Theses, Dissertations and Culminating Projects

1-2014

Green Remediation of Veterinary Antibiotics in Soil-Water Systems

Pravin Anraj Punamiya
Montclair State University

Follow this and additional works at: https://digitalcommons.montclair.edu/etd

GREEN REMEDIATION OF VETERINARY ANTIBIOTICS IN

SOIL-WATER SYSTEMS

A DISSERTATION

Submitted to the Faculty of

Montclair State University in partial fulfillment

of the requirements

for the degree of Doctor of Philosophy

by

PRAVIN ANRAJ PUNAMIYA

Montclair State University

Montclair, NJ

2015

Dissertation Chair: Dibyendu Sarkar, PhD, PG

MONTCLAIR STATE UNIVERSITY

THE GRADUATE SCHOOL

DISSERTATION APPROVAL

We hereby approve the Dissertation

GREEN REMEDIATION OF VETERINARY ANTIBIOTICS

IN SOIL-WATER SYSTEMS

of

Pravin Anraj Punamiya

Candidate for the Degree:

Doctor of Philosophy

Dissertation Committee:

Department of Earth and
Environmental Studies

Dr. Dibyendu Sarkar
Dissertation Chair

Certified by:

Dr. Joan C. Ficke
Dean of The Graduate School

Dr. Rupali Datta

5/6/15

Date

Dr. Nina Goodey

Dr. Dilrukshi Ramanathan

Dr. Ronald MacGillivray

ABSTRACT

GREEN REMEDIATION OF VETERINARY ANTIBIOTICS IN

SOIL-WATER SYSTEMS

by Pravin Anraj Punamiya

Veterinary antibiotics (VAs) are considered emerging contaminants of concern. Considerable efforts have been made to understand the fate and transport of VAs in soil and water environment and very few have attempted to develop novel remediation strategies to overcome the problem of antibiotic resistance and potential toxicity to aquatic species. To our knowledge, the present study is a pioneer study in which it attempts to develop a low-cost, "green" remediation technique utilizing a waste by-product of the drinking water treatment industry, namely, the Al-based water treatment residuals (Al-WTR), as a sorbent to stabilize tetracycline (TTC) and oxytetracycline (OTC) in aqueous medium, manure piles and manure-treated soil. The ultimate goal of the study was to evaluate the effectiveness of Al-WTR in treating manure, soils, and manure-amended soils to immobilize tetracyclines (TCs) to lower risk associated with TCs in environment. We conducted: i) laboratory batch sorption study followed by modeling and surface spectroscopic characterization to understand the extent and mechanism of TTC/OTC retention by Al-WTR, ii) short-term incubation study to evaluate the effectiveness of Al-WTR in immobilizing and stabilizing TTC and OTC in manure, soils, and manure-amended soils under static conditions, and iii) long-term greenhouse column study to evaluate the effectiveness of Al-WTR in immobilizing and stabilizing TTC and OTC in soils and manure amended soils under dynamic conditions in

a controlled environment. Results from the batch sorption study showed that Al-WTR has high sorption capacity for TTC and OTC as a function of solution properties. This, along with the rapid sorption kinetics and low release potential make them excellent sorbents for TCs removal from aqueous medium. Results from modeling and spectroscopic studies suggest that TTC and OTC are adsorbed on Al-WTR surface via strong inner-sphere mechanism, indicating permanent retention. Short term incubation and long term greenhouse column studies showed immobilization of TTC and OTC in Al-WTR amended soils and manure amended soils. LC/MS/MS analysis did not reveal any known detectable degradates or metabolites of TCs other than very low concentration of daughter compounds. Greenhouse column studies also showed that Al-WTR application significantly reduces plant available and water soluble TCs from soils and manure-amended soils. Overall, this research demonstrated the potential of Al-WTR to develop into an effective, low-cost, green remediation technology for TC-contaminated soil-water systems.

ACKNOWLEDGEMENTS

First and foremost, I would like to thank my advisor, chair, and mentor Dr. Dibyendu Sarkar for his encouragement, understanding, support, and personal guidance, which have been vital to the overall success of my PhD research. I would also like to thank my other committee members (Drs. Datta, Goodey, Ramanathan, and MacGillivray), who were always available to me; and without whose critique, encouragement, knowledge, and assistance this research would not have been successful. My special thanks go to Dr. Ramanathan for permitting me to use her laboratory and LC-MS/MS at the Kean University, New Jersey.

This research has been partially supported financially by student research grant awards from the Geological Society of America (GSA) and New Jersey Water Resources Research Initiatives (NJWRRI) -USGS program. SIROM Scientific Solutions, LLC is greatly acknowledged for curricular practical training (CPT) and paid summer internships. I am also thankful to the Montclair State University (MSU) for graduate assistantship, analytical facilities, and the Center for Writing Excellence for proofreading the dissertation chapters.

I would like to thank my family for their unending support and love. My Parents and sisters have always shown love and boundless inspiration from home. My wife, Pooja offered me encouragement and moral support, particularly during my difficult time and several recurring phases of frustration in my life as a graduate student. She was always there for me in spite of her busy schedule in her own graduate program. Last but not the least; I would like to thank my colleagues in the environmental geochemistry

laboratory who were always there for me and all my close friends who made my term at

MSU a very pleasant experience.

DEDICATION

To my loving parents, Anraj and Pavan, and my sisters, Bhawna and Kavita

And

To my wife, Pooja and to my daughter, Pahal

TABLE OF CONTENTS

Content	Page

Content	Page

Content	Page

Content	Page

Content	Page

LIST OF TABLES

Table   **Page**

LIST OF FIGURES

LIST OF ABBREVATIONS

2-(N-morpholino)ethanesulfonic acid (MES)
Aluminum (Al)
Carbon Hydrogen Nitrogen and Sulfur analyzer (CHNS analyzer)
Cation Exchange Capacity (CEC)
Chlortetracycline (CTC)
Clean Air Act (CAA)
Clean Water Act (CWA)
Code of Federal Regulations (CFR)
Concentrated Animal Feeding Operations (CAFOs)
Electrical Conductivity (EC)
Ethylenediaminetetraacetate (EDTA)
Extended X-ray absorption fine structure (EXAFS)
Fourier Transform Infrared Spectrometry (FTIR)
High Performance Liquid Chromatography (HPLC)
Inductively Coupled Plasma Mass Spectrometry (ICP-MS)
Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES)
Industrial Farm Animal Production (IFAP)
Iron (Fe)
Octanol-Water Partition Coefficient (K_{ow})
Organic Matter (OM)
Oxytetracycline (OTC)
Parts per million (ppm)
Phosphorus (P)
Piperazine-N,N′-bis(2-ethanesulfonic acid) (PIPES)
Quality Assurance and Quality Control (QA & QC)
Resources Conservation and Recovery Act (RCRA)
Solid Solution Ratio (SSR)
Tetracycline (TTC)
Tetracyclines (TCs)
Toxicity Characteristic (TC)
Toxicity Characteristic Leaching Procedure (TCLP)
United States Environmental Protection Agency (USEPA)
UV-Visible spectrophotometer (UV-Vis)
Veterinary antibiotics (VAs)
Water Treatment Residuals (WTR)
X-ray absorption near edge structure (XANES)
X-ray Diffraction (XRD)
Zero Point Charge (ZPC)

CHAPTER 1

INTRODUCTION

1. Introduction

Veterinary antibiotics (VAs) are the emerging contaminants of concern to the drinking water supplies, aquatic, and terrestrial ecosystems (Kümmerer, 2009; Aga, 2008; Kemper, 2008; Sarmah et al., 2006). Veterinary antibiotics are used in large amounts in concentrated animal feeding operations (CAFOs) to treat disease, protect the health of animals, and as feed supplements to improve their growth rate (Boxall et al., 2003; Sarmah et al., 2006). Livestock producers in the United States use more than 5,000 tons of antibiotics annually, even in the absence of any disease in the animals and much more in the event of outbreak of diseases (Kumar et al., 2005a, Sarmah et al., 2006). Several studies have shown that VAs are poorly adsorbed in the gut, of the animals and high amount ranging from 25% to 90% of the antibiotics are excreted in the urine and feces as active substances (Halling-Sorensen et al., 1998; Kumar et al., 2005a; Sarmah et al., 2006). Once excreted, VAs can enter into soils, surface- and/or groundwater via manure applied soils or via sludge storage at CAFOs. Most of the CAFOs store manure in the form of stockpiles or lagoons before land application (Moore et al., 1995). Land application of manure is a common practice in the U.S. since it serves the dual purpose of supplying nutrients to crops as well as serving as a means of disposal. These antibiotics are eventually released in to the environment through runoff streams from the CAFOs or leakage streams from the storage structures (Ostermann et al., 2013).

A nationwide reconnaissance study carried out by the U.S. Geological Survey showed the presence of low levels of pharmaceuticals, veterinary antibiotics, hormones, and other organic contaminants in a water and soil system from a network of 139 streams across 30 states (Kolpin et al., 2002). Among the VAs commonly used in the livestock industry are the tetracycline antibiotics (TCs) (e.g. tetracycline (TTC), oxytetracycline (OTC), and chlortetracycline (CTC)), which ranks second in production and usage worldwide, with OTC being the most popular for cattle in the U.S. Hamscher et al. (2005) reported concentrations of up to 270 μg kg^{-1} (OTC), 443 μg kg^{-1} (TTC), and 93 μg kg^{-1} (CTC) in manure impacted surface soils. Winckler and Grafe (2001) found TCs to persist in agricultural soils at concentrations of 450–900 μg kg^{-1}. Kay et al. (2004) observed that after manure was applied to a field in 2 consecutive years, OTC concentration in the soil was 1691 μg kg^{-1}, and concentrations as high as 613.2 μg L^{-1} were observed in drain flows, which end up in aquatic systems through agricultural effluent discharges. In addition, since the wastewater treatment process does not remove the antibiotics, biosolids produced during treatment contain antibiotics at concentrations several thousand folds higher than that of wastewater. About 50% of the biosolids are land applied, and the remaining is disposed of in landfills. Highly concentrated antibiotics persisting in the biosolids can be leached out, contaminating surface and ground water. Although there are restrictions on the use of raw manure in organic farming in the U.S. due to the risk of bacterial contamination, there are no guidelines regarding the presence of contaminants such as hormones and antibiotics in manure (Kumar et al., 2005a). Once the VAs are present in the soil, the fate is based on the binding/sorption of VAs to the soil

and its degradation behavior (Tolls, 2001). Based on binding and degradation behavior of VAs, they can degrade in soil with time (Blackwell et al., 2007), remain in parent form and via run-off enter into surface water (Topp et al., 2008), leach into groundwater (Topp et al., 2008), and/or be in available form to be taken up by plants (Kumar et al., 2005a)

Tetracyclines (TCs) are complex organic compounds with unique chemical characteristics and behaviors. Their structures contain connected ring systems with multiple ionizable functional groups. Tetracyclines have three pK_a's and hence, can exist as cationic, zwitterionic, or anionic species under acidic, moderately acidic to neutral, and alkaline conditions, respectively. The ionization behavior can be expected to significantly influence TCs sorption to soil components and other sorbents. The high polarity (e.g., log K_{ow} (octanol-water partition coefficient) = -1.97 to -0.47) and consequently high aqueous solubility (0.52−117 mM) of TCs portends that TCs could be highly mobile in soils (Tolls, 2001). Thus, effective means of immobilizing TCs in soils need to be employed to reduce the potential health hazards that could result from the presence of TCs in the soil environment, and consequently in surface and ground water.

In the U.S, agricultural pollution contributes close to 50% of surface water contamination from which 20% contribution is from livestock production (USEPA, 2002). The major human health and environmental concern of VAs in environment is the development and spread of bacterial resistance to VAs (Zhang et al., 2013; Oberle et al., 2012; Allen et al., 2010; Chee-Sanford et al., 2001; Sengelov et al., 2003) and toxicity to aquatic and terrestrial species (Gonzalez-Pleiter et al., 2013; Zhang et al., 2012, Wollenger et al., 2000; Park and Choi, 2008). There is limited information available on

ecotoxicological effects of antibiotics on terrestrial environment. However, there is a great concern regarding the impact of antibiotics on plant growth, soil fauna, soil enzyme activities, and nutrient cycling (Baguer et al., 2000; Kumar et al., 2005b; Sarmah et al., 2006). These impacts could be direct, such as antibiotics' toxicity to soil fauna and flora, or indirect effects, such as nutrient availability due to changed microfauna and microflora (Baguer et al., 2000; Westgaard et al., 2001; Muller et al., 2002; Kumar et al., 2005b;). Terrestrial ecotoxicity data for TCs used in agriculture show that they may be toxic to soil organisms and plants even at very low concentrations (Boxall et al., 2004; Kumar et al., 2005b; Sarmah et al., 2006; Kemper, 2008).

Some VAs such as tylosin degrades naturally over a period of time, and therefore are not a major cause for concern (De Liguoro et al., 2003). However, antibiotics such as TCs, are persistent in the environment (Kumar et al., 2005b; Hamscher et al., 2005; Wang and Yates, 2008). Currently, the only "remediation" process practiced is to allow the VAs to degrade in the lagoons or in the soil after amendment or composting. However, the extensive use and repeated detection of TCs and other VAs in the environment have increased the uncertainties over propagation of VAs-resistant bacteria, lower efficacy of VAs, and other possible antagonistic effects on human and environmental health (Kolpin et al., 2002; Thiele-Bruhn et al., 2003).

Recognizing the risk posed by VAs on ecological and human health, considerable efforts are being made to develop cost-effective treatment technologies to remove VAs from aqueous solutions. To find an optimal method for adsorption of TTCs and other VAs, sorbents such as soils, clays, minerals, humic acids, metal oxides (Fe/Al

hydroxides), carbon nanotubes, and plants have attracted great interest (Figueroa et al., 2004; Figueroa and MacKay, 2005; Gu and Karthikeyan, 2005; Gu et al., 2007; Pils and Laird, 2007; Ji et al., 2009; Rakshit et al., 2013a Rakshit et al., 2013b; Zhang et al., 2011; Zhao et al., 2012; Datta et al., 2013). The use of adsorbent materials provides an appealing substitute for soil-water remediation, particularly if the adsorbent is low-cost and does not involve any pretreatment process before application (Janos et al., 2007). However, the preparation procedures of some of these adsorbents are complicated and costly, which hinders their use.

In this study, utilization of a waste by-product from the drinking water treatment industry, surrogate of Al hydroxides, derived primarily from drinking water treatment residuals, or WTRs was proposed as an effective, low cost, green sorbent for TTC and OTC remediation. Drinking-water treatment residuals are generated from the drinking water treatment processes. The WTRs are primarily amorphous masses of aluminum (Al) or iron (Fe) hydroxides or calcium ($CaCO_3$), referred to as Al-WTRs (indicating the use of Al salt), Fe-WTRs (Fe salt), or Ca-WTRs ($CaCO_3$) (O'Connor et al., 2002; Ippolito et al., 2011), that also contain sediment and humic substances removed from the raw water as well as activated carbon and polymers (Elliott and Dempsey, 1991; Makris et al., 2005b; Ippolito et al., 2011). More than 2 million tons of WTRs are generated from the drinking-water treatment facilities in the U.S. every day (Prakash and Sengupta, 2003). Limited landfill space and increased emphasis on pollution control encourage plant operators to find alternative disposal methods, such as land application of the WTRs. State regulations (e.g., FL, OH) treat WTRs as non-hazardous wastes as long as they pass

the toxicity characteristic leaching procedure (TCLP) routine. Several studies have shown that the toxicity characteristic values for WTRs generated in the U.S. are typically well below the limit set by the USEPA (Elliott et al., 2002; Makris et al., 2006; Sarkar et al., 2007). In several states, WTRs are used as soil amendments or as a soil conditioner and to prevent excess P from leaching into surface water bodies. Benefits associated with use of WTRs as a soil amendment include improved soil structure (El-Swaify and Emerson, 1975), increased moisture-holding capacity (Bugbee and Frink, 1985), and increased availability of nutrients for various plants (Heil and Barbarick, 1989). Many studies have reported field application of the WTRs for agronomic and ecological benefits (e.g., Jacobs and Teppen, 2000; Agyin-Birikorang et al., 2007).

Previous research has demonstrated the high affinity of WTRs for other contaminants of environmental and public health concern, such as arsenate, phosphate, perchlorate, selenium, and mercury (Miller et al., 2011; Nagar et al., 2010; Ippolito et al., 2009; Hovsepyan and Bonzongo, 2009; Sarkar et al., 2007; Makris et al., 2004; Makris et al., 2006a, 2006b). Additionally, Makris et al. (2006a, 2006b) have shown that the toxicity characteristics leaching procedure (TCLP) values of WTRs are well below the hazardous waste toxicity characteristics criterion as defined in Title of the Code of Federal Regulations, part 261.24, permitting land application of WTR as an environmentally safe disposal method.

1.1 Research Objectives

The first objective of the study was to evaluate the effectiveness and efficiency of Al-WTR as a sorbent to remove TTC and OTC from an aqueous medium and study the

effect of solution properties (pH, ionic strength [IS], and sorbate: sorbent ratio [SSR]), reaction time, and initial TTC and OTC concentration on sorption of TTCs by Al-WTR.

The second objective of the study was evaluating the effect of competing ligands (phosphate P(V) and sulfate) and complexing metal (calcium) on TTC and OTC sorption envelopes at optimum SSR as a function of pH and different TTC/OTC ratios, and determine the effectiveness of Al-WTR to remove TCs from aqueous media in the presence of competing ligands (phosphate P(V) and sulfate) and complexing metal (calcium) under different conditions. After completion of the batch study, an incubation study was carried out.

The third objective of the study was to determine TTC and OTC sorption isotherm and adsorption kinetics in TTC/OTC rich cattle manure and two physico-chemically variant manure amended soil types from Immokalee and Belleglade series, respectively mixed with two Al-WTR application rates. Further the objective was to evaluate effectiveness of Al-WTR as a sorbent to immobilize and stabilize TTC and OTC in TTC/OTC rich manure and manure amended soils as a function of initial antibiotics concentration, reaction time, and the presence of competing agricultural species (phosphate and sulfate). Upon completion of the batch and incubation studies a one year greenhouse study was performed.

The fourth objective of the study was to assess the long term (one year) effect of Al-WTR on TTC and OTC immobilization and stabilization in two soils and manure amended soils with varying physico-chemical properties. Moreover the objective was to

evaluate the effect of Al-WTR on TTC and OTC uptake by plants and TCs concentration in leachates over that period of time.

The present study was aimed to conduct a focused, yet comprehensive "proof of concept" study to evaluate the potential of using the WTR-derived sorbent to treat manure and manure-amended soils to immobilize the most commonly used forms of TCs in the US cattle industry. The study will lead to development of a sustainable, inexpensive, in-situ remediation strategy for TCs-rich manures and manure-amended soils, which will help to reduce potential associated risk of TCs in soil-water systems and thus lower their impact on human and aquatic habitat health.

1.2 Organization of thesis

      The above-mentioned research objectives were accomplished and the results and research findings were organized in the form of various chapters in this dissertation. Each chapter covers one objective as follows:

- Chapter 2 entitled "Effectiveness of Aluminum-based Drinking Water Treatment Residuals as a Novel Sorbent to Remove Tetracyclines from Aqueous Medium", effectiveness and efficiency of Al-WTR as a sorbent to remove TTC and OTC from an aqueous medium and study the effect of solution properties (pH, ionic strength [IS], and sorbate: sorbent ratio [SSR]), reaction time, and initial TTC and OTC concentration on sorption of TTCs by Al-WTR. This study also reported the results of surface complexation modeling and Attenuated Total Reflectance-Fourier Transform Infrared (ATR-FTIR) experiments to predict potential mechanism of TTC and OTC adsorption at surface of Al-WTR

- Chapter 3 entitled "Effect of Solution Properties, Competing Ligands, and Complexing Metal on Sorption of Tetracyclines on Al-Based Drinking Water Treatment Residuals", evaluating the effect of competing ligands (phosphate P(V) and sulfate), and complexing metal (calcium) on TTC and OTC sorption envelopes at optimum SSR as a function of pH and different TTC/OTC ratios. The study also determined the effectiveness of Al-WTR to remove TCs from aqueous media in the presence of competing ligands (phosphate P(V) and sulfate), and complexing metal (calcium) under different conditions.

➢ Chapter 4 entitled "Al-based drinking water treatment residuals as an effective low-cost sorbent for immobilization and stabilization of tetracyclines in manure and manure amended soils: A short term incubation study", evaluated TTC and OTC sorption isotherm and adsorption kinetics in TTC/OTC rich cattle manure and two physico-chemically variant manure amended soil types from Immokalee and Belleglade series, respectively mixed with two Al-WTR application rates, and effectiveness of Al-WTR as a sorbent to immobilize and stabilize TTC and OTC in TTC/OTC rich manure and manure amended soils as a function of initial antibiotics concentration, reaction time, and the presence of competing agricultural species (phosphate and sulfate)

➢ Chapter 5 entitled "Immobilization and Stabilization of Tetracyclines in Soils and Manure Amended Soils: A Long-Term Greenhouse Column Study", aimed to assess the long term (one year) effect of Al-WTR on TTC and OTC immobilization and stabilization in two soils and manure amended soils with varying physico-chemical properties, and evaluate the effect of Al-WTR on TTC and OTC uptake by plants and TCs concentration in leachates over that period of time.

Appendices A, B, C, and D contain detailed experimental data for Chapters 2, 3, 4, and 5, respectively. Appendix E contains pictures from incubation and greenhouse studies. Appendix F is the preface for the journal article published from a portion of chapter 2 in Journal of Environmental Quality (2013) *42, 1449-1459.*

1.3 References

- Aga, D. S. 2008. Fate of pharmaceuticals in the environment and in water treatment systems. CRC Press: Boca Raton, FL.
- Agyin-Birikorang, S., O'Connor, G., Jacobs, L., Makris, K., Brinton, S., 2007. Long-term phosphorus immobilization by a drinking water treatment residual. Journal of Environmental Quality 36, 316-323.
- Allen, H.K., Donato, J., Wang, H.H., Cloud-Hansen, K.A., Davies, J., Handelsman, J., 2010. Call of the wild: antibiotic resistance genes in natural environments. Nature Reviews Microbiology 8, 251-259.
- Baguer, A.J., Jensen, J., Krogh, P.H., 2000. Effects of the antibiotics oxytetracycline and tylosin on soil fauna. Chemosphere 40, 751-757.
- Blackwell, P.A., Kay, P., Boxall, A.B.A., 2007. The dissipation and transport of veterinary antibiotics in a sandy loam soil. Chemosphere 67, 292-299.
- Boxall, A.B.A., Kolpin, D.W., Halling-Sørensen, B., Tolls, J., 2003. Peer Reviewed: Are Veterinary Medicines Causing Environmental Risks? Environmental Science & Technology 37, 286A-294A.
- Bugbee, G.J., Frink, C.R., 1985. Alum sludge as soil amendment: Effects on soil properties and plant growth. New Haven Connecticut Agric. Exp. Stn. Bull. 827.
- Chee-Sanford, J., Aminov, R., Krapac, I., Garrigues-Jeanjean, N., Mackie, R., 2001. Occurrence and diversity of tetracycline resistance genes in lagoons and groundwater underlying two swine production facilities. Applied and Environmental Microbiology 67, 1494-1502.
- Datta, R., Das, P., Smith, S., Punamiya, P., Ramanathan, D.M., Reddy, R., Sarkar, D., 2013. Phytoremediation potential of vetiver grass [Chrysopogon zizanioides (L.)] for tetracycline. *Internatl.* J. Phytorem. 15:343-351
- De Liguoro, M., Cibin, V., Capolongo, F., Halling-Sørensen, B., Montesissa, C., 2003. Use of oxytetracycline and tylosin in intensive calf farming: evaluation of transfer to manure and soil. Chemosphere 52, 203-212.
- Elliott, H.A., Dempsey, B.A., 1991. Agronomic effects of land application of water treatment sludges. J. AWWA, 83 126–131.
- Elliott, H.A., O'Connor, G.A., Lu, P., Brinton, S., 2002. Influence of water treatment residuals on phosphorus solubility and leaching. Journal of Environmental Quality 31, 1362-1369.
- El-Swaify, S.A., Emerson, W. W., 1975. Changes in the physical properties of soil clays due to precipitated aluminum and iron hydroxides: I. Swelling and aggregate stability after drying. Soil Sci. Soc. Am. J., 39, 1056-1063.
- Figueroa, R.A., Leonard, A., MacKay, A.A., 2004. Modeling Tetracycline Antibiotic Sorption to Clays. Environmental Science & Technology 38, 476-483.
- Figueroa, R.A., MacKay, A.A., 2005. Sorption of Oxytetracycline to Iron Oxides and Iron Oxide-Rich Soils. Environmental Science & Technology 39, 6664-6671.
- Gonzalez-Pleiter, M., Gonzalo, S., Rodea-Palomares, I., Leganes, F., Rosal, R., Boltes, K., Marco, E., Fernandez-Pinas, F., 2013. Toxicity of five antibiotics and

their mixtures towards photosynthetic aquatic organisms: Implications for environmental risk assessment. Water Research 47, 2050-2064.

- Halling-Sørensen, B., Nielsen, S.N., Lanzky, P.F., Ingerslev, F., Lutzhoft, H.C.H., Jorgensen. S.E., 1998. Occurrence, fate and effects of pharmaceutical substances in the environment − a review. Chemosphere **36,** 357–393.
- Hamscher, G., Pawelzick, H.T., Hoper, H., Nau, H., 2005. Different behaviour of tetracyclines and sulfonamides in sandy soils after repeated fertilisation with liquid manure, Environ. Toxicol. Chem. 24, 861–868.
- Heil, D.M., Barbarick, K.A., 1989. Water treatment sludge influence on the growth of sorghum-sudangrass. J. Environ. Qual., 18, 292-298.
- Hovsepyan, A., Bonzongo, J.C.J., 2009. Aluminum drinking water treatment residuals (Al-WTRs) as sorbent for mercury: Implications for soil remediation. J. Hazard Mater. 164:73-80.
- Ippolito, J., Barbarick, K., Elliott, H., 2011. Drinking Water Treatment Residuals: A Review of Recent Uses. Journal of Environmental Quality 40, 1-12.
- Ippolito, J.A., Scheckel, K.G., Barbarick, K.A., 2009. Selenium adsorption to aluminum-based water treatment residuals. Journal of Colloid and Interface Science 338, 48-55.
- Jacobs, L.W., Teppen, B.J., 2000. WTR as a soil amendment to reduce nonpoint source pollution from phosphorus-enriched soils. 9 p. *In* Proc. 14th Annual Residuals and Biosolids Management Conference, 27–29 Feb. 2000, MA. CD-ROM, Water Environment Federation, Alexandria, VA.
- Janos, P., Sypecká, J., Mlckovská, P., Kurán, P., Pilarová, V., 2007. Removal of metal ions from aqueous solutions by sorption onto untreated low-rank coal (oxihumolite). Separation and Purification Technology 53, 322-329.
- Ji, L., Chen, W., Duan, L., Zhu, D., 2009. Mechanisms for strong adsorption of tetracycline to carbon nanotubes: A comparative study using activated carbon and graphite as adsorbents. Environmental Science & Technology 43, 2322-2327.
- Kay, P., Blackwell, P., Boxall, A., 2004. Fate of veterinary antibiotics in a macroporous tile drained clay soil. Environmental Toxicology and Chemistry 23, 1136-1144.
- Kemper, N. 2008. Veterinary antibiotics in the aquatic and terrestrial environment.Ecol. Indicators 8:1-13.
- Kolpin, D., Furlong, E., Meyer, M., Thurman, E., Zaugg, S., Barber, L., Buxton, H., 2002. Pharmaceuticals, hormones, and other organic wastewater contaminants in US streams, 1999-2000: A national reconnaissance. Environmental Science & Technology 36, 1202-1211.
- Kumar, K., Gupta, S.C., Chander, Y., and Singh. A.K. 2005a. Antibiotic use in agriculture and its impact on the terrestrial environment. Adv. Agron 87, 1–54.
- Kümmerer, K. 2009. Antibiotics in the aquatic environment − A review − Part I.Chemosphere 75:417-434.

- Makris, K., Harris, W., O'Connor, G., Obreza, T., 2004. Phosphorus immobilization in micropores of drinking-water treatment residuals: Implications for long-term stability. Environmental Science & Technology 38, 6590-6596.
- Makris, K., Harris, W., O'Connor, G., Obreza, T., Elliott, H., 2005. Physicochemical properties related to long-term phosphorus retention by drinking-water treatment residuals. Environmental Science & Technology 39, 4280-4289.
- Makris, K., Sarkar, D., Datta, R., 2006. Evaluating a drinking-water waste by-product as a novel sorbent for arsenic. Chemosphere 64, 730-741.
- Makris, K., Sarkar, D., Datta, R., 2006a. Aluminum-based drinking-water treatment residuals: A novel sorbent for perchlorate removal. Environmental Pollution 140, 9-12.
- Miller, M.L., Bhadha, J.H., O'Connor, G.A., Jawitz, J.W., Mitchell, J., 2011. Aluminum water treatment residuals as permeable reactive barrier sorbents to reduce phosphorus losses. Chemosphere 83, 978-983.
- Moore, P.A. Jr., Daniel, T.C., Sharpley, A.N., Wood. C.W., 1995. Poultry manure management – Environmentally sound options. J. Soil Water Conserv. 50, 321-327.
- Nagar, R., Sarkar, D., Makris, K., Datta, R., 2010. Effect of solution chemistry on arsenic sorption by Fe- and Al-based drinking-water treatment residuals. Chemosphere 78, 1028-1035.
- Oberle, K., Capdeville, M.J., Berthe, T., Budzinski, H., Petit, F., 2012. Evidence for a Complex Relationship between Antibiotics and Antibiotic-Resistant Escherichia Coli: From Medical Center Patients to a Receiving Environment. Environmental Science & Technology 46, 1859-1868.
- O'Connor, G.A., Elliott, H.A., Lu, R., 2002. Characterizing water treatment residuals phosphorus retention. Soil and Crop Science Society of Florida Proceedings 61, 67-73.
- Ostermann, A., Siemens, J., Welp, G., Xue, Q., Lin, X., Liu, X., and Amelung, W.2013. Leaching of veterinary antibiotics in calcareous Chinese croplands, Chemosphere, 91 (7) 928-934.
- Park, S., Choi, K., 2008. Hazard assessment of commonly used agricultural antibiotics on aquatic ecosystems. Ecotoxicology 17, 526-538.
- Prakash, P., Sengupta, A.K., 2003. Selective coagulant recovery from water treatment plant residuals using donnan membrane process. Environmental Science & Technology 37, 4468-4474.
- Rakshit, S., Sarkar, D., Elzinga, E.J., Datta, R.., 2013b. In Situ ATR-FTIR study of Oxytetracycline Sorption on Magnetite . J. Environ. Qual. 42:822-827.
- Rakshit, S., Sarkar, D., Elzinga, E.J., Punamiya, P., Datta, R., 2013a. Mechanisms of ciprofloxacin removal by nano-sized magnetite. J. Hazard Mater. 246-247:221-226.

