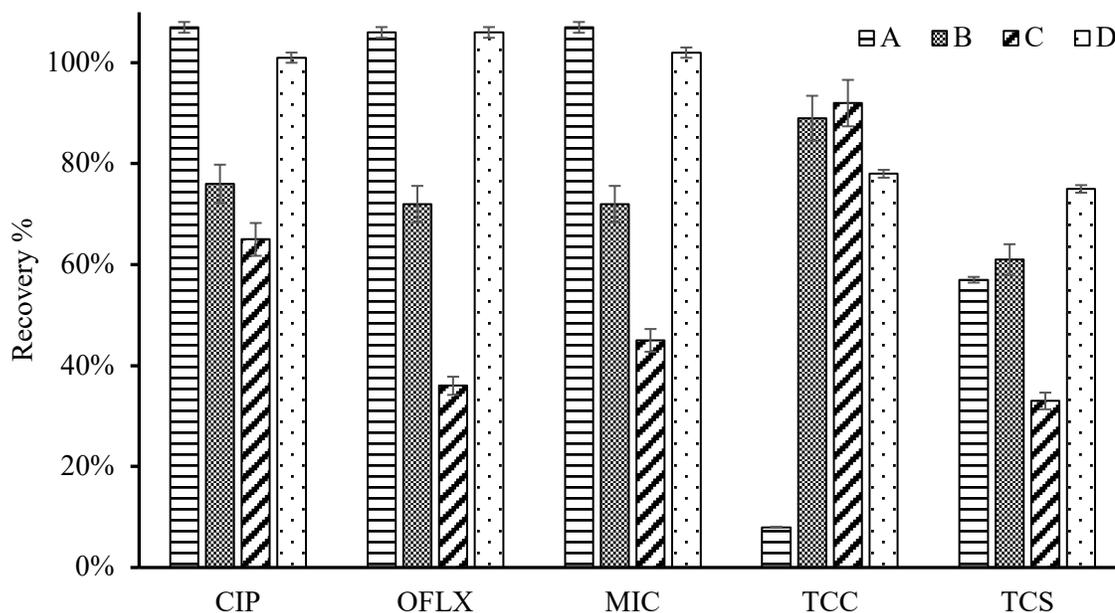
Figure 5.3: Effect of solvent and clean-up of five selected micropollutants on liquid phase extraction recovery

A: activated and eluted with methanol, without clean-up step;

B: activated and eluted with methanol and acetone, without clean-up step;

C: activated and eluted with methanol and acetone, clean-up step with hexane and DCM;

D: activated and eluted with methanol and acetone, clean-up step with hexane.

The extraction of PPCPs from the solid phase is more complex than the liquid phase and has rarely been documented (Abril et al., 2018). Targeted PPCPs were extracted from solid using acetonitrile and pH 2.0 water. The d-SPE (QuEchERS) method was also carried out for extraction due to its simplicity and easy operation. However, preliminary experiments indicated that compared to d-SPE, SPE extraction method was better, although needed optimization. SPE method and d-SPE (QuEchERS) method for analyzing the five micropollutants in sludge samples were used. As shown in Figure 5.3, both methods developed for five micropollutants were not successfully applied to the analysis of sludge samples. Despite MIC, TCC and TCS showing high recovery, CIP and OFLX (R% <10%) were considered unacceptable. Ultimately, an optimized SPE protocol was developed by

adjusting pH (500 µL 1M HCl) below 3 in the first extraction and removing organic extraction buffer by air dry for 1 h. Afterwards, adding water at pH 2 to reach a total volume of 20 mL, the diluted samples were passed through the HLB cartridge and then reconstituted, filtered through a 0.22 µm PTFE filter, transferred into a 2 mL amber vial and stored at -4 °C until LC-MS analysis.

The simultaneous liquid/solid phase extraction method was optimized to recover the five selected compounds of micropollutants from sludge samples. It can be seen in Figure 5.4 that the recovery of CIP, OFLX and MIC from the liquid phase was close to 100% and showed slightly lower recovery from the solid phase. Lowest recovery of 41% occurred for CIP, followed by 61% for OFLX from the solid phase, probably due to matrix complexity, and also due to their strong partition to the biosolids and sludge. Further optimization of the analytical protocol was not conducted as the concentration of CIP and OFLX could be revised using these recovery values. Overall, the single-column extraction method was suitable for optimum extraction of the selected PPCPs in both liquid and solid phases.
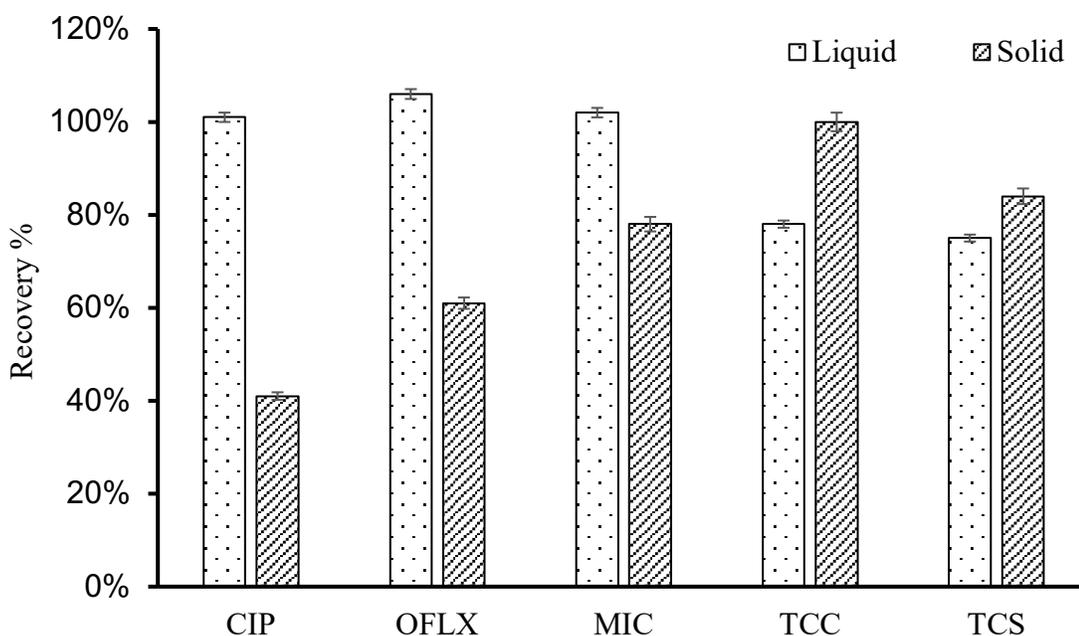
Figure 5.4: Optimized recoveries of five PPCPs in liquid and solid phase extractions

LC-MS/MS analyses

ILC was used as internal injection standards or isotope dilution quantitation standards to correct residual matrix effects during sample preparation and LC-MS/MS analysis, improving the quality of data quality. ILC generally improves average recovery and relative standard deviation (Hao et al., 2008).

The high-resolution Q-Exactive Orbitrap does not have actual baseline noise level in data generation. Therefore, the instrumental limits of detection (LOD) and quantification (LOQ) cannot be calculated using the traditional signal to noise method. The LOD of the selected compounds was evaluated on the basis of 5 times signal response of 5 times detection (N= 5/5), and LOQ was determined by lowest concentration detected with the relative standard deviation (RSD %) less than 25% (Morrison et al., 2018). The LOD and LOQ ranged from 0.5-2 ng/mL and 5-10 ng/mL, respectively, with a correlation coefficient greater than 0.99. The method detection limits, indicative of concentrations that can be observed within sample matrices were determined for each matrix with unique recovery efficiency (Re %) and signal suppression/enhancement (SSE %). Retention time and analysis criteria of select analytes are presented in Table 5.3.
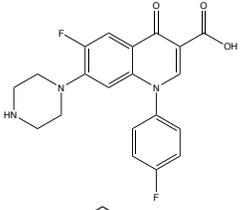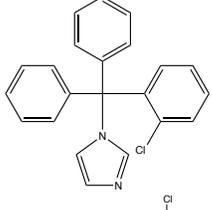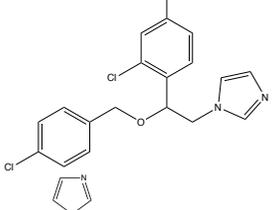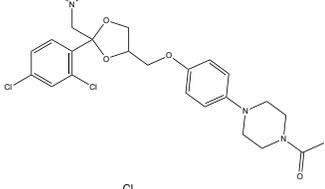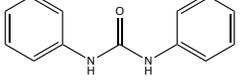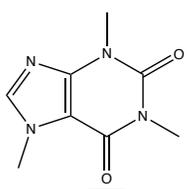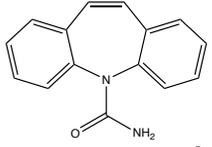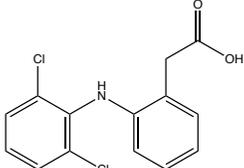
Table 5.3: LC-MS characteristics of the selected PPCPs

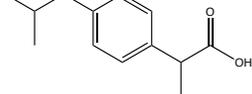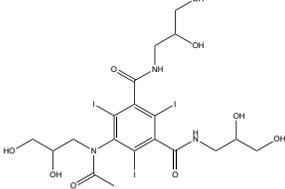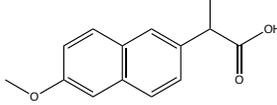| Compound	Method	LOD (ng/mL)	LOQ (ng/mL)
Ciprofloxacin	[M+H]:332.14099 MS/MS:288.15121@hcd33 RT:2.89	0.5	10
Ofloxacin	[M+H]:362.15000 MS/MS:318.16107@hcd32 RT:2.83	0.5	5
Miconazole	[M+H]:414.99390 MS/MS:158.97711@hcd35 RT:5.34	0.5	10
Triclosan	[M-H]:286.94250 RT:7.33	2	10
Triclocarban	[M-H]:312.96942 MS/MS:158.97073@hcd15 RT:7.26	0.5	0.5

In summary, simultaneous detection and quantification of five PPCPs from sludge and biosolids samples were achieved using a simple SPE method and analysis using LC-MS/MS. Recoveries, LOD and LOQ for the characterization protocol were satisfactory, indicating this extraction and analysis method can be applied for environmental samples. A list of compounds for which the optimized analytical method potentially can be used for detection and quantification in environmental samples is provided in Table 5.4. The PPCPs examined in this work represent a wide range of properties such as log K_{ow} and pKa, which dictate the hydrophobicity and dissociation of the compounds in water, respectively. It is envisaged that the analytical methods developed in this work will be applicable to other similar PPCPs such as fluoroquinolones antibiotics, azole antifungals and transformation products of triclosan and triclocarban (Table 5.4). The procedure was optimized to achieve satisfactory recoveries, LOD and LOQ for simultaneous detection of trace contaminants in environmental samples.

Table 5.4: The PPCPs with similar range of pKa and log K_{ow} as the test PPCPs, structure and detected environment concentration listed

Group	Compound	Structure	pKa	log K_{ow}	Environment concentration (ng/L)
Fluoroquinolone's antibiotics	Difloxacin (DIF)		pKa1: 5.64; pKa2: 6.45	0.89	n.d. -20.6 (He & Blaney, 2015)
	Enrofloxacin (ENR)		pKa1: 5.69; pKa2: 6.68	0.89	n.d.-15.6 (He & Blaney, 2015)
	Fleroxacin (FLE)		pKa1: 5.44; pKa2: 6.06	0.47	8.5-506 (He & Blaney, 2015)
	Gatifloxacin (GAT)		pKa1: 5.69; pKa2: 8.73	-0.58	n.d. (He & Blaney, 2015)
	Lomefloxacin (LOM)		pKa1: 5.64; pKa2: 8.70	-0.39	n.d. (He & Blaney, 2015)
	Moxifloxacin (MOX)		pKa1: 5.69; pKa2: 9.42	-0.50	n.d.-81.5 (He & Blaney, 2015)
	Norfloxacin (NOR)		pKa1: 5.77; pKa2: 8.68	-0.92	n.d.-86.8 (He & Blaney, 2015)
	Orbifloxacin (ORB)		pKa1: 5.49; pKa2: 8.77	0.25	n.d. (He & Blaney, 2015)

|					
	Sarafloxacin (SAR)		pKa1: 5.74; pKa2: 8.68	0.56	n.d. (He & Blaney, 2015)
Azole antifungals	Clotrimazole (CLO)		pKa: 6.62	4.1	n.d.-110 (Kahle et al., 2008)
	Econazole (ECO)		pKa: 6.77	5.5	n.d.-72.7 (Huang et al., 2012)
	Ketoconazole (KET)		pKa: 6.75	4.34	n.d.-122.9 (Huang et al., 2012)
Transformation products	Methyl triclosan (MeTCS)		-	-	n.d.-354 (Kantiani et al., 2008)
	Carbanilide (NCC)		pKa: 11.53	3.0	1.1-160 (Venkatesan et al., 2012)
Other PPCPs commonly found in WWTPs	Caffeine (CAF)		pKa: 10.4	-0.007	60-114 (Sui et al., 2010)
	Carbamazepine (CAR)		pKa: 7	2.47	156-188 (Clara et al., 2004)
	Diclofenac (DIC)		pKa: 4.2	4.51	359-6897 (H. R. Buser et al., 1998)
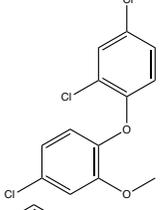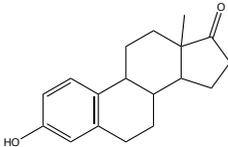
Compound		pKa		Range (Reference)
Estrone (EST)		pKa: 10.3	3.13	1-96 (Servos et al., 2005)
Ibuprofen (IBU)		pKa: 4.9	3.97	241-2724 (H.-R. Buser et al., 1999)
Iohexol (IOH)		pKa: 11.7	-3.05	14432-16008 (Deblonde et al., 2011)
Naproxen (NAP)		pKa: 4.2	3.5	n.d-5513 (Snyder et al., 2004)
Salicylic acid (SAL)		pKa: 2.3	1.13	100-33100 (Martín et al., 2012)
Sulfamethoxazole (SUL)		pKa: 5.7	0.89	117-400 (Snyder et al., 2004)
Trimethoprim (TRI)		pKa: 6.6	1.33	118-150 (Kim et al., 2005)
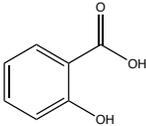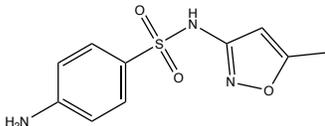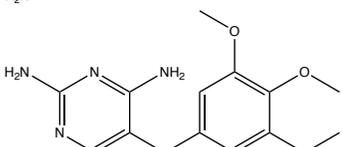
n.d. = not det

5.3.2 Partition of PPCPs in sludge

It is expected that the selected PPCPs with high octanol-water coefficients should partition into the solid phase. In order to separate the effect of background presence of the selected PPCPs, isotopically labelled compounds as surrogate standards were spiked into the sludge and their distributions between the liquid and solid phases of the sludge were determined over 1 hour. The absence of desorption during the experimental time implies that the sorption of the PPCPs in sludge and biosolids is strong. The five PPCPs, all partitioned predominately in the solid phase, ranging from 89% for TCS and 98% for CIP, respectively (Figure 5.5). Typically compounds with a higher octanol-water coefficient partition into solids phase with an organic fraction (f_{OC}), resulting higher solids phase concentration of MIC (log K_{ow}= 6.1), TCC (log K_{ow}= 4.6) and TCS (log K_{ow}= 4.76). However, FQs, which

also demonstrated very high partitioning within the solid phase, have much lower K_{ow} values (log K_{ow} of CIP = 0.28 and OFLX, log K_{ow} -0.39). While K_{ow} is a suitable measure to model partition of a compound in solids with high organic content, another commonly used parameter for estimating organic compounds partition is solid-liquid distribution coefficients (K_d). The reported solid-liquid K_d for FQs (CIP, 1435-4550 L/kg; OFLX, 4976 L/kg) is very high (Tran et al., 2018; Cheng et al., 2018; Tang et al., 2019). Additionally, acid dissociation constant (pKa) can be used to estimate the partition of organics in sludge by determining the different ionization forms of the compounds at environmental pH. In earlier research, the zwitterion of FQs showed strong sorption affinity, followed by cation and anion, and aerobic conditions facilitated FQ sorption (Wang et al., 2017). At pH 7-8, relevant to the wastewater, activated sludge and anaerobic digestion, FQs will be present mostly in their zwitterion form, and therefore will remain adsorbed to sludge.
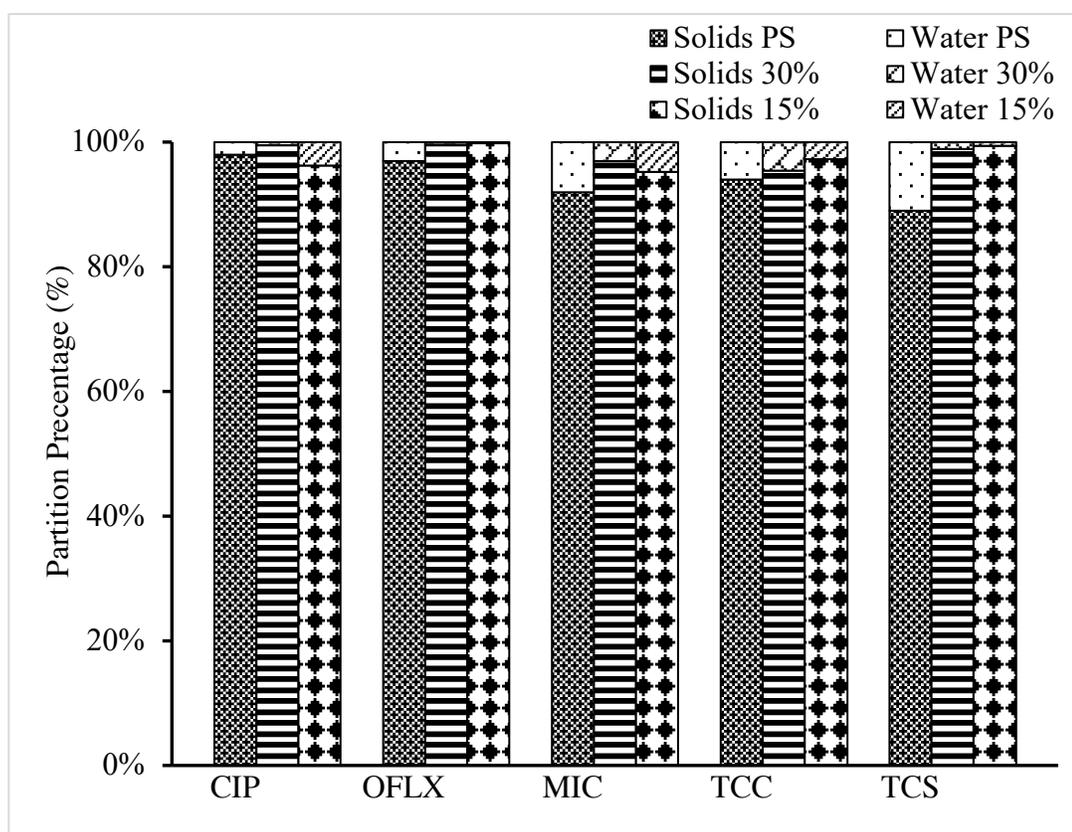
Figure 5.5: Partition of selected PPCPs in primary sludge, 15% :85% (PS-WAS: biosolids) and 30% :70% (PS-WAS: biosolids). (Solids means partition percentage in solids phase and water means in liquid phase)

The log K_d versus K_{ow} for the PPCPs from different literature are plotted in Figure 5.6. Most K_d values determined in this study are located in the interval of the box. The relationship between log K_d and K_{ow} did not follow linear relationship. K_d values are case-specific and the sorption of PPCPs to the solid phase is complex, involving several processes as adsorption, surface complexation, and partitioning (Prasad et al., 2019).

For FQs, higher K_d values were found in biosolids and varied from 1260 L/kg to 11226 L/kg and 1114 L/kg to 15980 L/kg for CIP and OFLX, respectively. The K_d values of MIC in PS and biosolids were 1581 L/kg, 968 L/kg and 1792 L/kg, respectively. The K_d values of TCC ranged from 929 L/kg to 1784 L/kg, while it was 1062-7589 L/kg for TCS. These results are in agreement with some of the reported values for different PPCPs. For example, Narumiya et al. (2013) reported log $K_d > 3$, and more than 90% PPCPs remained in the solid phase. The experimental K_d values of the selected compounds in three biosolids samples varied greatly, indicating sorption behavior substantially affected by the biosolids properties. Nonetheless, relatively high K_d values indicate that these PPCPs are likely to accumulate on both PS and biosolids. Therefore, it is imperative that these compounds need to be removed from biosolids prior to the land application or safe disposal. It requires comprehensive and systematic research on the fate of trace concentration PPCPs in stabilized biosolids such as raw anaerobic digestion and TAH biosolids.
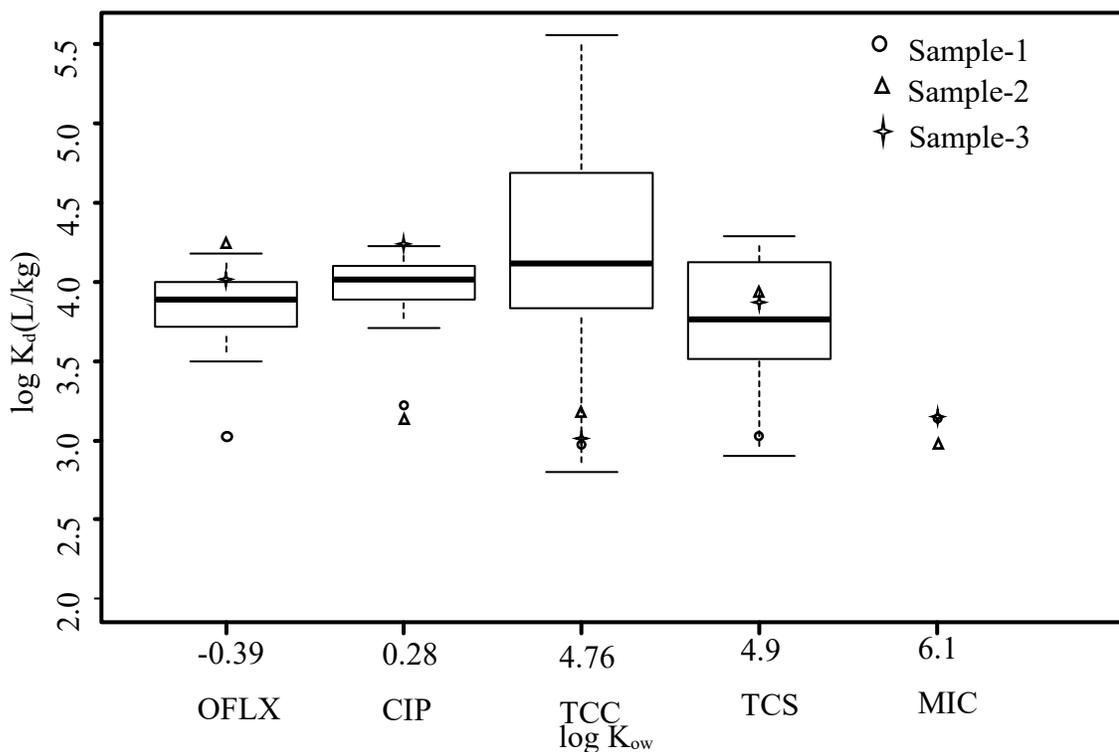
Figure 5.6: Boxplot of log K$_d$ versus log K$_{ow}$ of the selected PPCPs from literature and compared with the data from this study in sludge sample and biosolids (Sample -1: primary sludge, Sample-2: 15% : 85% biosolids : PS-WAS, v/v, and Sample-3: 30% : 70% biosolids

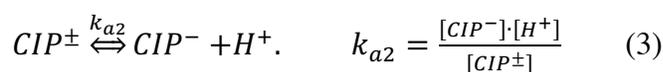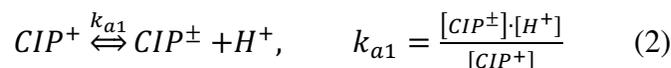## 5.3.3   Effect of TAH on PPCPs in DI water

The effect of TAH on PPCPs in DI water was conducted as a control study, and the results are shown in Figure 5.7. FQs (CIP and OFLX) showed the lowest but significant degradation of 43%, where MIC and TCs were more easily degraded by thermo-alkaline treatment.
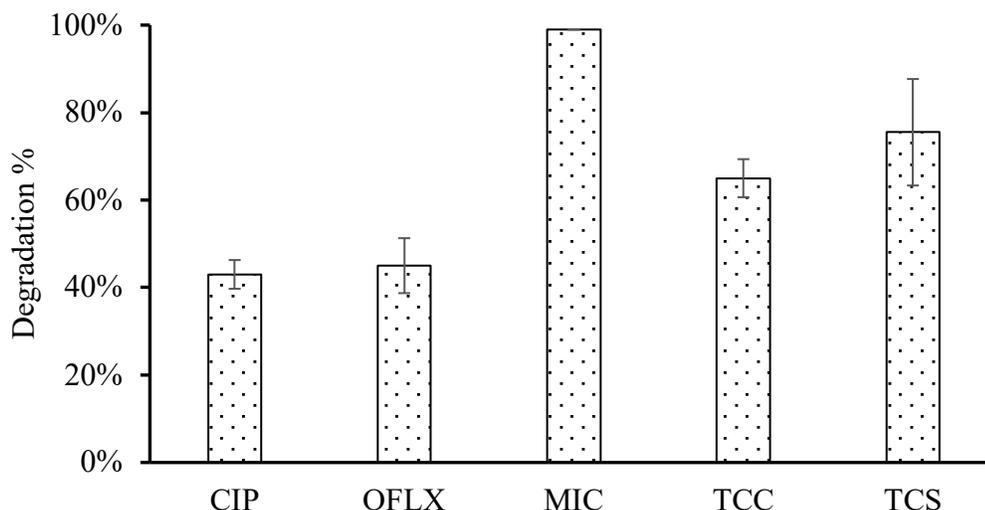
Figure 5.7: Degradation of PPCPs in DI water due to thermo-alkaline treatment

In an earlier study conducted by El-Gamel et al (2011), only 8.5% loss of CIP was reported due to thermal treatment at 40-200 °C. The pH is an important factor affecting the degree of ionization of these compounds. For example, three species (CIP+, CIP±, CIP-) of CIP are formed at different pH (Equation 2-3):

$$CIP^+ \overset{k_{a1}}{\Longleftrightarrow} CIP^\pm + H^+, \qquad k_{a1} = \frac{[CIP^\pm]\cdot[H^+]}{[CIP^+]} \qquad (2)$$

$$CIP^\pm \overset{k_{a2}}{\Longleftrightarrow} CIP^- + H^+. \qquad k_{a2} = \frac{[CIP^-]\cdot[H^+]}{[CIP^\pm]} \qquad (3)$$

The speciation of CIP with respect to pH is shown in Figure 5.8 (a):

$$\alpha_{CIP^+} = \frac{[H^+]^2}{[H^+]^2 + k_{a1}\cdot[H^+] + k_{a1}\cdot k_{a2}}; \qquad (4)$$

$$\alpha_{CIP^\pm} = \frac{k_{a1}\cdot[H^+]}{[H^+]^2 + k_{a1}\cdot[H^+] + k_{a1}\cdot k_{a2}}; \qquad (5)$$

$$\alpha_{CIP^-} = \frac{k_{a1}\cdot k_{a2}}{[H^+]^2 + k_{a1}\cdot[H^+] + k_{a1}\cdot k_{a2}}. \qquad (6)$$

At the alkaline condition (pH=9.5), CIP mainly exists in the anionic form (CIP⁻) and zwitterionic form (CIP±) with $-NH_2^+$ and $-COO^-$ groups. The highly polar functional

groups, such as C–O, C–O–C and COOH can serve as strong electron acceptors and conjugate with π electron-donating groups of N–H and O–H to form π-π electron donor-acceptor system forming a stable structure, which is not amenable to breakage at a moderate temperature of 75°C. Likewise, OFLX belonging to FQs group, shares a similar structure and functional group as CIP. OFLX also is present in anionic and zwitterionic forms as Figure 5.8 (b) shows. The apparent stability of zwitterions is the reason for the lower degradation of FQs.
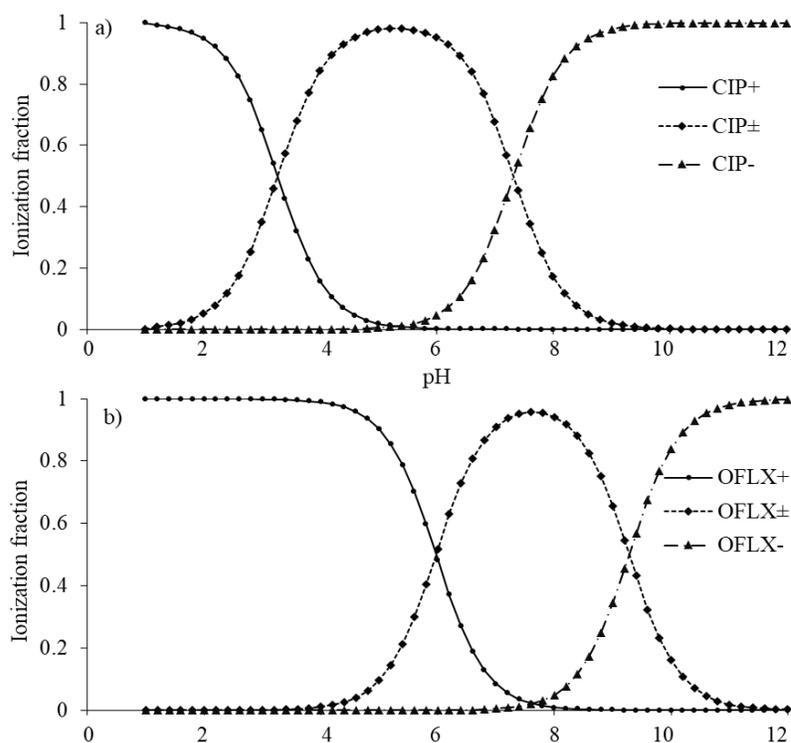
Figure 5.8: Ionization schemes of CIP (a) and OFLX (b)

In addition, pre-evaluation experiments were conducted to investigate the effect of thermal (room temperature and 75°C) and alkaline conditions (pH 6.5 and pH 9.5), separately. The experiments were carried out using 0.909 μg/mL mixture solution of five PPCPs in two 2.5 mL glass tubes adjusted to pH 6.5 and pH 9.5, respectively. About 150 μL spiked solution of pH 6.5 and 9.5 were taken into 250 μL polypropylene vials maintained at room temperature and 75 °C. The samples were taken at different time point: 0 hours, 1 hour, and 3 hours, after treatment. About 0.625 μg/ mL mixture solution of five PPCPs as internal standard was spiked into the polypropylene vials immediately. Afterward, the vials were

seated for 5 min and then analyzed using LC-MS. The results were shown in Table 5.5. Interestingly, no obvious degradation of CIP and OFLX was observed with pH variation and increased temperature, showing only thermal or alkaline treatment is not effective for these PPCPs. However, MIC, TCC and TCS degraded significantly in both thermal and alkaline conditions, almost fully degraded after seven days. In addition, the thermal treatment alone caused higher degradation than alkaline treatment of MIC, TCC and TCS.

Table 5.5: Percentage remaining of selected PPCPs in various thermal and alkaline treatment with time

Condition	Time/ hr	CIP	OFLX	MIC	TCC	TCS
pH 6.5	1	96%	93%	74%	46%	30%
Room Temperature	3	78%	78%	63%	45%	7%
pH 6.5 75°C	1	91%	86%	26%	-	1%
	3	82%	89%	3%	37%	0%
pH 9.5	1	88%	84%	84%	56%	143%
Room Temperature	3	88%	87%	67%	45%	53%
pH 9.5 75°C	1	90%	85%	6%	65%	0%
	3	81%	87%	2%	31%	0%

5.3.4   Effect of TAH on PPCPs in biosolids

To investigate the degradation of the PPCPs in the sludge, biosolids were spiked with ILC and thoroughly mixed, and an aliquot was removed. The remaining biosolids sample was treated in the TAH reactor. The concentration of both the spiked ILC and PPCPs that were naturally present in the untreated biosolids and TAH treated biosolids were determined and

compared. The concentrations of all five naturally occurring compounds before and after following TAH treatment of biosolids are shown in Table 3.6. The concentration of MIC in the biosolids was the highest at $10,382\pm534$ ng/g. TCS was also detected at a relatively high concentration of 6166 ± 532 ng/g. The high co-occurrence ($r=0\sim1$) between MIC and TCS was reported at $r = 0.75$, which indicated that they might have similar behavior in the environment (Zhang et al., 2018); their structural similarity (both have ether and Cl in their structure) also can be seen in Table 5.1. A high amount (4680-10900 ng/g) of TCS was found in biosolids in experimental fields in Canada (Gottschall et al., 2012; Sabourin et al., 2012). CIP belonged to fluoroquinolone antibiotics (FQs) and was detected in biosolids samples at 1835 ± 113 ng/g. CIP has been detected at high concentration of 2759 ng/g in sludge from the WWTP of southern Spain (Krzeminski et al., 2018). OFLX and TCC both were detected at a relatively lower concentration of 148 ± 5 ng/g and 86 ± 9 ng/g, respectively. FQs are detected at higher concentrations due to poor biodegradability during anaerobic digestion (Li et al., 2017). These compounds are recalcitrant to biodegradation and remains in sludge which directly dispersed into agriculture soil and leaching into surface water. Literature showed FQs antibiotics affect mainly microorganism and strongly inhabit microbial activities when they bound to soil (Githinji et al., 2011), and FQs are moderately toxic toward alga when they dispersed into aqueous system (Hernando et al., 2006). TCS and TCC were estimated have the potential risks to freshwater, terrestrial environments, and soil such as antibiotics resistance genes of microbial and endocrine disruption effects or birth effects on human (Musse, 2018). All the compounds tested showed some degree of degradation/removal following the TAH of the biosolids (Figure 5.6). However, significant differences can be seen in the results obtained from the experiments conducted in DI water. Although the degradation of the five PPCPs in DI ranged from 43-99%, the degradation in biosolids by TAH was only 36-41%. Similar to DI, the least amount of degradation occurred for the FQs. The highly polar carbon-fluorine bond is one of the strongest with average bond energy of around 480 kJ/mol, making fluoro organic compounds highly thermally and chemically stable (Pagliaro et al., 2005). In addition, biosolids matrix being considerably more complex, the polar functional groups like -COOH, -CHO and -NH$_2$ of these compounds are likely to interact with the biosolids

(Chen et al., 2013). Nonetheless, TAH treatment could remove the selected PPCPs to some extent.

In addition to the non-labelled PPCPs that were naturally present in the biosolids, the concentrations of ILC, applied directly prior to TAH were measured. As expected, the ILC showed similar behavior as the non-labelled compounds with the exception of CIP and OLFX, where the ILC showed significantly lower degradation in sludge (Figure 5.9). The results indicate that the matrix effect is more significant for the recalcitrant fluorinated compounds.