- Sarkar, D., Makris, K., Vandanapu, V., Datta, R., 2007. Arsenic immobilization in soils amended with drinking-water treatment residuals. Environmental Pollution 146, 414-419.
- Sarmah, A. K., Meyer, M. T.; Boxall, A. B. A., 2006. A global perspective on the use, sale, exposure pathways, occurrence, fate and effects of veterinary antibiotics (VAs) in the environment Chemosphere 65, 725-759.
- Sengelov, G., Agerso, Y., Halling-Sorensen, B., Baloda, S. B., Anderson, J. S., Jensen, L. B., 2003. Bacterial antibiotic resistance levels in Danish farmland as a result of treatment with pig manure slurry Environ. Int 28. 587-595.
- Sengelov, G., Agerso, Y., Halling-Sorensen, B., Baloda, S.B., Andersen, J.S., Jensen, L.B., 2003. Bacterial antibiotic resistance levels in Danish farmland as a result of treatment with pig manure slurry. Environment International 28, 587-595.
- Stoob, K., Singer, H.P., Mueller, S.R., Schwarzenbach, R.P., Stamm, C.H., 2007. Dissipation and transport of veterinary sulfonamide antibiotics after manure application to grassland in a small catchment. Environmental Science & Technology 41, 7349-7355.
- Thiele-Bruhn, S., 2003. Pharmaceutical antibiotic compounds in soils - a review. Journal of Plant Nutrition and Soil Science-Zeitschrift Fur Pflanzenernahrung Und Bodenkunde 166, 145-167.
- Tolls, J., 2001. Sorption of Veterinary Pharmaceuticals in Soils: A Review. Environmental Science & Technology 35, 3397-3406.
- Topp, E., Monteiro, S.C., Beck, A., Coelho, B.B., Boxall, A.B.A., Duenk, P.W., Kleywegt, S., Lapen, D.R., Payne, M., Sabourin, L., Li, H.X., Metcalfe, C.D., 2008. Runoff of pharmaceuticals and personal care products following application of biosolids to an agricultural field. Science of the Total Environment 396, 52-59.
- Wang, Q., Yates, S.R., 2008. Laboratory Study of Oxytetracycline Degradation Kinetics in Animal Manure and Soil. Journal of Agricultural and Food Chemistry 56, 1683-1688.
- Winckler, C., Grafe, A., 2001. Use of veterinary drugs in intensive animal production Evidence for persistence of tetracycline in pig slurry. Journal of Soil Sediment - 1, 67 - 70.
- Wollenberger, L., Halling-Sorensen, B., Kusk, K.O., 2000. Acute and chronic toxicity of veterinary antibiotics to Daphnia magna. Chemosphere 40, 723-730.
- Zhang, L., Song, X.Y., Liu, X.Y., Yang, L.J., Pan, F., Lv, J.N., 2011. Studies on the removal of tetracycline by multi-walled carbon nanotubes. Chemical Engineering Journal 178, 26-33.
- Zhang, Y., Zhang, C., Parker, D. B., Snow, D. D., Zhou, Z., Li, X., 2013. Occurrence of antimicrobials and antimicrobial resistance genes in beef cattle storage ponds and swine treatment lagoons. Sci Total Environ. 463-464C:631-638.

- Zhao, Y.P., Gu, X.Y., Gao, S.X., Geng, J.J., Wang, X.R., 2012. Adsorption of tetracycline (TC) onto montmorillonite: Cations and humic acid effects. Geoderma 183, 12-18.

CHAPTER 2

Effectiveness of Aluminum-based Drinking Water Treatment Residuals as a Novel Sorbent to Remove Tetracyclines from Aqueous Medium

Abstract

Low levels of various veterinary antibiotics (VAs) have been found in water resources across the United States as a result of point and nonpoint-source pollution. As the first phase of developing a potential green sorbent for tetracycline (TTC) and oxytetracycline (OTC), we examined the effects of solution chemistry, pH, ionic strength (IS), sorbate:sorbent ratio (SSR), and reaction time on TTC and OTC sorption by a waste byproduct of the drinking-water treatment process, namely, Al-based drinking-water treatment residuals (Al-WTR). The sorption of TTC and OTC on Al-WTR increased with increasing pH up to pH 7 and decreased in the pH range of 8 to 11. A concentration of 20 g L^{-1} was deemed as optimum SSR, where more than 95% of the initially added TTC and OTC were sorbed and equilibrium was reached in 2 h. A pseudo–second-order model (R^2 = 0.99) was used for Al-WTR sorption for TTC and OTC. The data best fit the linearized form of the Freundlich isotherm (R^2 = 0.98). No significant effect ($p > 0.05$) of IS on sorption of TTC and OTC was observed between 0.05 and 0.5 mmol L^{-1}. However, at higher initial concentrations (>1 mmol L^{-1}), IS dependence on TTC and OTC sorption was observed. Surface complexation modeling and Fourier transform infrared spectroscopy analysis indicated the possibility of TTC and OTC forming a mononuclear

monodentate surface complex through strong innersphere-type bonds on Al-WTR. The results show promising potential of Al-WTR for use as a "green" and cost-effective sorbent to immobilize and stabilize TTC in soils and waters.

2. Introduction

A nationwide reconnaissance study carried out by the U.S. Geological Survey showed presence of low levels of pharmaceuticals - veterinary antibiotics (VAs), hormones, and other organic contaminants in water systems from a network of 139 streams across 30 states (Kolpin et al., 2002). Veterinary antibiotics are being used increasingly to protect the health of farm animals and also to accelerate their growth (Blackwell et al., 2007; Boxall et al., 2003). Studies have shown that as much as 25 to 75 percent of the VAs administered orally may pass through the alimentary canal of cattle unchanged (Loffler et al., 2005; Kumar et al., 2005). Once excreted in urine and manure, VAs can enter into soils, surface water, and/or groundwater via manure applied soils or via manure storage at concentrated animal feeding operations (CAFOs). As a result, VAs are being considered emerging contaminants of concern to the drinking water supplies, aquatic ecosystems, and terrestrial ecosystems (Kümmerer, 2009; Aga, 2008; Kemper, 2008; Sarmah et al., 2006). This is because of the fact that even at ng L^{-1} levels, these molecules are biologically active and can affect critical development stages and endocrine systems of aquatic and terrestrial organisms (Aga, 2008; Daughton et al., 1999; Levy, 1998). Also, the widespread use and frequent detection of VAs in the environment have raised concerns over proliferation of antibiotic-resistant bacteria, decrease in the effectiveness of medical antibiotics, and other potential adverse human health and ecological effects (Oberle et al., 2012; Allen et al., 2010; Agersø et al., 2006; Thiele-Bruhn et al., 2003).

The tetracycline group of antibiotics ranks second in production and usage worldwide, with tetracycline (TTC) and oxytetracycline (OTC) being most popular for cattle in the US (Levy, 2002). Tetracyclines (TCs) are complex organic compounds with unique chemical characteristics and behaviors. The TC structure contains connected ring systems with multiple ionizable functional groups (Figure 2-1A). Tetracyclines have three pK_a's and hence, can exist as cationic, zwitterionic, or anionic species under acidic, neutral, and alkaline conditions, respectively (Figure 2-1B). The ionization behavior can be expected to significantly influence the sorption of TCs to soil components and other sorbents. The high polarity (e.g., log K_{ow} = -1.97 to -0.47) and high aqueous solubility (0.52−117 mM) of TCs portend that TCs could be highly mobile (Kulshrestha et al., 2004; Thiele-Bruhn et al., 2003). Thus, effective means of immobilizing TCs in soils need to be employed to reduce potential health hazards that could result from presence of TCs in the soil environment, and consequently in surface and ground water.

Considerable efforts are being made to develop cost-effective treatment technologies to remove VAs from aqueous solutions. In order to find an optimal method for adsorption of TCs and other VAs, sorbents such as soils, clays, minerals, humic acids, metal oxides (Fe/Al hydroxides), carbon nanotubes, and plants have attracted great interest (Figueroa et al., 2004; Figueroa and Mackay, 2005; Gu and Karthikeyan, 2005; Gu et al., 2007; Pils and Laird, 2007; Ji et al., 2009; Rakshit et al., 2013a Rakshit et al., 2013b; Zhang et al., 2011; Zhao et al., 2012; Datta et al., 2013). The use of adsorbent materials provides an attractive alternative for the treatment of contaminated waters, especially if the sorbent is inexpensive and does not require an additional pre-treatment

step before its application (Janos et al., 2007). However, the preparation procedures of some of these adsorbents are complicated and costly, which hinders their application in the real world.

In the current study, we evaluated the potential of drinking-water treatment residuals (WTRs) as a "green" sorbent and an inexpensive surrogate for Al-hydro(oxides), thus giving

a new life to a waste by-product which is generated in millions of tons every day in water treatment facilities in the US (Prakash and Sengupta, 2003) and typically landfilled. WTRs are a by-product of the drinking water treatment process and can be obtained free of charge from drinking water treatment facilities. The WTRs are primarily amorphous masses of aluminum (Al) or iron (Fe) hydroxides or Calcium ($CaCO_3$), referred to as Al-WTRs (use of Al salt), Fe-WTRs (Fe salt), or Ca-WTRs ($CaCO_3$) (O'Connor et al., 2001; Ippolito et al., 2011), that also contain sediment and humic substances removed from the raw water, as well as activated carbon and polymers (Elliott and Dempsey, 1991; Makris et al., 2005; Ippolito et al., 2011). Previous research has demonstrated the high affinity of WTRs for other contaminants of environmental and public health concern, such as arsenate, phosphate, perchlorate, selenium, and mercury (Miller et al., 2011; Nagar et al., 2010; Ippolito et al., 2009; Hovsepyan and Bonzongo, 2009; Sarkar et al., 2007; Makris et al., 2004; Makris et al., 2006a and 2006b). Additionally, Makris et al, (2006a and 2006b) have shown that the toxicity characteristics leaching procedure (TCLP) values of WTRs are well below the hazardous waste toxicity
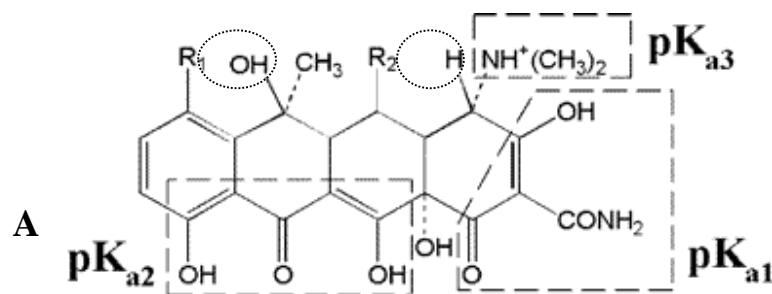
Antibiotic	R1	R2	pK_{a1}	pK_{a2}	pK_{a3}
TTC	H	H	3.3	7.7	9.7
OTC	H	OH	3.37	7.49	9.88
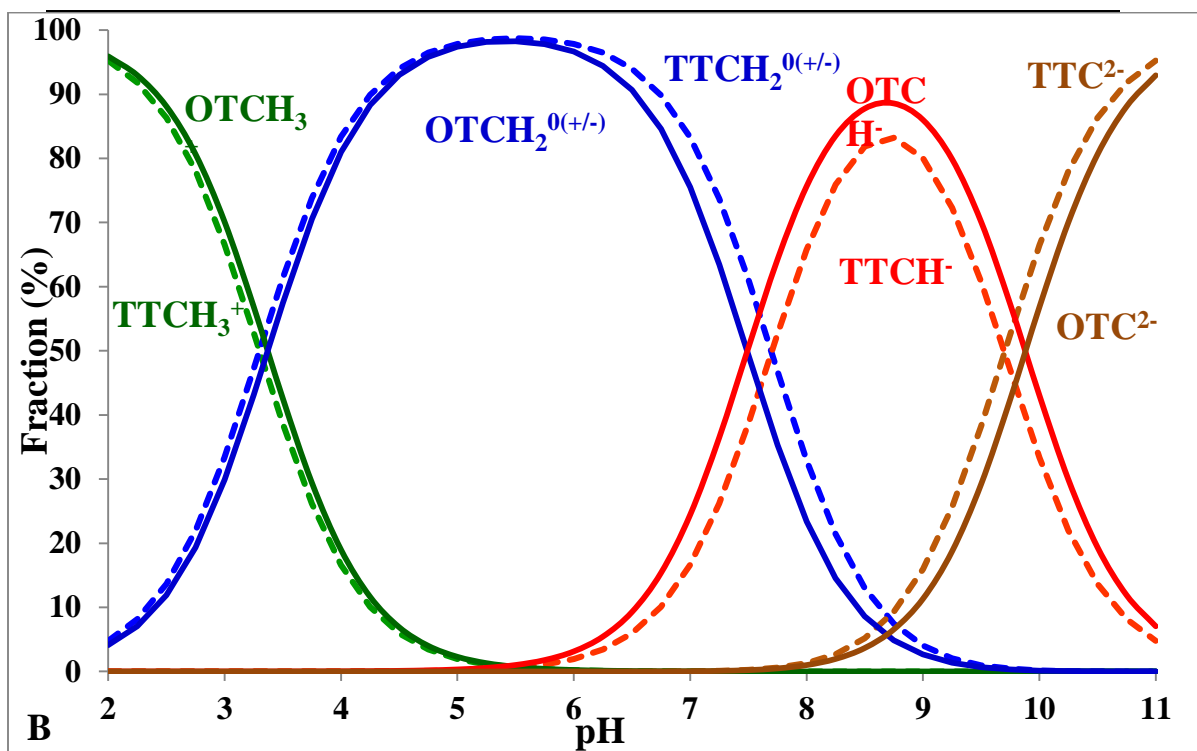
Figure 2-1: Structure (A) and pH dependent surface speciation (B) of TTC and OTC. The graph is plotted using the log K values (Gu and Karthikeyan, 2005; Figueroa and Mackay, 2005) of TTC and OTC, where $OTCH_3^+$, $OTCH_2^{0\,(+/-)}$, $OTCH^-$, and $OTCH^{-2}$ represent different species of OTC and $TTCH_3^+$, $TTCH_2^{0\,(+/-)}$, $TTCH^-$, and $TTCH^{-2}$ represent different species of TTC.

characteristics criterion as defined in Title of the Code of Federal Regulations (CFR), part 261.24, permitting land application of WTR as environmentally safe disposal method.

The main objectives of this study were i) to evaluate the effectiveness and efficiency of Al-based WTR as a sorbent to remove TTC and OTC from an aqueous medium; ii) to study the effect of solution properties (pH, ionic strength (*IS*), and sorbate: sorbent ratio (SSR)), reaction time, and initial TTC and OTC concentration on sorption of TCs by Al- WTR; and iii) to study the release potential of TTC and OTC from the spent WTR to determine possibility of TCs leaching.

2.1 Materials and Methods

2.1.1 Reagents and Materials

Tetracycline hydrochloride and oxytetracycline hydrochloride (USP grade, ≥98%) were obtained from Sigma-Aldrich chemical (St. Louis, MO). Trace-metal grade hydrochloric acid, sodium hydroxide, potassium chloride, and oxalic acid, all certified ACS grade were purchased from Fisher Scientific (Fair Lawn, NJ). Methanol and acetonitrile (HPLC grade, ≥99%) were purchased from Fisher Scientific (Fair Lawn, NJ). 1,4-piperazinebis(ethane sulfonic acid (PIPES) was purchased from Sigma–Aldrich (St. Louis, MO). The disodium salt of ethylenediaminetetraacetic acid (EDTA) was obtained from Acros Organics (Morris Plains, NJ). All solutions and working standards were freshly prepared in high-purity water (18 MΩ-cm, Barnstead nano-pure Diamond, Dubuque, IA).

2.1.2 Analytical method

A Finnigan surveyor plus HPLC system (Thermo Scientific, Somerset, NJ) equipped with quadruple pumps coupled with surveyor PDA plus detector (photodiode array) and a surveyor plus auto-sampler were used for all the analyses. A hypersil gold C_{18} column 150 x 4.6mm, 5μm (Thermo Scientific) with a corresponding hypersil gold guard column 10 x 4mm, 5μm (Thermo Scientific) at room temperature was used for all separations. Samples were eluted isocratically with a mobile phase consisting of 0.01 M aqueous oxalic acid: acetonitrile: methanol (150:20:20 by volume) (Fritz and Zuo, 2007). The mobile phase was mixed and sonicated for 5 min before use. The flow rate was maintained at 1.5 ml/min, with an injection volume of full loop (25 μl), while the UV detector was set at 360 nm. Linear calibration was used for quantification based on the curves between the concentration and peak area of known standard of TCs. Metal concentrations were quantified using ICP-MS (Thermo Electron, San Hose, CA). Surface charge measurements were performed using a Zeta Sizer nano-series ZS90 (Malvern, United Kingdom). FTIR spectra were acquired using Thermo Nicolet 4700 spectrometer (Thermo Scientific, Somerset, NJ) equipped with an attenuated total reflection (ATR) accessory with Zn-Se cell. The spectra were obtained accumulating 256 scans at a resolution of 4 cm^{-1} in range of 4000 – 850 cm^{-1}. In brief, samples from batch sorption experiments with higher TTC/OTC (5.0 x 10^{-3} M) concentration were reacted with 20 g L^{-1} Al-WTR. After centrifugation and filtration, samples were freeze dried and used for analysis. Reference spectra for TTC, OTC, and Al-WTR were also obtained.

2.1.3 WTR collection and characterization

Al-WTR was used in the batch sorption study. The Al-WTR was obtained from a drinking-water treatment plant in Bradenton, Florida. The WTR samples were air-dried and were subsequently passed through a 2-mm sieve before analyses. WTR samples were characterized for several physico-chemical properties as earlier discussed in Nagar et al. (2009). In brief, solution pH, electric conductivity, and organic matter content were measured using standard protocols (Ben-Dor and Banin, 1989, Hanlon et al., 1997a, 1997b). Total C and N were determined by combustion at 1100°C using an Elementar Vario EL CHNS/O analyzer (Elementar, Mount Laurel, NJ). Oxalate-extractable Fe and Al concentrations were determined by ICP-MS after extraction with Tamm's reagent (Loeppert and Inskeep, 1996). The TCLP was determined using USEPA SW-846 Method 1311 to ensure that the toxicity characteristics concentration of several metals and metalloids were below the EPA threshold limit for their safe use and land application.

2.1.4 Batch sorption experiments

All batch sorption experiments were carried out using 0.01M KCl as the background electrolyte except for the *IS* dependent sorption studies. The pH values of all the solutions were checked before and after the sorption experiment. Suspensions in the test tubes were shaken end-over-end on a reciprocating shaker at 250 rpm. The samples were centrifuged (4000 x g) followed by filtration through 0.25µm syringe filters. Sample

tubes were wrapped in Al-foils to inhibit photo-degradation of TCs. The concentrations of TTC and OTC in the filtrates were determined by using HPLC.

2.1.4.1 Effect of solid: solution ratio

Seven different solid: solution ratios (SSRs) ranging from 1 g L^{-1} to 100 g L^{-1} (1, 5, 10, 15, 20, 40, 75, and 100 g L^{-1}) (Al WTR: TTC/OTC) were employed to study the effect of the SSR on TCs sorption. The SSRs were based on the values from the previous studies on various environmental contaminants (Nagar et al., 2010; Sarkar et al., 2007; Makris et al., 2006a). TTC and OTC solutions were prepared by dissolving tetracycline hydrochloride and oxytetracycline hydrochloride, respectively in 0.01 M KCl (background solution) to achieve concentrations of 0.1, 0.5 and 1.0 mM. The concentrations represent the mean TCs concentration in the soil and water environment, 5 times higher, and 10 times higher than the mean concentration, respectively (Hamscher et al., 2005). Sorption of TCs was initiated by mixing Al-WTR with 50 ml of TTC/OTC solutions in 50-mL polypropylene centrifuge tubes to provide the desired SSRs. This resulted in a matrix of WTR x 7 SSR's x 3 TTC/OTC concentrations x 3 replicates, plus respective controls. The pH was maintained at 6.0 ± 0.2, value based on previous reports with Al hydroxides (Gu and Karthikeyan, 2005) with 0.01 M PIPES buffer. Sorbed TTC/OTC was inferred from the difference between the concentration of TTC/OTC added in the initial solution and the concentration of TTC/OTC in the solution at equilibrium.

2.1.4.2 Sorption kinetics experiment

Time intervals ranging from 5 min to 96 h (5min, 10 min, 20 min, 30 min, 1 h, 2 h, 5 h, 10 h, 24 h, 48 h, and 96 h) were examined to determine the effect of reaction time on sorption of TCs by Al-WTR. Al-WTR and TTC/OTC solutions (0.1, 0.5, and 1 mM TTC/OTC prepared in 0.01 M KCl) were added in to each test tube to make up the optimum SSR determined from the previous SSR experiment. The samples were withdrawn at desired time intervals, centrifuged, and filtered. Filtered solutions collected at each time interval were then analyzed for TCs using HPLC.

Following the sorption kinetics experiment, extraction treatments were conducted on the above TTC/OTC reacted samples. The centrifuge tube, WTR, and the entrained solution were weighed, and extraction was initiated by either adding DI water, KCl solution (10 mM), methanol and 0.25M EDTA (Figueroa and Mackay, 2005) to bring the sample back to the desired solid/solution ratio. The sample solutions were shaken at 250 rpm on a reciprocating shaker for 24 h. The samples were withdrawn, centrifuged, and filtered. The total percentage of TTC/OTC released was calculated by comparing the concentration of TTC/OTC released to the amount of TTC/OTC sorbed.

2.1.4.3 Sorption edge experiments

The effect of pH and *IS* on sorption of TCs on Al-WTR was assessed using sorption edge experiments. The optimized SSR (20 g L^{-1} for Al-WTR) and equilibration time (24 h) were used from the previous sorption experiments. The effect of pH on sorption was studied by determining the amount of the TTC/OTC adsorbed within the pH

range of 3-11 (1 unit increment). The suspensions/mixtures in the tubes were adjusted to

the desired pH values by adding 0.1 M HCl/0.1 M NaOH. The volume of the acid/base

used during the adjustment was recorded and added to the total volume for the use of

final calculations. PIPES (0.01 M) was used as a non-interfering buffer agent. Stock

TTC/OTC solution was added to obtain initial concentration of 0.1, 0.5, and 1.0 mM,

respectively. This experiment protocol was repeated to investigate the effect of *IS* on the

sorption efficiency Al-WTR. KCl was chosen as a background solution. Three *IS's* of

KCl - 0.01M, 0.1M, and 0.5M were employed for this experiment using optimized SSR,

equilibration time, and pH values.

2.1.4.4 Sorption isotherms

Sorption isotherms were obtained to evaluate TCs distribution between the Al-

WTR and the TTC/OTC solution as a function of sorbent concentration. Equilibration

time, SSR's, pH, and *IS* were selected on the basis of the above sorption experiments. A

batch sorption experiment was conducted using similar protocol used in the sorption edge

experiments. Sorption isotherms were obtained by mixing a fixed amount of Al-WTR (20

g L^{-1}) with varying TCs concentrations (0.05, 0.1, 0.2, 0.5, 1.0, and 2.0 mM TTC/OTC)

maintaining optimum pH (6.0 ± 0.2) and varying *IS* (0.01, 0.1, and 0.5 M KCl).

2.1.5 Surface complexation modeling

TTC and OTC sorption on Al-WTR as a function of pH was modeled to predict

possible surface complexation reactions. A diffuse layer model with least number of

input parameters was used. The surface parameters for the Al-WTR used in the model

simulations were obtained from the literature listed in Table 2-2 (Makris et al., 2004).

Data was fitted using a nonlinear optimization protocol (FITEQL 4.0, Herbelin and

Westall, 1999). The surface complexation reactions used for modeling the sorption-edge

of TTC and OTC sorption on WTR are:

$$XOH + TTC^{2-} + H^+ = XTTC^- + H_2O \quad (2\text{-}1)$$

$$XOH + OTC^{2-} + H^+ = XOTC^- + H_2O \quad (2\text{-}2)$$
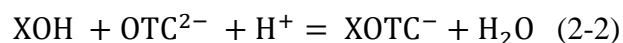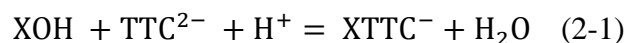
The surface functional group is XOH, where X represents a reactive hydroxyl bound to a

metal ion Al in the oxide mineral, such as, the Al-hydroxide components of WTR. We

only used Al reactive sites in Al-WTR to model the data as Al-sites in Al-WTR are

predominant.

Protonation/deprotonation reactions with equilibrium constants used in the model of TCs

sorption by Al-WTR. The log k values for amorphous Al oxide were obtained from

Goldberg and Johnston (2001). The log k values were used by Nagar et al, (2010) to

successful model sorption of As(V) on Al-WTR using FITEQL 4.0 program.

Additionally, Makris et al, (2004) have also shown that Al-WTR is highly amorphous in

nature.

$$XOH \rightarrow XO^- + H^+ \quad \textit{Log k of Al-WTR (-9.09)} \ (2\text{-}3)$$

$$XOH_2^+ \rightarrow XOH + H^+ \ \textit{Log k of Al-WTR (7.38)} \ (2\text{-}4)$$
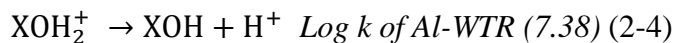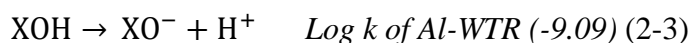
Aqueous speciation of TTC and OTC were also considered following the reactions

below:

$$TTC_T = TTCH_3^+ + TTCH_2^0 + TTCH^- + TTC^{2-} \quad (2\text{-}5)$$

$$\text{TTCH}_3^+ = \text{TTCH}_2^0 + \text{H}^+ \quad log\ ka_1 = \text{-}3.3 \qquad (2\text{-}6)$$

$$\text{TTCH}_2^0 = \text{TTCH}^- + \text{H}^+ \quad log\ ka_2 = \text{-}7.7 \qquad (2\text{-}7)$$

$$\text{TTCH}^- = \text{TTC}^{2-} + \text{H}^+ \quad log\ ka_2 = \text{-}9.7 \qquad (2\text{-}8)$$

$$\text{OTC}_\text{T} = \text{OTCH}_3^+ + \text{OTCH}_2^0 + \text{OTCH}^- + \text{OTC}^{2-} \quad (2\text{-}9)$$

$$\text{OTCH}_3^+ = \text{OTCH}_2^0 + \text{H}^+ \quad log\ ka_1 = \text{-}3.37 \qquad (2\text{-}10)$$

$$\text{OTCH}_2^0 = \text{OTCH}^- + \text{H}^+ \quad log\ ka_2 = \text{-}7.49 \qquad (2\text{-}11)$$

$$\text{OTCH}^- = \text{OTC}^{2-} + \text{H}^+ \quad log\ ka_2 = \text{-}9.88 \qquad (2\text{-}12)$$
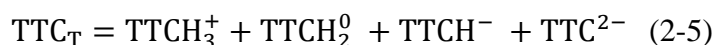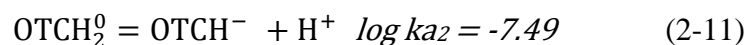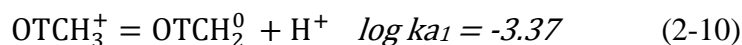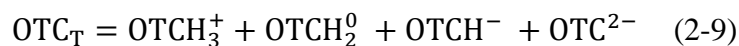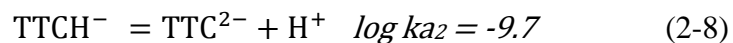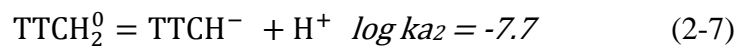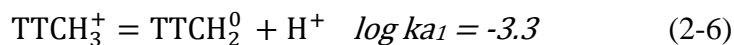
TTC_T and OTC_T in reactions represent all possible solutions species for TTC and OTC, respectively. The log k values of the reaction were obtained from the literature (Gu and Karthikeyan, 2005; Figueroa and Mackay, 2005).

2.1.6 Statistical analysis

Data obtained were statistically analyzed using JMP IN version 9.0 (Sall et al., 2005). To examine the effect of pH, contact time, and initial TCs loads on TCs sorption by Al-WTR two-way analysis of variance (ANOVA) was performed. To evaluate significant differences among treatment means Tukey–Kramer HSD test was used. The data is reported as the mean of three replicates with one standard deviation.

2.2 Results and Discussion

2.2.1 General Chemical Properties of the Al-WTR

Al-WTR used in the study was acidic in nature (Table 2-1). Oxalate (0.2M) extraction was carried out to determine the non-crystalline (amorphous) phase of Al

Table 2-1. Physicochemical properties of Al-based WTR. Data are expressed as the mean of three replicates ± one standard deviation.

	Al-WTR
Source	Bradenton, FL
pH	6.1 ± 0.2
Electrical conductivity (dS m^{-1})	0.84 ± 0.1
Solid organic matter (%)	33 ± q1.4
Carbon (g/kg)	184 ± 4.2
Nitrogen (g/kg)	6 ± 0.5
(Al) Total (g/kg)	115.3 ± 1.7
(Al) Oxalate extractable (g/kg)	98.1 ± 1.2
(Fe) Total (g/kg)	12.5 ± 1.9
(Fe) Oxalate extractable (g/kg)	4.5 ± 1.1

hydroxides. Oxalate-extractable Al concentrations for Al-WTR was in the range of 97.5 to 99.2 g kg^{-1}, close to 85 to 90 % of total Al, which is consistent with the amorphous nature of the Al-WTR reported earlier (Ippolito et al., 2011). The total C values obtained for Al-WTR was in the range of organic C found in various WTRs nationwide (23 to 205 g kg^{-1}) (Dayton and Basta, 2005; Ippolito et al., 2011). The TCLP values (Table 2-3) of Al-WTR were compared to biosolid land application USEPA limits as there are no threshold values set for WTRs. Based on the TCLP results, Al-WTR did not exhibit any hazardous waste characteristics as defined in Title 40 of CFR, Part 261.24 for the parameters evaluated. The TCLP values obtained for Al-WTR are consistent with results documented by other researchers (Makris et al., 2006a and 2006b; Ippolito et al., 2011), which means land application of the WTRs would be permitted as an environmentally sound disposal method.

2.2.2 Effect of Solid: Solution Ratio (SSR) on TTC Sorption by the WTR

Three different initial concentrations of TTC and OTC (0.1, 0.5, and 1.0 mM) and eight different SSRs ranging from 1 to 100 g L^{-1} were used to evaluate the optimum SSR for TTC and OTC sorption by Al-WTR. Results showed significant interaction ($p<0.001$) between the amount of TTC and OTC sorbed at different SSRs load for Al-WTR (Figure 2-2). Al-WTR showed higher sorption affinity for TTC (Figure 2-2A) as compared to OTC (Figure 2-2 B) at all the SSR tested at different initial concentrations. However, there was significant ($p<0.001$) interaction between the SSR and the initial TTC and OTC

Table 2-2. Solid and suspension properties used in modeling TTC and OTC sorption by Al-WTR

Parameter	Al-WTR
Surface area (m^2 g^{-1})[a]	104.9
Site density (site nm^{-2})[a]	5.4
Total surface sites (mol L^{-1}) [b]	0.018
Suspension density (g L^{-1})	20

Note: Initial TTC/OTC concentrations: 0.1, 0.5, and 1.0 mM, background electrolyte: 0.01 M KCl.
[a] Makris et al. (2004).
[b] Calculated using suspension density of Al-WTR in reaction mixture.

Table 2-3. Toxicity characteristics values of several metals and metalloids measured in Al-WTR, using the EPA 1311 TCLP extraction method. The results for Al-WTR are compared to biosolids land application USEPA limits in mg L^{-1} and normalized values to mg kg^{-1}. Data are expressed as mean of three replicates ± one standard deviation.

Analyte	------mg L^{-1}--------		------Normalized to mg kg^{-1}-----	
	Al-WTR	USEPA regulatory limit	Al-WTR	USEPA limit
As	<MDL † (< 0.007)	5.0	<MDL † (< 0.9)	75
Al	204.02	NR*	4082 ± 70.2	15,000§
Cd	0.0007	1.0	0.0153 ± 0.0120	89
Cr	0.011	5.0	2.334 ± 0.0570	3,000
Cu	0.003	10.0	0.566 ± 0.0746	4,300
Fe	0.06	NR	11.85± 0.586	15,000§
Pb	0.0001	5.0	0.0180 ± 0.0109	840
Hg	< 0.05$^{¥}$	0.2	11.0$^{¥}$	57
Ni	0.002	NR	0.408 ± 0.0148	420
Se	< 0.05$^{¥}$	1.0	15.0$^{¥}$	100
Zn	0.01	NR	2.269 ± 0.0963	7,500

†<MDL † (Below Method Detection Limit for As 0.9 mg kg^{-1} using ICP-MS)
§ Al and Fe does not fall under USEPA hazardous waste criteria. The limits are for solid industrial waste.
*NR: Not regulated under the Toxicity Characteristics; values not given by USEPA
$^{¥}$Data obtained from Sarkar et al., (2007)

concentrations on the overall amount of TTC and OTC sorbed by the Al-WTR (Figure 2-2A and B). Increasing the SSR to 20 g L^{-1}, showed no significant difference in the total amount of TTC and OTC sorbed up to SSR of 100 g L^{-1}. 20 g L^{-1} was deemed to be optimum SSR for Al-WTR where more than 95% of the initially added TTC (98%) and OTC (96%) concentration were sorbed at all the three concentrations (0.1, 0.5, and 1.0 mM) tested. (Figure 3). The seen behavior can be explained by greater external and internal specific surface area of the Al-WTR and due to sorption of TTC/OTC in the macropores and micropore of Al-WTR (Makris et al., 2004 and 2006). Similar nature of Al-WTR and sorption behavior has been reported by Makris et al. (2006). Also, in previous reports, similar sorption behavior by Al hydrous oxides for TCs removal from aqueous medium have been observed (Gu and Karthikeyan, 2005; Chen, and Huang; 2010). Based on above TTC and OTC, SSR experiments, we selected 20 g L^{-1} as the optimum SSR. The optimum SSR value was further used in the sorption edge, sorption isotherm, and sorption kinetic experiments.

2.2.3. Sorption edge experiment

The control experiment showed no significant ($p>0.05$) change in the initially added TCs concentration (recovery > 97%) over the entire pH range (3-11) tested (data not shown). Therefore, no loss of TCs is assumed to have occurred owing to sorption to the glassware, photochemical and/or the other reactions in solution under experimental conditions. There was no significant changed observed in the pH value after the sorption
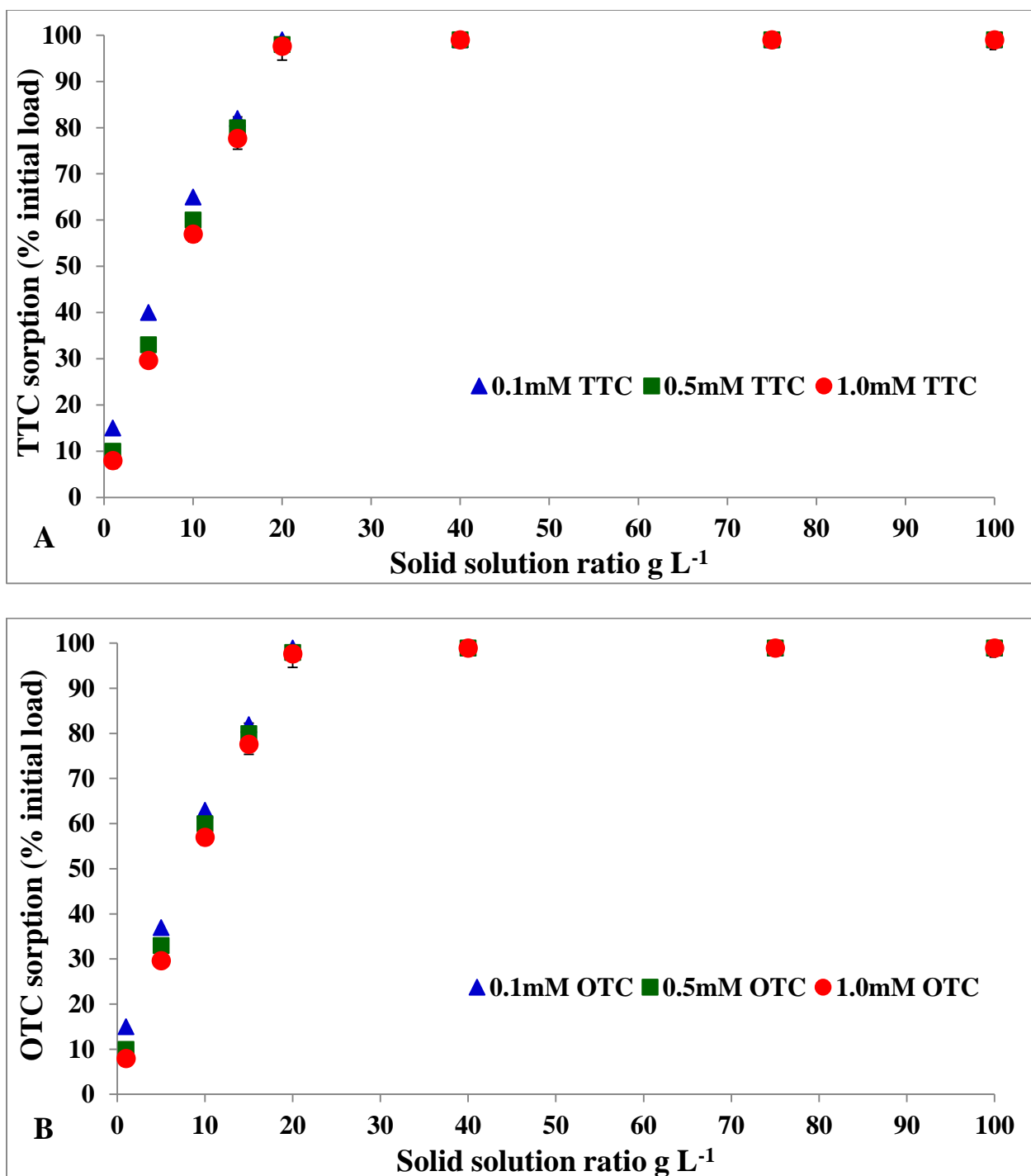
Figure 2-2: Effect of SSR on the amount of TTC (3A) and OTC (3B) sorbed by Al-WTR as a function of different initial TTC and OTC concentrations (0.1, 0.5, and 1.0 mM). Experimental parameters: reaction time 24 h, ionic strength 0.01 M KCl, and pH 6.0 ± 0.1. Data are expressed as mean of three replicates ± one standard deviation.

of TTC/OTC on Al-WTR. The observed change was less than ± 0.2 initial pH value. The

final pH values were recorded and used in the experiment. PIPES buffer at 0.01M

concentration was use to maintain the pH of the solution. Sorption behavior of TTC

(Figure 2-3A) and OTC (Figure 2-3 B) onto Al-WTR showed pH dependence at the

optimum SSR and all the TTC and OTC concentration tested (0.1, 0.5, and 1.0 mM).

Sorption of TTC and OTC on Al-WTR increased from 85% to 97% and 82% to 95%,

respectively with increase in pH from 3 to 7. However, increasing the pH above 8, the

sorption significantly ($p<0.05$) decreased for both TTC (97 % to 76 %) and OTC (95% to

72%) (Figure 2-3 A and B). At all the three TTC and OTC

concentrations tested (0.1, 0.5, and 1.0 mM), more than 90% of the initially added TTC

(97%) and OTC (95%) was sorbed by Al-WTR from the solution in the pH range 5-7.

The TTC and OTC sorption edge obtained in the current study with Al-WTR is similar to

those of previous studies on Al hydrous oxides. (Gu and Karthikeyan, 2005; Chen and

Huang, 2010). The pH dependence sorption showed by Al-WTR is consistent with pH-

dependent surface speciation of TTC and OTC (Figure 2-1B), and surface charge

characteristics of Al-WTR (Figure 2-4). This was verified by plotting different species of

TTC, OTC, and Al-WTR from their respective log K and pK_a values (Gu and

Karthikeyan, 2005; Figueroa and MacKay, 2005; Nagar et al., 2010). Tetracyclines have

three pKa's, hence, they can exist as cationic, zwitterionic, or anionic species under

acidic, moderately acidic to neutral, and alkaline conditions, respectively. In the pH range

of 3.3 to 7.7, TTC and OTC exists as zwitterions and therefore the net charge on the

surface is zero (+/-). The pH zpc (zero point of charge) for Al-WTR was in pH range 8.5
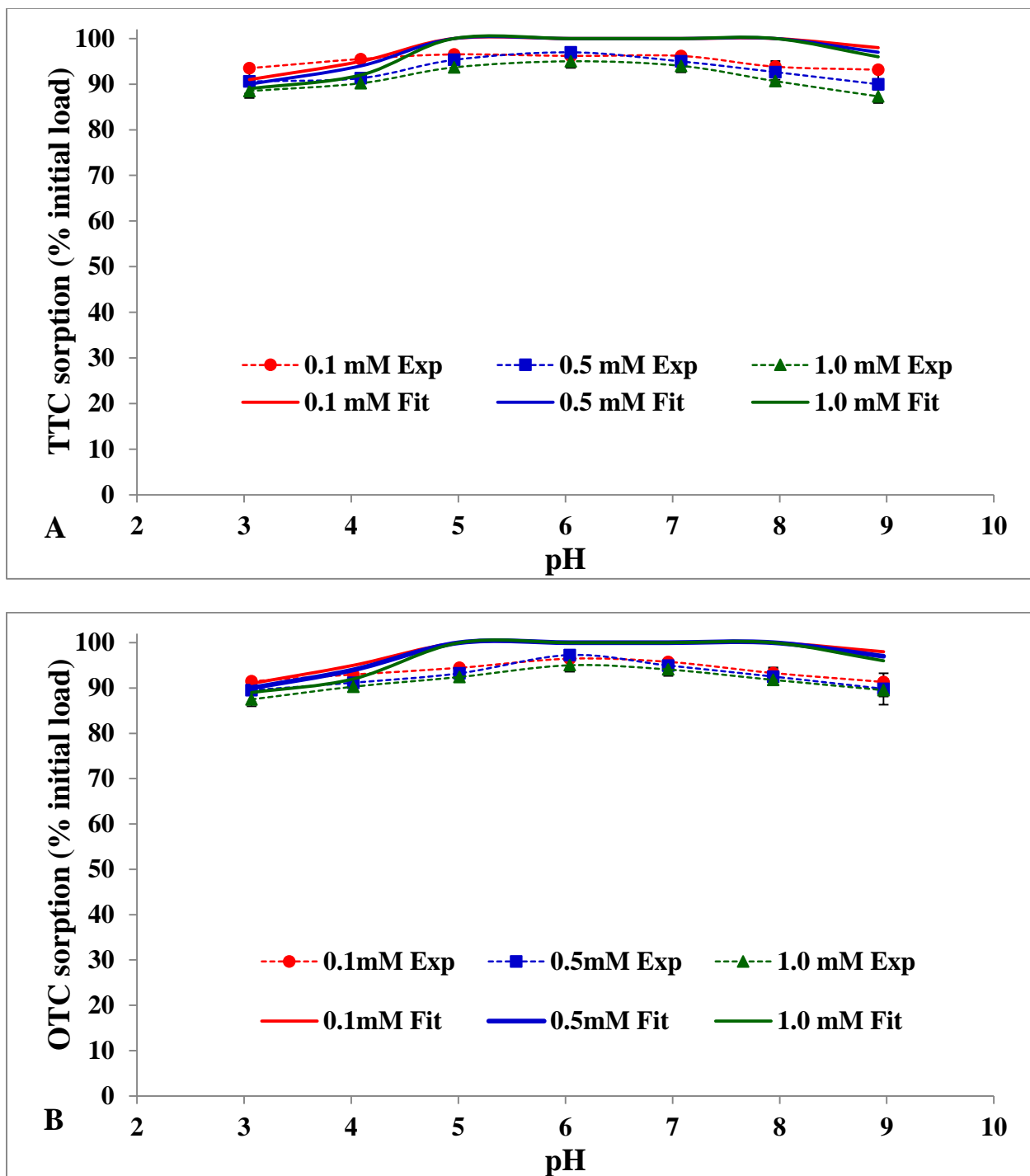
Figure 2-3: Sorption edge of TTC (A) and OTC (B) sorption by Al-WTR as a function of different pH and initial TTC and OTC concentrations (0.1, 0.5, and 1.0 mM). Experimental parameters: reaction time 24 h, ionic strength 0.01 M KCl, and SSR 20 g L^{-1}. Data are expressed as mean of three replicates ± one standard deviation.
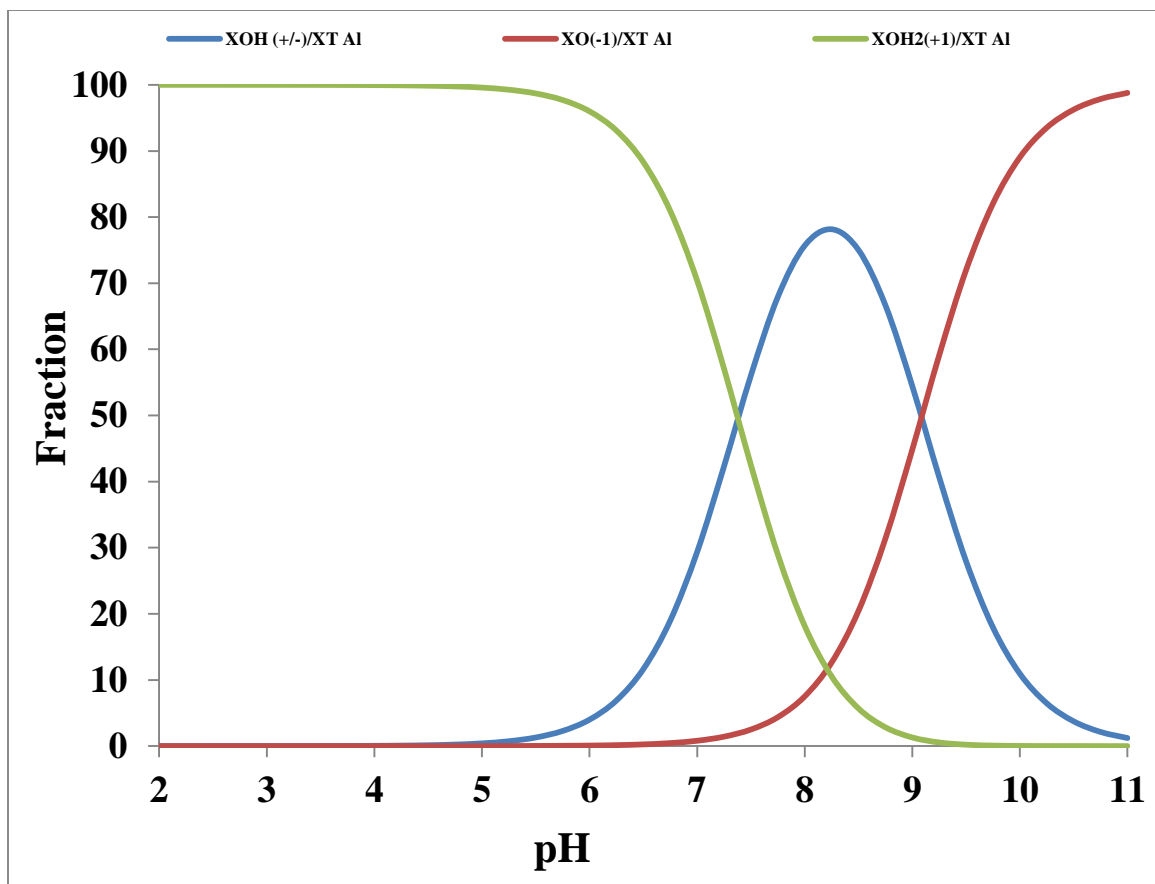
Figure 2-4: pH dependent surface speciation of Aluminum (Al) hydroxide. The graph is plotted using the log K values (Nagar et al., 2010) of Al hydroxide where XOH_2^+, XOH, and XO^-, represent different species of Al. XT indicates total possible surface species of Al.

to 9. The zpc values obtained are consistent with previous reported values for amorphous

Al (Sparks, 1995). The gradual decrease of sorption with increasing pH is consistent

with the proposed surface complexation reactions (reactions 1 and 2), in which proton is

consumed (Figure 2-3A and B). Generally ligand exchange reactions are governed by

strong inner-sphere adsorption and unlikely to be controlled by surface charge. The Al-

WTR is redox stable and hence the surface does not easily disintegrate or lose sites for

sorption. Therefore there is not much decrease in TTC and OTC sorption with increase

in pH above 7 and higher TTC/OTC concentrations. Nagar et al, (2010) reported similar

pH dependent behavior of Al-WTR above pH 7 for sorption of Arsenic.

2.2.4 Ionic strength (IS) effect

No significant effect (p>0.05) of *IS* on sorption of TTC (Figure 2-5A) and OTC

(Figure 2-5B) was observed between 0.05-0.5 mM initial TTC and OTC concentration

for Al-WTR, respectively. However, at the higher initial TTC and OTC concentrations

(>0.5 mM), *IS* dependence on sorption was observed (Figure 2-5). Increasing *IS* from

0.01 to 0.5 M KCl resulted in a pronounced and significant effect ($p<0.05$) on the

sorption of both TTC and OTC (initial concentration > 0.5 mM) by Al-WTR indicating

that the sorption of TTC and OTC on Al-WTR is dependent on surface coverage. The

decrease in the sorption of TTC and OTC with increase in *IS* can be also be attributed to

the competition from K^+ and Cl^- with TCs (at pH 6 TTC/OTC occurs as zwitterions and

the net charge on the surface is zero) (Zhao et al., 2012). At low surface coverage there is

no competitive effect and weak *IS* dependence on sorption, indicating initial complex
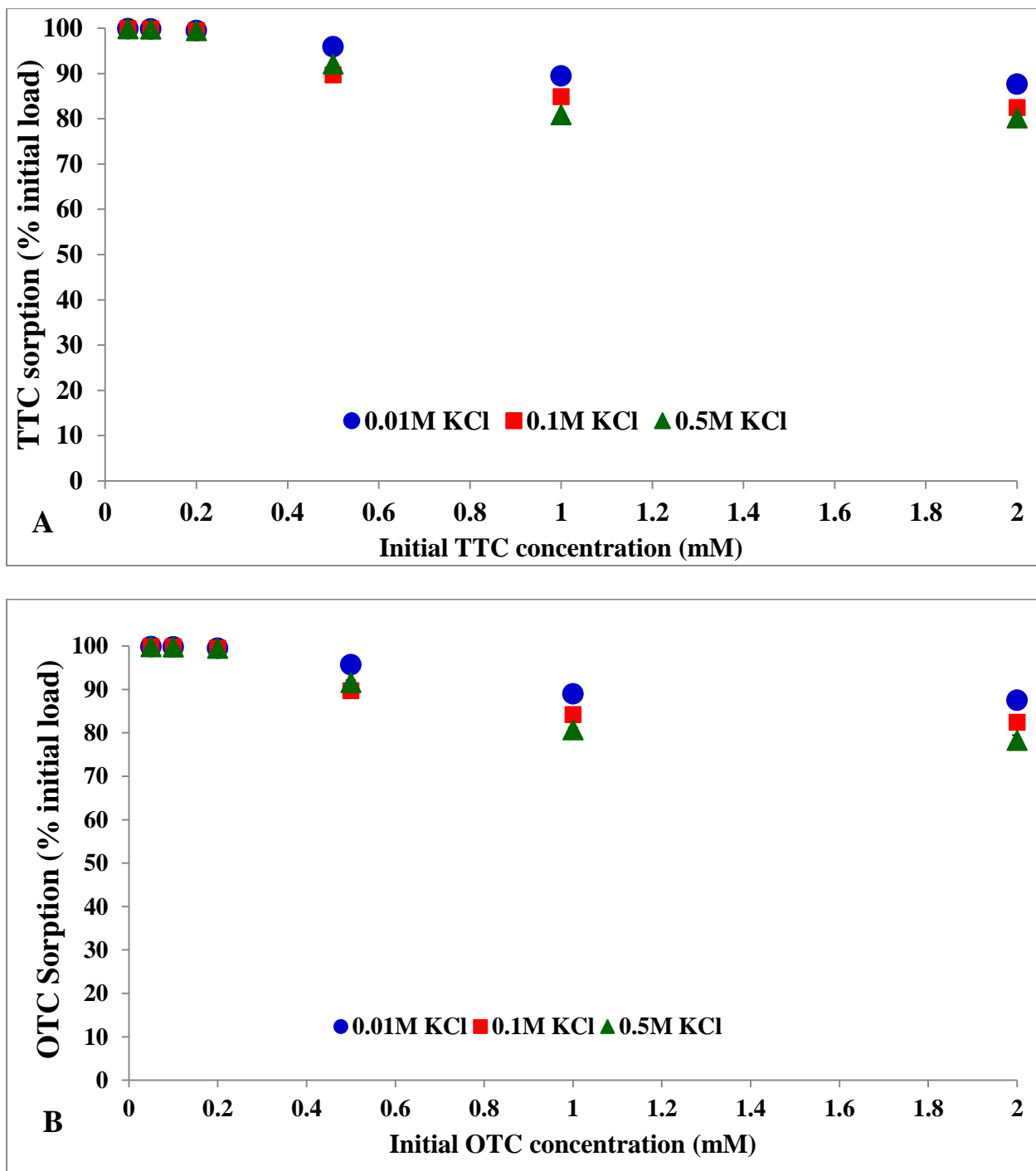
Figure 2-5: Effect of ionic strength on the amount of TTC (A) and OTC (B) sorbed by Al-WTR as a function of different initial TTC and OTC concentrations (0.05 to 2 mM). Experimental parameters: reaction time 24 h, pH 6.0 ± 0.1, and SSR 20 g L^{-1}. Data are expressed as mean of three replicates ± one standard deviation.

formation (Goldberg and Johnston, 2001). However, at the highest surface coverage *IS*

dependence on sorption was observed suggesting possible presence of both inner-sphere

and weak outer-sphere type of complexes.

Isotherm for TTC (Figure 2-6A) and OTC (Figure 2-6B) sorption on Al-WTR

indicates that *IS* effect on TTC and OTC is dependent on surface coverage. The best data

fits for both TTC and OTC by Al-WTR was obtained using the linearized form of the

Freundlich isotherm equation and the parameters are listed in Table 2-4. The distribution

coefficient (K_d) values decreased with increasing *IS* suggesting the effect of K^+ and Cl^- on

the sorption of TTC and OTC as an outer sphere complexes. L-type isotherms were

obtained for both TTC and OTC, indicating high affinity of sorbent (Al-WTR) for the

sorbate (TCs) at low surface coverage, and a decreasing affinity with increasing surface

coverage (Essington, 2004). Further spectroscopic results are required to interpret the

surface sorption mechanism.

2.2.5 Surface complexation modeling

The surface complexation reactions indicated a ligand exchange reaction where a

mononuclear monodentate surface complex was formed. Assumptions of diffuse layer

model predict inner-sphere surface complexation.   The model assumptions can be

justified based on the results from effect of *IS* on TTC and OTC sorption on Al-WTR.

Results show no significant dependence of *IS* on sorption, indicating weak *IS* effect on

sorption. In addition, our IR data indicated inner-sphere nature of the bonding (See

section *2.2.8*).   The fit of the models is shown in Figure 2-3.
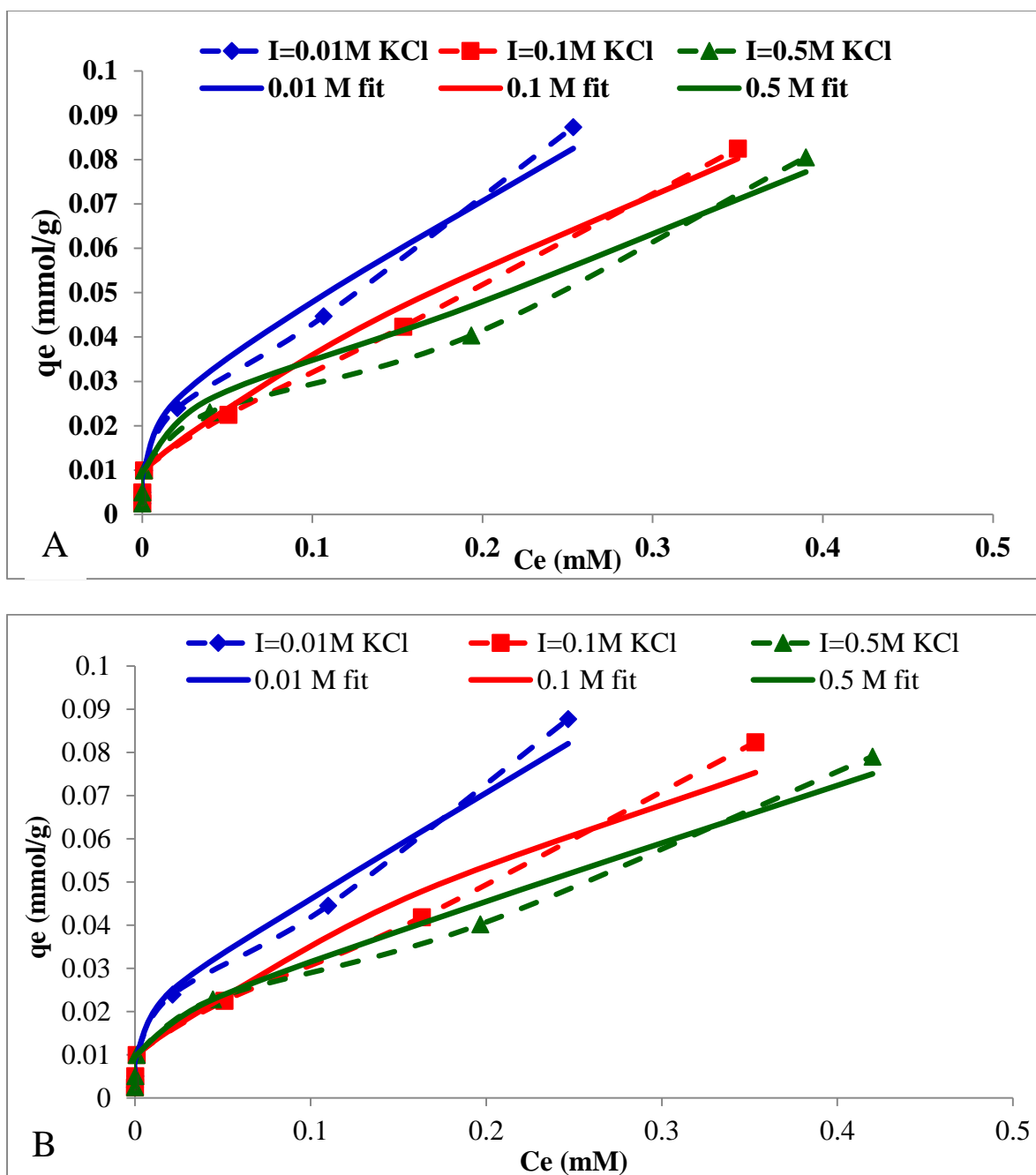
Figure 2-6: TTC (A) and OTC (B) sorption isotherm with simulated Freundlich fit for Al-WTR at different ionic strength (0.01, 0.1 and 0.5 M KCl) as a function of different initial TTC and OTC concentrations. Experimental parameters: reaction time 24 h, pH 6.0 ± 0.1, and SSR 20 g L^{-1}. q_e is the amount of TTC/OTC sorbed onto the Al-WTR in mol g^{-1}; C_e is the equilibrium tetracycline concentration in mM. Data are expressed as mean of three replicates ± one standard deviation.