Table 5.6: Concentrations of selected PPCPs in biosolids before and following TAH treatment (n=3)

	Pre-TAH (ng/g)	Post-TAH (ng/g)
CIP	1835±113	1155±134
OFLX	148±5	93±8
MIC	10382±534	6881±360
TCC	86±9	54±2
TCS	6165±531	3664±132
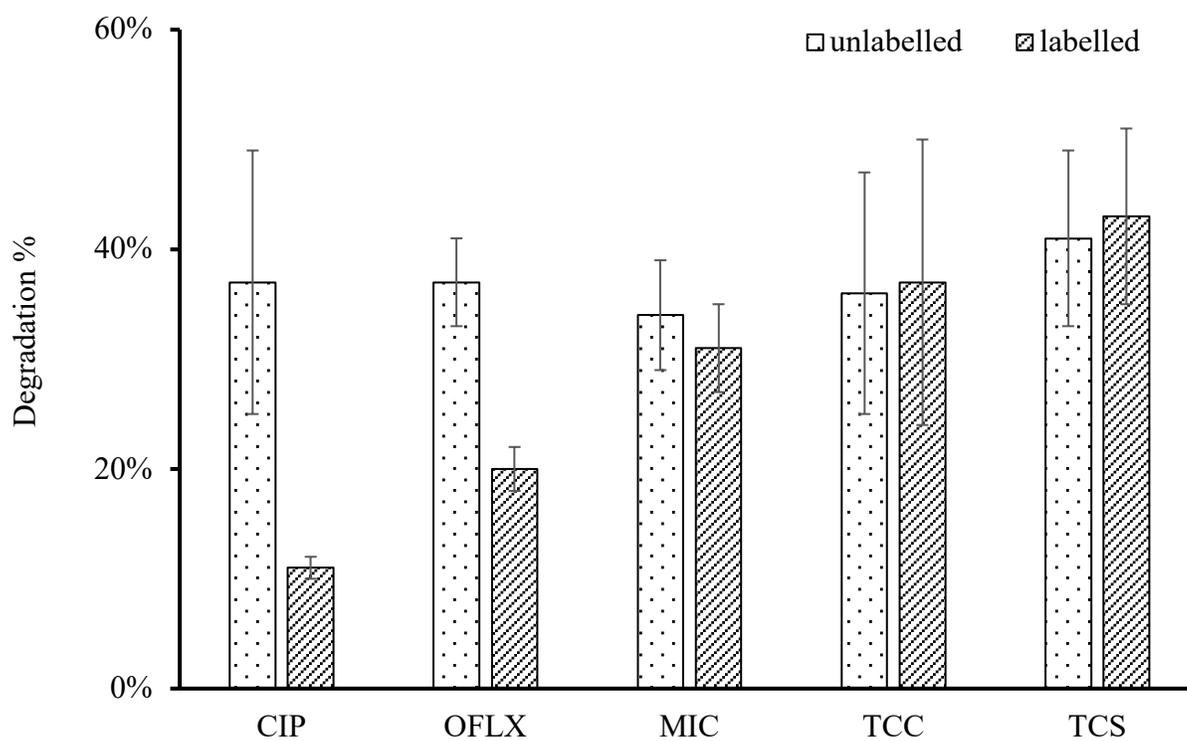
Figure 5.9: Percent degradation of isotopically labelled PPCPs spiked into biosolids and unlabeled PPCPs (naturally occurring in biosolids) before and after TAH treatment

5.4   Conclusion

Optimum extraction methods for simultaneous maximum recovery of five targeted PPCPs from water, municipal sludge and biosolids, and their detection methods using LC-MS were established. Both liquid and solids samples were extracted using acetonitrile and water at pH 2.0 (using HCl) in the ratio of 60:40 and cleaned using HLB cartridge. The selected PPCPs were detected at the concentration range of 54±3 ng/g to 6166±532 ng/g in biosolids collected from two local wastewater treatment plants at Ontario, Canada. A proprietary TAH (pH 9.5, 75ºC, 45 min), which is used for post-treatment of biosolids for better stabilization was evaluated in this work for the removal of the target PPCPs.   The average removal of target PPCPs from sludge due to TAH was around 40%, where fluoroquinolone compounds showed significantly lower degradation due to their structural stability. This study indicated that TAH is an effective process to remove micropollutants in biosolids, which is an additional advantage when TAH is used as a post-treatment method for greater stabilization of biosolids. Further research is being conducted to investigate the ability of TAH to enhance biodegradation of micropollutants in sludge.

5.5 References

Abril, C., Santos, J. L., Malvar, J. L., Martín, J., Aparicio, I., & Alonso, E. 2018. Determination of perfluorinated compounds, bisphenol A, anionic surfactants and personal care products in digested sludge, compost and soil by liquid-chromatography-tandem mass spectrometry. Journal of Chromatography A, 1576: 34-41. https://doi.org/10.1016/j.chroma.2018.09.028

Ahmad, M., Abbott, T., Cimon, C., Abel-Denee, M., & Eskicioglu, C. 2019. Post treatment of anaerobically digested municipal sludge for enhanced ammonia, odors, coliforms, and micropollutants removal. IWA Publishing, Chapter 11. https://doi.org/10.2166/9781780409740_0153

Ashfaq, M., Li, Y., Rehman, M. S. U., Zubair, M., Mustafa, G., Nazar, M. F., Yu C.P., & Sun, Q. 2019. Occurrence, spatial variation and risk assessment of pharmaceuticals and personal care products in urban wastewater, canal surface water, and their sediments: A case study of Lahore, Pakistan. Science of The Total Environment, 668: 653-663: https://doi.org/10.1016/j.scitotenv.2019.06.285

Carrere, H., Antonopoulou, G., Affes, R., Passos, F., Battimelli, A., Lyberatos, G., & Ferrer, I. (2016). Review of feedstock pretreatment strategies for improved anaerobic digestion: from lab-scale research to full-scale application. Bioresource Technology, 199: 386-397. https://doi.org/10.1016/j.biortech.2015.09.007

Cheng, Dengmiao, Yao Feng, Yuanwang Liu, Jinpeng Li, Jianming Xue, and Zhaojun Li. 2018. Quantitative models for predicting adsorption of oxytetracycline, ciprofloxacin and sulfamerazine to swine manures with contrasting properties, Science of The Total Environment, 634: 1148-56 https://doi.org/10.1016/j.scitotenv.2018.04.114.

Chen, Y., Yu, G., Cao, Q., Zhang, H., Lin, Q., & Hong, Y. 2013. Occurrence and environmental implications of pharmaceuticals in Chinese municipal sewage sludge. Chemosphere, 93(9): 1765-1772:https://doi.org/10.1016/j.chemosphere.2013.06.007

Das, Sreejon, Nillohit Mitra Ray, Jing Wan, Adnan Khan, Tulip Chakraborty, and Madhumita B Ray. 2017. Micropollutants in wastewater: fate and removal processes, Physico-Chemical Wastewater Treatment and Resource Recovery: 75: http://doi.org/10.5772/65644.

Elbeshbishy, Elsayed, Saad Aldin, George Nakhla, Ajay Singh, and Bill Mullin. 2014. Impact of alkaline-hydrolyzed biosolids (Lystek) addition on the anaerobic digestibility of TWAS in lab–And full-scale anaerobic digesters, Waste Management, 34: 2090-97: http://doi.org/10.1016/j.wasman.2014.07.022.

El-Gamel, N. E., Hawash, M. F., & Fahmey, M. A. 2011. Structure characterization and spectroscopic investigation of ciprofloxacin drug. Journal of Thermal Analysis and Calorimetry, 108(1):253-262: https://doi.org/10.1007/s10973-011-1584-8.

Githinji, L. J. M., Musey, M. K., & Ankumah, R. O. 2011. Evaluation of the Fate of Ciprofloxacin and Amoxicillin in Domestic Wastewater,Water, Air, & Soil Pollution, 219(1): 191–201:https://doi.org/10.1007/s11270-010-0697-1.

Gottschall, N, E Topp, C Metcalfe, M Edwards, M Payne, S Kleywegt, P Russell, and DR Lapen. 2012. Pharmaceutical and personal care products in groundwater, subsurface drainage, soil, and wheat grain, following a high single application of municipal biosolids to a field, Chemosphere, 87: 194-203:https://doi.org/10.1016/j.chemosphere.2011.12.018.

Guerra, P., Teslic, S., Shah, A., Albert, A., Gewurtz, S. B., & Smyth, S. A. 2019. Occurrence and removal of triclosan in Canadian wastewater systems. Environmental Science and Pollution Research, 26(31): 31873-31886.https://doi.org/10.1007/s11356-019-06338-w

Hao, C., Zhao, X., Tabe, S., & Yang, P. 2008. Optimization of a multiresidual method for the determination of waterborne emerging organic pollutants using solid-phase extraction and liquid chromatography/tandem mass spectrometry and isotope

dilution mass spectrometry. Environmental Science & Technology, 42(11): 4068-4075. https://doi.org/10.1021/es7028125

Hernando, M. D., Mezcua, M., Fernández-Alba, A. R., & Barceló, D. 2006. Environmental risk assessment of pharmaceutical residues in wastewater effluents, surface waters and sediments, Talanta, 69(2): 334–342. https://doi.org/https://doi.org/10.1016/j.talanta.2005.09.037

Huang, Q., Zhang, J., Xiong, S., Peng, X., Wei, G., Liu, L., Sun, X., & Li, L. 2018. Development of ultrasound-assisted extraction of commonly used azole antifungals in soils. Analytical Methods, 10(44): 5265-5272. https://doi.org/10.1039/C8AY01585F

Khadra, A., Ezzariai, A., Merlina, G., Capdeville, M. J., Budzinski, H., Hamdi, H., Pinelli, E., & Hafidi, M. 2019. Fate of antibiotics present in a primary sludge of WWTP during their co-composting with palm wastes. Waste Management, 84: 13-19. https://doi.org/10.1016/j.wasman.2018.11.009

Kodešová, Radka, Aleš Klement, Oksana Golovko, Miroslav Fér, Martin Kočárek, Antonín Nikodem, and Roman Grabic. 2019. Soil influences on uptake and transfer of pharmaceuticals from sewage sludge amended soils to spinach, Journal of Environmental Management, 250: 109407:

https://doi.org/10.1016/j.jenvman.2019.109407

Kosma, C. I., Lambropoulou, D. A., & Albanis, T. A. 2010. Occurrence and removal of PPCPs in municipal and hospital wastewaters in Greece. Journal of Hazardous Materials, 179: 804-817: https://doi.org/10.1016/j.jhazmat.2010.03.075

Krzeminski, Pawel, Maria Concetta Tomei, Popi Karaolia, Alette Langenhoff, C Marisa R Almeida, Ewa Felis, Fanny Gritten, Henrik Rasmus Andersen, Telma Fernandez, and Celia Manaia. 2018. Performance of secondary wastewater treatment methods for the removal of contaminants of emerging concern implicated in crop uptake and antibiotic resistance spread: A review, Science of The Total Environment:

https://doi.org/10.1016/j.scitotenv.2018.08.130.

Li, H., Zou, S., Li, C., & Jin, Y. 2013. Alkaline post-treatment for improved sludge anaerobic digestion. Bioresource Technology, 140: 187-191: https://doi.org/10.1016/j.biortech.2013.04.093

Li, Ning, Huajie Liu, Yonggang Xue, Hongyang Wang, and Xiaohu Dai. 2017. Partition and fate analysis of fluoroquinolones in sewage sludge during anaerobic digestion with thermal hydrolysis pretreatment, Science of The Total Environment, 581: 715-21: http://doi.org/10.1016/j.scitotenv.2016.12.188.

Morrison, Lucas M, Justin B Renaud, Lyne Sabourin, Mark W Sumarah, Ken KC Yeung, and David R Lapen. 2018. High-Throughput Quantitation of Neonicotinoids in Lyophilized Surface Water by LC-APCI-MS/MS, Journal of AOAC International, 101: 1940-47: https://doi.org/10.5740/jaoacint.17-0486.

Musee, N. 2018. Environmental risk assessment of triclosan and triclocarban from personal care products in South Africa, Environmental Pollution, 242: 827–838:https://doi.org/https://doi.org/10.1016/j.envpol.2018.06.106

Narumiya, M., Nakada, N., Yamashita, N., & Tanaka, H. 2013. Phase distribution and removal of pharmaceuticals and personal care products during anaerobic sludge digestion. Journal of hazardous materials, 260: 305-312:https://doi.org/10.1016/j.jhazmat.2013.05.032

Pagliaro, M., & Ciriminna, R. 2005. New fluorinated functional materials. Journal of Materials Chemistry, 15(47): 4981-4991: https://doi.org/10.1039/b507583c

Prasad, M. N. V., Vithanage, M., & Kapley, A. (Eds.). 2019. Pharmaceuticals and Personal Care Products: Waste Management and Treatment Technology: Emerging Contaminants and Micro Pollutants. Butterworth-Heinemann An imprint of Elsevier. Chapter 15.

Sabourin, Lyne, Peter Duenk, Shelly Bonte-Gelok, Michael Payne, David R Lapen, and Edward Topp. 2012. Uptake of pharmaceuticals, hormones and parabens into

vegetables grown in soil fertilized with municipal biosolids, Science of The Total Environment, 431: 233-36: https://doi.org/10.1016/j.scitotenv.2012.05.017.

Salveson, A., Rauch-Williams, T., Dickenson, E., Drewes, J., Drury, D., McAvoy, D., & Snyder, S. 2012. Trace organic compound indicator removal during conventional wastewater treatment. Alexandria, IWA Publishing.

Sherburne, Jessica J, Amanda M Anaya, Kim J Fernie, Jennifer S Forbey, Edward T Furlong, Dana W Kolpin, Alfred M Dufty, and Chad A Kinney. 2016. Occurrence of triclocarban and triclosan in an agro-ecosystem following application of biosolids, Environmental Science & Technology, 50: 13206-14. https://doi.org/10.1021/acs.est.6b01834

Semreen, M. H., Shanableh, A., Semerjian, L., Alniss, H., Mousa, M., Bai, X., & Acharya, K. 2019. Simultaneous determination of pharmaceuticals by solid-phase extraction and liquid chromatography-tandem mass spectrometry: A case study from Sharjah sewage treatment plant. Molecules, 24(3): 633. https://doi.org/10.3390/molecules24030633

Tang, Jinpeng, Sai Wang, Jingjing Fan, Shengxin Long, Lin Wang, Chen Tang, Nora Fungyee Tam, and Yang Yang. 2019. Predicting distribution coefficients for antibiotics in a river water–sediment using quantitative models based on their spatiotemporal variations, Science of The Total Environment, 655: 1301-10: https://doi.org/10.1016/j.scitotenv.2018.11.163.

Tessier, Judith. 2017. Biosolids Management and Disposal Practices in Canada: 9.

Torri, Silvana Irene, and Marisol Natalia Cabrera. 2017. The environmental impact of biosolids land application, Organic waste: management strategies, environmental impact and emerging regulations: 185-208, Mark Collins (Editor), Nova Science Publishers.

Tran, Ngoc Han, Martin Reinhard, and Karina Yew-Hoong Gin. 2018. Occurrence and fate of emerging contaminants in municipal wastewater treatment plants from

different geographical regions-a review, Water Research, 133: 182-207: https://doi.org/10.1016/j.watres.2017.12.029.

Xia, K., Bhandari, A., Das, K., & Pillar, G. (2005). Occurrence and fate of pharmaceuticals and personal care products (PPCPs) in biosolids. Journal of Environmental Quality, 34: 91-104: https://doi.org/10.2134/jeq2005.0091

Yang, Yun-Ya, Gurpal S Toor, and Clinton F Williams. 2015. Pharmaceuticals and organochlorine pesticides in sediments of an urban river in Florida, USA, Journal of Soils and Sediments, 15: 993-1004: https://doi.org/10.1007/s11368-015-1077-7.

Zhang, Xiangyu, and Ruying Li. 2018. Variation of antibiotics in sludge pretreatment and anaerobic digestion processes: Degradation and solid-liquid distribution, Bioresource Technology, 255: 266-72: https://doi.org/10.1016/j.biortech.2018.01.100.

Zhou, Li-Jun, Guang-Guo Ying, Jian-Liang Zhao, Ji-Feng Yang, Li Wang, Bin Yang, and Shan Liu. 2011. Trends in the occurrence of human and veterinary antibiotics in the sediments of the Yellow River, Hai River and Liao River in northern China, Environmental Pollution, 159: 1877-85: http://doi.org/10.1016/j.envpol.2011.03.034.

<div align="center">Chapter 6</div>

6 pH-dependence molar absorptivity of selected micropollutants and effect on UV photolysis

6.1 Introduction

Wastewater is the most significant point source of discharging micropollutants to freshwater bodies. Driven by water scarcity, treating municipal wastewater for beneficial reuse is becoming a cost-effective alternative (Bilińska et al., 2019; Caicedo et al., 2019). The regulations and limit levels of pollutants in wastewater are significant concerns in reuse applications (Kellis et al., 2013; Rizzo et al., 2020). The presence of micropollutants, including pharmaceuticals and personal care products (PPCPs), pesticide and endocrine modulating compounds in environment, is receiving significant attention due to their potential threat to human and ecosystem health (Ashraf, 2017). In recent years, research shows micropollutants' physiological effects on aquatic organisms even at trace concentrations ranging from ng/L to μg/L (Yan et al., 2018). PPCPs are a kind of typical micropollutants that have received greater attention due to their widespread use and recalcitrance in wastewater treatment units (Mao et al., 2020; Yang et al., 2017). Therefore, robust and eco-efficient technologies are urgently needed to remove PPCPs to protect public health and environmental safety.

Generally, typical wastewater treatment plants (WWTP) are not designed to remove PPCPs. After the primary and secondary processes, the treated effluent contains many PPCPs, removal efficiency ranging from 17%-40% and 38%-90%, respectively (de Jesus Gaffney et al., 2017; Yang et al., 2017). Many WWTP uses UV-disinfection as a final step of water treatment to remove pathogens. Additionally, UV treatment is also practiced after membrane processes for tertiary treatment of water for reuse purposes (Cvetnic et al., 2017). UV-254 nm radiation used in the disinfection process can cause photolysis of the PPCPs depending on their molar absorptivity in this wavelength (Carlson et al., 2015). High-energy UV photons excite electronic states of the organic compounds, producing reactive radicals which combine with oxidative species to form degradation products (NAS et al., 2017).

Table 6.1 summarizes the PPCPs degradation efficiencies via UV photolysis published in the literature by various researchers. Multiple literature revealed that different PPCPs have different degradation efficiencies depending on molecular structure and absorption spectrum (Lian et al., 2015). For example, Kim et al. (2009) found that the degradation of theophylline was only 3%, while ketoprofen removed by 90%. It is indicated that direct UV photolysis significantly degraded certain species of PPCPs, such as amoxicillin, sulfamethoxazole, sulfamonomethoxine. The main reason attributed to higher degradation efficiencies is the relatively high molar extinction coefficients. Molar absorptivity is a crucial parameter in evaluating the rate of a photochemical reaction of the compound, which could be used for designing or controlling UV system operation for potable reuse of wastewater. However, the molar absorptivity is very limited by the pH of PPCPs based on molecular structure, especially for some basic and acidic functional groups (Cai et al., 2020; Fang et al., 2013). Therefore, it is of great importance to study the effect of pH variations on molar absorptivity of some commonly found PPCPs of different molecular properties.

Table 6.1: A literature review of PPCPs degradation efficiencies by UV photolysis

Compound	Sample type	Degradation (%)	Reference
amoxicilline	water	>90	(Hoehne et al., n.d.)
carbamazepine	Milli-Q water	25	(Alharbi et al., 2017)
chloramphenicol	deionized water	25	(Dong et al., 2017)
ciprofloxacin	wastewater effluent	48	(De la Cruz et al., 2012)
diclofenac, sulfamethoxazole	Milli-Q water	>90	(Alharbi et al., 2017)
ibuprofen	wastewater effluent	34	(De la Cruz et al., 2012)
ketoprofen	pure water	90	(Kim et al., 2009)
sulfathiazole	distilled water	26	(Yun et al., 2018)
tetracycline	distilled water	35	(Yun et al., 2018)
theophylline	pure water	3	(Kim et al., 2009)

The objective of this work was to determine the pH dependence molar absorptivity of selected micropollutants such as industrial byproducts, pharmaceuticals, and pesticides from wastewater treated for potable reuse. Firstly, the pH-dependent molar absorptivity of the 12 micropollutants was determined at different pH (pH 5.0, pH 6.0, pH 7.0, and pH 8.0). Then, the relationship between molar extinction coefficient (ε) and quantum yield (QY) with pH and acid dissociation constant (pKa) was investigated using a lab-scale UV collimated beam system. This work provides relevant information for UV disinfection reactor design to remove the selected PPCPs and to predict the characteristics of similar functional group compounds for controlling the UV photolysis process.

6.2   Material and method

6.2.1   Chemicals

The pharmaceuticals used in this study include: (1) antipyrine (AP, analytical standard), (2) sulfathiazole (STZ, ≥98%), (3) sulfamethoxazole (SMX, analytical standard), (4) diclofenac(DFC, analytical standard), (5)fluoxetine (FLX, pharmaceutical secondary standard), (6) hydrochlorothiazide (HTZ, crystalline), (7) amoxicillin (AMX, ≥900 μg per mg), (8) doxycycline (DXC, ≥98%), (9) chlortetracycline (CTC, analytical standard), (10) ciprofloxacin (CFX, ≥98%), (11) chloramphenicol (CPC, ≥98%) and (12) sulfisoxazole (SXZ, analytical standard) were obtained from Sigma-Aldrich, Canada. The chemical ($KHPO_4$; ≥98%) used to make up buffer solutions, was obtained from EMD Millipore, Canada, while KH_2PO_4 (≥99.3%) was obtained from Fisher Scientific, Canada.

6.2.2   Determination of molar extinction coefficients in different pH

The pH values were measured using a pH meter (Thermo Scientific Orion Star A111) calibrated with solutions of pH 4.0, 7.0, and 10.0. Stock solutions of PPCPs at pH 5.0, 6.0, 7.0 and 8.0 were prepared by dissolving them in phosphate buffers (10 mM). Different mass ratios of monobasic and dibasic potassium phosphate (Table 6.2) were used to prepare a 10 mM buffer with the pH range from 5.0-8.0 solution according to the Henderson–Hasselbalch equation (Equation 6.1).

$$pH=pKa+\log\frac{B_{base}}{A_{acid}} \qquad\qquad (6.1)$$

Table 6.2: mass ratios of monobasic and dibasic potassium phosphate (pH 5.0, pH 6.0, pH 7.0, pH 8.0)

pH	KHPO$_4$ (mL)	KH$_2$PO4 (mL)	Milli-Q (mL)
5	0.14	9.86	990
6	1.3	8.7	990
7	4.2	5.8	990
8	9.32	0.68	990

A UV-vis spectrophotometer (UV-3600, Shimadzu) was used to analyze the molar absorbance of selected PPCPs in the wavelength range of 200-800 nm. The molar absorbance was calculated using the Beer-Lambert equation (Equation 6.2).

$$A = \varepsilon l C \ or \ \varepsilon = A/(l \times C) \qquad\qquad (6.2)$$

where A is the absorbance, l is path length of the sample (1 cm), C is concentration of sample (mol·L^{-1}), ε is molar absorptivity or molar extinction coefficient (L•mol^{-1}•cm^{-1}) and is a measure of the probability of the electronic transition.

6.2.3   Analytical methods

The quantifications of the PPCPs were monitored by a high-performance liquid chromatography (HPLC) system (Dionex ICS-3000, Thermo Scientific, USA), with an Agilent C18 column at a UV wavelength of 260 and 276 nm. The mobile phase was a mixture of ammonium acetate (40 mM) and acetonitrile in the ratio of 40/60 (v/v). The flow rate was set at 0.4 - 0.6 mL min^{-1}, and the injection volume was 100 μL. The column temperature was set at 25 °C with 3 min running sampling time. The details of HPLC conditions for targeted PPCPs are listed in Table 6.3.

**Table 6.3: Parameters of targeted PPCPs in HPLC**

	SMX	DCF	AP	FLX
Column	C-18			
Mobile phase	ammonium acetate (40mM) and acetonitrile in the ratio of 40/60 (v/v)			
Flow rate	0.6 mL/min	0.4 mL/min	0.6 mL/min	0.4 mL/min
Injection volume	100 μL			
Detection wavelength	260 nm	276nm	260 nm	260 nm
Retention time	1.33min	1.47min	1.40min	1.21 min
Run time	3min	3min	3min	3min

The TOC of UV degradation samples was measured using a Shimadzu TOC-L analyzer (CPH TOC, Shimadzu Scientific Instruments Ltd., Japan), calibrated with a standard glucose solution to obtain the limit of detection (LOD) at 0.1 mg/L. TOC is a water quality parameter that is commonly applied in water treatment processes. The TOC is the sum of the dissolved and particulate organic carbon, of which the inorganic carbon is removed via acidification.

6.2.4 UV collimated beam experiment

UV photodegradation of selected PPCPs was carried out in a UV collimated beam apparatus, established by Darby et al. (Darby, 1995). The configuration of a lab-scale collimated beam system is described in Figure 6.1. The system is constructed with an LP-UV lamp (20.4 W) in a horizontal copper pipe. A vertical collimating tube extends downward from the middle of the lamp pipe to attain the collimation of the UV light. 50 mL PPCPs sample is placed into a sterilized petri dish (internal diameter 5.6 cm) containing a stir bar to keep a distance to the UV lamp at 34 cm. The Petri-factor (PF) is a correction factor to reflect the ratio of the incident irradiance at the center of the sample surface to the incident irradiance over the sample surface. Bolton and Linden suggest that PF values

should be higher than 0.9 to achieve a well-designed collimated beam system. The average PF was consistently calculated around 0.94 before and after these collimated beam experiments, achieving the recommended value.
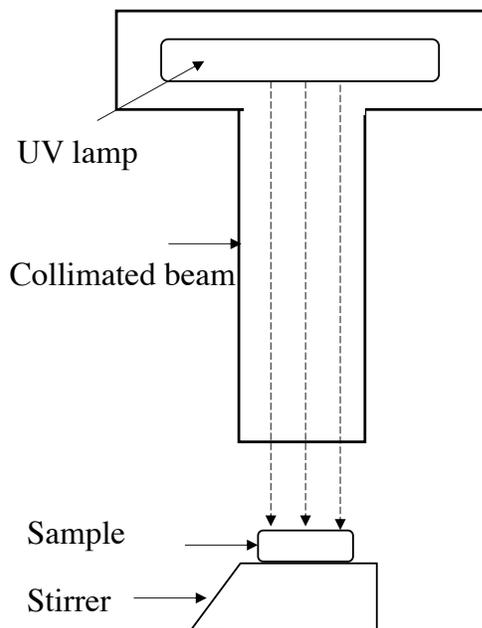
Figure 6.1: Typical UV collimated beam system

Four PPCPs (antipyrine/AP,diclofenac/DFC,fluoxetine/FLX, and sulfamethoxazole/SMX) were prepared with phosphate buffer at pH 5.0, 6.0, 7.0 and 8.0. Initial concentrations of the four PPCPs with different pH were individually kept at 5 ppm. Six Petri-dishes at some conditions were prepared for different degradation times (10 min, 30 min, 60 min, 90 min, 120 min) to obtain different UV fluences. Samples are collected after those time intervals of irradiation and analyzed via HPLC and TOC for degradation efficiency of the PPCPs. All experiments and analysis were conducted triplicated, and the standard deviation were shown in error bar.

6.2.5   Determination of photodegradation kinetics

The photodegradation of selected PPCPs was evaluated considering a pseudo-first-order kinetics. The kinetic constants (k) were determined according to Equation 6.3

$$\ln\left(\frac{C}{C_0}\right) = -kt \qquad\qquad (6.3)$$

where C is the final PPCPs concentration (μg/L), C_0 is the initial PPCPs concentration (μg/L), and t is the UV irradiation time (min).

Quantum yield is usually defined as the moles of a chemical change per Einstein of photon energy absorbed, which was calculated using Equations 6.4 (Bolton & Stefan, 2002). In this work, the fluence-based pseudo-first-order rate constants, k′ (cm^2 mJ^{-1}), are obtained from the slope of ln (C/C$_0$) vs. UV dose plot.

$$\phi_C = \frac{k' * U_\lambda}{\ln(10) * \varepsilon_C} \qquad\qquad (6.4)$$

where U_λ is the molar photon energy given in unit of J Einstein^{-1} at the irradiation wavelength λ (254 nm), which equals to 471527.7 J/Einstein, ε_C is the molar extinction coefficient, which has the unit of M^{-1} cm^{-1} for the targeted compound at the irradiation wavelength (254 nm), and ϕ_c is the quantum yield for the targeted compound.

6.3 Results and discussion

6.3.1 Determination of molar extinction coefficients at varying pH

Measurement of 12 PPCPs molar extinction coefficient at 254 nm in different pH shows that pH influences molar extinction coefficient (Figure 6.2). The experimental results imply that ciprofloxacin (CFX) and sulfisoxazole (SXZ) are more pH-sensitive than other compounds in terms of molar absorptivity at pH 8.0. Sulfisoxazole (SXZ) is a much higher molar absorptivity at pH 5.0, pH 6.0 and pH 7.0 compared with other compounds. Amoxicillin (AMX) and fluoxetine (FLX) have the lowest molar absorptivity among those micropollutants samples.
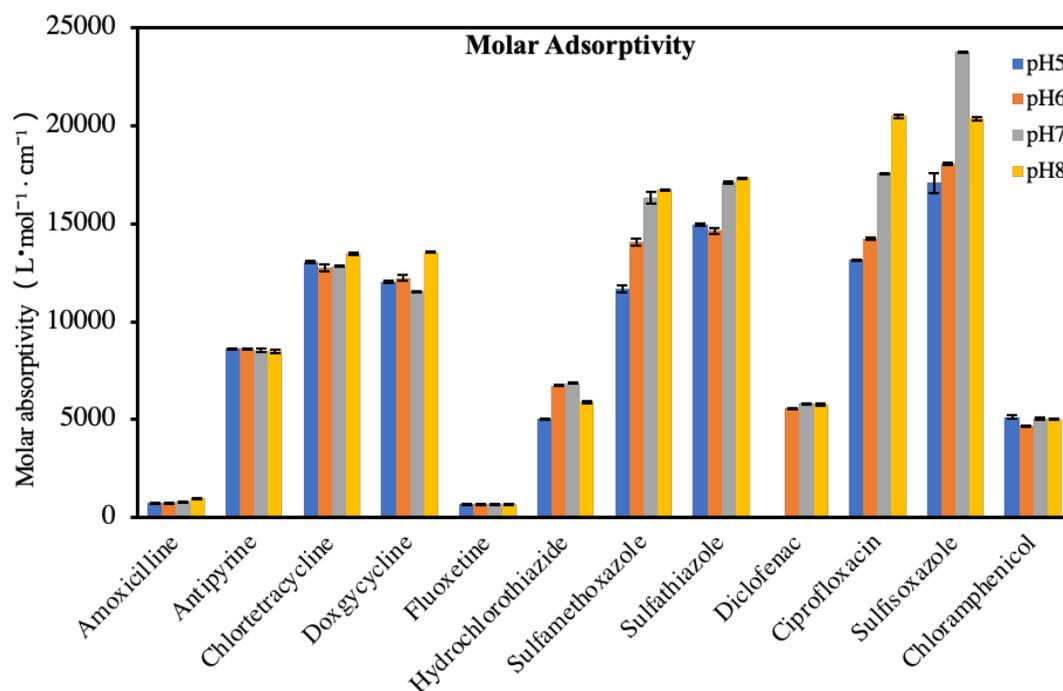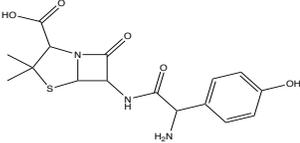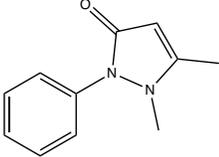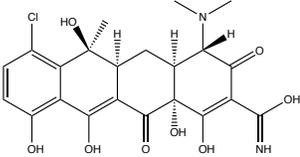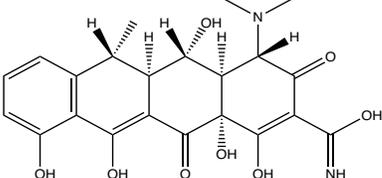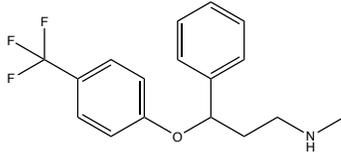
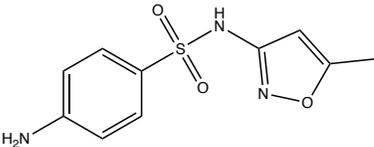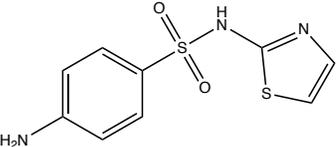**Figure 6.2: Molar extinction coefficient of 12 selected PPCPs at pH 5.0; pH 6.0; pH 7.0; pH 8.0**
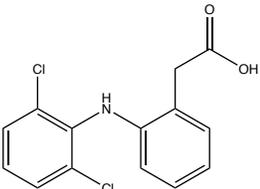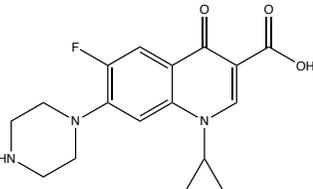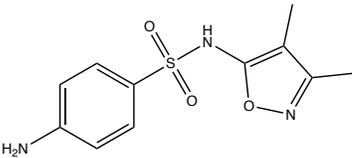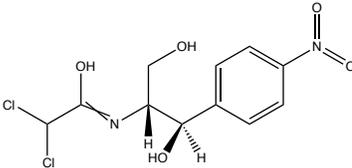
The molar extinction coefficients of 12 compounds have different performance towards pH changes due to their chemical structure and pKa value (Table 6.4). The molar absorptivity of AP, DFC, and FLX was not sensitive to pH changes with less than 2.5 % in the target pH range. The pKa values for AP, DFC and FLX are all at the pH range at 5-8, and are, respectively 1.45, 4.15 and 10.1. As pH increased, the ionization form kept stable for these compounds, resulting in the non-sensitive to pH behavior. For tetracycline-class drug CTC and DFC, the molar extinction coefficient decreased until pH 7 then increased in the range of 12751 to 13450 and 11535 to 13529 M^{-1} cm^{-1}, respectively. Tetracycline group compounds are amphoteric molecules with three pKa values for conjugated tricarbonyl (pKa 1: 2.8-3.4), conjugated phenolic diketone (pKa 2: 7.2-7.8), and dimethylamine (pKa 2: 9.1-9.8). Different ionic species will predominate as a function of the pH (Wei et al., 2019). Hence, the increase in the monoanionic CTC and DFC fraction can explain the results. The molar absorptivity of sulfinol group SMX, STZ and SXZ increased from 11668 to 16712, 14939 to 17313, and 17091 to 23764 M^{-1} cm^{-1}, respectively, with increasing pH 5 to 8. For the sulfa-drugs, with increasing pH, the sulfinol group is

deprotonated to release the lone-pair electrons of its nitrogen atom, thus resulting in a hyperchromic effect on the absorption peaks (Lian et al., 2015). With the increase in pH above the second pKa value, sulfa-drugs mainly exist in anionic form. In the anionic form, the sulfinol-nitrogen atom obtains two lone-pairs of electrons and achieves p-π conjugation with sulfinol and penta-heterocycle groups. The two lone-pairs of electrons produce strong repulsion and increase the energy difference between HOMO (highest occupied molecular orbital) and LUMO (lowest unoccupied molecular orbital), which results in a blue-shift in the UV–Vis absorption spectra and higher absorbance for the anions (Lian et al., 2015).