Table 2-4. Freundlich Isotherm model* parameters for TTC/OTC sorption by Al WTR. pH was maintained at 6.0 ± 0.1, SSR 20 g L^{-1}, and reaction time 24h.

TCs	Ionic Strength (KCl)	K_f	n	N (1/n)	R^2
TTC	0.01 M	0.178	1.81	0.55	0.99
	0.1 M	0.166	1.45	0.68	0.98
	0.5 M	0.130	1.61	0.62	0.98
OTC	0.01 M	0.172	1.74	0.57	0.99
	0.1 M	0.161	1.40	0.71	0.98
	0.5 M	0.127	1.56	0.64	0.98

*Freundlich isotherm: $q_e = K_f \times C_e{}^n$; where q_e is the amount of TTC/OTC sorbed onto the Al-WTR in mol g^{-1}; C_e is the equilibrium tetracycline concentration in mM; and K_f and n are dimensionless Freundlich isotherm constants.

Table 2-5. Pseudo-second-order reaction rate constants in WTR suspensions after a 0.5 and 1.0 mM TTC/OTC initial concentration, pH 6.0 ± 0.1, IS 0.01M KCl, SSR 20 g L^{-1} for Al-WTR and contact time ranged from 0.083 to 48 h.

TCs	WTR Form	TTC mM	1st order rate fit (R^2)	2nd order rate fit (R^2)	2nd order reaction rate k (1 h^{-1} M^{-1})[a]
TTC	Al-WTR	0.5	0.86	0.99	4.019×10^{-3}
TTC	Al-WTR	1.0	0.83	0.99	2.004×10^{-3}
OTC	Al-WTR	0.5	0.83	0.99	4.028×10^{-3}
OTC	Al-WTR	1.0	0.81	0.99	2.015×10^{-3}

[a] Where the slope of a linear fit to a n-order reaction equals: $(n-1) * kn * C_0^{n-1}$

The fit of the model to TTC (Figure 2-3A) and OTC (Figure 2-3B) sorption data on Al-WTR was excellent with mononuclear monodentate surface complex formation. Usually for ligand exchange reaction, more OH^- ions are generated upon adsorption and hence the pH value increases. However, in our experiments, a buffer (0.01M PIPES) is used for controlling the pH value of the system. Therefore, although for adsorption reaction (monodentate surface complexation) the pH is supposed to increase, the buffer kept the pH value nearly constant. In modeling, dinegative species of TTC and OTC were used. Other researchers also modeled OTC adsorption on iron oxide using a single dinegative OTC species (OTC^{2-}) (Figueroa and Mackay, 2005). The modeling exercises were also performed using other species of TTC and OTC, but the mode did not converge. The theoretical insight of this phenomenon can be drawn from the fact that although TTC^{2-} and OTC^{2-} were not the dominant species at lower pH, even a small percentage of this reactive species could have driven the speciation equilibrium of OTC and TTC towards OTC^{2-} and TTC^{2-}, and all other species of TTC and OTC (such as $TTCH_3^+$ / $OTCH_3^+$, $TTCH_2^0$ / $OTCH_2^0$, and $TTCH^-$ / $OTCH^-$) would act as a reservoir of TTC^{2-} and OTC^{2-}. In fact, Wessels et al. (1998) and Dos Santos et al. (2000) noted that surface complexation could force ionization of TC even at low pH.

The log k^{int} for TTC (equation 2-5) and OTC (equation 2-9) sorption on Al-WTR was close to 15 and the error term (WSOS/DF) for the computer program FITEQL 4.0 modeling was less than 20 for both TTC and OTC sorption, indicating a good fit (Herbelin and Westall, 1999). Further studies using spectroscopic technique are required to isolate our assumed surface complex by surface complexation modeling.

2.2.6 Sorption kinetics

Kinetic experiments were carried out to study the effect of reaction time on the sorption of TTC and OTC by Al-WTR. These experiments were conducted under the optimum conditions (SSR, pH, and *IS*) obtained from previously described studies. The kinetic measurements were carried out under maximum sorption capacity conditions. A kinetically-driven TTC (Figure 2-7A) and OTC (Figure 2-7B) sorption was observed for Al-WTR. The TTC and OTC sorption at Al-WTR-water interface was biphasic, showing an initially rapid, followed by a slow sorption rate (Figure 2-7). Sorption of TTC by Al-WTR was nearly complete (>95%) within 2 h regardless of the initial concentration of TTC, and slowly proceeded to 100% sorption by the end of the 48-h period (Figure 2-7A). Similar sorption behavior was observed for OTC by Al-WTR, where more than 90% of initially added OTC was sorbed within 2h and preceded to 100% sorption by the end of the 48-h period (Figure 2-7B).   The sorption behavior of Al-WTR is consistent with the previous reports on sorption of various environmental contaminants (P, As, ClO^{-4}, etc.) by Al-WTR (Nagar et al., 2010; Sarkar et al., 2007; Makris et al., 2006a, 2006b). The high sorption capacity of Al-WTR observed for TTC and OTC is comparable to that of common industrial sorbents such as clays, minerals, humic acids, metal oxides (Fe/Al hydroxides), and carbon nanotubes; suggesting that Al-WTR can be effective sorbent for TCs (Figueroa et al., 2004; Figueroa and Mackay, 2005; Gu and Karthikeyan, 2005; Gu et al., 2007; Ji et al., 2009; Zhang et al., 2011; Zhao et al., 2012).

The kinetic data was fitted to various kinetic models. A pseudo-second-order kinetic model fitted the data best (Table 2-5). The "rapid" phase of TTC (Figure 2-7A)
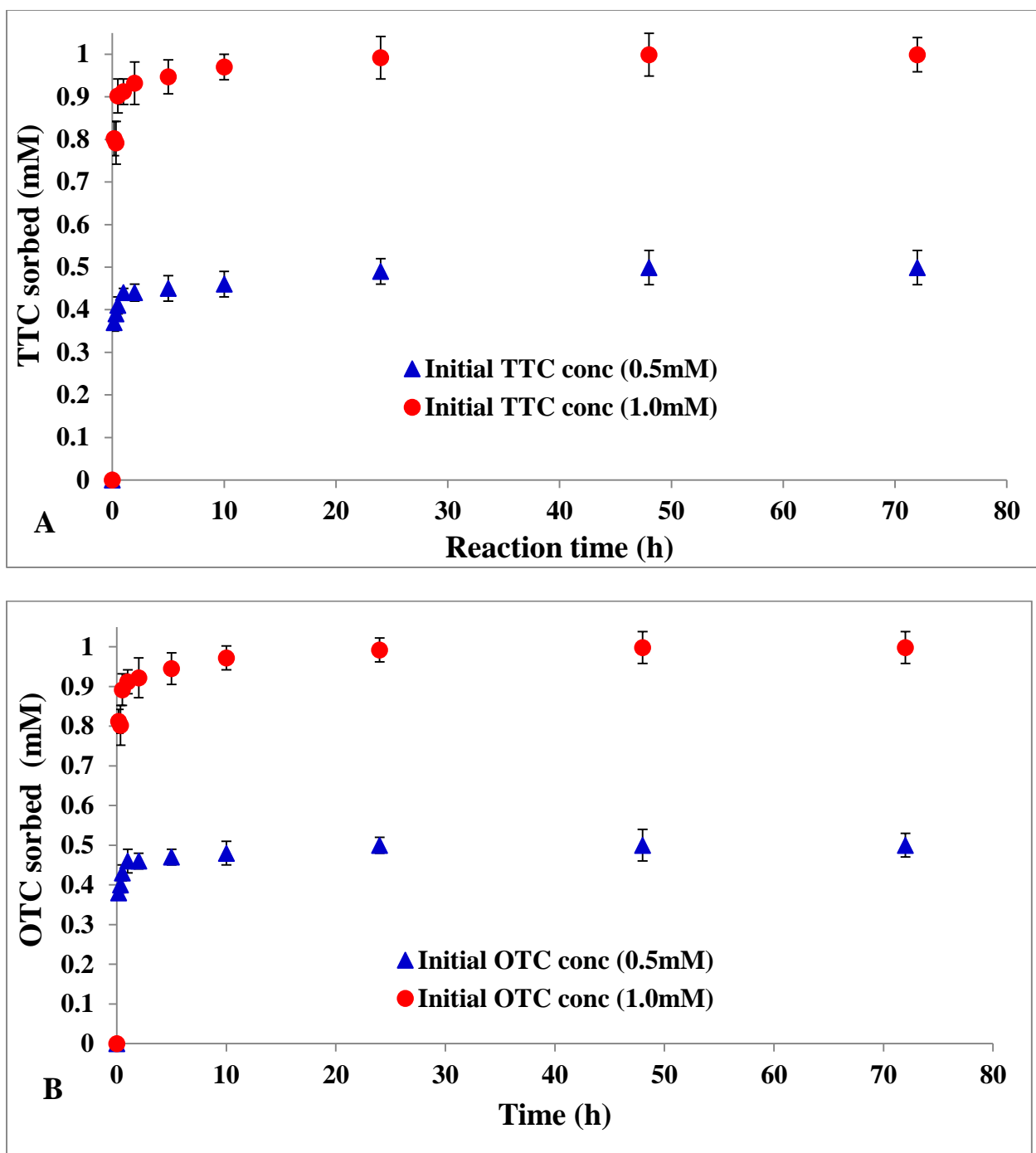
Figure 2-7: Effect of reaction time on the amount of TTC (7A) and OTC (7B) sorbed by Al-WTR as a function of different initial TTC and OTC concentrations (0.5 and 1 mM). Experimental parameters: SSR 20 g L^{-1}, pH 6.0 ± 0.1, and ionic strength 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.

Table 2-6. Comparison of the reaction rate and total surface site available for sorption with current and previous studies.

Study	Sorbent	Reaction time (h)	SSR g L^{-1}	Surface area m^2 g^{-1}	Total surface area m^2 g^{-1}	Total available surface sites
Current	Al-WTR	2	20	104.9	2098	18.8×10^{-3} mol L^{-1}
Gu and Karthikeyan, (2005)	Al-hydrous oxide	8	2	322	644	---
Gu and Karthikeyan, (2005)	Fe- hydrous oxide	8	2	386	772	---
Figueroa and Mackay, (2005)	Goethite	72	10	17.8	178	2.21×10^{-3} mol L^{-1}
Figueroa and Mackay, (2005)	Hematite	24	10	11.1	111	1.54×10^{-3} mol L^{-1}
Rakshit et al., (2010)	Magnetite	>5	20	40	800	6.9×10^{-3} mol L^{-1}

--- (data was not available for calculation from the report)

and OTC (Figure 2-7B) sorption would most probably include highly accessible surfaces (particle exteriors and macropores) (Van Riemsdijk and Lyklema, 1980), while the "slow" phase could be related with diffusion in micropores of the Al-WTR, as was with P and As (Makris et al., 2004; Nagar et al., 2010). The second-order rate coefficient for Al-WTR were higher at 0.5mM than1 mM for both TTC and OTC concentration, consistent with there being less TTC and OTC sorption per unit time for the second biphasic (longer-term) sorption stage (Table 2-5). It is evident from the sorption data that Al-WTRs have high affinity for TTC and OTC regardless of the initial TTC and OTC concentration and the equilibration was rapid (Figure 2-7A and B). A report by Gu and Karthikeyan (2005) on interaction of TTC with Al and Fe hydrous oxides reported an equilibrium sorption time of 8h for 0.1 mM TTC. Figueroa and Mackay (2005) used an equilibration time ranging from 24h-72h for OTC sorption on Fe-oxides and Fe- oxide rich soils. In comparison with some published reports (Gu and Karthikeyan, 2005; Figueroa and Mackay, 2005; Rakshit et al., 2010) the total available surface sites of Al and Fe-WTR for sorption of TTC based on the SSR was the highest for the sorption of TTC and OTC (Table 2-6).

2.2.7 Potential release of sorbed TTC and OTC

Experiments were carried out to determine potential release of TTC and OTC from the treated Al- WTR (Table 2-7). Four different treatments (DI water, 1M KCl, methanol, and 0.25M EDTA) were used. Methanol treatment was used to check the role of hydrophobicity on sorption, KCl was used to see the effect of competing ion on

sorption, and EDTA was used as a chelating agent to evaluate competitively displaced sorbed TTC and OTC from Al-WTR. TTC release from Al -WTR with all the four treatments was minimal, suggesting irreversible sorption (Table 2-7). There was no TTC release when Al-WTR was treated with DI water by Al-WTR. Less than 4% of the sorbed TTC was removed in the methanol phase suggesting that hydrophobic interaction between Al-WTR and TTC was minimal. With the treatment of 1 M KCl, less than 5% of the sorbed TTC was removed, indicating that the TTC sorption mechanism was stronger than just non-specific electrostatic interaction. Less than 15% of the total sorbed TTC was desorbed by EDTA suggesting strong TTC-Al-WTR complexes (Nowack et al., 1996). Similar release behavior was seen for OTC with the abovementioned four treatments (Table 2-7). There was no OTC release (below method detection) for samples treated with DI water, less than 4% for methanol, less than 5% for KCl, and less than 15% with 0.25M EDTA of the total sorbed OTC. Further to confirm the retention and release behavior we checked into the possibility of transformation of TTC and OTC by Al-WTR. However, no unknown peaks were seen during the analysis of TTC and OTC by HPLC. Further, LC/MS/MS analyses were performed of one sample each for TTC and OTC to confirm if there is any transformation or degradation of TTC and OTC. No known daughter compounds or metabolites of TTC and OTC were observed, expect for low intensity peak for epi-tetracycline/oxytetracycline (epimerization) but in very low and insignificant concentrations. The TTC and OTC release behavior observed is encouraging with respect to stability of TCs sorbed to the retentive surfaces of the Al-WTR.

Table 2-7. Potential release (%) of TTC/OTC by various treatments on the sorbed TTC/OTC after equilibration time (24 h). Data are expressed as the mean of three replicates ± one standard deviation.

Extractant/ Treatments	TTC	OTC
DI water	<MDL**	<MDL**
Methanol	3.6 ± 0.3	3.75 ± 0.4
1M KCl (pH 6.0 ± 0.1)	4.8 ± 0.2	4.9 ± 0.25
0.25M EDTA (pH 6.0 ± 0.1)	14.1 ± 0.5	14.4 ± 0.5

** Below method detection limit (1×10^{-3} mM).

2.2.8 FTIR analysis

The ATR-FTIR spectra of crystalline TTC, crystalline OTC, untreated Al-WTR, and TTC and OTC (5 x 10^{-3} M) sorbed on to Al-WTR (20 g L^{-1}) at pH 6.0 ± 0.15 are shown in Figure 2-8 (a-e). There were no major vibration bands observed for untreated Al-WTR in the spectra, except for minor bending vibrations (Figure 8e). Characteristic IR bands associated with the functional groups of crystalline OTC/TTC molecule resulted peaks near 1679 cm^{-1} for amide I (the C=O group of the –$CONH_2$ moiety), 1618 & 1583 cm^{-1} for amide II (two N-H bonds of –$CONH_2$), and 1535 & 1446 cm^{-1} for skeletal C=C vibrations (aromatic ring) (Rakshit et al., 2003b). The FTIR spectra of TTC treated Al-WTR showed band shift compared to that of crystalline TTC, indicating potential interaction between TTC and Al- WTR (Kang et al., 2011; Zhao et al., 2012, Rakshit et al., 2013b) (Fig.8c). The IR bands (1679, 1618, 1583, 1535, and 1446 cm^{-1}) of OTC/TTC treated Al-WTR shifted to lower wave numbers (1664, 1596, 1560, 1516 & 1432 cm^{-1}) (Figure 2-8). Generally, lower wave number shifts in IR bands are characterized by weakening of the chemical bonds. In fact, strong bonding through the N-atom of –$CONH_2$ group would weaken two N-H bonds (IR bands 1618 & 1583 cm^{-1}). The lower wave number shift of 1679 cm^{-1} band pertaining to C=O moiety of –$CONH_2$ group probably indicates the loss of intermolecular H-bonding due to less electron density on O atom (Rakshit et al., 2013b). A strong covalent interaction of N atom with Al-WTR would result the unavailability of the lone pair of electron on N atom. Therefore, this electron pair cannot be delocalized in the –$CONH_2$ system causing less electron density on the O atom. A change of delocalization of electron density in the
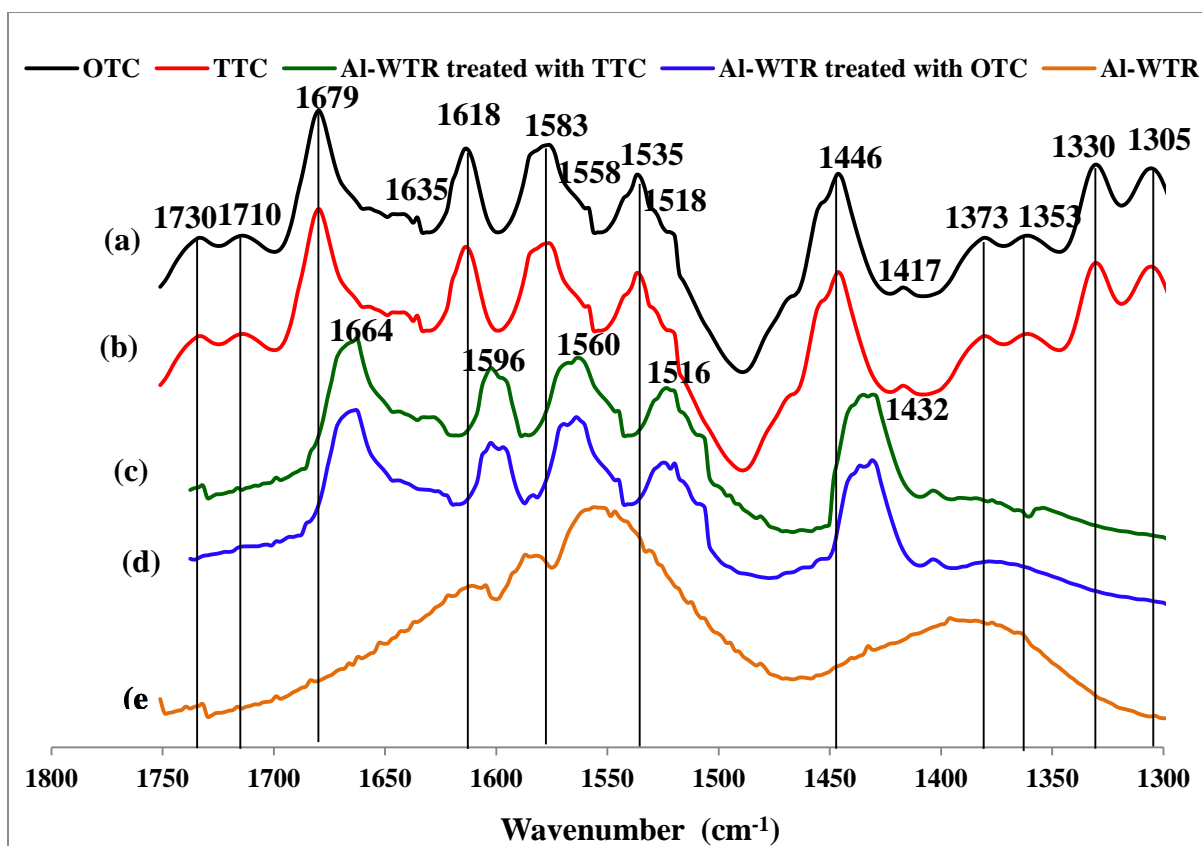
Figure 2-8: ATR-FTIR spectra of (a) crystalline TTC, (b) crystalline OTC, (c) Al-WTR (20 g L^{-1}) equilibrated for 24 h with TTC (5 x 10^{-3} M) at pH 6.0 ± 0.15, (d) Al-WTR (20 g L^{-1}) equilibrated for 24 h with OTC (5 x 10^{-3} M) at pH 6.0 ± 0.15, and (e) untreated Al-WTR. All spectra are normalized with the highest peak observed.

skeletal C=C in the aromatic ring is possible due to the conjugated C-OH system. If O-H group forms strong covalent bond with Al-WTR, the pair of electron on O atom cannot be delocalized with the ring C=C. This would cause a peak shift in IR bands pertaining to the skeletal C=C region. Since our IR data could not be collected below wave number 1300 cm^{-1}, the change of peak (~1180 cm^{-1}) due to bonding via O atom of –OH group cannot be directly verified. Overall, the IR band shifts are similar to other studies in which surface complexation of TTC/OTC was studied and a strong inner-sphere type interaction was inferred (Gu and Karthikeyan 2005; Kang et al., 2011, Rakshit et al., 2013b). This is consistent with our surface complexation modeling results (see section 3.5). The verification of the stoichiometry of the surface complexes proposed in SCM modeling by IR data may be limited in the sense that IR data presented here can be used to understand the participation of functional groups of TTC and OTC at a given pH, but IR data do not indicate how many molecules of TTC and OTC participate in bonding or each TTC and OTC molecule binds with the number of metal centers. Similarly, formation of mononuclear (i.e. 1:1 stoichiometry between TTC /OTC and WTR) surface species by SCM modeling does not specify any specific structure of the surface complex (Evanko and Dzombak, 1999).

2.3 Conclusions

　　To the knowledge of the authors, this study is the first attempt to use an industrial waste byproduct in the form of drinking water treatment residuals to remove TTC and OTC from aqueous medium. Results show that Al-WTR has superb sorption capacity for

both TTC and OTC, rapid sorption kinetics, and low release potential with different treatments/extractant, making them excellent low-cost and green sorbents for TCs removal from aqueous medium. Results from the study also provide a comprehensive understanding of the TTC and OTC sorption by Al-WTR at varying solution chemistry, which is vital in designing potential remediation systems. Results from the FTIR study and SCM modeling suggest that TTC and OTC are adsorbed on Al-WTR surface through strong inner-sphere type bonds. The research presented here provides evidence that Al-WTR has the potential to be used as a cost-effective medium for the immobilization TCs in wastewater treatment facilities and in manure storage lagoons in CAFOs. Further studies are needed to document the efficacy of Al-WTR in remediation of manure-amended soil systems contaminated with TCs under different conditions. On-going studies in our laboratory is focused on looking into the effect of competing ligands and complexing metals on TTC and OTC removal by WTRs in aqueous medium and removal efficiency of Al-WTR from TCs rich manure and manure amended soils under laboratory incubation and greenhouse settings.

2.4 References

- Aga, D. S. 2008. Fate of pharmaceuticals in the environment and in water treatment systems. CRC Press: Boca Raton, FL.
- Agerso, Y., Wulff, G., Vaclavik, E., Halling-Sorensen, B., Jensen, L., 2006. Effect of tetracycline residues in pig manure slurry on tetracycline-resistant bacteria and resistance gene tet(M) in soil microcosms. Environment International 32, 876-882.
- Allen, H.K., Donato, J., Wang, H.H., Cloud-Hansen, K.A., Davies, J., Handelsman, J., 2010. Call of the wild: antibiotic resistance genes in natural environments. Nature Reviews Microbiology 8, 251-259.
- Aristilde, L., Marichal, C., Miehe-Brendle, J., Lanson, B., Charlet, L., 2010. Interactions of Oxytetracycline with a Smectite Clay: A Spectroscopic Study with Molecular Simulations. Environmental Science & Technology 44.
- Ben-Dor E., Banin. A., 1989. Determination of organic matter content in arid-zone soils using a simple "loss-on-ignition" method. Commun. Soil Sci. Plant Anal. 20:1675–1695
- Blackwell, P.A., Kay, P., Boxall, A.B.A., 2007. The dissipation and transport of veterinary antibiotics in a sandy loam soil. Chemosphere 67, 292-299.
- Boxall, A.B.A., Kolpin, D.W., Halling-Sørensen, B., Tolls, J., 2003. Peer Reviewed: Are Veterinary Medicines Causing Environmental Risks? Environmental Science & Technology 37, 286A-294A.
- Chen, W.-R., Huang, C.-H., 2010. Adsorption and transformation of tetracycline antibiotics with aluminum oxide. Chemosphere 79, 779-785.
- Datta, R., Das, P., Smith, S., Punamiya, P., Ramanathan, D.M., Reddy, R., Sarkar, D., 2013. Phytoremediation potential of vetiver grass [Chrysopogon zizanioides (L.)] for tetracycline. *Internatl.* J. Phytorem. 15:343-351
- Daughton, C., Ternes, T., 1999. Pharmaceuticals and personal care products in the environment: Agents of subtle change? Environmental Health Perspectives 107, 907-938.
- Dayton, E.A., Basta, N.T., 2005. A method for determining the phosphorus sorption capacity and amorphous aluminum of aluminum-based water treatment residuals. J. Environ. Qual.
- Dos Santos, H.F., Xavier, E.S., Zerner, M.C., De Almeida, W.B., 2000. Spectroscopic investigation of the Al(III)-anhydrotetracycline complexation process. Journal of Molecular Structure-Theochem 527, 193-202.
- Elliott, H.A., Dempsey, B.A., 1991. Agronomic effects of land application of water treatment sludges. J. AWWA, 83 126–131.
- Essington, M. E., 2004. Soil and Water Chemistry; CRC Press: Boca Raton, FL.
- Evanko, C.R., Dzombak. D.A., 1999. Surface Complexation Modeling of Organic Acid Sorption to Goethite. J. Colloid Interf. Sci. 214:189-206.

- Figueroa, R.A., Leonard, A., MacKay, A.A., 2004. Modeling Tetracycline Antibiotic Sorption to Clays. Environmental Science & Technology 38, 476-483.
- Figueroa, R.A., MacKay, A.A., 2005. Sorption of Oxytetracycline to Iron Oxides and Iron Oxide-Rich Soils. Environmental Science & Technology 39, 6664-6671.
- Fritz, J.W., Zuo, Y., 2007. Simultaneous determination of tetracycline, oxytetracycline, and 4-epitetracycline in milk by high-performance liquid chromatography. Food Chemistry 105, 1297-1301.
- Goldberg, S., Johnston. C., 2001. Mechanisms of arsenic adsorption on amorphous oxides evaluated using macroscopic measurements, vibrational spectroscopy, and surface complexation modeling. J. Colloid Interf. Sci. 234:204-216.
- Gu, C., Karthikeyan, K., 2005. Interaction of tetracycline with aluminum and iron hydrous oxides. Environmental Science & Technology 39, 2660-2667.
- Gu, C., Karthikeyan, K.G., Sibley, S.D., Pedersen, J.A., 2007. Complexation of the antibiotic tetracycline with humic acid. Chemosphere 66, 1494-1501.
- Hamscher, G., Pawelzick, H.T., Hoper, H., Nau, H., 2005. Different behaviour of tetracyclines and sulfonamides in sandy soils after repeated fertilisation with liquid manure, Environ. Toxicol. Chem. 24, 861−868.
- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997a. Soil pH (1:2v/v). IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL) chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p15.
- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997b. Electrical Conductivity. IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL) chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p24.
- Herbelin, A.L., Westall, J.C., 1999. FITEQL 4.0: A computer program for determination of chemical equilibrium constants from experimental data. Department of Chemistry, Oregon State University: Corvallis, Oregon.
- Hovsepyan, A., Bonzongo, J.C.J., 2009. Aluminum drinking water treatment residuals (Al-WTRs) as sorbent for mercury: Implications for soil remediation. J. Hazard Mater. 164:73-80.
- Ippolito, J., Barbarick, K., Elliott, H., 2011. Drinking Water Treatment Residuals: A Review of Recent Uses. Journal of Environmental Quality 40, 1-12.
- Ippolito, J.A., Scheckel, K.G., Barbarick, K.A., 2009. Selenium adsorption to aluminum-based water treatment residuals. Journal of Colloid and Interface Science 338, 48-55.
- Jacobs, L.W., Teppen, B.J., 2000. WTR as a soil amendment to reduce nonpoint source pollution from phosphorus-enriched soils. 9 p. *In* Proc. 14th Annual Residuals and Biosolids Management Conference, 27−29 Feb. 2000, MA. CD-ROM, Water Environment Federation, Alexandria, VA.