Table 6.4: Chemical structure and reported pKa values of 12 PPCPs

Compound	Structure	pKa	Reference for pKa
AMX		2.4; 7.4; 9.6	(Rolinson, 1974)
AP		1.45	(Baeza & Knappe, 2011)
CTC		3.57; 7.49; 9.88	(Shaojun et al., 2008)
DC		3.50; 7.07; 9.13	(Bolobajev et al., 2016)
FLX		10.1	(Nakamura et al., 2008)
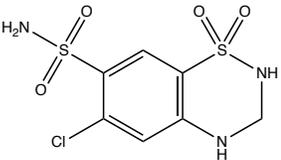
HCT		7.9; 9.2	(Real et al., 2010)
SMX		1.85, 5.6	(Qiang & Adams, 2004)
STZ		2.2, 7.2	(Boreen et al., 2004)
DFC		4.15	(Hung et al., 2001)
CFX		5.9, 8.89	(Peng et al., 2015)
SXZ		1.5, 5	(Boreen et al., 2004)
CPC		11.03	(Qiang & Adams, 2004)

Other compounds such AMX, CPF, CPC, and HCT belong to various class drugs; the molar extinction coefficient has changed around their pKa value when their pKa value is located in the range of pH 5-8. Therefore, knowledge of molar absorptivity in UV-254 and the pKa

value of functional groups through structure-activity relationship (SAR) are the intimate understanding of characteristics of selected probe compounds. The analysis of SAR enables determining the chemical groups sensitive to UV-254 wavelength in different pH. It is also revealed how the relationship between the molar absorptivity and the pH and at what wavelength each pH absorbed best. The results imply that specific micropollutants are removed efficiently at a particular pH; therefore, controlling pH during wastewater treatment may be required to remove micropollutants from wastewater by photolysis processes. Moreover, potential models can be developed to predict the characteristics of similar functional group compounds for controlling photolysis.

6.3.2   Behavior of absorption scans at varying pH

The data from these scans are likely to be useful both for understanding the potential for photolysis using alternative sources of radiation (medium-pressure Hg lamp, excimers, UV LEDs, solar radiation), but also for understanding the roles of various functional groups in defining absorption behavior. The behavior of absorption scans from 200 nm to 800 nm at varying pH for 12 target micropollutants were illustrated by UV-vis spectrophotometer in Figure. 6.3a. Most of these compounds have absorption peaks from 200 nm to 400 nm, which are well overlapped with the solar irradiation in the range of 290 nm to 400 nm (Cheng et al., 2019).

As shown in Figure. 6.3, AP, DFC, FLX have maximum absorbance in the UV range at 244 to 256 nm, 274 to 278 nm, and 261 to 265 nm, respectively. Also, pH did not affect light absorption of these three compounds, consistent with their non-sensitive performance at 254 nm wavelength. CTC and DC are two typical tetracycline group antibiotics, which showed two absorption peaks around 275 nm and 370 nm. The second absorption peaks also well overlapped with the solar irradiation wavelength range. These results agree with the previous report on tetracycline-class drugs (Wei et al., 2019). Moreover, the characteristic peaks of CTC and DC have a slight right shift with higher pH values (Figure 6.3 b). A similar redshift phenomenon has been reported in a recent publication due to protonation or deprotonation under pH changes (Xu et al., 2020). It should be noted that pH could strongly affect the hydroxyl bond (-OH) bound to the phenolic diketone functional group of CTC and DC. Protonation or deprotonation affects the spectrum shift

both in molar absorbance intensities and absorption peaks, resulting in the difference in quantum yields and direct photolysis rate constants at various pH.
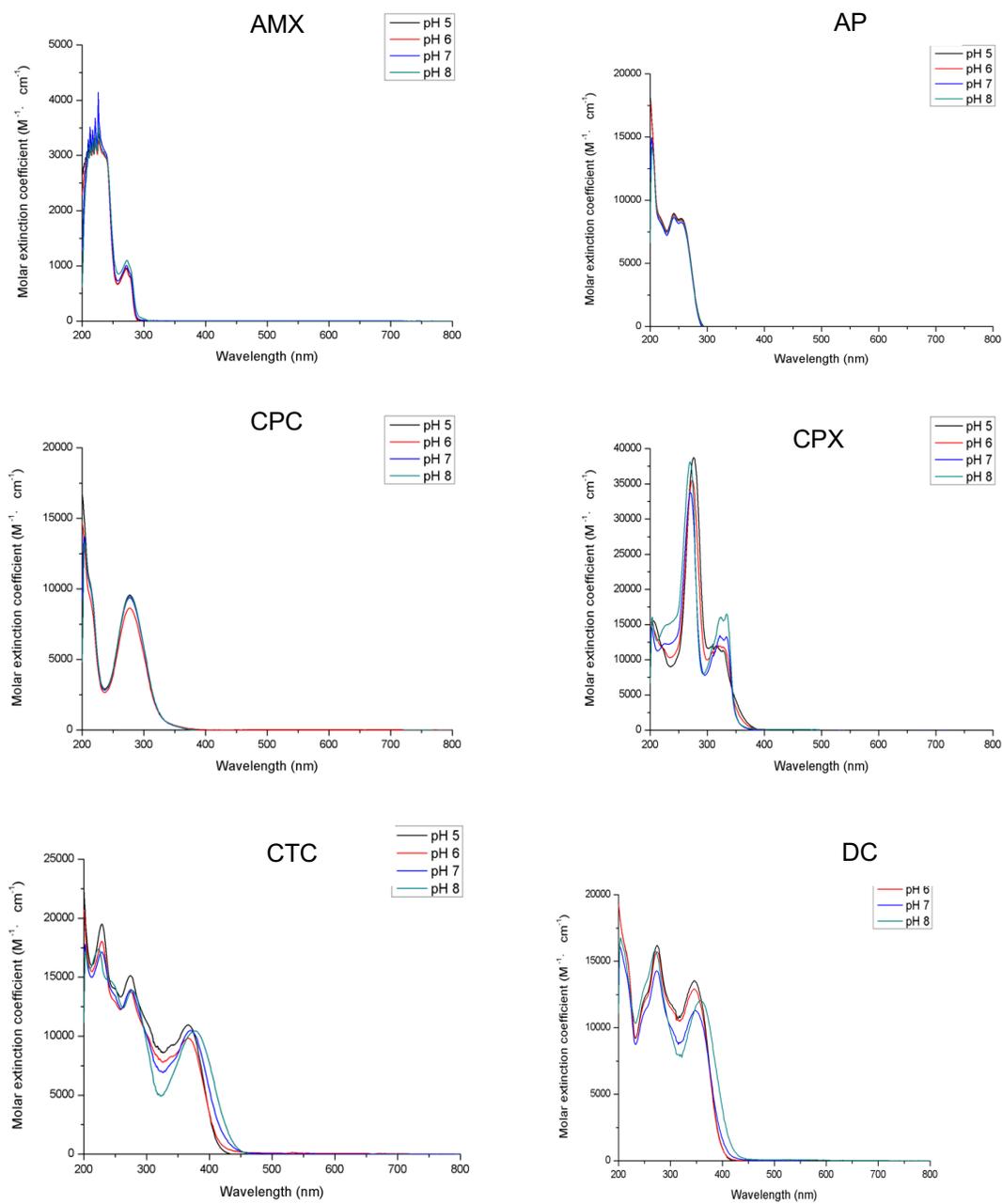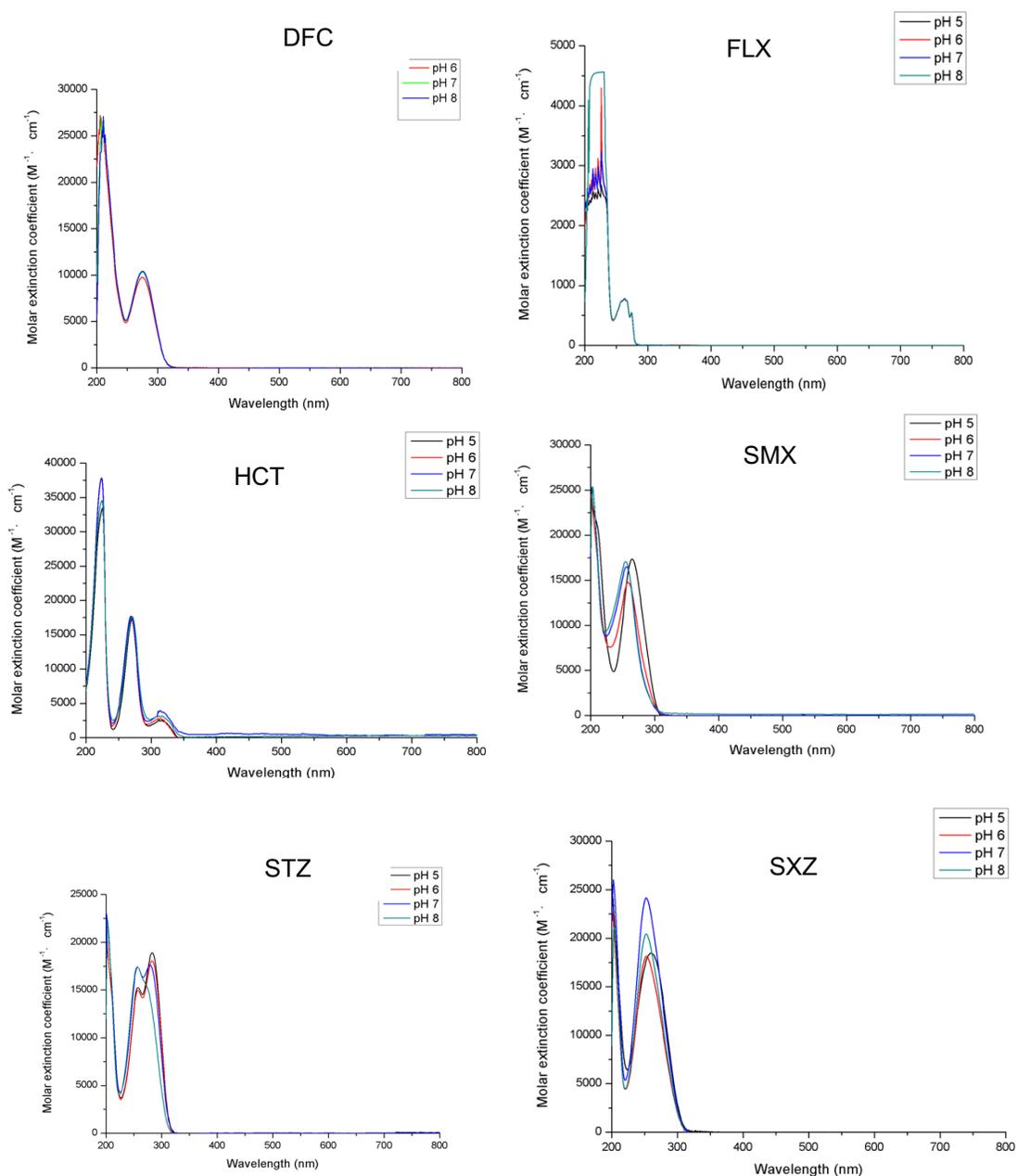
Figure 6.3 a): UV spectra of 12 micropollutants at certain concentration at pH 5.0-8.0; (AMX 400 mg/L; AP 15 mg/L; CPC 40 mg/L; CPX 24 mg/L; CTC 32 mg/L;DC 32 mg/L; DFC 40 mg/L; FLX 400 mg/L; HCT 8 mg/L; SMX 12 mg/L; STZ 12 mg/L; SXZ 12 mg/L)
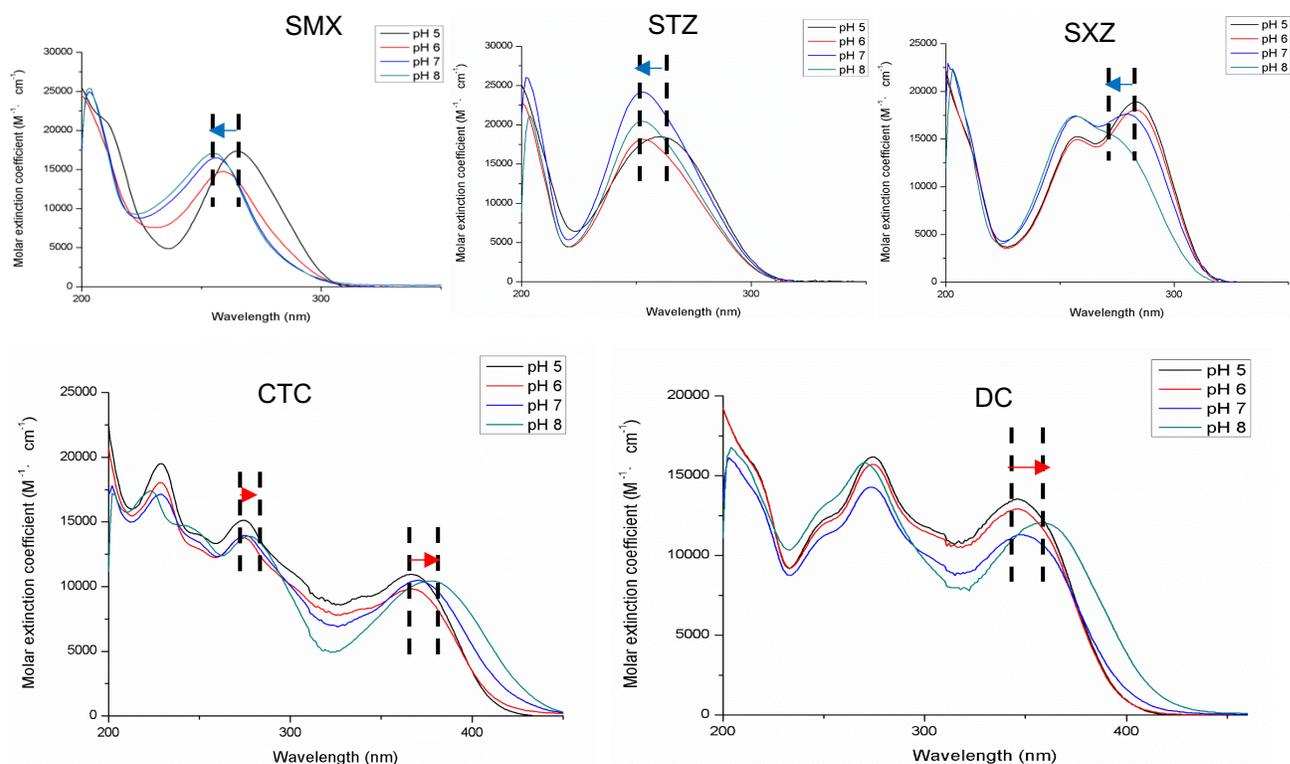
**Figure 6.3 b):UV spectra shift for tetracycline and sulfinol group compound
at pH 5.0-8.0**

The differences in sulfonyl group in both the molar absorptivity and positions of the absorption peaks were illustrated. The SMX, STZ, and SXZ have one maximum absorption peak around the wavelength range from 256 to 265 nm, 284 to 288 nm, and 252 to 261 nm. Only a small wavelength overleap with solar irradiation (290 to 400 nm) can be seen from the molar absorbance spectra, which indicated sulfinyl group drugs could be sensitive to the irradiance UV range. Moreover, the maximum absorption peaks of sulfa group antibiotics have a slightly left shift with higher pH values (Figure 6.3 b). The blue shift phenomenon happened due to the protonation state and molecular orbital (MO) changed with pH variety. With the increasing pH, the molar ratio of the negative and the neutral forms between SMX species increased, and hence the most percentage of SMX is

deprotonated, the electrons of single bond N on SMX⁻ led to this blue shift phenomenon(Luo et al., 2018).

6.3.3   Effect of pH on SMX, AP, DFC and FLX photodegradation

Direct UV photodegradation experiments were conducted at a bench scale with an LP-UV lamp (20.4 W) for different degradation times (10 min to 120 min) to achieve different UV fluences. Four micropollutants SMX, AP, DFC and FLX, are studied at the initial concentration of 5 ppm with varying pH. The UV dose (mJ/cm^2) calculation equation (6.5-6.7) for bench test is (Chen et al., 2006):

$$I_x(\lambda) = I_o(\lambda)e^{-x\alpha(\lambda)} \tag{6.5}$$

$$I_{avg}(\lambda) = \frac{\int_0^D I_o(\lambda)e^{-x\alpha(\lambda)}}{D} = I_o(\lambda)\left[\frac{1-e^{-\alpha(\lambda)D}}{\alpha(\lambda)D}\right] \tag{6.6}$$

$$D = t \times \sum_\lambda I_{avg}(\lambda) \tag{6.7}$$

D = UV dose, mJ/cm^2 , I = UV light intensity in the bulk solution, mW/cm^2 (I_{ave} = average intensity within the suspension, and I_0 = UV intensity measured at the surface of suspension, mW/cm^2), α is the absorbance coefficient (cm^{-1}) at λ (254 nm) wavelength, x is the path length (cm), and t = exposure time, s. The UV dose of all the compounds for different degradation times is presented in Table 6.5.

Table 6.5: UV dose for different compounds degradation at pH 5.0-8.0

Time (min)		10	30	60	90	120
SMX UV dose (mJ/cm²)	pH 5	67.60	202.77	405.54	608.31	915.1
	pH 6	70.68	212.04	424.08	636.117	848.16
	pH 7	65.67	197.01	394.02	591.03	788.04
	pH 8	68.07	204.22	408.44	612.66	816.88
AP UV dose (mJ/cm²)	pH 5	60.8	121.61	182.41	364.82	547.22
	pH 6	58.13	116.26	174.38	348.77	523.15
	pH 7	60.23	120.46	180.69	361.37	542.06
	pH 8	61.34	122.67	184.01	368.02	552.03
DFC UV dose (mJ/cm²)	pH 6	76.67	153.34	230.01	460.02	690.03
	pH 7	76.05	152.1	228.15	456.2	684.3
	pH 8	75.64	151.28	226.93	453.86	924.50
FLX UV dose (mJ/cm²)	pH 5	80.73	242.2	484.39	726.59	968.78
	pH 6	80.28	240.83	481.67	722.5	963.34
	pH 7	80.12	240.35	480.71	721.06	961.41
	pH 8	80.55	241.64	483.29	724.93	966.58

The direct photolysis data of SMX, AP, DFC and FLX at pH 5 to pH 8 were fitted to the pseudo-first–order kinetic model. The degradation kinetics of all the compounds under 254 nm UV irradiation are presented in Figure 6.4.
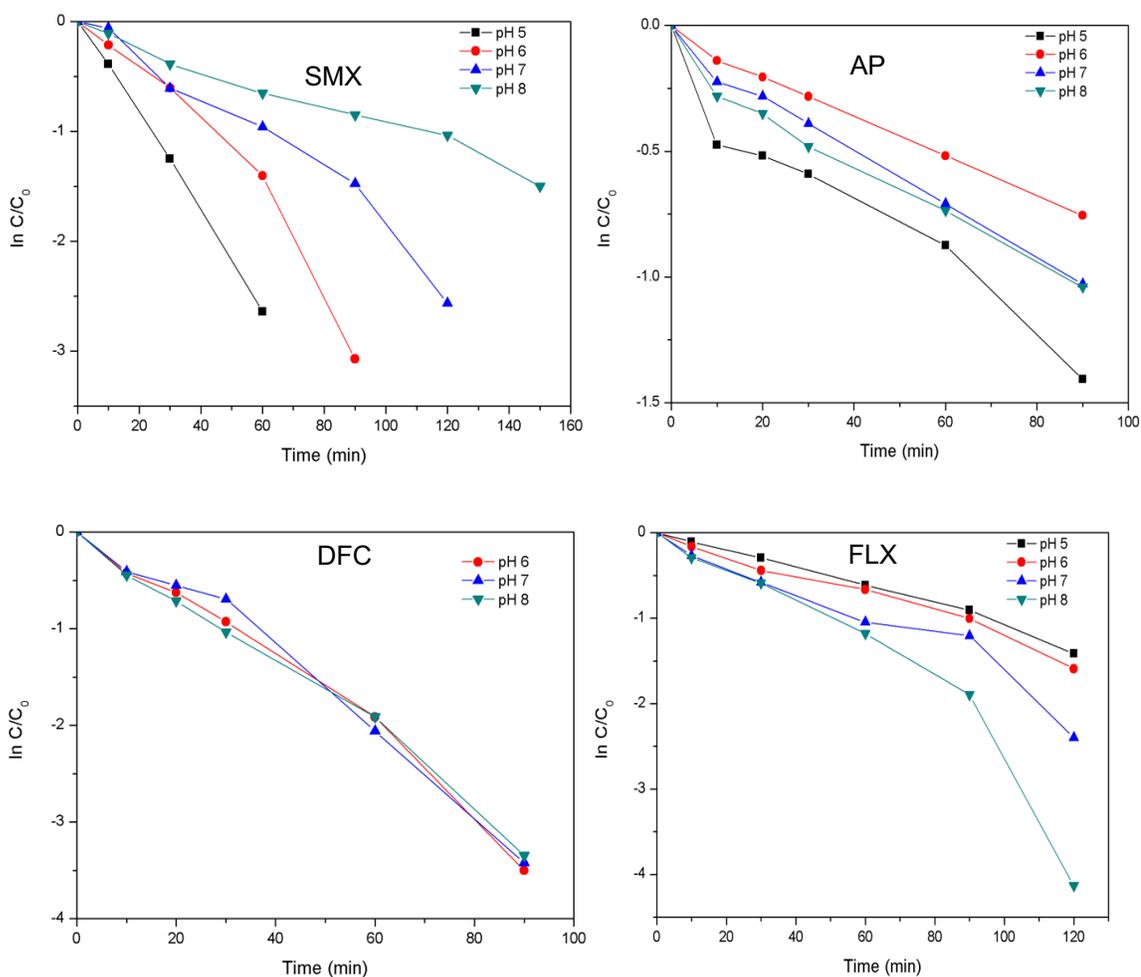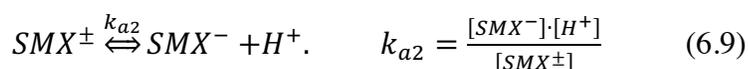
Figure 6.4: Time–dependent UV direct degradation kinetics of SMX, AP, DFC, and FLX (5ppm) at pH 5-8

The pseudo-first-order rate (k') of AP and DFC only have a slight change with pH change at the range of 1.39×10^3 to 1.80×10^3 and 4.71×10^3 to 4.98×10^3 cm^{-2} mJ, respectively (Table 6.6). The UV photolysis rates for SMX decreased with pH increasing from 6.54×10^3 to 1.36×10^3 cm^{-2} mJ. On the contrary, FLX presented an opposite trend; the degradation rate of FLX increased from 1.40×10^3 to 3.81×10^3 cm^{-2} mJ with pH 5 to 8. At pH 5, SMX has the highest rate of UV photolysis, while the rates of both AP and FLX were prolonged. Both SMX and DFC degraded at a faster rate at pH 6 compared with AP and FLX. At pH 7, the photolysis rates of the compounds followed the order DFC > SMX > FLX > AP. When pH increased to 8, the degradation of SMX decreased to the lowest among all compounds, while FLX increased 2.7 times higher than its rate at pH 5.

Table 6.6: Photochemical values (k′ and QY) for SMX, AP, FLX and DFC at pH 5.0-8.0

Compound	k′ (cm^{-2} mJ) × 10^3				QY (mol Einstein^{-1})			
	pH 5	pH 6	pH 7	pH 8	pH 5	pH 6	pH 7	pH 8
SMX	6.54	4.54	3.08	1.36	0.11	0.07	0.04	0.02
AP	1.79	1.39	1.80	1.73	0.04	0.03	0.04	0.04
FLX	1.40	1.53	2.19	3.81	0.42	0.45	0.65	1.13
DFC	-	4.89	4.98	4.71	-	0.18	0.18	0.17

It is interesting to note that SMX decreased QY as the pH increased from 5 to 8. Similar photolysis results for sulfonamide group antibiotics have been reported recently (Wei et al., 2019). The different QY values for SMX again confirm the effect of solution pH on direct UV photolysis by varying the forms of ionized species. Previous research has also demonstrated that various chemical speciation sulfa group drugs affect the photochemistry because each species will have its quantum yield and light absorption properties (Boreen et al., 2004; Werner et al., 2006). Using the pKa values from the literature, the fractions of each ionic species in the mixture for the SMX at different pH were calculated using Equation (6.8-6.9). The compound SMX has two pKa values of 1.85 and 5.6, respectively (Qiang & Adams, 2004). Figure. 6.5 a showed the percentage of SMX species and the fraction contribution of each species with respect to pH.

$$SMX^+ \overset{k_{a1}}{\Longleftrightarrow} SMX^\pm + H^+, \qquad k_{a1} = \frac{[SMX^\pm]\cdot[H^+]}{[SMX^+]} \qquad (6.8)$$

$$SMX^\pm \overset{k_{a2}}{\Longleftrightarrow} SMX^- + H^+. \qquad k_{a2} = \frac{[SMX^-]\cdot[H^+]}{[SMX^\pm]} \qquad (6.9)$$
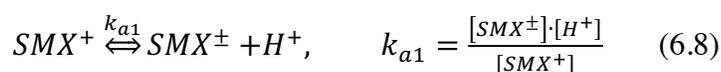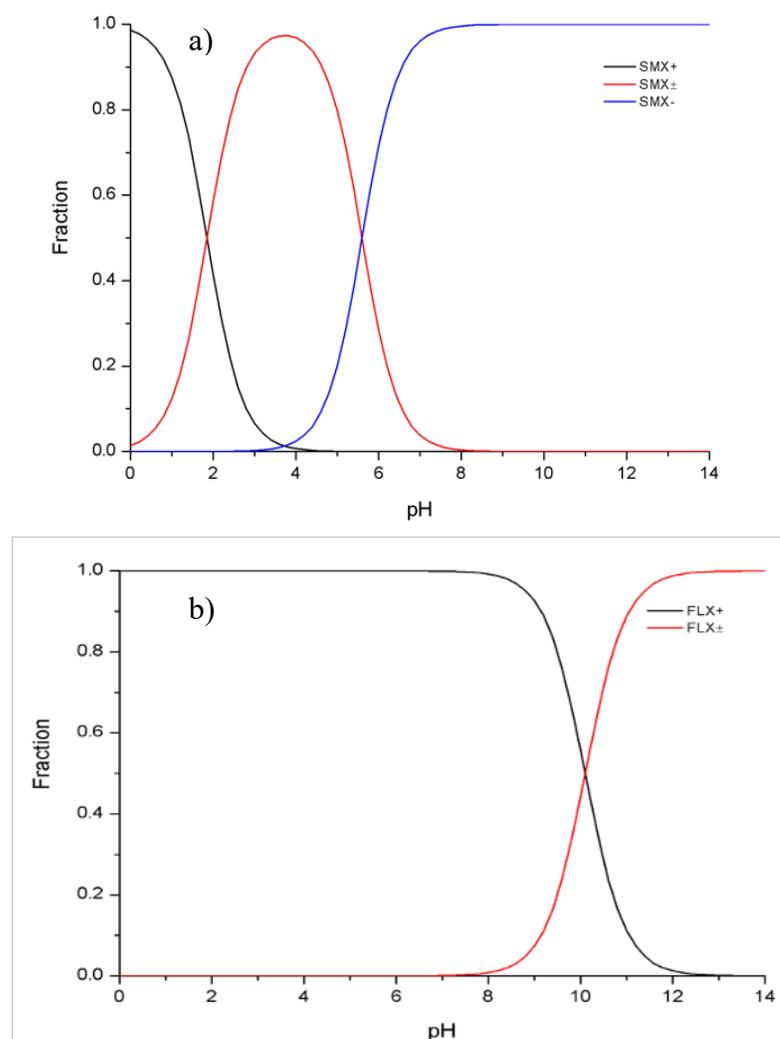
Figure 6.5: Speciation fraction of SMX and FLX at different pH

The acid condition exhibits slower photolysis rates for SMX than in alkaline and neutral conditions. With the increase in pH, the existing form of SMX converted from cation form (SMX^+) or neutral form ($SMX^\pm$) to anion form (SMX^-) increased. This result indicated that deprotonated SMX^- was much more difficult to undergo photolysis than neutral ($SMX^\pm$). Similar results were also described in the most recent publication (Liu et al., 2021).

For FLX, the photolysis rate of FLX was increased as pH increased from 5 to 8. These results are in accordance with previous findings that fluoxetine was most persistent at pH

4 under irradiation of visible light from pH 2-12 (Yin et al., 2017). Similarly, the photodegradation characteristic could be in accord with the ionization form of FLX at different pH (Figure 6.5 b). The photolysis rate did not change much for low pH due to lower amount of FLX⁻ existing under pH 6. However, as the pH increased closer to pKa value, the photolysis increased by 73% compared to pH 7 and pH 8, probably due to deprotonation. The increasing pH was assumed to contribute to the ammonium group's deprotonation, resulting in enhanced photodegradation ability (Lam et al., 2005). In summary, solution pH played an essential role in certain micropollutants' photolysis, depending on their physical and chemical characteristics.

6.4 Conclusion

In this study, the effect of pH dependence on the molar extinction coefficient of 12 PPCPs and the photodegradation effectiveness of selected PPCPs with an LP-UV lamp that emits at 254 nm were investigated. No notable variations were found on the molar absorptivity of three compounds (AP, AMX, and FLX), while sulfa group compounds (SMX, STZ, and SXZ), fluoroquinolones group compound (CIP), and tetracycline group compounds (CTC and DC) showed considerable pH-dependence in the tested pH range. These results in the pH-dependent effect of molar absorptivity are closely related to their pKa, in which pH alters the ionic species forms of PPCPs. Similarly, the behavior of absorption scans from 200 nm to 800 nm show the effect of pH, which may explain the mechanism of photodegradation under different operating conditions. Furthermore, the absorption peaks of 12 PPCPs are overlapped with the solar irradiation; therefore, solar light can be used for their degradation.   The lab-scale direct UV photolysis experiment by a collimated beam apparatus determined four compounds' photodegradation rate constants (k') and quantum yields (QY). SMX and DFC showed a high degradation rate compared to AP and FLX. The pH-dependent photodegradation information in this work could shed light on removing those PPCPs, or compounds from similar groups in actual water treatment at different pH conditions.

6.5　Reference

Alharbi, S. K., Kang, J., Nghiem, L. D., van de Merwe, J. P., Leusch, F. D. L., & Price, W. E. (2017). Photolysis and UV/H2O2 of diclofenac, sulfamethoxazole, carbamazepine, and trimethoprim: Identification of their major degradation products by ESI–LC–MS and assessment of the toxicity of reaction mixtures. *Process Safety and Environmental Protection*, *112*, 222–234. https://doi.org/https://doi.org/10.1016/j.psep.2017.07.015

Ashraf, M. A. (2017). *Persistent organic pollutants (POPs): a global issue, a global challenge*. Springer.

Bilińska, L., Blus, K., Gmurek, M., & Ledakowicz, S. (2019). Coupling of electrocoagulation and ozone treatment for textile wastewater reuse. *Chemical Engineering Journal*, *358*, 992–1001. https://doi.org/https://doi.org/10.1016/j.cej.2018.10.093

Bolobajev, J., Trapido, M., & Goi, A. (2016). Effect of iron ion on doxycycline photocatalytic and Fenton-based autocatalytic decomposition. *Chemosphere*, *153*, 220–226. https://doi.org/https://doi.org/10.1016/j.chemosphere.2016.03.042

Bolton, J. R., & Stefan, M. I. (2002). Fundamental photochemical approach to the concepts of fluence (UV dose) and electrical energy efficiency in photochemical degradation reactions. *Research on Chemical Intermediates*, *28*(7), 857–870. https://doi.org/10.1163/15685670260469474

Boreen, A. L., Arnold, W. A., & McNeill, K. (2004). Photochemical fate of sulfa drugs in the aquatic environment: sulfa drugs containing five-membered heterocyclic groups. *Environmental Science & Technology*, *38*(14), 3933–3940.

Cai, A., Deng, J., Xu, M., Zhu, T., Zhou, S., Li, J., Wang, G., & Li, X. (2020). Degradation of tetracycline by UV activated monochloramine process: Kinetics, degradation pathway, DBPs formation and toxicity assessment. *Chemical Engineering Journal*, *395*, 125090.

Caicedo, C., Rosenwinkel, K.-H., Exner, M., Verstraete, W., Suchenwirth, R., Hartemann, P., & Nogueira, R. (2019). Legionella occurrence in municipal and industrial wastewater treatment plants and risks of reclaimed wastewater reuse: Review. *Water Research*, *149*, 21–34. https://doi.org/https://doi.org/10.1016/j.watres.2018.10.080

Carlson, J. C., Stefan, M. I., Parnis, J. M., & Metcalfe, C. D. (2015). Direct UV photolysis of selected pharmaceuticals, personal care products and endocrine disruptors in aqueous solution. *Water Research*, *84*, 350–361.

Chen, J. P., Yang, L., Wang, L. K., & Zhang, B. (2006). Ultraviolet radiation for disinfection. In *Advanced Physicochemical Treatment Processes* (pp. 317–366). Springer.

Cheng, S., Zhang, X., Song, W., Pan, Y., Lambropoulou, D., Zhong, Y., Du, Y., Nie, J., & Yang, X. (2019). Photochemical oxidation of PPCPs using a combination of solar irradiation and free available chlorine. *Science of The Total Environment*, *682*, 629–638. https://doi.org/https://doi.org/10.1016/j.scitotenv.2019.05.184

Cvetnic, M., Ukic, S., Kusic, H., Bolanca, T., & Bozic, A. L. (2017). Photooxidative degradation of pesticides in water; response surface modeling approach. *Journal of Advanced Oxidation Technologies*, *20*(1).

Darby, J. (1995). *Comparison of UV irradiation to chlorination: guidance for achieving optimal UV performance*. Water Environment Research Foundation.

de Jesus Gaffney, V., Cardoso, V. V., Cardoso, E., Teixeira, A. P., Martins, J., Benoliel, M. J., & Almeida, C. M. M. (2017). Occurrence and behaviour of pharmaceutical compounds in a Portuguese wastewater treatment plant: Removal efficiency through conventional treatment processes. *Environmental Science and Pollution Research*, *24*(17), 14717–14734.

De la Cruz, N., Giménez, J., Esplugas, S., Grandjean, D., de Alencastro, L. F., & Pulgarín, C. (2012). Degradation of 32 emergent contaminants by UV and neutral

photo-fenton in domestic wastewater effluent previously treated by activated sludge. *Water Research*, *46*(6), 1947–1957. https://doi.org/https://doi.org/10.1016/j.watres.2012.01.014

Dong, H., Qiang, Z., Hu, J., & Qu, J. (2017). Degradation of chloramphenicol by UV/chlorine treatment: Kinetics, mechanism and enhanced formation of halonitromethanes. *Water Research*, *121*, 178–185. https://doi.org/https://doi.org/10.1016/j.watres.2017.05.030

Fang, J.-Y., Ling, L., & Shang, C. (2013). Kinetics and mechanisms of pH-dependent degradation of halonitromethanes by UV photolysis. *Water Research*, *47*(3), 1257–1266.

Hoehne, L., Vettorello, G., Ethur, E. M., Kuhn, D., De Freitas, E. M., & Steffens, C. (n.d.). *Analysis of degradation of the micro pollutant amoxicillin by photolysis and evaluation of the degradation products*.