- Janos, P., Sypecká, J., Mlckovská, P., Kurán, P., Pilarová, V., 2007. Removal of metal ions from aqueous solutions by sorption onto untreated low-rank coal (oxihumolite). Separation and Purification Technology 53, 322-329.
- Ji, L., Chen, W., Duan, L., Zhu, D., 2009. Mechanisms for strong adsorption of tetracycline to carbon nanotubes: A comparative study using activated carbon and graphite as adsorbents. Environmental Science & Technology 43, 2322-2327.
- Kang, J., Liu, H.J., Zheng, Y.M., Qu, J.H., Chen, J.P., 2011. Application of nuclear magnetic resonance spectroscopy, Fourier transform infrared spectroscopy, UV-Visible spectroscopy and kinetic modeling for elucidation of adsorption chemistry in uptake of tetracycline by zeolite beta. Journal of Colloid and Interface Science 354, 261-267.
- Kemper, N. 2008. Veterinary antibiotics in the aquatic and terrestrial environment.Ecol. Indicators 8:1-13.
- Kolpin, D., Furlong, E., Meyer, M., Thurman, E., Zaugg, S., Barber, L., Buxton, H., 2002. Pharmaceuticals, hormones, and other organic wastewater contaminants in US streams, 1999-2000: A national reconnaissance. Environmental Science & Technology 36, 1202-1211.
- Kulshrestha, P., Giese, R.F., Aga, D.S., 2004. Investigating the Molecular Interactions of Oxytetracycline in Clay and Organic Matter: Insights on Factors Affecting Its Mobility in Soil. Environmental Science & Technology 38, 4097-4105.
- Kumar, K., Gupta, S.C., Chander, Y., and Singh. A.K. 2005. Antibiotic use in agriculture and its impact on the terrestrial environment. Adv. Agron 87, 1–54.
- Kümmerer, K. 2009. Antibiotics in the aquatic environment – A review – Part I.Chemosphere 75:417-434.
- Levy, S. B., 1998. The challenge of antibiotic resistance. Sci. Am., 278:46-53.
- Levy, S. B., 2002. The Antibiotic Paradox; Perseus Publishing: Cambridge,MA.
- Loeppert, R.H., Inskeep, W.P., 1996. Iron. In: Sparks, D.L., Page, A.L., Helmke, P.A., Loeppert, R.H., Soltanpour, P.N., Tabatabai, M.A., Johnston, C.T., Summer, M.E. (Eds.), Method of Soil Analysis Part 3 Chemical methods, SSSA Book Series, pp. 639–664.
- Loffler, D., Rombke, J., Meller, M., Ternes, T.A., 2005. Environmental fate of pharmaceuticals in water/sediment systems. Environmental Science & Technology 39, 5209-5218.
- Makris, K., Harris, W., O'Connor, G., Obreza, T., 2004. Phosphorus immobilization in micropores of drinking-water treatment residuals: Implications for long-term stability. Environmental Science & Technology 38, 6590-6596.
- Makris, K., Harris, W., O'Connor, G., Obreza, T., Elliott, H., 2005. Physicochemical properties related to long-term phosphorus retention by drinking-water treatment residuals. Environmental Science & Technology 39, 4280-4289.
- Makris, K., Sarkar, D., Datta, R., 2006b. Evaluating a drinking-water waste by-product as a novel sorbent for arsenic. Chemosphere 64, 730-741.

- Makris, K., Sarkar, D., Datta, R., 2006a. Aluminum-based drinking-water treatment residuals: A novel sorbent for perchlorate removal. Environmental Pollution 140, 9-12.
- Miller, M.L., Bhadha, J.H., O'Connor, G.A., Jawitz, J.W., Mitchell, J., 2011. Aluminum water treatment residuals as permeable reactive barrier sorbents to reduce phosphorus losses. Chemosphere 83, 978-983.
- Nagar, R., Sarkar, D., Makris, K., Datta, R., 2010. Effect of solution chemistry on arsenic sorption by Fe- and Al-based drinking-water treatment residuals. Chemosphere 78, 1028-1035.
- Nowack, B., Lützenkirchen, J., Behra, P., Sigg, L., 1996. Modeling the adsorption of metal-EDTA complexes onto oxides. Environmental Science and Technology 30, 2397- 2405.
- O'Connor, G.A., Elliott, H.A., Lu, R., 2002. Characterizing water treatment residuals phosphorus retention. Soil and Crop Science Society of Florida Proceedings 61, 67-73.
- Oberle, K., Capdeville, M.J., Berthe, T., Budzinski, H., Petit, F., 2012. Evidence for a Complex Relationship between Antibiotics and Antibiotic-Resistant Escherichia Coli: From Medical Center Patients to a Receiving Environment. Environmental Science & Technology 46, 1859-1868.
- Pils, J., Laird, D., 2007. Sorption of tetracycline and chlortetracycline on K- and Ca-saturated soil clays, humic substances, and clay-humic complexes. Environmental Science & Technology 41, 1928-1933.
- Prakash, P., Sengupta, A.K., 2003. Selective coagulant recovery from water treatment plant residuals using donnan membrane process. Environmental Science & Technology 37, 4468-4474.
- Rakshit, S., Sarkar, D., Elzinga, E.J., Datta, R.., 2013b. In Situ ATR-FTIR study of Oxytetracycline Sorption on Magnetite . J. Environ. Qual. 42:822-827.
- Rakshit, S., Sarkar, D., Elzinga, E.J., Punamiya, P., Datta, R., 2013a. Mechanisms of ciprofloxacin removal by nano-sized magnetite. J. Hazard Mater. 246-247:221-226.
- Rakshit, S., Punamiya, P., Datta, R., Sarkar, D., 2010. Sorption of oxytetracycline on magnetite-water interface. Geochimica Et Cosmochimica Acta 74, A846-A846.
- Sarkar, D., Makris, K., Vandanapu, V., Datta, R., 2007. Arsenic immobilization in soils amended with drinking-water treatment residuals. Environmental Pollution 146, 414-419.
- Sarmah, A. K., Meyer, M. T.; Boxall, A. B. A., 2006. A global perspective on the use, sale, exposure pathways, occurrence, fate and effects of veterinary antibiotics (VAs) in the environment Chemosphere 65, 725-759.
- Sparks, D. L. 1995. Environmental soil chemistry, 2nd ed. Academic Press, San Diego, CA.

- Thiele-Bruhn, S., 2003. Pharmaceutical antibiotic compounds in soils - a review. Journal of Plant Nutrition and Soil Science-Zeitschrift Fur Pflanzenernahrung Und Bodenkunde 166, 145-167.
- Van Riemsdijk, W.H., Lyklema, J., 1980. Reaction of phosphate with gibbsite $(AI(OH)_3)$ beyond the adsorption maximum. J. Colloid Interf. Sci. 76:55-66.
- Wessels, J.M., Ford, W.E., Szymczak, W., Schneider, S., 1998. The complexation of tetracycline and anhydrotetracycline with $Mg2+$ and $Ca2+$: A spectroscopic study. Journal of Physical Chemistry B 102, 9323-9331.
- Zhang, D., Pan, B., Wu, M., Wang, B., Zhang, H., Peng, H., Wu, D., Ning, P., 2011a. Adsorption of sulfamethoxazole on functionalized carbon nanotubes as affected by cations and anions. Environmental Pollution 159, 2616-2621.
- Zhao, Y.P., Gu, X.Y., Gao, S.X., Geng, J.J., Wang, X.R., 2012. Adsorption of tetracycline (TC) onto montmorillonite: Cations and humic acid effects. Geoderma 183, 12-18.

CHAPTER 3

Effect of Solution Properties, Competing Ligands, and Complexing Metal on Sorption of Tetracyclines on Al-Based Drinking Water Treatment Residuals

Abstract

In the current batch study, we investigated the effect of solution pH (3-11) on tetracyclines (TCs); mainly tetracycline (TTC) and oxytetracycline (OTC) by Al-based drinking water treatment residuals (Al-WTR) as a function of sorbate: sorbent ratio (SSR) and initial TTC/OTC concentrations. The effects of competing ligands (phosphate (P(V)) and sulfate) and complexing metal (calcium (Ca^{2+})) on TTC and OTC sorption at optimum SSR as a function of pH and different TTC/OTC ratios were also evaluated. The sorption behavior for both TTC and OTC on Al-WTR was pH-dependent. The sorption in absence of competing ligands and complexing metal increased with increasing pH up to circum-neutral pH and then decreased at higher pH. The presence of P(V) when added simultaneously had a significant negative effect ($p<0.001$) on the sorption of TTC and OTC adsorbed by Al-WTR at higher TTC/OTC:P ratios. However, when P(V) was added after the equilibration of TTC and OTC by Al-WTR the effect was minimal and insignificant ($p > 0.1$) The presence of sulfate had a minimal/negligible effect on the sorption of TCs by Al-WTR. A significant negative effect ($p<0.001$) on the adsorption of

TCs by Al-WTR was observed in the pH range below 5 and at higher TCs: Ca^{2+} ratios, probably due to TCs-Ca^{2+} complex formation. FTIR analysis indicated the possibility of inner-sphere type bonding by the functional groups of OTC/ TTC on the surface of Al-WTR. Results from the batch sorption study indicate high affinity of Al-WTR for TCs in the pH range 4-8 (majorly encountered pH in the environment) in the presence of competing ligands and complexing metal. Our research will help to develop a cost-effective medium for TCs removal in wastewater treatment facilities and at concentrated animal feeding operations.

Keywords: Tetracyclines, Sorption, Drinking water treatment residuals, Competing ligands, Complexing metal, Remediation

3. Introduction

Veterinary antibiotics (VAs) are widely used for therapeutic (prevent and treat diseases) and sub-therapeutic (growth promotion and prophylaxis) purposes in the livestock industry. A review of the literature suggests presence of low levels (ng-µg/L) of VAs in the aquatic and terrestrial environments (Netthisinghe et al., 2013; Watkinson et al., 2009; Kümmerer, 2009; Kemper, 2008). The presence of VAs in the environment is of great concern because, even at low levels, these molecules are biologically active and can affect critical development stages of organisms (Aga, 2008). The widespread use and frequent detection of VAs in the environment have also raised concerns over proliferation of antibiotic-resistant bacteria (Netthisinghe et al., 2013; Oberle et al., 2012; Allen et al., 2010).

Most of the VAs, such as tetracyclines, macrolides, quinolones, sulfonamides, etc. have multiple functional groups and can exist as different dissociation species based on the environmental conditions. For instance, tetracyclines (TCs) are complex organic compounds with unique chemical characteristics and behaviors. TCs structure contains connected ring systems with multiple ionizable functional groups (Figure 2-1A). TCs have three pK_a's and hence, can exist as cationic, zwitterionic, or anionic species under acidic, moderately acidic to neutral, and alkaline conditions, respectively (Figure 2-1B). The surface speciation of these antibiotics leads to a particular and unique environmental behavior compared with traditional hydrophobic compounds. Due to the complicated environmental behavior of these VAs, it is difficult to assess their environmental risks.

Considerable efforts are being made to develop cost-effective treatment technologies (use of clays, minerals, metal oxides, carbon nanotubes, waste byproducts, plants, etc.) to remove VAs from aqueous solutions (Punamiya et al., 2013; Rakshit et al., 2013a; Rakshit et al., 2013b;; Datta et al., 2013; Kang et al., 2010; Ji et al., 2009; Wang et al., 2008; Figueroa and Mackay, 2005; Gu and Karthikeyan, 2005). However, little emphasis has been given to looking into the effect of competing ligands and complexing metals on adsorption. In the environment, anions and cations normally compete for sorption sites with environmental contaminants on the sorbent. Thus, it is very important to study the competitive adsorption of different anions and cations with the emerging environmental contaminants. In addition, investigations on the competition and complexation can provide insights into the reaction occurring at the surface of sorbent. The presence of competing ligands and complexing metals also make the environmental behavior of VAs more complicated. Studies have shown an increase as well as a decrease in the sorption of antibiotics in the presence of various competing ligands and complexing metals on clays, sediments, soils, oxides, and various sorbents (Zhang et al., 2011; Ji et al., 2009; Kang et al., 2010; Wang et al., 2008;). Reports from Wang et al. (2008), Ji et al. (2009), and Kang et al. (2010) showed that the presence of Cu^{2+} increased the adsorption of tetracycline on montmorillonite, carbon nanotubes and chitosan, respectively. Zhang et al. (2011) studied adsorption of sulfamethoxazole on carbon nanotubes in the presence of cations (Ca^{2+} and Cs^+) and anion (P(V)). Their study emphasized that both increase and decrease in adsorption of sulfamethoxazole could be observed with addition of cations/anions, depending on the environmental conditions (pH

dependent behavior in this study). Studies have also shown a pH dependent effect on increasing and decreasing sorption behavior of TCs in the presence of cations and anions (Pei et al., 2011; Chen and Huang, 2010; Teer Laak et al., 2006; Figueroa and Mackay, 2005; Gu and Karthikeyan, 2005).

An earlier batch sorption experiment in our laboratory demonstrated high affinity of Al- drinking water treatment residuals (Al-WTR) in removing TCs from aqueous solutions (Punamiya et al., 2013). The WTRs are primarily amorphous masses of aluminum (Al) or iron (Fe) hydroxides, or calcium ($CaCO_3$), referred to as Al-WTRs (use of Al salt), Fe-WTRs (Fe salt), or Ca-WTRs ($CaCO_3$) (O'Connor et al., 2001), which also contain sediment and humic substances removed from the raw water, as well as activated carbon and polymers (Elliott and Dempsey, 1991; Makris et al., 2005; Ippolito et al., 2011). Within 5 h of reaction time, nearly complete removal of TTC was achieved with minimum release from the Al-WTR (Punamiya et al., 2013). However, there is no detailed study on the pH dependent effect of competing ligands or complexing metals on TCs sorption by Al-WTR.

Solution properties, for instance, pH, initial TCs concentration, and simultaneous presence of other ions significantly affect the binding dynamics of TCs to Al/Fe hydr(oxide) and other surfaces (Punamiya et al., 2013; Zhang et al., 2012; Rakshit et al., 2010; Chen and Huang, 2010; Teer Laak et al., 2006; Figueroa and Mackay, 2005; Gu and Karthikeyan, 2005). An understanding of phosphate and sulfate competition and calcium complexation will be beneficial in the development of an effective treatment process for TCs removal from aqueous medium.

The present study is aimed at i) investigating the effect of solution pH (3-11) on TTC and OTC by Al-WTR as a function of sorbate: sorbent ratio (SSR) and initial TTC/OTC concentration; ii) evaluating the effect of competing ligands (phosphate P(V) and sulfate) and complexing metal (calcium) on TTC and OTC sorption envelopes at optimum SSR as a function of pH and different TTC/OTC ratios; and iii) determine the effectiveness of Al-WTR to remove TCs from aqueous media in the presence of competing ligands (phosphate P(V) and sulfate) and complexing metal (calcium) under different conditions.

3.1 Materials and Methods

3.1.1 Reagents and Materials

Tetracycline hydrochloride (USP grade, $\geq$98%) and oxytetracycline hydrochloride (USP grade, $\geq$98%) were obtained from Sigma-Aldrich chemical (St. Louis MO). Reagent grade phosphate monobasic monohydrate ($NaH_2PO_4.H_2O$), sodium sulfate decahydrate ($Na_2SO_4.10H_2O$), and calcium nitrate tetrahydrate ($Ca(NO_3)_2.4H_2O$) were purchased from Fisher Scientific (Fair Lawn, NJ). Trace-metal grade hydrochloric acid, sodium hydroxide, potassium chloride, and oxalic acid all certified ACS grade were purchased from Fisher Scientific (Fair Lawn, NJ). Methanol (HPLC grade, $\geq$99%) and acetonitrile (HPLC grade, $\geq$99%) were purchased from Fisher Scientific (Fair Lawn, NJ). PIPES (1,4-piperazinebis(ethane sulfonic acid) was purchased from Sigma–Aldrich (St. Louis, MO). All solutions and working standards were freshly prepared in high-purity water (18 MΩ-cm, Barnstead NANO-pure Diamond, Dubuque, IA).

3.1.2. WTR collection and characterization

Al-based WTR was used in the batch sorption study. It was obtained from drinking-water treatment plant in Bradenton Florida. Al-WTR samples were air-dried and were subsequently passed through a 2-mm sieve before analyses. WTR samples were characterized for several physico-chemical properties as earlier discussed in Punamiya et al. (2013). In brief, solution pH, electric conductivity, and organic matter content were measured using standard protocols (Ben-Dor and Banin, 1989, Hanlon et al., 1997a, and Hanlon et al., 1997b). Total C, N, and S were determined by combustion at 1100 °C using an Elementar Vario EL CNS/O analyzer (Elementar, Mount Laurel, NJ). Oxalate-extractable Al and P concentrations after extraction with Tamm's reagent (Loeppert and Inskeep, 1996) and total Al, P, S, and Ca were determined by ICP-MS.

3.1.3 Analytical method

A Finnigan surveyor plus HPLC system (Thermo Scientific, Somerset, NJ), equipped with quadruple pumps, coupled with surveyor PDA plus detector (photodiode array), and a surveyor plus auto-sampler were used for all the analyses. A hypersil gold C_{18} column 150 x 4.6mm, 5µm (Thermo Scientific) with a corresponding hypersil gold guard column 10 x 4mm, 5µm (Thermo Scientific) at room temperature was used for all separations. Samples were eluted isocratically with a mobile phase consisting of 0.01 M aqueous oxalic acid: acetonitrile: methanol (150:20:20 by volume) (Fritz and Zuo, 2007.). The mobile phase was mixed and sonicated for 5 min before use. The flow rate was

maintained at 1.5 ml/min, with an injection volume of full loop (25 µl), while the UV detector was set at 360 nm. Linear calibration was used for quantification based on the curves between the concentration and peak area of known standard of TTC and OTC. Metal concentrations were quantified using ICP-MS (Thermo Electron, San Hose, CA). X-ray diffraction analysis was performed using Philips X' Pert (PANalytical, Westborough, MA).

Surface charge measurements were performed using a Zeta Sizer nano-series ZS90 (Malvern, United Kingdom). FTIR spectra were acquired using Thermo Nicolet 4700 spectrometer (Thermo Scientific, Somerset, NJ) equipped with an attenuated total reflection (ATR) accessory with Zn-Se cell. Spectra for the pH envelope experiment were collected. The spectra were obtained accumulating 256 scans at a resolution of 4 cm^{-1} in range of $4000 - 850$ cm^{-1}. In brief, samples from batch sorption experiments with higher TTC/OTC (5.0×10^{-3} M) concentration were reacted with 20 g L^{-1} Al-WTR at three different pH (4.1, 6.3, and 9.1) . After centrifugation and filtration, samples were freeze dried and used for analysis. Reference spectra for TTC, OTC, and Al-WTR were also obtained. All spectra were normalized with the highest peak observed.

3.1.4 Experimental Design

3.1.4.1 Batch sorption experiment

All the batch sorption experiments were conducted in 50 mL polypropylene centrifuge tubes. KCl (0.01M) was used as a background electrolyte for preparation of stock solutions for competing ligands (P(V) and sulfate) and complexing metal (calcium).

Stock of TTC and OTC were freshly prepared before the start of the experiment and immediately covered with Al-foil to prevent any photodegradation. PIPES buffer (0.01M) was used to control the pH of the solution. The suspensions in the test tubes were shaken end-over-end on a reciprocating shaker (MaxQ, Thermo Scientific) at 250 rpm for 24 h (equilibration time optimized from previous study). The samples were centrifuged (Allegra 21R, Beckman Coulter) at 4000 rpm, followed by filtration through 0.25µm syringe filters. During the time of shaking, the sample tubes were wrapped in Al-foil to inhibit photodegradation of TCs. The concentration of TCs in the filtrates was determined using HPLC. Adequate blanks, duplicates, and matrix spikes were used to meet quality assurance and quality control requirements.

3.1.4.2 TCs sorption experiment in absence of competing ligands and complexing metal

The effect of pH and SSR's on sorption of TCs on Al-WTR were assessed using batch sorption experiment. Effect of pH on sorption was studied by determining the amount of the TTC/OTC adsorbed within the pH range of 3-11. The suspensions/mixtures in the tubes were adjusted to the desired pH values by adding 0.1 M HCl/0.1 M NaOH. The volume of the acid and base used during the adjustment were recorded and added to the total volume for the use of final calculations. The pH values of all the solutions were checked before and after the sorption experiment. The final pH values were used for the sorption-edge experiment. Three SSR's 5, 10, and 20 g L^{-1} were employed to study the effect of the SSR on TCs sorption. Samples were reacted with four

different initial concentration of TTC/OTC (0.1, 0.5, 1.0, and 5.0 mM). The suspensions

in the test tubes were shaken, centrifuged, and filtered and analyzed using HPLC.

3.1.4.3 TCs sorption experiment in the presence of competing ligands and complexing metal

Sorption experiments were conducted to obtain sorption envelopes of TCs

sorption on Al-WTR as a function of pH (3-11), constant initial concentration of TCs

(1mM OTC and TTC, respectively), and in the presence of varying concentration of

competing ligands (P(V) and sulfate) and complexing metal (calcium). Equilibration

time, SSR's, and ionic strength were selected on the basis of the previous sorption

experiments. In brief, 24 h of equilibration time (Punamiya et al., 2013), 20 g L^{-1} SSR,

and 0.01M KCl ionic strength were used. Initial ratios (molar) of TCs to competing

ligands and complexing metals were 1:1, 1:5, 1:10, and 1:20. Initial TTC (1.0 mM L^{-1})

and OTC concentration (1.0 mM L^{-1}) for competing ligands was chosen to provide near

maximum surface coverage, according to previous specific area measurement for the

WTR (Makris et al., 2004). Phosphate monobasic monohydrate ($NaH_2PO_4.H_2O$) and

sodium sulfate decahydrate ($Na_2SO_4.10H_2O$) were used as source of P(V) and sulfate,

respectively for the competing ligands batch sorption experiment. Calcium nitrate

tetrahydrate ($Ca(NO_3)_2.4H_2O$) was used as a source of calcium for complexing metal

batch sorption experiment. Visual MINTEQ version 4.0.3 (chemical equilibrium model)

calculations were performed to check highest concentration of Ca^{2+} to be used in the

complexing metal experiment to avoid onset of precipitation and/or formation of any Ca-

mineral. Solutions for the sorption study were prepared by adding TTC/OTC and the competing ligand or the complexing metal simultaneously to avoid any pre-sorption on the Al-WTR. The suspensions in the test tubes were shaken, centrifuged, and filtered and analyzed using HPLC.

3.1.5 Statistical analysis

Data obtained were statistically analyzed using JMP IN version 9.0 (Sall et al., 2005). To examine the effect of competing ligands, complexing metal, and initial TTC and OTC loads on sorption by Al-WTR two-way ANOVA was performed. To evaluate significant differences among treatment means Tukey–Kramer HSD test was used. The data is reported as the mean of three replicates with one standard deviation.

Results and Discussion

3.2.1 Effect of pH and SSR on TTC/OTC sorption

General physicochemical properties of the Al-WTR are given in Table 3-1 and have been discussed in detail elsewhere (Punamiya et al., 2013). In brief, Al-WTR was acidic and amorphous in nature. The total C values (185 g kg^{-1}) for Al-WTR are in the range of organic C found in various WTRs nationwide (Ippolito et al., 2011). The Toxicity Characteristics Leaching Procedure (TCLP) values of Al-WTR were well below USEPA threshold limits (compared to values of biosolids as no limits are set for WTRs).

We attempted to identify the optimum SSR and pH for TTC and OTC sorption by Al-WTR using four different TTC/OTC initial concentrations ranging from 0.1 to 5 mM (Figures 3-1 and 3-2). SSR exerted a strong effect on sorption of TTC and OTC by the

Al-WTR. Increasing the SSR resulted in a significant ($p<0.05$) increase in the amount of

TTC and OTC sorbed by Al-WTR in the pH range (3-11) tested, regardless of the initial

TTC or OTC concentrations. At lower SSR (5 g L^{-1}), an increase in the initial

concentration of TTC and OTC had a negative effect on the amount of TTC and OTC

sorbed ranging between 35-65% of the initial concentration added (Figure 3-1C and 3-

2C). However, increasing the SSR to 20 g L^{-1}, there was no significant difference in the

total amount of TTC and OTC sorbed, removing nearly 90% of initially added TTC/OTC

(Figure 3-1A and 3-2A). At the intermediate SSR (10 g L^{-1}), TTC and OTC sorption

ranged between 60 to 80% (initially added concentration). There was no significant

($p>0.1$) effect between sorption of TTC and OTC on Al-WTR at all the concentrations

and SSRs tested, showing species' independent sorption behavior (because OTC and

TTC have similar functional groups). Significant ($p < 0.01$) interaction was seen between

the SSR and initial TTC and OTC concentrations on the overall amount of TTC/OTC

sorbed by Al-WTR (Figure 3-1 and 3-2). Based on both TTC and OTC SSR experiments,

we selected 20 g L^{-1} ratio as the optimum SSR that would maximize TTC/OTC

adsorption. The effect of pH on the sorption of TTC and OTC was examined using Al-

WTR at different SSRs and varying initial TTC and OTC concentrations (0.1-5 mM).

There was no significant ($p>0.1$) change in the initially added TTC/OTC concentration

(recovery > 98%) in the controls (without Al-WTR) over the entire pH range (3-11)

tested, indicating no loss of TTC/OTC due to the reactions other than sorption. Final pH

values were used for the sorption-edge experiment. TTC sorption envelopes for the Al-
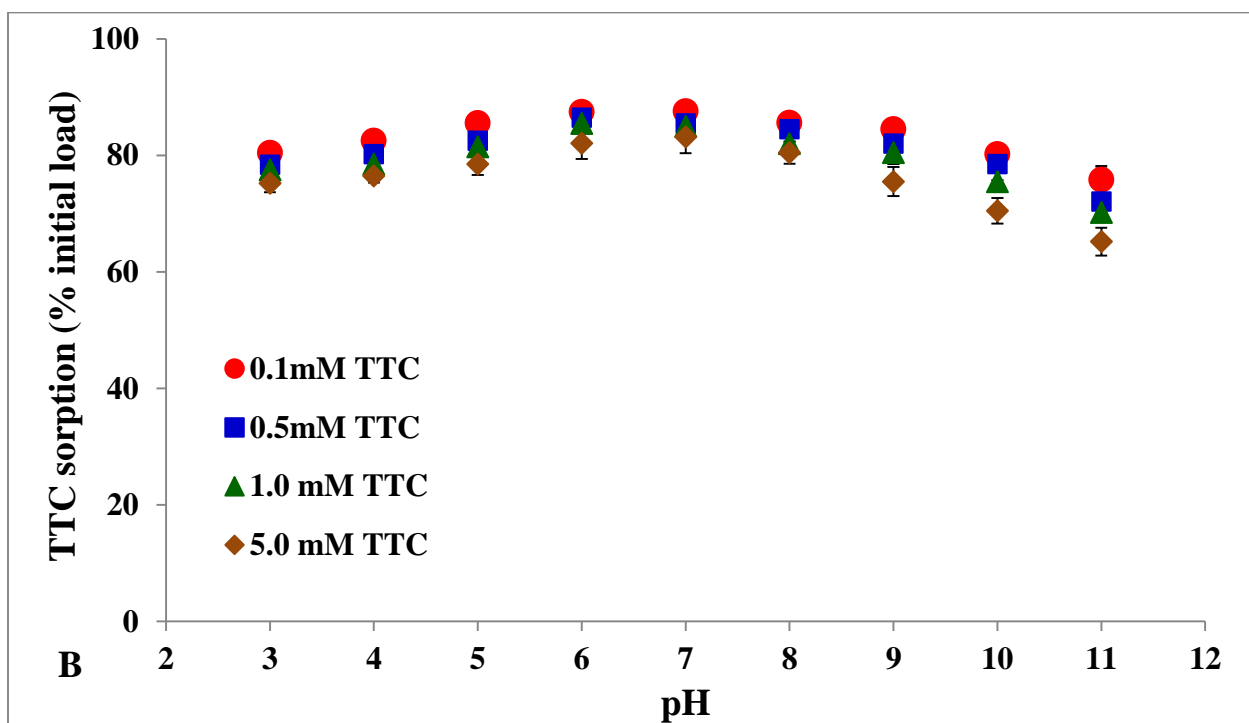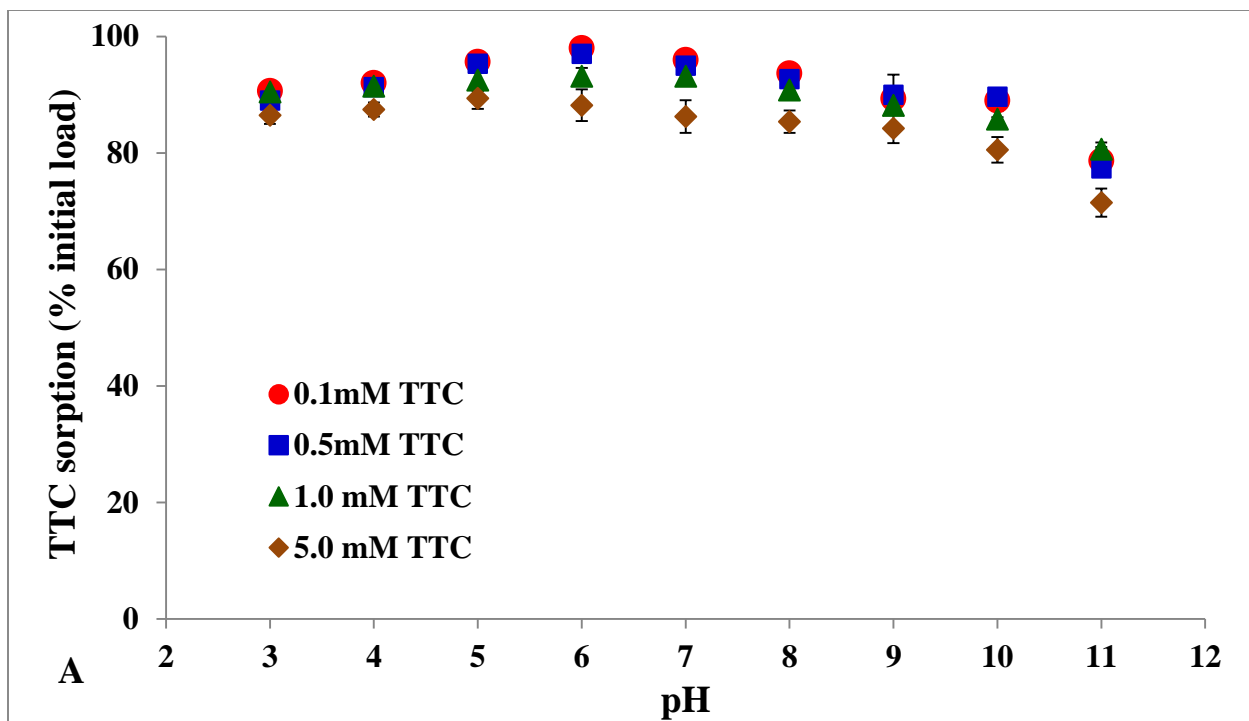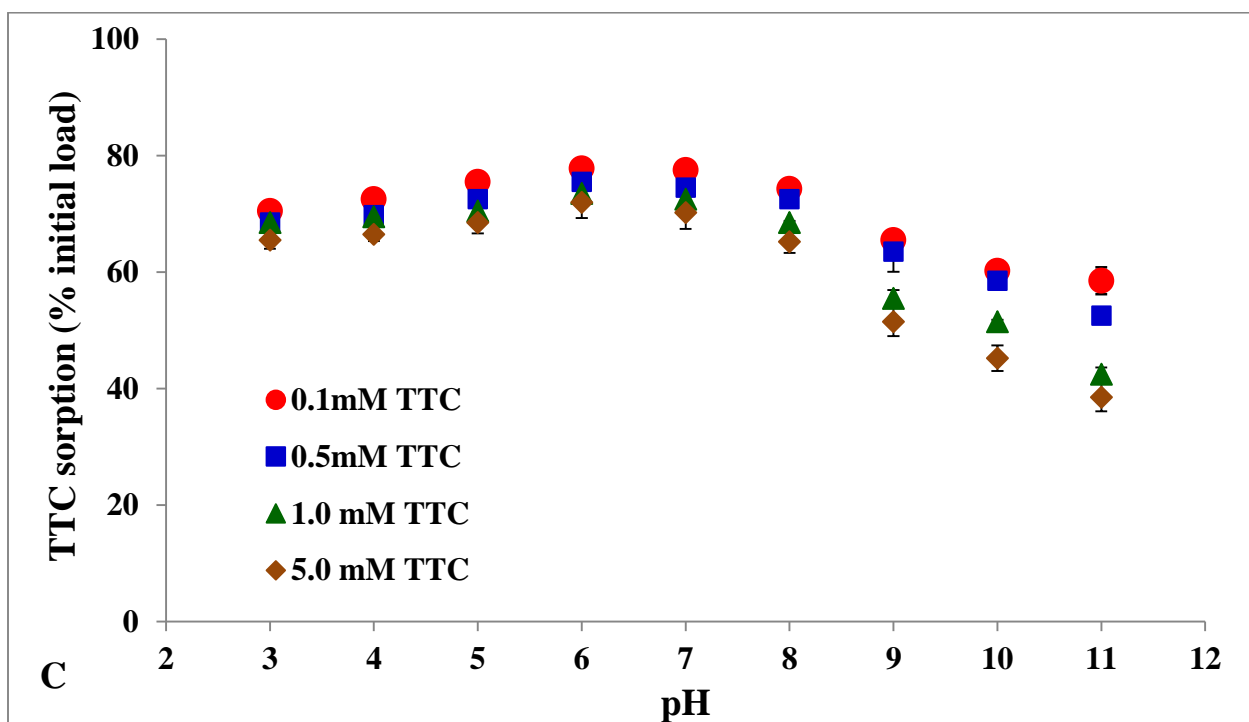
Figure 3-1: The effect of pH on the amount of TTC sorbed by Al-WTR (as a function of different initial TTC concentrations (0.1, 0.5, 1.0, and 5.0 mM TTC) at different SSR's: 20 g L^{-1} (A), 10 g L^{-1} (B), and 5 g L^{-1} (C), reaction time 24 h, and ionic strength buffer 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.
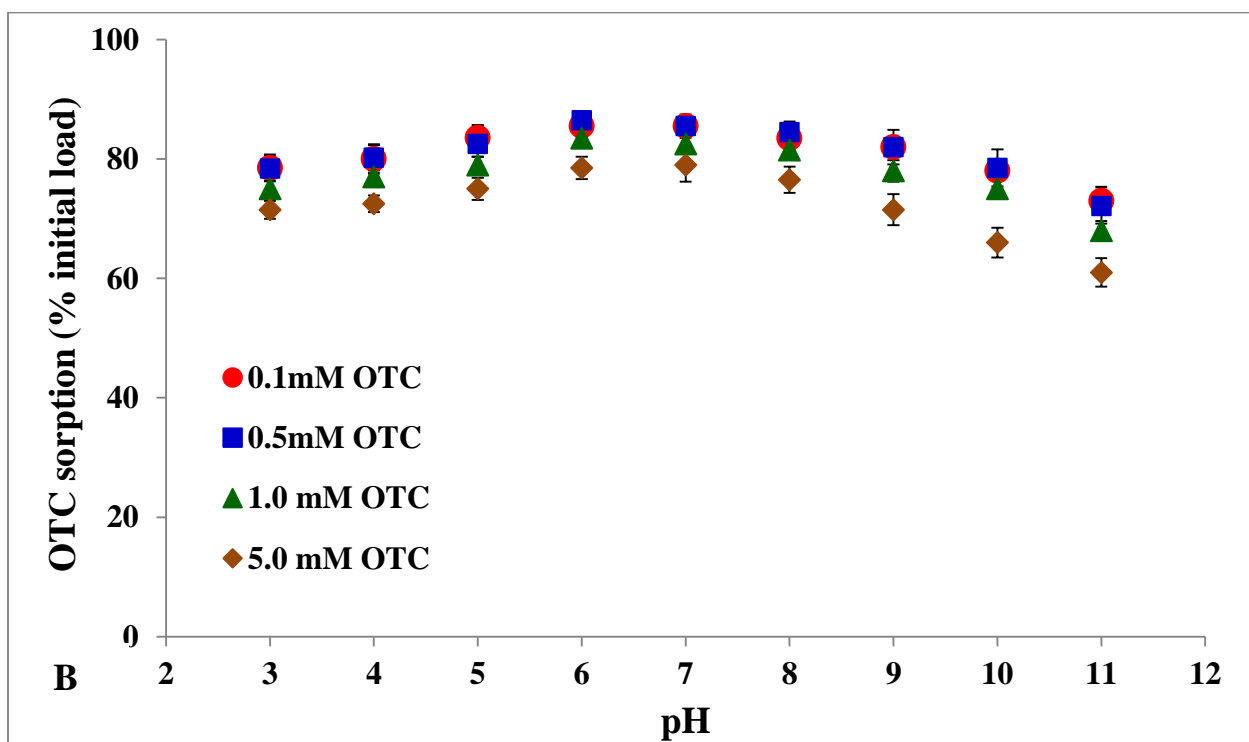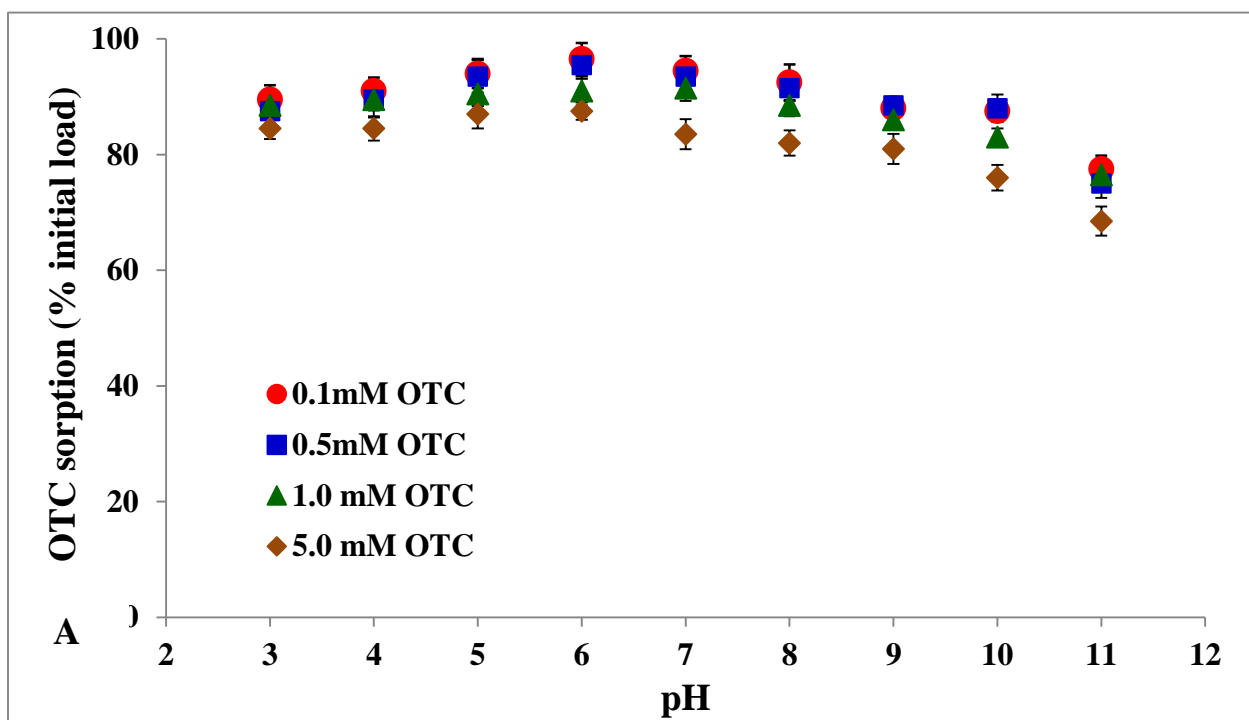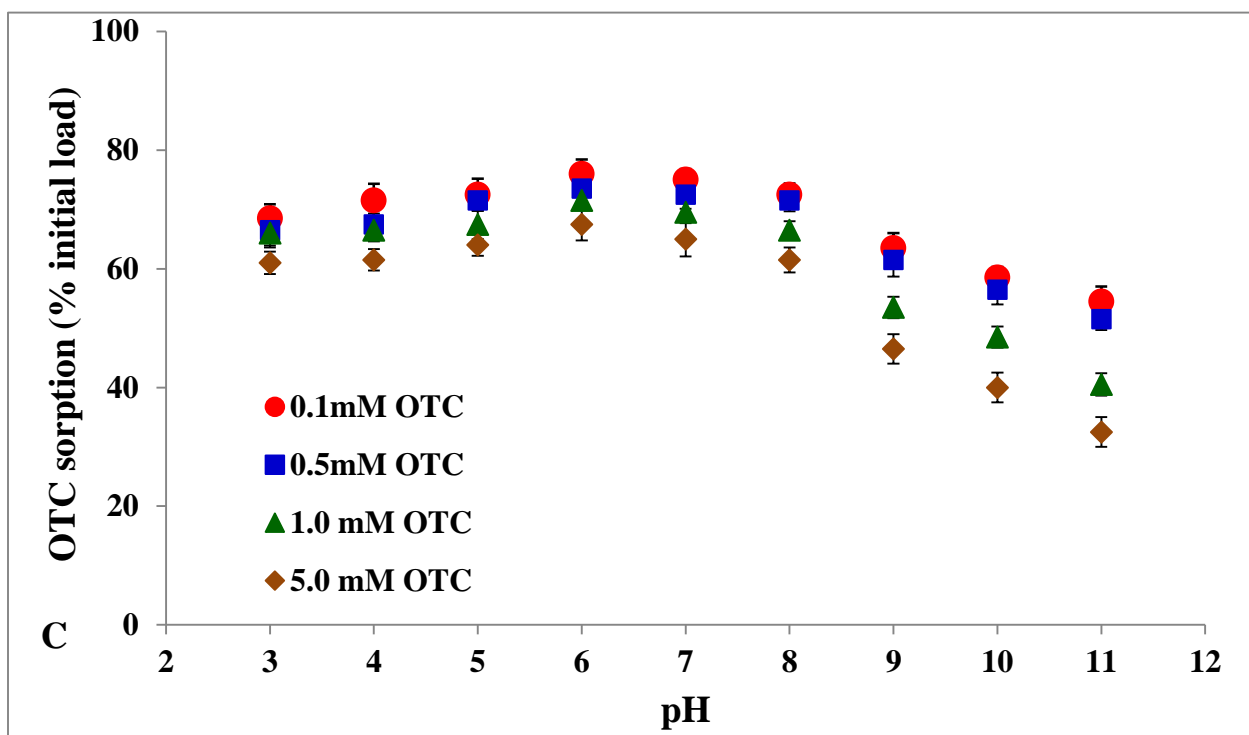
Figure 3-2: The effect of pH on the amount of OTC sorbed by Al-WTR (as a function of different initial TTC concentrations (0.1, 0.5, 1.0, and 5.0 mM OTC) at different SSR's: 20 g L^{-1} (A), 10 g L^{-1} (B), and 5 g L^{-1} (C), reaction time 24 h, and ionic strength buffer 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation

Table 3-1. Physicochemical properties of Al-based WTR. Data are expressed as the mean of three replicates ± one standard deviation.

	Al-WTR
Source	Bradenton, FL
pH	6.3 ± 0.2
Electrical conductivity (dS m^{-1})	0.85 ± 0.1
Solid organic matter (%)	33.2 ± 1.5
Carbon (g/kg)	182 ± 4.6
Nitrogen (g/kg)	6.2 ± 0.5
Total Al (g/kg)	115.9 ± 2.6
Oxalate extractable Al (g/kg)	87.4 ± 1.7
Total Fe (g/kg)	11.8 ± 1.4
Oxalate extractable Fe (g/kg)	4.2 ± 0.9
Total P (g/kg)	3.1 ± 0.4
Oxalate extractable P(g/kg)	2.75 ± 0.2
Total Ca (g/kg)	10.5 ± 0.51
Total S (g/kg)	6.6 ± 0.31

WTR at 20 g L^{-1} and at the lowest initial TTC concentration (0.1 mM) indicated more

than 90% sorption in the pH range between 5 and 7 (Fig. 3-1A). As the pH was increased

above 7, sorption of TTC on Al-WTR (TTC concentration: 0.1 mM) and SSR: 20 g L^{-1})

decreased from 95% to 80% showing pH dependent sorption behavior. This trend

remained unchanged even after increasing the initial TTC concentration to 5.0 mM (Fig.

3-1A). The high TTC sorption affinity of Al-WTR is mainly due to the high external and

internal surface area (Makris et al., 2006). At the intermediate SSR (10 g L^{-1}), TTC

sorption was close to 80-85% in the pH range of 5-7 and decreased with an increase in

pH above 7 to 65-73% (Fig. 3-1B). As the initial TTC concentrations were increased, a

decrease in the amount of sorption of TTC by Al-WTR was observed in the entire pH

range tested (Fig. 3-1B). At the lowest SSR (5 g L^{-1}) tested, there was a significant

(p<0.01) negative effect on the sorption of TTC by Al-WTR. However, the pH dependent

sorption behavior remained unchanged; maximum sorption was observed in pH range 5-7

and decrease in sorption with increase in pH above 7(Fig. 3-1C).

Similar behavior was observed for OTC at all concentrations and SSRs tested in

the pH range (Fig. 3-2). In brief, at 20 g L^{-1} maximum OTC sorption was observed by

Al-WTR at pH 6 with 95, 92, 91, and 87 % for 0.1, 0.5, 1.0, and 5.0 mM, respectively

(Fig. 3-2A). At the intermediate SSR (10 g L^{-1}) and pH 6, OTC sorption ranged from

77% to 84 % of the initial added OTC concentrations. At the lowest SSR (5 g L^{-1}) tested,

initial OTC concentration had a significant (p<0.01) negative effect on the sorption. At

highest OTC concentration (5.0 mM) and pH 6.0, only 67% sorption was observed,

followed by 71, 73, and 76 % at 1.0, 0.5, and 0.1 mM initial OTC concentrations.

TTC/OTC sorption envelopes for the Al-WTR are in agreement to those of previous studies (Punamiya et al., 2013; Chen and Huang, 2010; Gu and Karthikeyan, 2005; Figueroa et al., 2004). Gu and Karthikeyan (2005) observed an increase in the sorption of TTC on aluminum hydrous oxide up to pH 7 and a decrease at higher pH (above 7). Chen and Huang (2010) observed greater sorption of TTC on aluminum oxide (Al_2O_3) near pH 7 and sorption decreased when solution pH was increased or decreased. Previous studies on TTC/OTC sorption by clays (montmorillonite and kaolinite), clays and organic matter, and soils also showed gradual decrease in the sorption coefficient as the solution pH was raised from close to neutral to alkaline conditions (Ter Laak et al., 2006; Kulshrestha et al., 2004; Figueroa et al., 2004).

The observed pH dependence can be explained by the interplay between surface charge on the TTC/OTC and Al-WTR surfaces, ligand exchange reaction (Punamiya et al., 2013) and inner-sphere type chemical bond formation (IR data; section *3.2.4 FTIR analysis*). The pH-dependent surface speciation of Al-WTR shows that the surface of Al-WTR is positively charged when the pH is below its pH_{zpc} (8.5) and negatively charged when pH is above the pH_{zpc} (Figure 2-4). Considering aqueous speciation at pH < 3.3 or pH > 7.7 there is greater electrostatic repulsion (positive-positive or negative–negative respectively), due to similar surface charges of TTC and Al-WTR and electrostatic attraction in-between pH range (zero point charge). The decrease in the sorption coefficient with increase in the pH was explained as an adsorption mechanism by the abundance of zwitterionic species between pH 5 to 8, indicating zwitterionic form of OTC as a sorbate at environmental pH (Ter Laak et al.,

2006; Figueroa et al., 2004). However, at a higher initial concentration the trend is

different; i.e. the near highest sorption occurred at lower pH. This can be further

explained by the interaction between the surface charge and the chemical bond formation

(from IR data; section 3.4 FTIR analysis). The gradual decrease of sorption with

increasing pH is consistent with the proposed surface complexation reactions 3-1 and 3- 2

(surface complexation reaction and modeling discussed in details in Punamiya et al.,

2013a), in which protons are consumed. Generally, ligand exchange reactions are

governed by strong inner-sphere adsorption and unlikely to be controlled by surface

charge. Further, Al-WTR is redox stable and hence the surface does not easily

disintegrate or lose sites for sorption.

$$XOH + TTC^{2-} + H^+ = XTTC^- + H_2O \quad (3\text{-}1)$$

$$XOH + OTC^{2-} + H^+ = XOTC^- + H_2O \quad (3\text{-}2)$$
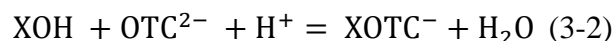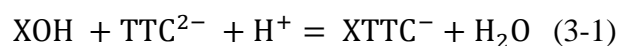
XOH= surface functional group, X = reactive hydroxyl bound to a metal ion (Al)

We have referred Al-hydroxide as a major component of Al-WTR

3.2.2 Effect of competing ligands (phosphate (P(V)) and sulfate) on TTC/OTC sorption

The competitive sorption studies for TTC and OTC and P(V) added

simultaneously and after equilibration of TTC/OTC on Al-WTR were conducted at pH

values between 3 and 11 at optimum SSR (20 g L^{-1}) to obtain sorption envelopes in the

presence of competing ligand. Different initial molar ratios of TTC/OTC to ligands were

chosen, and the sorption was measured after 24 h reaction time for simultaneously added

TTC/OTC and P(V) and 48h for P(V) added after equilibration of TTC/OTC. The total concentrations were chosen to provide near maximum surface coverage, according to previous specific surface area measurements for Al-WTR (Makris et al., 2004). MINTEQ calculations showed no secondary precipitation and/or formation of any mineral at the highest P(V) concentration used (data not shown). The concentrations of P(V) used were based on literature for agricultural runoff of total P (stormwater runoff 30-50 mM, snowmelt 1 mM, and base flow 15-25 mM for pasture with dairy operations, stream water, grassland, and tile drains) (Zaimes and Schultz, 2002).

The presence of P(V) anions in the solution resulted in the significant ($p < 0.005$) decrease in TTC and OTC sorption by Al-WTR at higher initial molar ratios of TTC/OTC:P (1:10 and 1:20) throughout the pH range tested (Fig. 3-3A and B). There was no significant difference ($p > 0.1$) observed at equimolar (1:1) and low molar ratios (1:5) on sorption of TTC/OTC in the presence of P(V). At equimolar (1:1) concentration of TTC/OTC (1mM) and P (1mM), more than 90% of initially added TTC and OTC were sorbed by Al-WTR in the pH range of 3-7. With an increase in TTC:P molar ratio to 1:10, sorption of TTC decreased to 80-82% (pH 3-7) and 74-78% (pH >7) of initially added TTC. At TTC:P molar ratio of 1:20, sorption of TTC further decreased to 73-76% (pH 3-7) and 61-63% (pH >7) of initially added TTC (Fig. 3-3A). The pH dependent sorption behavior for TTC on Al-WTR remained unchanged in the presence of P (V); maximum sorption was observed in pH range 5-7 and a decrease in sorption with an increase in pH above 7. Sorption behavior of OTC on Al-WTR was somewhat different than the sorption behavior of TTC in the presence of P(V) (Fig. 3-3A and B).The

presence of P(V) in the solution had a pronounced negative effect on the sorption of

OTC by Al-WTR compared to the sorption of TTC in the presence of P(V) (Fig. 3-3B).

At molar ratios of 1:10 and 1:20 (OTC:P), sorption of OTC decreased to 80-78% (pH 3-

7) , 73-76% (pH >7), 72-75% (pH 3-7), and 58-61% (pH >7) , respectively of initially

added OTC concentration. However, the pH dependent sorption behavior remained

unchanged in the presence of P(V); maximum sorption was observed in pH range 5-7,

and a decrease in sorption with an increase in pH above 7. In the presence of an

equimolar concentration of TTC/OTC and P(V), P(V) sorption by Al-WTR was higher

than that of TTC/OTC over the entire pH range tested. These results are in accordance

with previous studies, showing a slightly higher affinity of P(V) for Al-WTR (Caporale et

al., 2013; Makris et al., 2004 and 2005; Nagar et al., 2010). However, with the increase in

the molar concentrations of P(V) in the solution, the sorption of P(V) decreased

significantly (Fig. 3-3A and B).

The results obtained in the study are in agreement with previous studies that showed

decrease in the sorption coefficient of TTC/OTC and other antibiotics in presence of

phosphate (Wang et al., 2010; Zhang et al., 2011). Wang et al. (2010) studied the effect

of phosphate on adsorption of TTC on soils. The results from the study showed that the

addition of phosphate significantly decreased the adsorption of TTC on the soils. Zhang

et al. (2011) studied the adsorption of sulfamethoxazole on functionalized carbon

nanotubes. The results from the study showed that the presence of anion (phosphate)

decreased sulfamethoxazole adsorption on carbon nanotubes. The sorption behavior

observed was pH-dependent. The negative effect observed in presence of phosphate on

TTC/OTC sorption by Al-WTR can be explained by the increase in negative charge on the surface and the competition of P(V) for available surface sties on Al-WTR. It may be also explained by the presence of same surface sites or an increase in the surface negativity resulting from P(V) sorption.

Further, competitive sorption effect of P(V) added after equilibration of TTC/OTC on Al-WTR was conducted at pH values between 3 and 11 at optimum SSR (20 g L^{-1}) to obtain sorption envelopes in the presence of different P(V) concentrations (1, 10, and 20mM). Different initial molar ratios of TTC/OTC to P(V) were chosen, and the sorption was measured after 48h for P(V) added after equilibration (24h reaction time) of TTC/OTC on Al-WTR. Results showed there was a decrease in the amount of TTC (Fig. 3-4A) and OTC (Fig. 3-4B) sorbed by Al-WTR in presence of P(V). A decrease in sorption of TTC and OTC by Al-WTR was observed with increase in P(V) molar ratios. However, the decrease in sorption of TTC and OTC by Al-WTR was insignificant (P > 0.1) at all the tested molar ratios (Fig. 3-4 A and B) indicating P(V) can only desorb the weakly bound TTC/OTC in the outer sphere and/or more accessible and then more easily desorbed. The presence of P (V) in the solution after the equilibration of TTC/OTC on Al-WTR was not able to release the strongly bound TTC/OTC as evident from the surface complexation modeling (SCM) and fourier transform infrared spectroscopy (FTIR) analysis which indicates the possibility of TTC and OTC forming a mononuclear monodentate surface complex through strong innersphere-type bonds on Al-WTR (chapter 2 and section 3.2.4 on FTIR analysis). In the entire pH range and
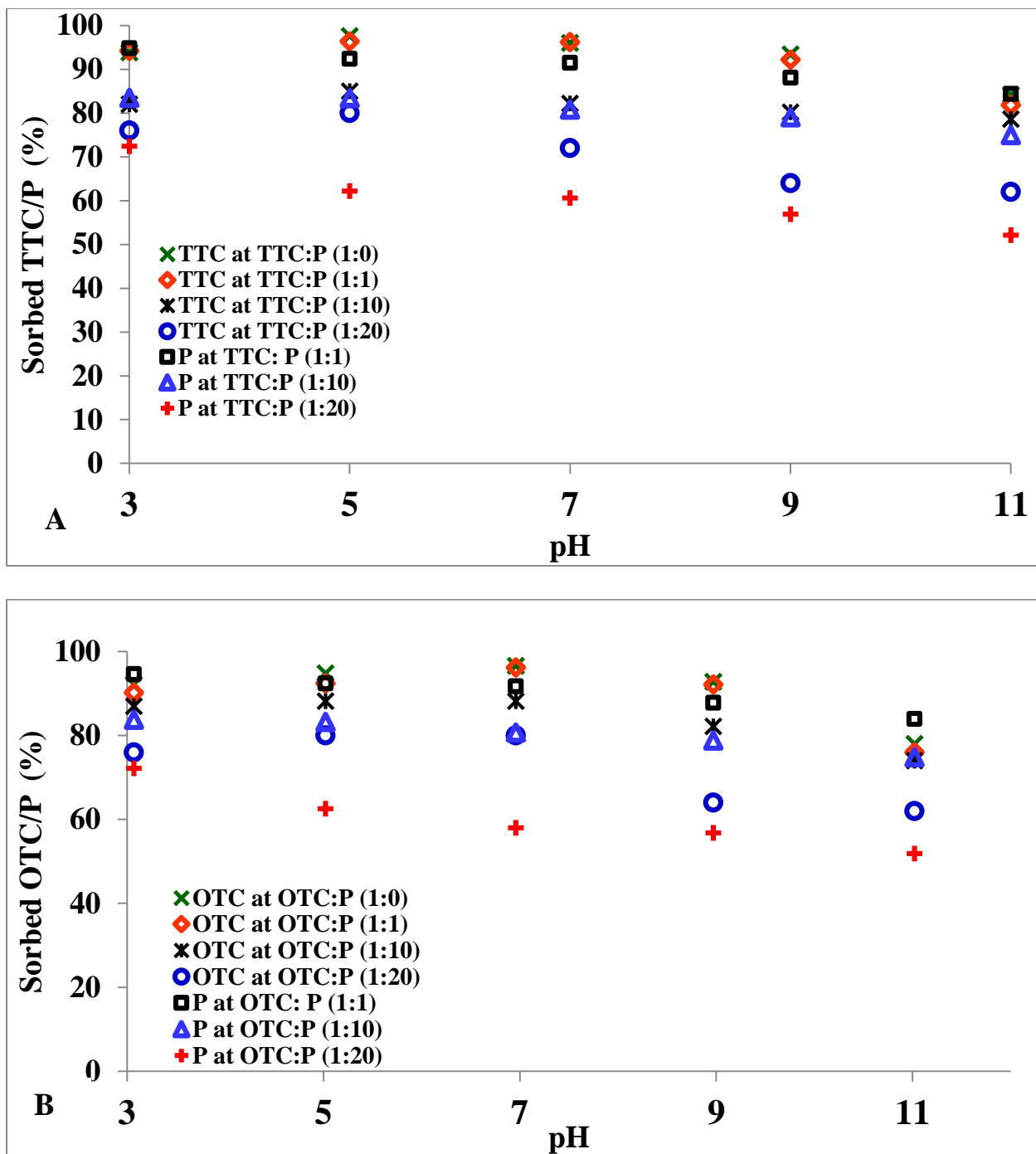
Figure 3-3: The effect of phosphate on the amount of TTC (A) and OTC (B) sorbed by Al-WTR as a function of pH and different TTC/OTC:P ratios. Initial TTC concentration 1.0 mM L^{-1}, reaction time 24 h, and ionic strength buffer 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.
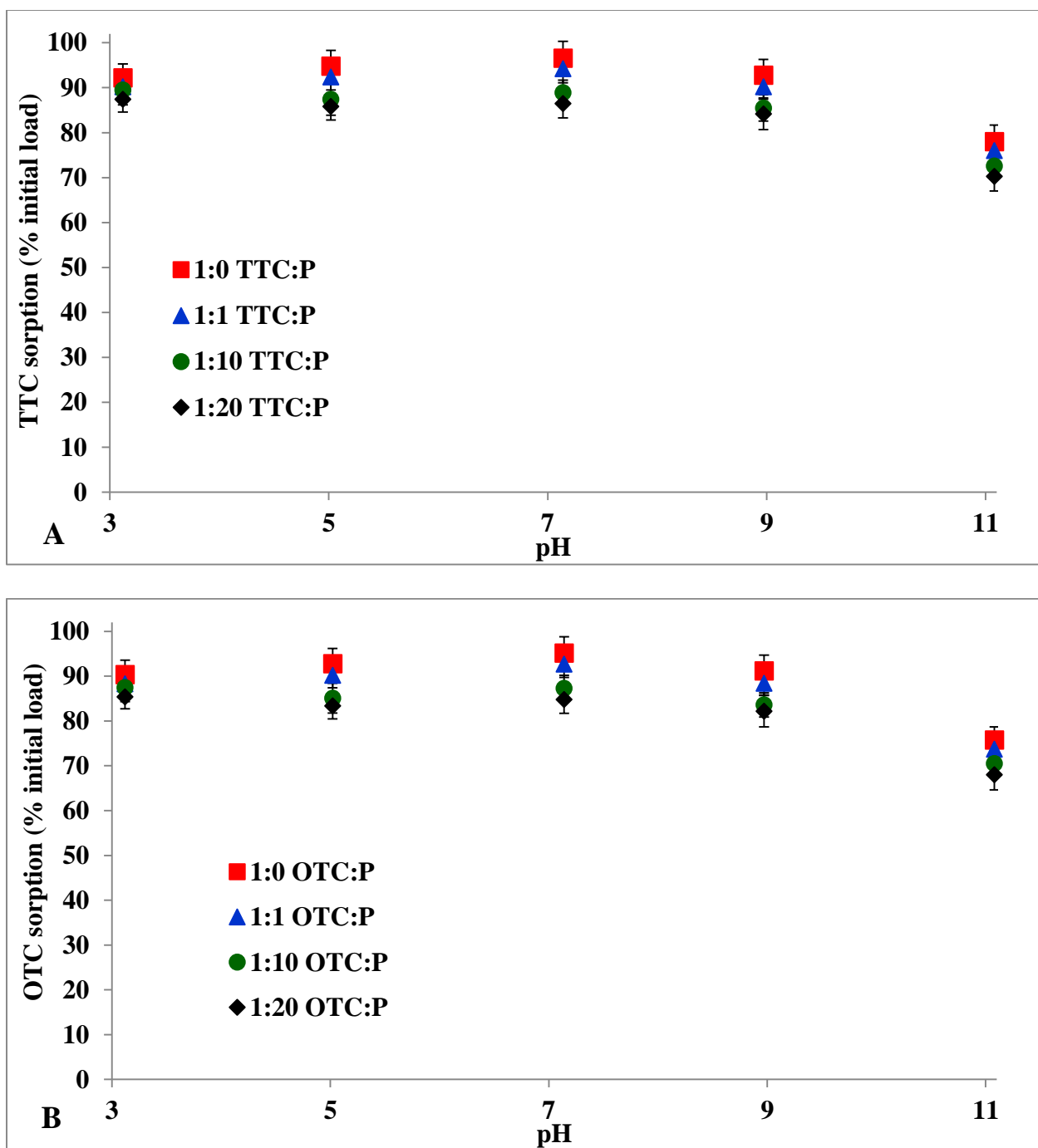
Figure 3-4: The effect of phosphate on the amount of TTC (A) and OTC (B) sorbed by Al-WTR as a function of pH and different TTC/OTC:P ratios. P(V) was added after TTC/OTC equilibration. Initial TTC/OTC concentration 1.0 mM L^{-1}, P(V) concentrations 1, 10, and 20mM, reaction time 24 h, and ionic strength buffer 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.
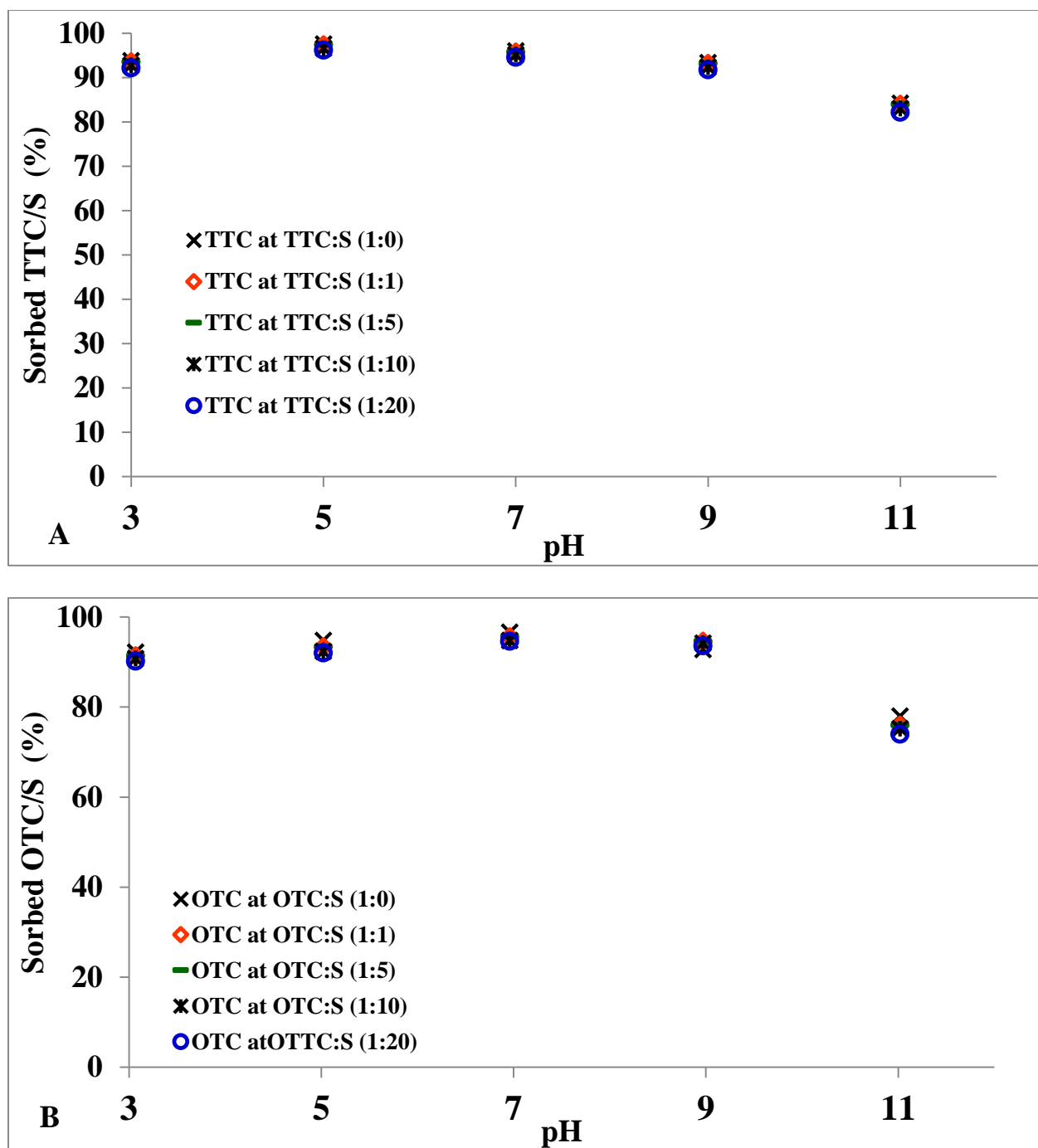
Figure 3-5: The effect of sulfate on the amount of TTC (A) and OTC (B) sorbed by Al-WTR as a function of pH and different TTC/OTC:S ratios. Initial TTC concentration 1.0 mM L^{-1}, reaction time 24 h, and ionic strength buffer 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.