Kellis, M., Kalavrouziotis, I. K., & Gikas, P. (2013). Review of wastewater reuse in the Mediterranean countries, focusing on regulations and policies for municipal and industrial applications. *Global NEST Journal*, *15*(3), 333–350.

Kim, I., Yamashita, N., & Tanaka, H. (2009). Photodegradation of pharmaceuticals and personal care products during UV and UV/H2O2 treatments. *Chemosphere*, *77*(4), 518–525. https://doi.org/https://doi.org/10.1016/j.chemosphere.2009.07.041

Lam, M. W., Young, C. J., & Mabury, S. A. (2005). Aqueous photochemical reaction kinetics and transformations of fluoxetine. *Environmental Science & Technology*, *39*(2), 513–522.

Lian, J., Qiang, Z., Li, M., Bolton, J. R., & Qu, J. (2015). UV photolysis kinetics of sulfonamides in aqueous solution based on optimized fluence quantification. *Water Research*, *75*, 43–50.

Liu, S., Fu, Y., Wang, G., & Liu, Y. (2021). Degradation of sulfamethoxazole by UV/sulfite in presence of oxygen: Efficiency, influence factors and mechanism.

Separation and Purification Technology, *268*, 118709.
https://doi.org/https://doi.org/10.1016/j.seppur.2021.118709

Luo, S., Wei, Z., Spinney, R., Zhang, Z., Dionysiou, D. D., Gao, L., Chai, L., Wang, D., & Xiao, R. (2018). UV direct photolysis of sulfamethoxazole and ibuprofen: An experimental and modelling study. *Journal of Hazardous Materials*, *343*, 132–139. https://doi.org/https://doi.org/10.1016/j.jhazmat.2017.09.019

Mao, H., Li, H., Li, Y., Li, L., Yin, L., & Yang, Z. (2020). Four typical personal care products in a municipal wastewater treatment plant in China: Occurrence, removal efficiency, mass loading and emission. *Ecotoxicology and Environmental Safety*, *188*, 109818. https://doi.org/https://doi.org/10.1016/j.ecoenv.2019.109818

NAS, B., DOLU, T., ATEŞ, H., ARGUN, M. E., & YEL, E. (2017). TREATMENT ALTERNATIVES FOR MICROPOLLUTANT REMOVAL IN WASTEWATER. *Selçuk Üniversitesi Mühendislik Bilim ve Teknoloji Dergisi*, *5*(2), 133–143.

Peng, X., Hu, F., Lam, F. L. Y., Wang, Y., Liu, Z., & Dai, H. (2015). Adsorption behavior and mechanisms of ciprofloxacin from aqueous solution by ordered mesoporous carbon and bamboo-based carbon. *Journal of Colloid and Interface Science*, *460*, 349–360.

Qiang, Z., & Adams, C. (2004). Potentiometric determination of acid dissociation constants (pKa) for human and veterinary antibiotics. *Water Research*, *38*(12), 2874–2890.

Real, F. J., Acero, J. L., Benitez, F. J., Roldán, G., & Fernández, L. C. (2010). Oxidation of hydrochlorothiazide by UV radiation, hydroxyl radicals and ozone: Kinetics and elimination from water systems. *Chemical Engineering Journal*, *160*(1), 72–78.

Rizzo, L., Gernjak, W., Krzeminski, P., Malato, S., McArdell, C. S., Perez, J. A. S., Schaar, H., & Fatta-Kassinos, D. (2020). Best available technologies and treatment trains to address current challenges in urban wastewater reuse for irrigation of crops in EU countries. *Science of the Total Environment*, *710*, 136312.

Rolinson, G. N. (1974). Laboratory evaluation of amoxicillin. *Journal of Infectious Diseases*, *129*(Supplement_2), S139–S145.

Shaojun, J., Zheng, S., Daqiang, Y. I. N., Lianhong, W., & Liangyan, C. (2008). Aqueous oxytetracycline degradation and the toxicity change of degradation compounds in photoirradiation process. *Journal of Environmental Sciences*, *20*(7), 806–813.

Wei, C., Li, X., Xie, Y., & Wang, X. (2019). Direct photo transformation of tetracycline and sulfanomide group antibiotics in surface water: Kinetics, toxicity and site modeling. *Science of The Total Environment*, *686*, 1–9. https://doi.org/https://doi.org/10.1016/j.scitotenv.2019.04.041

Werner, J. J., Arnold, W. A., & McNeill, K. (2006). Water hardness as a photochemical parameter: tetracycline photolysis as a function of calcium concentration, magnesium concentration, and pH. *Environmental Science & Technology*, *40*(23), 7236–7241.

Xu, M., Deng, J., Cai, A., Ma, X., Li, J., Li, Q., & Li, X. (2020). Comparison of UVC and UVC/persulfate processes for tetracycline removal in water. *Chemical Engineering Journal*, *384*, 123320. https://doi.org/https://doi.org/10.1016/j.cej.2019.123320

Yan, Z., Yang, H., Dong, H., Ma, B., Sun, H., Pan, T., Jiang, R., Zhou, R., Shen, J., Liu, J., & Lu, G. (2018). Occurrence and ecological risk assessment of organic micropollutants in the lower reaches of the Yangtze River, China: A case study of water diversion. *Environmental Pollution*, *239*, 223–232. https://doi.org/https://doi.org/10.1016/j.envpol.2018.04.023

Yang, Y., Ok, Y. S., Kim, K.-H., Kwon, E. E., & Tsang, Y. F. (2017). Occurrences and removal of pharmaceuticals and personal care products (PPCPs) in drinking water and water/sewage treatment plants: A review. *Science of the Total Environment*, *596*, 303–320.

Yin, L., Ma, R., Wang, B., Yuan, H., & Yu, G. (2017). The degradation and persistence

of five pharmaceuticals in an artificial climate incubator during a one year period. *RSC Advances*, *7*(14), 8280–8287.

Yun, S. H., Jho, E. H., Jeong, S., Choi, S., Kal, Y., & Cha, S. (2018). Photodegradation of tetracycline and sulfathiazole individually and in mixtures. *Food and Chemical Toxicology*, *116*, 108–113. https://doi.org/https://doi.org/10.1016/j.fct.2018.03.037

Chapter 7

7 Conclusions and recommendations

7.1 Summary and conclusion

The thesis investigated the effect and removal performance of micropollutants and microplastics during selected wastewater treatment processes.

The fate of microfibers in traditional wastewater processes is not clearly understood. In this study, the effect of coagulation on microfibers suspended in pure water and real domestic laundry effluent was investigated using ferric chloride at a concentration range of 10-30 mg/L. The effect of microfibers' size on their coagulation was determined in the size range: < 90 μm, 90-125 μm, and > 125 μm. The microfibers removal efficiency due to coagulation ranged from 86%-96% and 30%-94% in pure water and laundry wastewater, respectively, with higher efficiency in pure water. The presence of surfactant in detergent in laundry wastewater reduced the removal efficiency of microfibers by coagulation. Further addition of a low dosage of PACl (around 2 mg/L) enhanced the removal efficiency up to over 90% in laundry wastewater in the presence of detergent, which provides a reference for the process improvement. Based on these control studies, it can be concluded that the coagulation process in conventional WWTPs can effectively remove microfibers with a typical dosage of Fe-based coagulant or without ferric. However, the addition of a small amount of PACl would be required to remove microfibers associated with surfactants. The study also highlights that with 90% microfibers removed by settling/coagulation, the fate of microfibers in the settled sludge and final removal need to be further investigated.

Subsequently, the fate of the microfibers and their impact on the wastewater treatment plants' solid processing units were investigated. More specifically, the effect of microfibers concentration and pretreatment on anaerobic digestion was investigated. This study revealed for the first time that the behavior of real microplastics from a domestic laundry water affected the methane production in primary sludge anaerobic digestion. The results showed that microfibers at 20 mg/L insignificantly affected methane production, while

ozone pretreatment enhanced gas production by 28% in the same concentration level. However, ozone pretreatment at a higher level (100 mg/L to 1000 mg/L) had no effect on methane production compared with the same level microfibers not treated with ozone. In comparison to control, methane production was significantly increased by 32% and 35%, with increasing microfibers concentration from 100 mg/L to 1000 mg/L. In addition, dewatering ability of sludge was enhanced in the presence of microfibers, while phosphorus removal reflected a weak correlation with microfibers. Overall, addition of microfibers had a positive effect on anaerobic digestion.

Like microfibers, a significant portion of micropollutants with more hydrophobicity end up in the primary sludge making their way to anaerobic digestion and the biosolids, posing potential concern regarding the land application of biosolids. Thus, the third objective investigated the effect of TAH on five selected PPCPs, including fluoroquinolone antibiotics, ciprofloxacin (CIP), and ofloxacin (OFLX), and three commonly used antimicrobial agents, miconazole (MIC), triclosan (TCS) and triclocarban (TCC). At the onset, extraction and analytical methods were optimized for maximum simultaneous recovery and LC-MS quantification of the target PPCPs from water and biosolids for improved accuracy. The compounds were detected in the range of 54 ± 3 to 6166 ± 532 ng/g in raw biosolids collected from a local WWTP. Next, batch control adsorption experiments of the selected PPCPs were conducted in various sludges, which indicated about 89%–98% sorption of the PPCPs onto the solid phase due to their high octanol-water coefficients. Subsequently, thermo-alkaline (pH 9.5, 75 °C, 45 min) hydrolysis (TAH) was conducted to determine the extent of degradation of these compounds in deionized (DI) water and biosolids due to treatment. The degradation of these compounds due to TAH ranged from 42% to 99% and 37%–41% in pure water and biosolids, respectively, potentially lowering their risk in the environment due to land application.

While micropollutants of hydrophobic nature made their way to the sludge fraction, the molar absorptivity of twelve micropollutants was studied at varying pH 5-8 and their degradation using a UV collimated beam apparatus with an LP-UV lamp (20.4 W). Sulfa and tetracycline group compounds were found to have higher molar extinction coefficients among these compounds, indicating their higher degradation potential under UV-254

radiation. Also, pH affects molar absorption due to changes in ionic form, resulting in a blue or red shift in their peak absorption. Furthermore, direct UV photolysis removed four selected micropollutants, and the pseudo-first-order rate constant ranged from 1.40- 6.54 × 10^3 cm^{-2} mJ. The lab-scale direct UV photolysis experiment in a collimated beam apparatus proved that the effect of pH on quantum yield depends on the chemical characteristics of micropollutants.

7.2   Future Recommendations

Based on the scientific investigation of the micropollutants and microplastics, this thesis could be aggregated into two main projects. Chapter 3 involved work on removing microplastics from water via coagulation into sludge, and then the effect of microplastics in the solid stream on aerobic digestion was studied in Chapter 4. Chapter 5 and Chapter 6 described the removal of micropollutants in the solid and liquid stream by post-treatment after AD and direct UV photolysis, respectively.

1) As simultaneous detection methods have been established for targeted micropollutants, these methods require further investigation and validation for numerous non-targeted micropollutants in environmental samples;

2) TAH and UV photolysis are both efficient treatment processes for micropollutants; further study needs to investigate the transformation products and their toxicity to the environment.

3) It is recommended to study the mechanism for methane production enhanced by microfibers, and the effects of microplastics on microorganisms need to be clarified;

4) As microplastics can be vectors for micropollutants adsorption, this needs to be ascertained in future studies.

Appendices

Appendix A: Supplementary material of chapter 3

Table S1: Particles to mass result

	1	2	3	4	5
Initial W $_{fiber}$ (mg)	1.5	1.5	1.5	1.5	1.5
W_1 (mg)	90.0	90.5	89.6	89.2	90.3
W_2 (mg)	90.8	91.2	90.3	90.3	91.0
Number of Particles in 1/8 Area (N_1)	74	81	77	106	73
Relationship between mass and number	592=0.8 (740/mg)	648=0.7 (925/mg)	616=0.7 (880/mg)	848=1.1 (770/mg)	584=0.7 (834/mg)
Average number of fibers per mg particle	830±75 particles/mg				

Table S2: Chemical composition of liquid detergents (provided by the manufacturer from Smart Label)

Ingredient name	Function
Water	Solvent
Sodium and MEA Laureth Sulfate	Surfactant; clean agent
Sodium and MEA C10-16 Alkylbenzenesulfonate	Surfactant; clean agent
Polyethyleneimines Alkoxylated	Suspends soils

Propylene Glycol	Solvent
Sodium and MEA Citrate	Water Softener
Alcohol	Solvent
Sodium Borate	Stabilizer
Sodium and MEA Salts of C12-18 Fatty Acids	Suds Reducer
Lauramide MEA	Surfactant; clean agent
Pentasodium Pentetate	Cleaning Aid
C10-16 Alkyldimethylamine Oxide	Surfactant; clean agent
Fluorescent Brightener 71	Whitening Agent
Calcium Formate	Stabilizer
Sodium Cumenesulfonate	Stabilizer
C10-16 Pareth	Surfactant; clean agent
Subtilisin	Enzyme
Ethanolamine	Stabilizer
Amylase Enzyme	Enzyme
Polyoxyalkylene Substituted Chromophore	Colorant
Mannanase Enzyme	Enzyme
Fragrances	Perfume
Sodium Formate	Process Aid
Diethylenetriamine	Odor Remover

| Methyl Di-T-Butyl Hydroxyhydrocinnamate	Odor Remover

Table S3: Turbidity of laundry wastewater coagulation by PACl and FeCl₃

Coagulants	PACl (mg/L)/ FeCl$_3$(mg/L)	0/0	1/10	2/15	3/20	4/25	5/30
PACl	Initial (NTU)	174	176	174	175	170	171
	Final (NTU)	150	12.8	10.7	2.52	6.3	21.4
	Removal (%)	12%	95%	97%	99%	98%	94%
without detergent	Initial (NTU)	169	174	173	174	171	174
	Final (NTU)	140	141	145	146	150	148
	Removal (%)	17%	19%	16%	16%	16%	16%

Table S4: Turbidity of different cycles during coagulation

Laundry cycles No.	FeCl$_3$ (mg/L)	0	10	15	20	25	30
1 (without detergent)	Initial (NTU)	20.87±0.15	20.97±0.25	21.83±0.21	21.80±0.46	21.77±0.71	21.63±0.35
	Final (NTU)	14.60±0.10	11.37±0.15	5.21±0.02	2.65±0.03	1.87±0.01	1.38±0.02
	Removal (%)	30%	46%	76%	88%	91%	94%

|							
2 (without detergent)	Initial (NTU)	63.90±0.10	64.10±0.20	63.77±0.45	63.47±0.50	63.63±0.15	63.93±0.25
	Final (NTU)	44.97±0.12	40.87±0.15	39.67±0.06	30.90±0.10	9.42±0.02	6.42±0.02
	Removal (%)	30%	36%	38%	51%	85%	90%
3 (with detergent)	Initial (NTU)	287.67±2.08	283.33±1.53	284.00±1.00	286.67±0.58	294.67±4.16	302.00±3.61
	Final (NTU)	271.00±2.65	264.00±1.00	269.33±0.58	268.67±3.79	280.00±2.00	289.00±4.36
	Removal (%)	6%	7%	5%	7%	6%	4%
4 (with detergent)	Initial (NTU)	84.03±0.81	84.13±0.21	84.17±0.70	84.93±0.50	84.40±0.46	84.70±0.44
	Final (NTU)	71.40±0.10	73.37±0.61	72.10±0.44	72.87±0.44	72.40±0.40	72.67 ±0.35
	Removal (%)	15%	13%	14%	14%	14%	14%

SLS
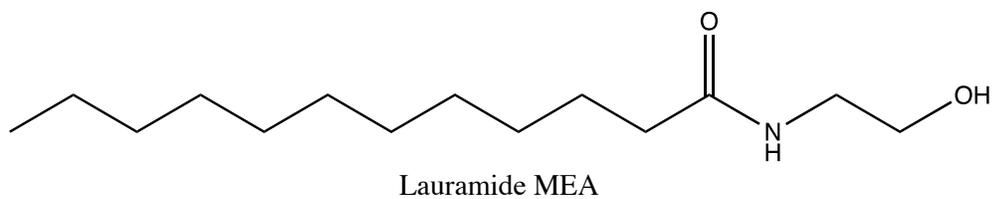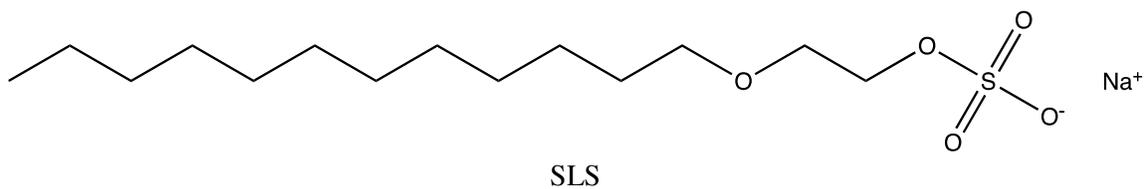
Lauramide MEA