different TTC/OTC molar ratios tested the decrease in TTC and OTC sorption was minimal and ranged from 3-9% of initially added TTC/OTC concentration.

The effect of sulfate (SO_4^{2-}) as a competing ligand was investigated on TTC/OTC adsorption by Al-WTR at optimum SSR (20 g L^{-1}) in commonly encountered environmental pH range (3-11). TTC/OTC and sulfate were added simultaneously in the solution and sorption envelopes were obtained. Presence of sulfate had no significant ($p > 0.1$) effect on TTC and OTC sorption by the Al-WTR (Fig. 3-5A and B). The pH dependent sorption behavior for TTC and OTC on Al-WTR remained unchanged in the presence of sulfate. Maximum sorption was observed between pH 5 to 7 and adsorption decreased with an increase in pH above 7. In the pH range of 5-7, 92-96% of initially added TTC was sorbed, followed by 87-90 % in pH range 3-5, and 82-85% in pH range 7-11, respectively (Fig. 3-5A) .The sorption of OTC was slightly lower but not significantly different than that of TTC by Al-WTR in the presence of sulfate. The maximum OTC sorption was observed in pH range 5-7 (89-93%), followed by 84-87% in pH range 3-5 and 72-80% in pH range 7-11 (Fig. 3-5B). The sorption behavior for TTC and OTC by Al-WTR remained unchanged despite the wide variation in added sulfate concentrations (1-20 mmol L^{-1}). This insignificant effect of sulfate on TTC and OTC sorption on Al-WTR could be explained by strong binding sites for TTC and OTC on Al-WTR surfaces (Punamiya et al., 2013). Also, study by Yang et al (2006) has shown that sulfate may form weaker or outer-sphere complexes.

3.2.3 Effect of complexing metal/cation (calcium) on TTC/OTC sorption

The presence of bivalent or trivalent metal ions in the solution has shown to alter the distribution of TTC and OTC by competing for charge sites on the surface or via complexation (Bao et al., 2010; Ter Laak et al., 2006). TTC and OTC have a strong tendency to form a complex with various bivalent and trivalent metal ions (Figueroa and Mackay, 2004; Ter Laak et al., 2006). Previous studies have shown that TTC and OTC forms a strong 2:1 metal ligand complex with multivalent cations via chelation (Schmitt and Schneider, 2000; Gu and Karthikeyan, 2005). It is important to study complexation of TTC and OTC with Ca^{2+} because of its abundance in water and soil systems. In current study, we investigated the effect of calcium on TTC/OTC sorption by Al-WTR at optimum SSR (20 g L^{-1}) in the environmental pH range (3-11). TTC/OTC and Ca^{2+} were added simultaneously in the solution and sorption envelopes were obtained.

There was a significant positive, as well as a negative effect ($p < 0.05$) on the sorption of TTC and OTC by Al-WTR in the pH range tested (Fig 3-5A and B). At an equimolar ratio of TTC and Ca^{2+}, sorption decreased by 12% in the pH range 3-5 and 5-6% in the pH range 5-7 when compared to the control (only TTC) (Fig. 3-6A). However, there was no significant effect on TTC sorption at pH 9, and slightly higher sorption was observed at pH 11. Similar behavior was observed by increasing the molar ratios to 1:5 and 1:10 (TTC: Ca) in the entire pH range tested. Increasing the molar ratio to 1:20 (TTC: Ca) had a significant negative effect at pH 3 where 20% decrease in sorption was observed. A significant positive effect at pH 11 was seen, where sorption of TTC by Al-WTR increased by 9% (Fig. 3-6A). Similar sorption behavior was observed for OTC by
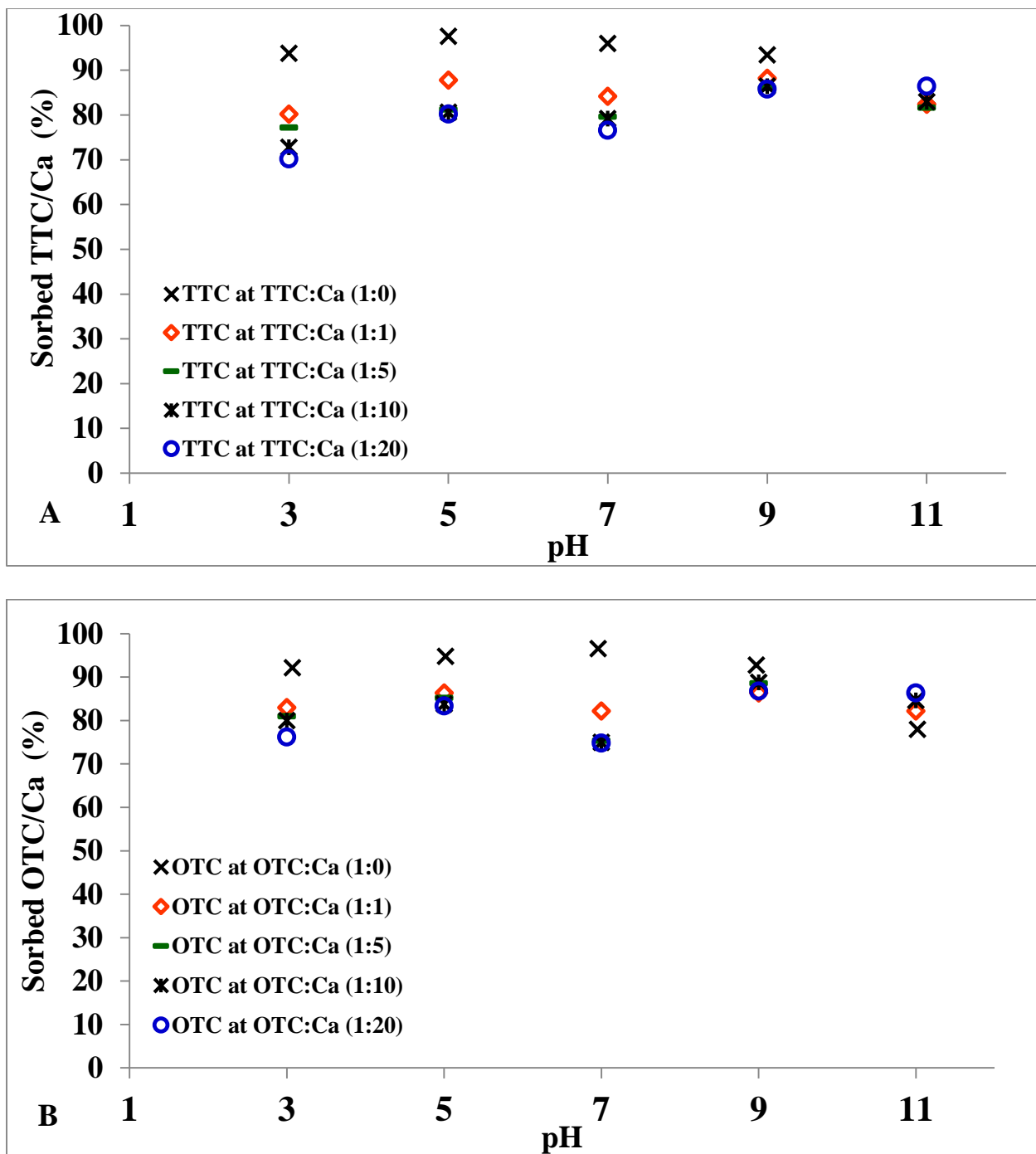
Figure 3-6: The effect of calcium on the amount of TTC (A) and OTC (B) sorbed by Al-WTR as a function of pH and different TTC/OTC:Ca ratios. Initial TTC concentration 1.0 mM L^{-1}, reaction time 24 h, and ionic strength buffer 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.
the presence of a cation (Ca^{2+}).

Al-WTR in presence of Ca in the solution (Fig. 3-6B). In brief, at an equimolar ratio of OTC and Ca, in the pH range 3-7, sorption of OTC decreased by 5-11% of the initial OTC concentration. Increasing the molar ratios to 1:5 and 1:10 (OTC: Ca) did not have a significant effect on sorption. The sorption behavior observed was similar to equimolar ratio of OTC and Ca (Fig. 3-6B). However, a significant effect was observed at molar ratio of 1:20 (OTC: Ca), at pH 11 the sorption of OTC by Al-WTR increased by 7%, whereas at pH 3 the sorption decreased by 23%.

Results indicated that calcium enhanced the sorption of both TTC and OTC by Al-WTR under alkaline conditions. This can be explained by the reversal of surface charge in the presence of a cation (Ca^{2+}). For example, when WTR surfaces are positively charged, Ca^{2+} complexation with OTC/TTC will increase their positive charges; therefore, there will be strong repulsion towards surface at that pH. However, at higher pH, WTR surface charge will be modified by Ca^{2+} to make it positive rather than negative. Therefore, at higher pH, since OTC/ TTC has negative charge, the sorption will increase. However, these increase and decrease due to surface charge modifications are minor because the sorption process is inner-sphere. Similar Ca-induced increase in TTC and OTC sorption at higher pH (pH >7) have been reported for clays, minerals, oxides, and soils (Zhao et al., 2012; Bao et al., 2010; Jia et al., 2008., Pils and Laird, 2007; Teer Laak et al., 2006; Figueroa et al., 2004). The calcium enhances sorption under alkaline pH conditions can be also explained by the surface bridging mechanism or surface charge modification. Further, it can be explained by the cation acting as the bridge between the surfaces of the TCs and Al-WTR. Cationic bridging as one of the mechanism of sorption

of TCs on different sorbents (clays, minerals, oxides, and soils) has been advocated by various studies (Zhao et al., 2012; Pils and Laird, 2007; Gu et al., 2007; Teer Laak et al., 2006; Figueroa et al., 2004).

3.2.4 FTIR analysis

The ATR-FTIR spectra of crystalline TTC at pH 5.5 , Al-WTR (20 g L^{-1}) equilibrated for 24 h with TTC (5 x 10^{-3} M) at pH 4.12 , pH 6.3 , pH 9.11, and untreated Al-WTR are shown in figure 3-7A (a-e) and spectra of crystalline OTC at pH 5.52 , Al-WTR (20 g L^{-1}) equilibrated for 24 h with OTC (5 x 10^{-3} M) at pH 4.15 , pH 6.32 , pH 9.13, and untreated Al-WTR are shown in figure 3-7B (a-e). A comparison of the characteristic IR bands of crystalline OTC/ TTC with OTC/TTC treated Al-WTR at various pH values indicated several key aspects of the surface interaction mechanism. Detailed assignments of the IR peaks for standard OTC / TTC molecule are discussed elsewhere (Boyd et al., 1995; Aristilde et al., 2010; Rakshit et al., 2013b). Briefly, the IR peak near 1678 cm^{-1} represents amide I (the C=O group of –CONH$_2$), peaks at 1619 & 1584 cm^{-1} indicates the presence of amide II (two N-H bonds of –CONH$_2$), and peaks near 1534 & 1445 cm^{-1} verifies the presence of skeletal C=C vibrations. The peak near 1445 cm^{-1} also represents C-H vibrations of the methyl in –N(CH$_3$)$_2$ groups. The change of IR bands in TTC and OTC treated Al-WTR compared to that of crystalline TTC/OTC revealed a similar feature. It is noted that the IR bands (1679, 1619, 1584, 1534, and 1446 cm^{-1}) in the TTC/OTC treated Al-WTR shifted to lower wave numbers (1664, 1596,
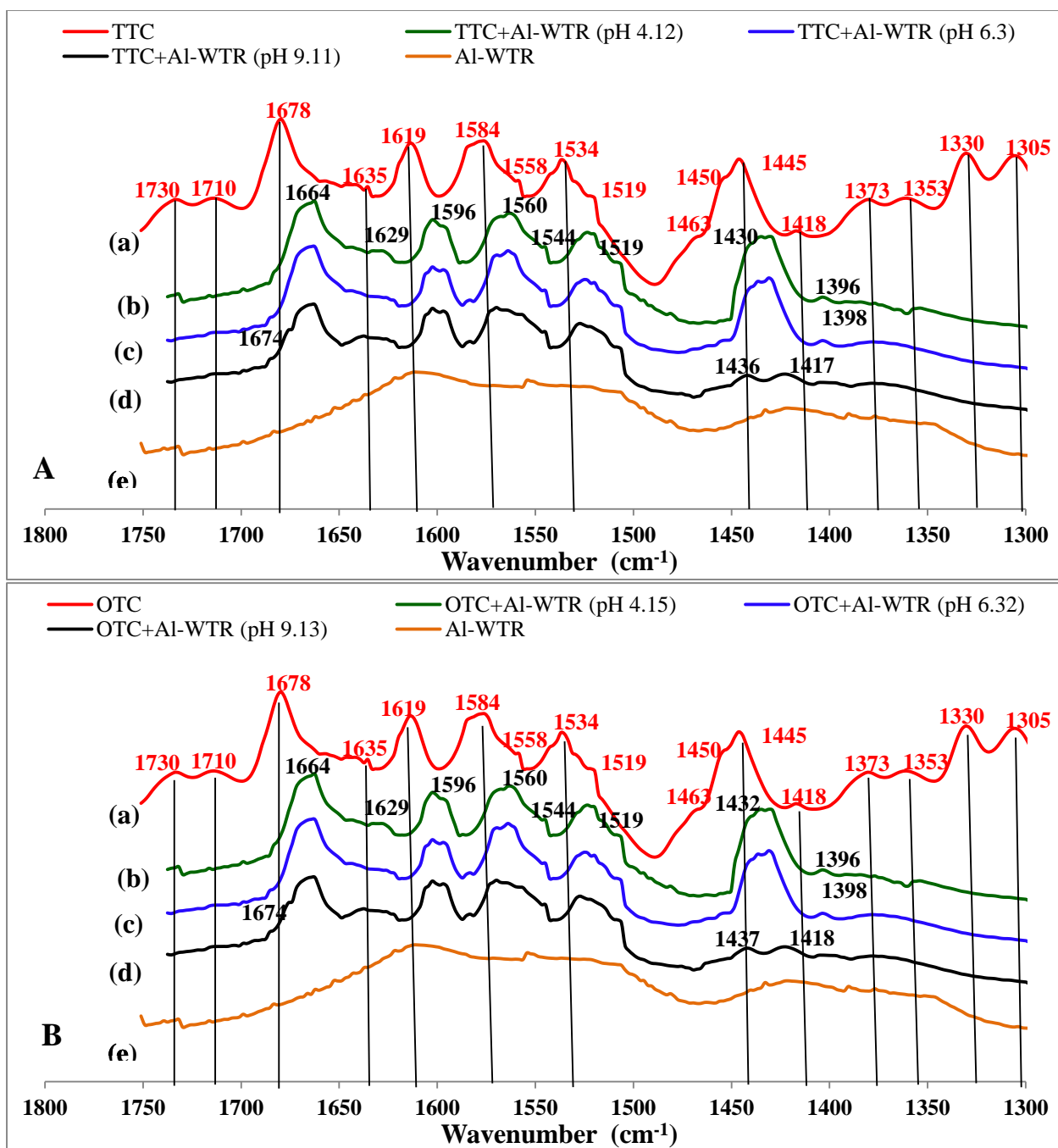
Figure 3-7: ATR-FTIR spectra; (8A): (a) crystalline TTC at pH 5.5 , Al-WTR (20 g L^{-1}) equilibrated for 24 h with TTC (5 x 10^{-3} M) at pH 4.12 (b) , pH 6.3 (c) , pH 9.11 (d), and (e) untreated Al-WTR; (8B) : a) crystalline OTC at pH 5.52 , Al-WTR (20 g L^{-1}) equilibrated for 24 h with OTC (5 x 10^{-3} M) at pH 4.15 (b) , pH 6.32 (c) , pH 9.13 (d), and (e) untreated Al-WTR. All spectra are normalized with the highest peak observed.

1560, 1519, and 1430 cm^{-1}) (Figure 3-7). These types of lower wave number shifts occur from weakening of the chemical bonds, which in turn is resulting from strong covalent interactions of specific functional groups of TTC/OTC molecule with the Al-WTR surface. For example, a strong covalent interaction of the N atom of the $-CONH_2$ group with Al-WTR would weaken the N-H bonds; therefore, a lower wave number shift (1584 $\rightarrow$ 1560 cm^{-1}) was observed (Rakshit et al., 2013b). A detailed discussion on the possible shifts pertaining to strong covalent interactions of OTC /TTC molecule with surface is noted in our earlier work (Rakshit et al., 2013b; Punamiya et al., 2013a). Overall, the IR data indicated that TTC/OTC molecule interacted with Al-WTR surface through strong covalent bond formation with $-CONH_2$ and $-N(CH_3)_2$ functional groups.

A comparison of the IR data collected for OTC/TTC treated Al-WTR at different pH values resulted in some additional aspects of the surface interaction mechanism. The IR peak at 1432 cm^{-1} in the spectra of adsorbed OTC/TTC on Al-WTR collected at pH values 4.12 & 6.3 was almost absent at pH 9.11 (Figure 3-7). IR bands in this region are characterized by skeletal C=C vibrations and C-H vibrations. At higher pH values $-N(CH_3)_2$ group of OTC/ TTC molecule exerts stronger interaction with the surface because at lower pH values $-N(CH_3)_2$ would be protonated; hence, the lone pair of electrons on N atom would be unavailable for covalent bonding. The disappearance of IR band (1432 cm^{-1}) at pH 9.11 most likely resulted from the change of C-H vibrations upon strong covalent interactions of N atom of $-N(CH_3)_2$ group with Al-WTR.

3.3 Conclusions

The batch sorption study demonstrated the effectiveness of Al-WTR to remove TTC and OTC from aqueous medium in the environmental pH range in the presence of commonly encountered competing ligands (phosphate and sulfate) and complexing metal (calcium). Al-WTR exhibited high sorption affinity for both TTC and OTC irrespective of high initial concentrations of TCs (up to 5mM), presence of anions, and cation, when added simultaneously in solution at optimum SSR (20 g L^{-1}) at pH ranging from 3-11. A pH-dependent behavior was observed for all the treatments tested. ATR-FTIR spectroscopic data revealed that the interaction of OTC and TTC with Al-WTR surface is a strong inner-sphere type and occurred via $–CONH_2$ and $–N(CH_3)_2$ functional groups. The presence of P(V) when added simultaneously decreased adsorption and removal efficiency of TTC and OTC by Al-WTR but minimal effect was observed when added after equilibration of TTC and OTC. Maximum adsorption of TTC and OTC by Al-WTR in presence of P(V) was seen between pH 5- 7. The effect of sulfate on TTC and OTC sorption by Al-WTR was minimal. Calcium promoted adsorption of TTC and OTC sorption on Al-WTR in alkaline pH condition (pH > 7); however, it decreased adsorption when pH was below 5. Results from the study show that a change in SSR, environmental pH, or the presence of competing ligands and complexing metal can significantly change the sorption behavior and distribution of TTC and OTC in aqueous medium. Therefore, it is necessary to take into account these environmental conditions while developing effective wastewater treatment and remediation strategies for TCs. Results from the

current study provide a detailed understanding of TTC and OTC sorption by Al-WTR at varying solution chemistry. The results obtained will be useful in developing a potential in situ "green" sorbent for treatment of TCs contaminated water at municipal wastewater treatment plants and CAFO's. Advanced experimental and modeling studies are required to better understand the sorption mechanism of TCs by Al-WTR. Ongoing incubation and greenhouse studies focus on evaluating the efficacy of Al-WTR in manure and manure amended soil.

3.4 References

- Aga, D. S. 2008. Fate of pharmaceuticals in the environment and in water treatment systems. CRC Press: Boca Raton, FL.
- Allen, H.K., Donato, J., Wang, H.H., Cloud-Hansen, K.A., Davies, J., Handelsman, J., 2010. Call of the wild: antibiotic resistance genes in natural environments. Nature Reviews Microbiology 8, 251-259.
- Aristilde, L., Marichal, C., Miehe-Brendle, J., Lanson, B., Charlet, L., 2010. Interactions of Oxytetracycline with a Smectite Clay: A Spectroscopic Study with Molecular Simulations. Environmental Science & Technology 44, 7839-784.
- Bao, Y.Y., Zhou, Q.X., Wan, Y., Yu, Q.A., Xie, X.J., 2010. Effects of soil/solution ratios and cation types on adsorption and desorption of tetracycline in soils. Soil Sci. Soc. Am. J. 74, 1553-1561.
- Ben-Dor E., Banin. A., 1989. Determination of organic matter content in arid-zone soils using a simple "loss-on-ignition" method. Commun. Soil Sci. Plant Anal. 20:1675–1695
- Boyd, S.L., Boyd, R.J., Bossonnette, P.W., Kerdaron, D.I., Aucoin, N.T., 1995. A theorrtical study of the effects of protonation and deprotonation on bond dissociation energies. J. Am. Chem. Soc. 117, 8816-8822.
- Caporale, A. G., Punamiya, P., Pigna, M., Violante, A., Sarkar, D., 2013. Effect of particle size of drinking-water treatment residuals on the sorption of arsenic in the presence of competing ions. J. Hazard Mater.260, 644-651
- Chen, W.-R., Huang, C.-H., 2010. Adsorption and transformation of tetracycline antibiotics with aluminum oxide. Chemosphere 79, 779-785.
- Datta, R., Das, P., Smith, S., Punamiya, P., Ramanathan, D.M., Reddy, R., Sarkar, D., 2013. Phytoremediation potential of vetiver grass [Chrysopogon zizanioides (L.)] for tetracycline. *Internatl.* J. Phytorem. 15:343-351
- Elliott, H.A., Dempsey, B.A., 1991. Agronomic effects of land application of water treatment sludges. J. AWWA, 83 126–131.
- Figueroa, R.A., Leonard, A., MacKay, A.A., 2004. Modeling Tetracycline Antibiotic Sorption to Clays. Environmental Science & Technology 38, 476-483.
- Figueroa, R.A., MacKay, A.A., 2005. Sorption of Oxytetracycline to Iron Oxides and Iron Oxide-Rich Soils. Environmental Science & Technology 39, 6664-6671.
- Fritz, J.W., Zuo, Y., 2007. Simultaneous determination of tetracycline, oxytetracycline, and 4-epitetracycline in milk by high-performance liquid chromatography. Food Chemistry 105, 1297-1301.
- Gu, C., Karthikeyan, K., 2005. Interaction of tetracycline with aluminum and iron hydrous oxides. Environmental Science & Technology 39, 2660-2667.
- Gu, C., Karthikeyan, K.G., Sibley, S.D., Pedersen, J.A., 2007. Complexation of the antibiotic tetracycline with humic acid. Chemosphere 66, 1494-1501.
- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997a. Soil pH (1:2v/v). IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL)

chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p15.

- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997b. Electrical Conductivity. IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL) chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p24.

- Ippolito, J., Barbarick, K., Elliott, H., 2011. Drinking Water Treatment Residuals: A Review of Recent Uses. Journal of Environmental Quality 40, 1-12.

- Ji, L., Chen, W., Duan, L., Zhu, D., 2009. Mechanisms for strong adsorption of tetracycline to carbon nanotubes: A comparative study using activated carbon and graphite as adsorbents. Environmental Science & Technology 43, 2322-2327.

- Jia, D.A., Zhou, D.M., Wang, Y.J., Zhu, H.W., Chen, J.L., 2008. Adsorption and cosorption of Cu(II) and tetracycline on two soils with different characteristics. Geoderma 146 (1–2), 224–230.

- Kang, J., Liu, H.J., Zheng, Y.M., Qu, J.H., Chen, J.P., 2011. Application of nuclear magnetic resonance spectroscopy, Fourier transform infrared spectroscopy, UV-Visible spectroscopy and kinetic modeling for elucidation of adsorption chemistry in uptake of tetracycline by zeolite beta. Journal of Colloid and Interface Science 354, 261-267.

- Kemper, N. 2008. Veterinary antibiotics in the aquatic and terrestrial environment.Ecol. Indicators 8:1-13.