Figure S1: Chemical structure of sodium laureth sulfat (SLS) and lauramide monoethanolamin (MEA)
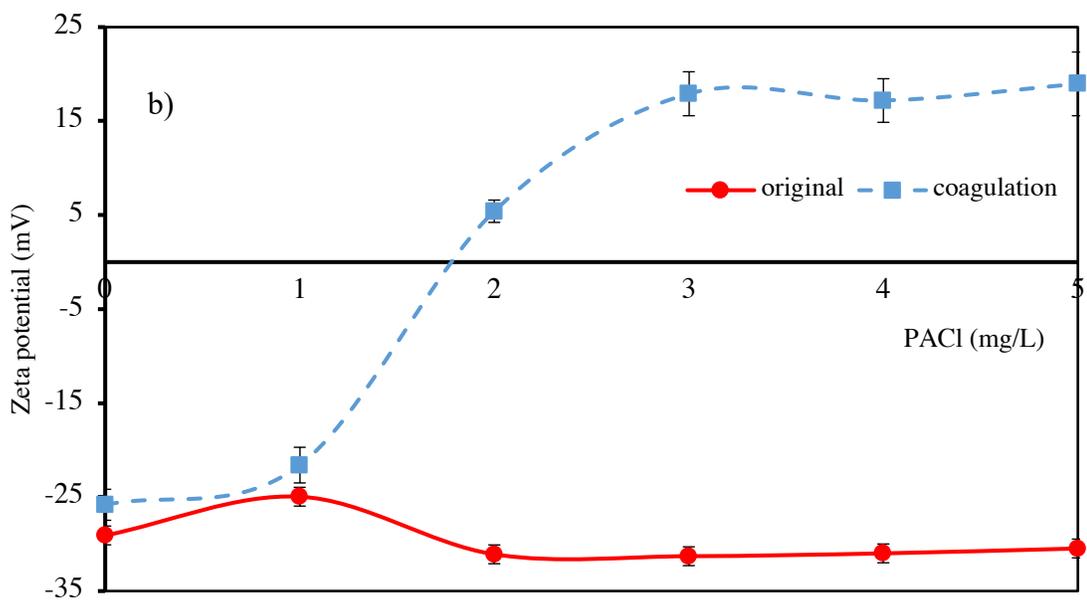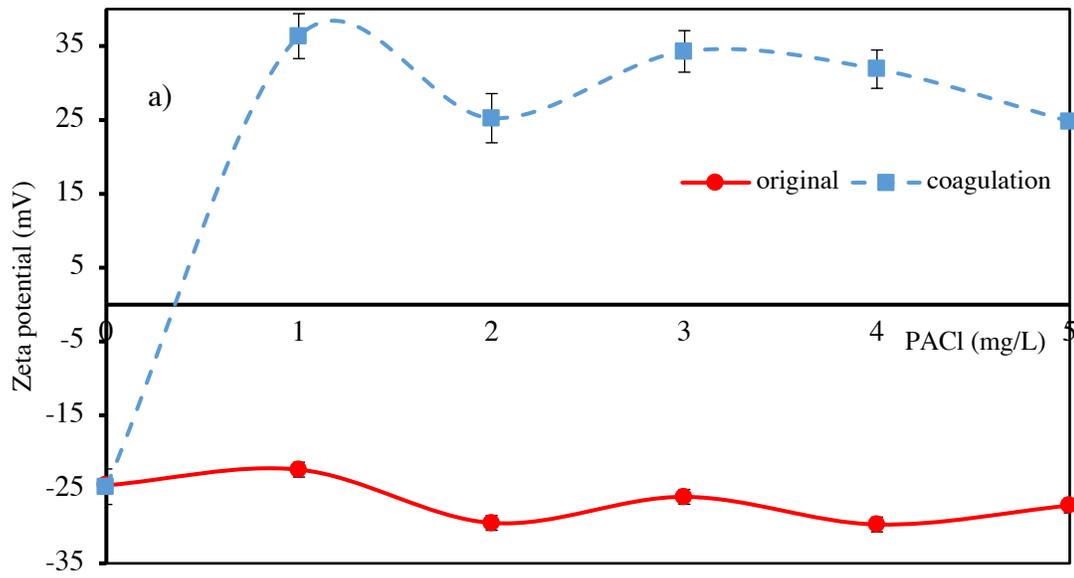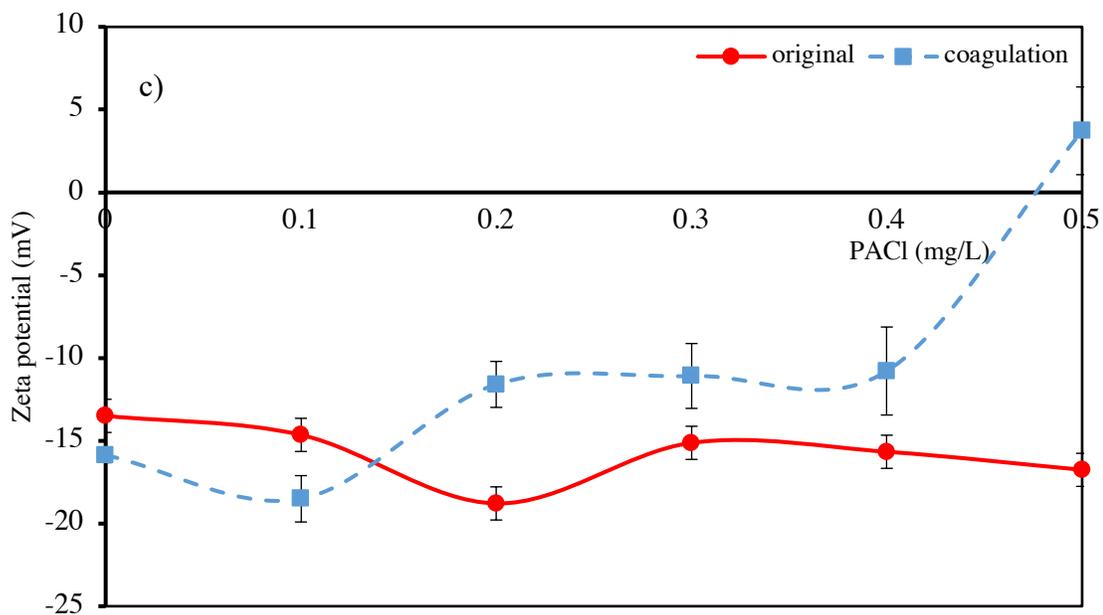
Figure S2: Changes of Zeta potential during coagulation (a) laundry wastewater coagulation without detergent; (b) laundry wastewater coagulation with detergent; (c) laundry wastewater coagulation at low dosage.
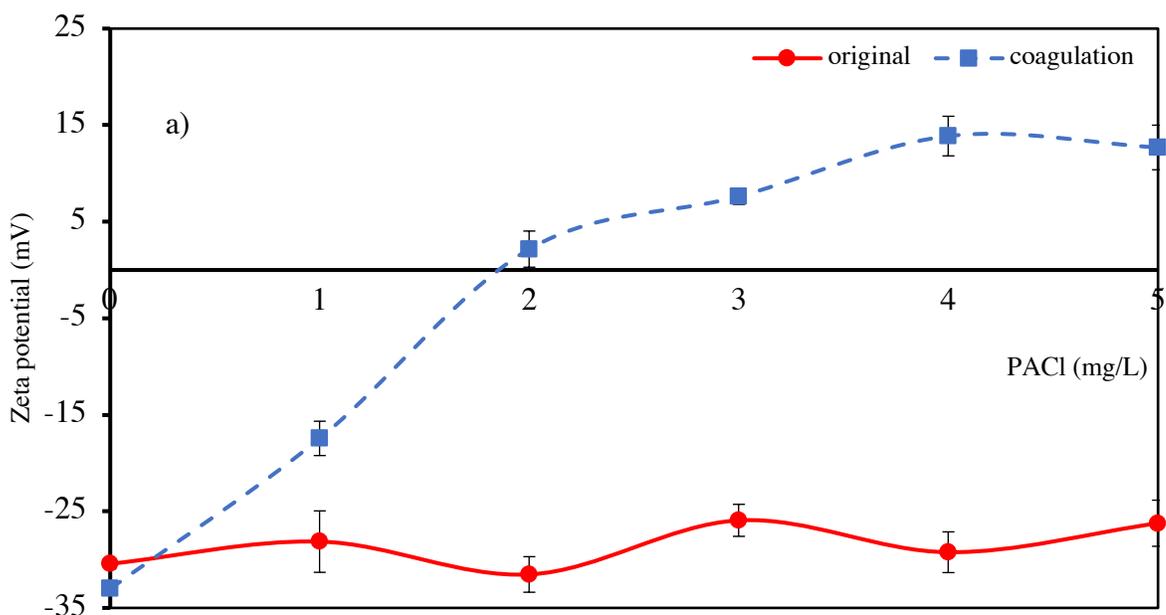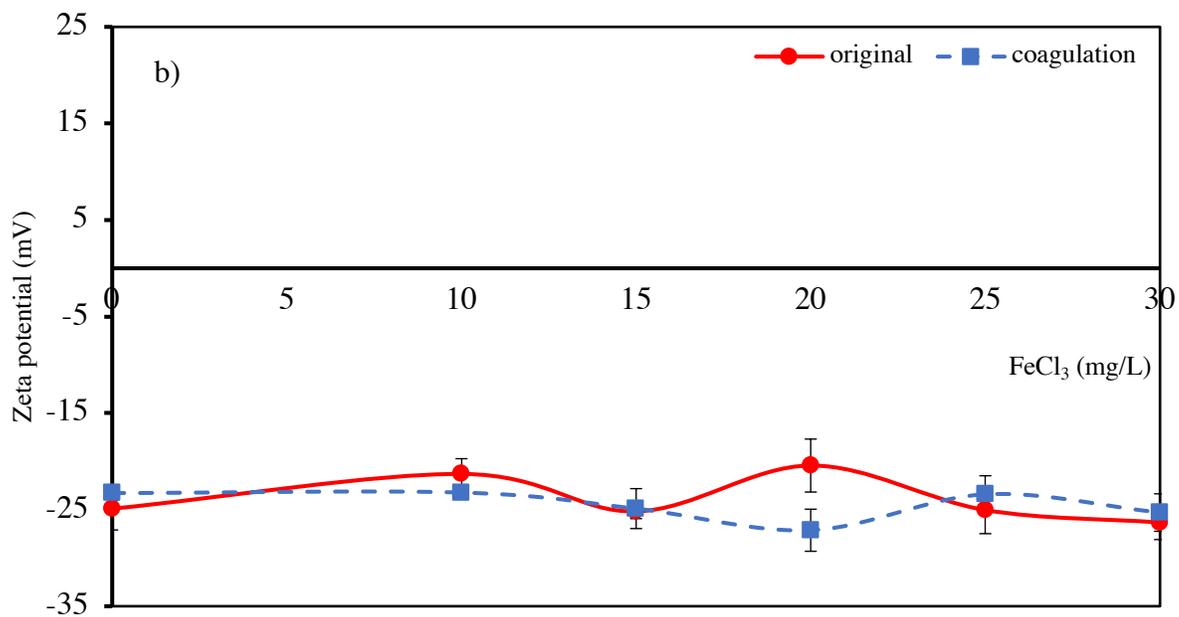
Figure S3: Changes of Zeta potential during coagulation (a) laundry wastewater coagulation with PACl in presence of detergent and (b) laundry wastewater coagulation with FeCl3 in presence of detergent.

Appendix B: Supplementary material of chapter 5
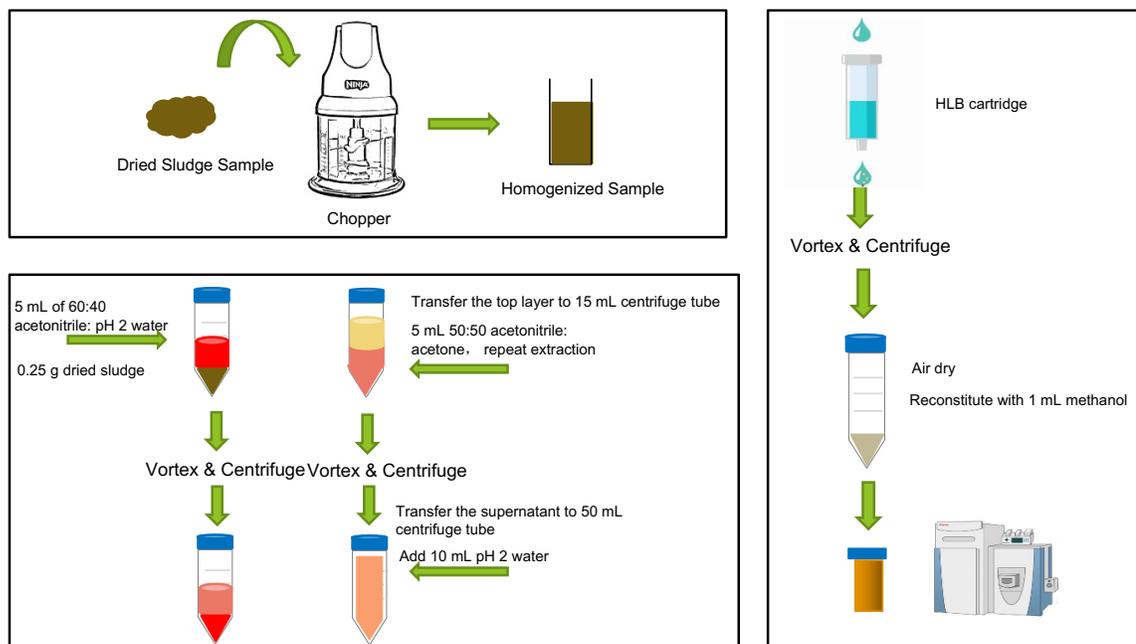
Figure S1: Schematic of solid phase extraction
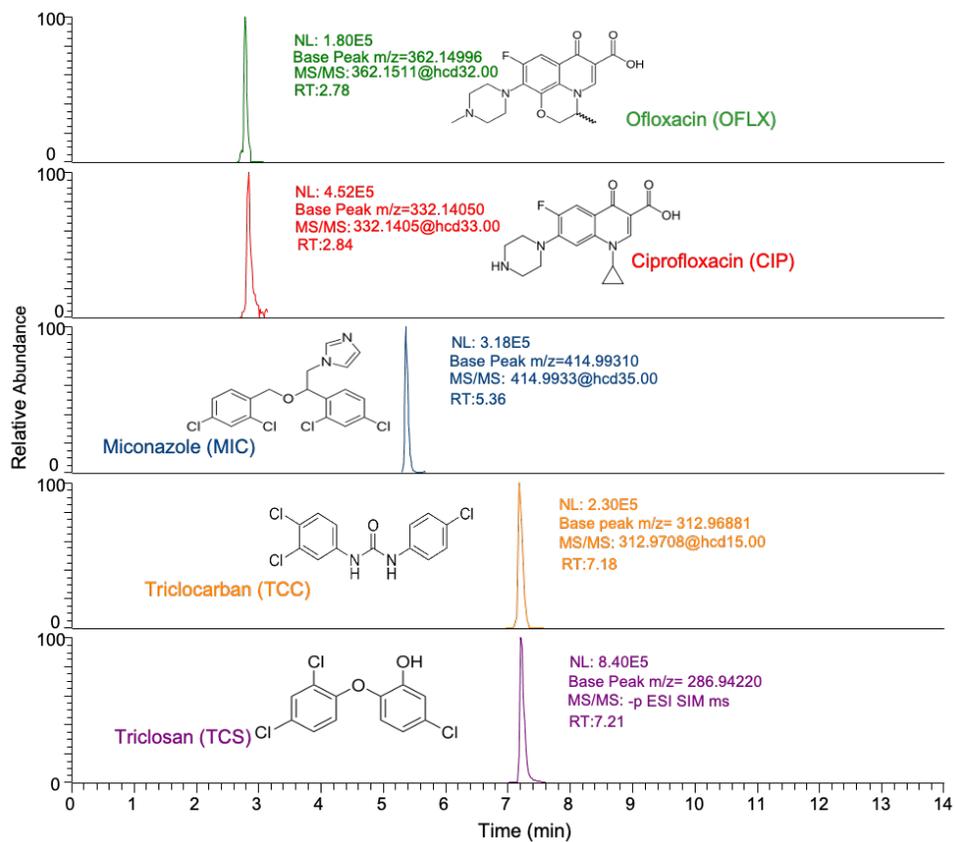
Figure S2: Diagram of LC-MS spectrum for targeted micropollutants
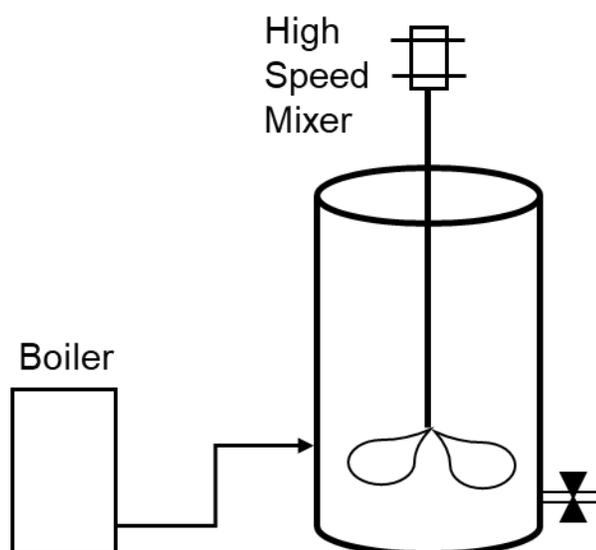
Figure S3: Schematic of thermal alkaline hydrolysis reactor

Curriculum Vitae

Name: Juan Li

Post-secondary   China University of Petroleum
Education and   Changping, Beijing, China
Degrees: 2010-2014 B.A.

China University of Petroleum
Changping, Beijing, China
2014-2017 M.A.

The University of Western Ontario
London, Ontario, Canada
2017-2021 Ph.D.

Honours and   Western Graduate Research Scholarship
Awards: 2017-2021

Related Work   Teaching Assistant
Experience   The University of Western Ontario
2017-2021

Research participant
Agriculture and Agri-Food Canada, London Research and
Development Centre
2018-2019

Publications:
Li, J., Sabourin, L., Renaud, J., Halloran, S., Singh, A., Sumarah, M., ... & Ray, M. B. (2021). Simultaneous quantification of five pharmaceuticals and personal care products in biosolids and their fate in thermo-alkaline treatment. Journal of Environmental Management, 278, 111404.

Chowdhury, P., Sarathy, S. R., Das, S., **Li, J**., Ray, A. K., & Ray, M. B. (2020). Direct UV photolysis of pharmaceutical compounds: Determination of pH-dependent quantum yield and full-scale performance. Chemical Engineering Journal, 380, 122460.