- Kulshrestha, P., Giese, R.F., Aga, D.S., 2004. Investigating the Molecular Interactions of Oxytetracycline in Clay and Organic Matter: Insights on Factors Affecting Its Mobility in Soil. Environmental Science & Technology 38, 4097-4105.

- Kümmerer, K. 2009. Antibiotics in the aquatic environment – A review – Part I.Chemosphere 75:417-434.

- Loeppert, R.H., Inskeep, W.P., 1996. Iron. In: Sparks, D.L., Page, A.L., Helmke, P.A., Loeppert, R.H., Soltanpour, P.N., Tabatabai, M.A., Johnston, C.T., Summer, M.E. (Eds.), Method of Soil Analysis Part 3 Chemical methods, SSSA Book Series, pp. 639–664.

- Makris, K., Harris, W., O'Connor, G., Obreza, T., 2004. Phosphorus immobilization in micropores of drinking-water treatment residuals: Implications for long-term stability. Environmental Science & Technology 38, 6590-6596.

- Makris, K., Harris, W., O'Connor, G., Obreza, T., Elliott, H., 2005. Physicochemical properties related to long-term phosphorus retention by drinking-water treatment residuals. Environmental Science & Technology 39, 4280-4289.

- Nagar, R., Sarkar, D., Makris, K., Datta, R., 2010. Effect of solution chemistry on arsenic sorption by Fe- and Al-based drinking-water treatment residuals. Chemosphere 78, 1028-1035.

- Netthisinghe, A. M. P., Cook, K. L., Rice, C., Gilfillen, R. A., Sistani, K. R., 2013. Soil Nutrients, Bacteria Populations, and Veterinary Pharmaceuticals across a Backgrounding Beef Feedlot. J. Environ. Qual. 42, 532–544

- O'Connor, G.A., Elliott, H.A., Lu, R., 2002. Characterizing water treatment residuals phosphorus retention. Soil and Crop Science Society of Florida Proceedings 61, 67-73.

- Oberle, K., Capdeville, M.J., Berthe, T., Budzinski, H., Petit, F., 2012. Evidence for a Complex Relationship between Antibiotics and Antibiotic-Resistant Escherichia Coli: From Medical Center Patients to a Receiving Environment. Environmental Science & Technology 46, 1859-1868.

- Pei, Z.G., Shan, X.Q., Zhang, S.Z., Kong, J.J., Wen, B., Zhang, J., Zheng, L.R., Xie, Y.N., Janssens, K., 2011. Insight to ternary complexes of co-adsorption of norfloxacin and Cu(II) onto montmorillonite at different ph using EXAFS. Journal of Hazardous Materials 186, 842-848.

- Pils, J., Laird, D., 2007. Sorption of tetracycline and chlortetracycline on K- and Ca-saturated soil clays, humic substances, and clay-humic complexes. Environmental Science & Technology 41, 1928-1933.

- Prakash, P., Sengupta, A.K., 2003. Selective coagulant recovery from water treatment plant residuals using donnan membrane process. Environmental Science & Technology 37, 4468-4474.

- Punamiya, P., Sarkar, D., Rakshit, S., Datta, R., 2013. Effectiveness of Al- based drinking water treatment residuals as a novel sorbent to remove tetracyclines from aqueous medium. J. Environ. Qual. *doi:10.2134/jeq2013.03.0082*.

- Rakshit, S., Sarkar, D., Elzinga, E.J., Datta, R.., 2013b. In Situ ATR-FTIR study of Oxytetracycline Sorption on Magnetite . J. Environ. Qual. 42:822-827.

- Rakshit, S., Sarkar, D., Elzinga, E.J., Punamiya, P., Datta, R., 2013a. Mechanisms of ciprofloxacin removal by nano-sized magnetite. J. Hazard Mater. 246-247:221-226.

- Rakshit, S., Punamiya, P., Datta, R., Sarkar, D., 2010. Sorption of oxytetracycline on magnetite-water interface. Geochimica Et Cosmochimica Acta 74, A846-A846.

- Sarkar, D., Makris, K., Vandanapu, V., Datta, R., 2007. Arsenic immobilization in soils amended with drinking-water treatment residuals. Environmental Pollution 146, 414-419.

- Sall, J., Creighton, L., Lehman, A., 2005. JMP start statistics, 8th edition. Cary, NC, SAS Institute.

- Schmitt, M.O., Schneider, S., 2000. Spectroscopic investigation of complexation between various tetracyclines and Mg^{2+} or Ca^{2+}. Phys. Chem. Comm. 9, 1–14.

- Ter Laak, T.L.,Wouter, A.G., Tolls, J., 2006. The effect of pH and ionic strength on the sorption of sulfachloropyridazine, tylosin, and oxytetracycline to soil. Environ. Toxicol. Chem. 25, 904–911.

- Wang, Y.J., Jia, D.A., Sun, R.J., Zhu, H.W., Zhou, D.M., 2008. Adsorption and cosorption of tetracycline and copper(II) on montmorillonite as affected by solution pH. Environmental Science & Technology 42 (9), 3254–3259.

- Wang, Y. J., Sun,R. J., Xiao, A. Y., Wang, S. Q., Zhou, D. M., 2010: Phosphate affects the adsorption of tetracycline on two soils with different characteristics. Geoderma, 156, 237-242.

- Watkinson, A. J., Murby, E. J., Kolpin, D. W., Costanzo, S. D., 2009. The occurrence of antibiotics in an urban watershed: From wastewater to drinking water. Sci. Total Environ. 407 (8), 2711−2723.

- Yang, Y., Zhao, Y.Q., Babatunde, A.O., Wang, L., Ren, Y.X., Han, Y., 2006. Characteristics and mechanisms of phosphate adsorption on dewatered alum sludge. Sep. Purif. Technol. 51, 193-200.

- Zaimes, G. N., Schultz, R, C., 2002. Phosphorus in Agricultural Watersheds: A Literature Review. Iowa State University, Ames, Department of Forestry

- Zhang, D., Pan, B., Wu, M., Wang, B., Zhang, H., Peng, H., Wu, D., Ning, P., 2011. Adsorption of sulfamethoxazole on functionalized carbon nanotubes as affected by cations and anions. Environmental Pollution 159, 2616-2621.

- Zhang, Y., Cai, X.Y., Lang, X.M., Qiao, X.L., Li, X.H., Chen, J.W., 2012. Insights into aquatic toxicities of the antibiotics oxytetracycline and ciprofloxacin in the presence of metal: Complexation versus mixture. Environmental Pollution 166, 48-56.

- Zhao, Y.P., Gu, X.Y., Gao, S.X., Geng, J.J., Wang, X.R., 2012. Adsorption of tetracycline (TC) onto montmorillonite: Cations and humic acid effects. Geoderma 183, 12-18.

CHAPTER 4

Al-Based Drinking Water Treatment Residuals as an Effective Low-Cost Sorbent for Immobilization and Stabilization of Tetracyclines in Manure And Manure Amended Soils: A Short Term Incubation Study

[Portion of this chapter has been submitted to Environ. Sci. Pollut. Res. for Publication]

Abstract

In the current incubation study, we evaluated the effectiveness of Al-based drinking water treatment residuals (Al-WTR) in immobilizing and stabilizing tetracycline (TTC) and oxytetracycline (OTC) in manure and manure amended soils as a function of initial antibiotics concentration, reaction time, and the presence of competing agricultural species (phosphate and sulfate). Cattle manure and two physico-chemically variant soil types (from Immokalee and Belleglade series) were chosen based on their potential differences vis-a-vis TTC/OTC reactivity. Manure and soil samples were spiked with various concentrations of TTC/OTC (0.22 to 22.5 mg/kg), amended at three rates (0, 25, and 50 g/kg) of Al-WTR, and incubated for a total of 90d with periodic sampling and analysis. The equilibration time to reach maximum TTC/OTC sorption for Al-WTR-amended soil, manure, and soil-amended manure was 7, 10, and 14d, respectively. Kinetic data was best described by the pseudo-second order rate model ($R^2 = 0.99$). The presence of phosphate in manure-soil system resulted in significant ($p<0.001$) decrease in TTC/OTC sorption by Al-WTR, with a strong dependence on initial phosphate concentration. Addition of sulfate had negligible or minimal effect on TTC/OTC sorption. Fourier transform infrared spectroscopy analysis indicated the possibility of

TTC and OTC forming surface complex through strong innersphere-type bonds on soils, manure, and manure applied soils amended with Al-WTR. Results obtained from the current incubation study showed high and rapid capacity of Al-WTR to immobilize and stabilize TTC and OTC in manure and manure amended soils. This research will be helpful in optimizing Al-WTR as a low cost green sorbent for remediation of TTC/OTC-contaminated manure and manure amended soils.

Keywords: Tetracyclines, Drinking Water Treatment Residuals, Soil, Manure, Remediation

4. Introduction

Veterinary antibiotics (VAs) are extensively used in concentrated animal feeding operations (CAFOs) to therapeutically, prophylactically, and sub-therapeutically treat exiting animal disease, mitigate bacterial pathogens , and as a supplement to boost growth, respectively (Sarmah et al., 2006). About 27.8 million pounds of antibiotics were administered to farm and companion animals annually from 2009 to 2011 in the U.S. (AHI 2011). Kulshrestha et al. (2004) and Kumar et al (2005) reported that 25 to 90% of administered VAs are excreted in an antimicrobially active form by means of urine and manure. Although accidental leakage or leaching from CAFOs waste storage can be a source of VAs in the environment, they are generally introduced to agricultural fields via land application of manure as low cost fertilizer.

USDA/ERS estimated about 132 million metric tons (dry weight) of manure of generated annually from confined livestock and poultry animals in the U.S. (USDA/ERS 2005). Due to enormous amount of manure generated every year, there has been increasing attention and concern from the regulatory, public, and scientific communities towards disposal of VAs-contaminated manure in the environment. Land application of manure is a common practice in the U.S. since it serves the dual purpose of supplying nutrients to crops as well as serving as a means of disposal. Studies has shown that due to various socio-economic reasons, significant amount of manure is usually land applied as a source of fertilizer to the adjoining arable lands and farms without manure been transported after they are stock piled, lagooned, or composed at CAFOs (Hakk et al., 2005; Wang and Yates, 2008b). The repetitive land application of VAs rich manure that

contains persistent antibiotics (e.g. tetracyclines group) has led to increasing

concentration of VAs in the soils and development of antibiotic resistant bacteria in the

environment (Zhang et al., 2013; Oberle et al., 2012; Munir and Xagoraraki, 2011).

Researchers have shown that antibiotic resistant bacterial genes may be transferred to

humans and animals through drinking water and food chains, resulting in diminished

success in antibiotic treatment (Zhang et al., 2013; Chee-Sanford et al., 2001; Sengelov et

al., 2003). The VAs present in the manure and soil can also affect the soil microbial

system, thus disturbing the ecosystem function in nutrient recycling and decomposition

(Westgaard et al., 2001; Muller et al., 2002). The VAs may also be transported to the

streams and rivers via runoff and drain flow through VAs rich land applied manure to

soils (Kay et al., 2004; Stoob et al., 2007; Topp et al., 2008; Ostermann et al., 2013) to

ground water via leaching (Blackwell et al., 2007) and may also enter food chain (Boxall

et al., 2006).

   Tetracyclines (TCs) including tetracycline (TTC), oxytetracycline (OTC),

Chlortetracycline (CTC), and doxycycline are the most widely used VAs at CAFO's in

the U.S. (AHI 2011). Batch laboratory studies, incubation, and field experiments have

shown that TCs are absorbed and remain persistent in manure and manure amended soils

(Bassil et al., 2013; McClellan and Halden, 2010; Hamscher et al., 2005.). The high

polarity (e.g., log K_{ow} (octanol-water partition coefficient) = -1.97 to -0.47) and

consequently high aqueous solubility (0.52−117 mM) of TCs portends that TCs could be

highly mobile in soils (Tolls 2001). TCs have been found in the surface soils amended

with manure in concentration ranging from micrograms to milligrams per kilogram (Aga

et al., 2005, Hamscher et al., 2002). A review by Kumar et al. (2005) reported trace levels to > 200 mg kg^{-1} or L^{-1} antibiotic concentration in manure with typical concentration in the range of 1 to 10 mg kg^{-1} or L^{-1}. Hamscher et al. (2002) detected high levels (20 mg kg^{-1}) of TTC in the top soil fertilized with liquid manure, Winckler and Grafe (2001) found TCs to persist in agricultural soils at concentrations of 450–900 μg kg^{-1}, and Hamscher et al. (2005) reported concentrations of up to 270 μg kg^{-1} (OTC), 443 μg kg^{-1} (TTC), and 93 μg kg^{-1} (CTC) in manure impacted surface soils. Further, Kay et al. (2004) observed that after manure was applied to a field in 2 consecutive years, OTC concentration in the soil was 1691 μg kg^{-1}, and concentrations as high as 613.2 μg L^{-1} were observed in drain flows, which end up in aquatic systems through agricultural effluent discharges. Sarmah et al. (2006) also reported high possibility of TCs, being release from soil surface to mineral horizon to the aquifer, causing surface and groundwater contamination. Thus, proper treatment of TCs rich animal manure is required before its application in the agricultural land as a source of nutrient to avoid its transportation, leaching, and release in to the aqueous environment.

On-farm manure management strategies such as on-site storage and composting (Dijk and Keukens, 2000), composting with straw and hardwood chips and heating (Arikan et al., 2007), composting with addition of water, aeration, mixing, and heat treatment (Dolliver et al., 2008), commercial hardwood biochars (Teixidó et al., 2013), etc. have been used with relative success. However, effective means of immobilizing VAs in soils needs to be employed to reduce the potential health hazards that could result from the presence of VAs in the soil environment, and consequently in surface and

ground water. In the current study we evaluated the effectiveness of Al-based drinking water treatment residuals (Al-WTR) as a "green" sorbent and an inexpensive surrogate for Al-hydro(oxides) to immobilize and stabilize TCs in manure and manure applied soils to lower the release of TCs in aqueous medium. Studies have shown that adsorption of VAs on minerals in soils decrease their antibacterial activity and thereby decrease adverse effect on biota (Chander et al., 2005).

Millions of tons of WTR are generated every day in water treatment facilities in the U.S. and typically landfilled (Prakash and Sengupta, 2003). Water treatment residuals are a by-product of the drinking water treatment process and can be obtained free of charge from drinking water treatment facilities. The Al-WTR are primarily amorphous masses of aluminum (Al) hydroxides (indicating the use of Al salt) that contain sediment and humic substances removed from the raw water as well as activated carbon and polymers (Elliott and Dempsey, 1991; O'Connor et al., 2002; Makris et al., 2005; Ippolito et al., 2011). Our previous research has demonstrated high affinity of Al-WTR for TCs in aqueous medium under different environmental conditions (Punamiya et al., 2013a and 2013b) .Additionally, Punamiya et al. (2013a) has shown that the toxicity characteristics leaching procedure (TCLP) values of Al-WTR are well below the hazardous waste toxicity characteristics criterion as defined in Title of the Code of Federal Regulations, part 261.24, permitting land application of WTR as environmentally safe disposal method.

The objectives of the current laboratory incubation study were; i) to determine TTC and OTC sorption isotherm and adsorption kinetics in TTC/OTC rich cattle manure

and two physico-chemically variant manure amended soil types from Immokalee and Belleglade series, respectively mixed with two Al-WTR application rates; and ii) to evaluate effectiveness of Al-WTR as a sorbent to immobilize and stabilize TTC and OTC in TTC/OTC rich manure and manure amended soils as a function of initial antibiotics concentration, reaction time, and the presence of competing agricultural species (phosphate and sulfate).

4.1 Materials and methods

4.1.1 Reagents and Materials

Tetracycline hydrochloride (USP grade, $\geq 99\%$) and oxytetracycline hydrochloride (USP grade, $\geq 99\%$) were obtained from Sigma-Aldrich chemical (St. Louis MO). Reagent grade phosphate monobasic monohydrate ($NaH_2PO_4.H_2O$) and sodium sulfate decahydrate ($Na_2SO_4.10H_2O$) were purchased from Fisher Scientific (Fair Lawn, NJ). Trace-metal grade hydrochloric acid, sodium hydroxide, potassium chloride, and oxalic acid, all certified ACS grade were purchased from Fisher Scientific (Fair Lawn, NJ). Methanol (HPLC grade, $\geq 99\%$) and acetonitrile (HPLC grade, $\geq 99\%$) were purchased from Fisher Scientific (Fair Lawn, NJ). PIPES (1,4-piperazinebis(ethane sulfonic acid) was purchased from Sigma–Aldrich (St. Louis, MO). All solutions and working standards were freshly prepared in high-purity water (18 MΩ-cm, Barnstead NANO-pure Diamond, Dubuque, IA).

Manure, soil, and Al-WTR collection

Fresh cattle manure samples were systematically collected from surface, adjacent corners, and bottom of the manure pit at Rutgers Cook Campus, NJ, USA. The fresh manure samples were thoroughly mixed into a single batch and refrigerated at 4°C until further use. Two types of soils were used in this study—the Immokalee Spodosols series and Belleglade Pahokee Muck series. The Immokalee series Spodosols soils were collected from surface horizons in the Southwest Florida Research and Education Center, Immokalee, Florida and the Belleglade Pahokee Muck series soils were collected from the surface horizons from Everglades Research and Education Center at Belle Glade, Florida. The Al-based WTR were obtained from the drinking-water treatment plant in Bradenton, FL, USA. Manure, WTR, and Soil samples were air-dried and then sieved with a 2-mm sieve before the Al-WTR being subjected to characterization and sorption experiments.

4.1.2 Manure and soil amendment and Al-WTR application

One hundred grams of each soil and manure were spiked with TTC and OTC to achieve target concentrations of 0, 2.25 and 22.5 mg/kg (mass of TTC or OTC /mass of dry soil or manure), which represent background TTC/OTC concentrations typically found at agricultural soils resulting from 1-5 years of continuous manure application and 10 times higher concentration, respectively (Kay et al., 2004; Hamscher et al., 2005). Further, as separate treatment conditions, both soils were amended with TTC/OTC rich manure at a rate of 11.2 Mg ha^{-1} to simulate a realistic field loading rate. All the treatments were kept in a bag under aerobic condition in darkness at room temperature

$(25 \pm 2°C)$. Al-based WTR was thoroughly mixed with the 100 g of manure, soils, and

manure amended soils, respectively at 25 and 50 g kg^{-1} rates. The moisture content was

adjusted to 70 % of water holding capacity and equilibrated for 7 days (7d) in aerobic

condition with sufficient amount of mixing every 24 h. After equilibration, manure, soils,

and manure amended soils were aerated bi-weekly and constant water content was

maintained. The application rates of Al-WTR were chosen based on practical application

rates of biosolids in agricultural fields (Elliott et al. 2002). No pH control was imposed.

However, pH of all the solutions was measured before and after the sorption experiments.

4.1.3 Manure, soil, and Al-WTR collection characterization

Manure, soils and Al-WTR were characterized for selected physicochemical

properties before the initiation of incubation study. The pH, electrical conductivity, water

content, and particle size (soils and Al-WTR) were measured using standard protocols

(Hanlon et al., 2002). Organic matter was measured using the loss-on-ignition method

(Ben-Dor and Banin, 1989; Klute 1996). Total C and N in the samples were determined

by combustion at 1100°C using an Elementar Vario EL CHNS/O analyzer (Elementar,

NJ USA). Concentrations of Oxalate-extractable Al and Fe of the soils and WTRs

samples were determined using Tamm's reagent (Klute 1996). Acid digestion was used

for total-recoverable Fe and Al concentrations following the USEPA 3050B method

(USEPA, 2000). Toxicity Characteristic Leaching Procedure (TCLP) using SW-846

Method 1311 was used for the WTRs to assess their potential for any waste leaching in a

landfill environment (EPA method 131). The concentrations of RCRA metals and

metalloids in the extract of the TCLP method were then assessed against regulatory levels to check whether WTRs exhibits the Toxicity Characteristic (TC). Phosphorus concentration was colorimetrically measured with a UV/vis-spectrophotometer, using the molybdate–ascorbic acid method (Watanabe and Olsen, 1965). Texture of the soils was determined by pipette method and Malvern particle size analyzer (Gee and Bauder, 1986).

4.1.4 Analytical analysis

All the elemental analyses were performed using Thermo X-series Inductively Coupled Plasma Mass Spectrometer (ICP-MS) (Thermo Electron). Detailed HPLC of TCs analysis has been discussed in details elsewhere (Punamiya et al., 2013). In brief, Finnigan surveyor plus HPLC system (Thermo Scientific) with surveyor PDA plus detector (photodiode array) and a surveyor plus auto-sampler were used for all analyses. A hypersil gold C_{18} column (150 × 4.6 mm, 5 μm) (Thermo Scientific) with a corresponding hypersil gold guard column (10 × 4 mm, 5 μm) (Thermo Scientific) at room temperature was used for all separations. Samples were eluted isocratically with a mobile phase consisting of 0.01 mol L^{-1} aqueous oxalic acid:acetonitrile:methanol (150:20:20 by volume) (Fritz and Zuo, 2007). Needed QA/QC procedures were followed to assure a recovery of 90-110% of spikes and standards.

4.1.5 Experimental Design

4.1.5.1 TTC/ OTC sorption kinetics

Kinetic experiments were conducted at selected time intervals (1, 2, 4, 7, 10, 14, 30, 60, and 90 d) to determine the effect of contact time on TTC and OTC sorption by Al-WTR-amended manure and soils equilibrated for 7 days. Samples were reacted with TTC/OTC solutions to attain initial TTCs loads of 0, 2.25 and 22.5 mg kg^{-1}. Three Al-WTR application rates were used 0, 25, and 50 g kg-^{1}. The concentration were selected based on the TTC/OTC concentrations typically found at agricultural soils resulting from 1-5 years of continuous manure application (Kay et al., 2004; Hamscher et al., 2005). One gram of sample was taken at the specified time period and extraction was performed using 20 mL 0.01 M KCl. The samples were shaken at 120 rpm on a reciprocal shaker, centrifuged at 4,000g for 15 min, filtered and analyzed for total soluble TTC/OTC Using HPLC. No pH control was imposed during kinetic experiments but the pH of the samples was measured before and after shaking. Optimum contact time obtained from this experiment was utilized for further sorption experiments.

4.1.5.2 Sorption kinetics in presence of competing agricultural species (phosphate and sulfate)

Sorption kinetics at different time intervals (1, 2, 4, 7, 10, 14, 30, 60, and 90 d) were conducted in presence of competing agricultural species (phosphate and sulfate) in manure, manure amended soils, and soils mixed with 50 g kg^{-1} Al-WTR. The initial concentration of TTC/OTC used were 2.25 and 22.5 mg kg^{-1}. Visual MINTEQ version 4.0.3 (chemical equilibrium model) calculations were performed to check highest concentration of phosphate and sulfate to be used in the experiment to avoid onset of

precipitation and/or formation of any mineral. The ratios of TTC/OTC to competing

species were 1:1, 1:5, and 1:10. Stock solutions of phosphate and sulfate were prepared in

0.01 M KCl using phosphate monobasic monohydrate ($NaH_2PO_4.H_2O$) and sodium

sulfate decahydrate ($Na_2SO_4.10H_2O$), respectively. TTC/OTC and the competing

agricultural species (phosphate and sulfate) were added simultaneously to soils to avoid

any pre-sorption on the Al-WTR.

4.1.6 Statistical Analysis

Statistical analysis was performed using JMP IN version pro 10 (Sall et al. 2005).

Two-way analysis of variance (ANOVA) was performed to examine the effect of contact

time, initial TTC and OTC concentration, and competing agricultural species (phosphate

and sulfate) on TTC and OTC sorption by Al-WTR amended manure, soils and manure

amended soils. Tukey–Kramer honest significant difference (HSD) test was used to

evaluate differences among treatment means. Treatment differences were deemed

significant at $P = \leq 0.05$ or 0.01. Adsorption data were fit to a linear and two non-linear

models namely Freundlich and Langmuir Isotherm models. Kinetics data were validated

using pseudo first and second order reaction rate models. All data were expressed as

mean ($n = 2$) along with standard deviation.

4.2 Results and Discussion

4.2.1 Manure, soil, and Al-WTR properties

Studies have shown that physico-chemical properties such as pH, CEC, clay content, (Kulshreshta et al., 2004; Sassman and Lee, 2005; Ter Laak et al., 2006; Bao et al., 2010), Al and Fe hydrous oxide (Figueroa et al., 2004; Figueroa and MacKay, 2005; Gu and Karthikeyan 2005), soil organic matter (Bao et al., 2009), humic materials (Gu et al., 2007; Pils and Laird, 2007;) presence of competing ligands and complexing metals (Jia et al ., 2009; Wang et al., 2010; Zhang et al., 2011; Zhao et al., 2012) etc. of manure, soil, and sorbent may highly influence the fate and transport of TCs in the environment. Selected physiochemical properties of soils (Immokalee series Spodosols and Belleglade -Pahokee Muck series), manure, and Al-WTR used in the incubation study are listed in Table 4-1. Immokalee soil is a sandy (99%) spodosol, acidic (pH 5.9) in nature with low Fe/Al, Ca/Mg, and P contents. Due to the sandy nature and lack of positive charged surfaces (e.g., amorphous Fe/Al oxides), the Immokalee soil is likely to have minimal retention capacity (Datta and Sarkar, 2005; Figueroa and MacKay, 2005; Gu and Karthikeyan 2005), thereby potentially releasing high content of TCs. The Belleglade soil was slightly alkaline (pH 7.85) with high SOM (85%) and much higher concentrations of Fe, Al, Ca, and Mg compared to Immokalee soil indicating higher TCs retention potential (Datta and Sarkar, 2005; Gu and Karthikeyan, 2005; Figueroa and MacKay, 2005;).The soils also varied widely in their salinity (measured as electrical conductivity [EC]) and the CEC, both likely to affect TCs sorption (Datta and Sarkar, 2005; Sassman and Lee,

Table 4-1: Selected general physico-chemical properties of Immokalee and Belleglade soils, cattle manure, and Al-WTR used in the greenhouse study. Data are expressed as mean of three replicates ± one standard.

	Al-WTR	Immokalee	Belleglade	Manure
pH	5.1 ± 0.34	5.9 ± 0.42	7.85 ± 0.12	6.2 ± 0.1
EC[a] (µs/cm)	363 ± 12.3	59.5 ± 4.5	503 ± 13	240 ± 5.5
CEC[b] (C mol/kg)	Nd	774 ± 28[§]	18,908 ± 1204[§]	Nd
OM[c] (g/kg)	240 ± 8.78	8.40 ± 0.2[§]	80.0 ± 1.5	250.5± 2.5
Sand (%)	65 ± 6	99 ± 0.02	84.2 ± 1.5	Nd
Clay (%)	15 ± 3	0.57 ± 0.01	4.58 ± 2.2	Nd
Silt (%)	13 ± 3	0.35 ± 0.05	7.02 ± 1.4	Nd
$(Al+Fe)_{Total}$ (g/kg)	122.2 ± 8.5	0.08 ± 0.001	5.42 ± 0.46	<MDL
$(Al+Fe)_{ox}$ (g/kg)	92.3 ± 4.5	0.02 ± 0.001	1.20 ± 0.005	<MDL
Total P g kg^{-1}	2.5 ± 0.3	0.23 ± 0.006	6.8 ± 0.058	4.5 ± 2.1
Total Ca + Mg (g kg^{-1})	12 ± 4.2	1.178 ± 0.1[§]	40.8 ± 2.3[§]	0.13 ± 0.02
TCs (mM)	<MDL†	<MDL	<MDL	<MDL

Nd; Not determined
[a] EC = Electrical conductivity
[b] CEC = Cation exchange capacity
[c] OM = Organic matter
†= Below Method detection limit
[§] Datta and Sarkar, 2005

2005; Ter Laak et al., 2006; Bao et al., 2010). General physicochemical properties of the Al-WTR are given in Table 4-1 and have been discussed in details elsewhere (Punamiya et al., 2013). In brief, Al-WTR was acidic (6.1) and amorphous (about 75% of total Al) in nature. The total C values (185 g kg^{-1}) for Al-WTR are in the range of organic C found in various WTRs nationwide (Ippolito et al., 2011). The toxicity characteristics leaching procedure (TCLP) values of Al-WTR were well below USEPA threshold limits (compared to values of biosolids as no limits are set for WTRs, Table 2-3). Total carbon and nitrogen content for Al-WTR was higher than soils (Table 4-1). The manure was also acidic (pH 6.2) in nature with highest EC (240 ± 5.5 μs/cm) and organic matter (250 ± 2.5) compared to soils and Al-WTR. No detectable background levels (method detection limit 1×10^{-3} mmol L^{-1}) of TTC and OTC were found in the soils, manure, and Al-WTR used in the incubation study.

4.2.2 Sorption kinetics in absence of competing agricultural species (phosphate and sulfate)

Adsorption kinetics studies were performed to evaluate the adsorption affinity of Al-WTR applied at three different rates (0, 25, and 50 g kg^{-1}) with varying initial TTC and OTC concentrations (0, 2.25, and 22.5 mg kg^{-1}) for manure, soils, and manure amended soils at room temperature (25 ± 2 °C) under aerobic conditions (Figure 4-1 to 4-6). The kinetic effect was observed on TTC and OTC sorption by Al-WTR-amended manure, soils, and manure amended soils as a function of contact time. A kinetically driven TTC and OTC sorption was observed for Al-WTR amended manure (Figure 4-1

and 4-2), soils (Figure 4-3 to 4-6), and manure amended soils (Figure 4-3 to 4-6).

Kinetics data for Al-WTR amended manure, soils, and manure amended soil (Table 4-2)

best fits the pseudo second order reaction rate model ($R^2 \leq 0.98$). The adsorption kinetics

was biphasic, showing an initially rapid, followed by a slow, sorption rate for all the

treatments and experimental conditions tested in the study. The kinetics behavior

observed is similar with our previous kinetic study on TTC and OTC sorption by Al-

WTR in absence of soil and manure (Punamiya et al., 2013). The reason for the better fit

of pseudo second order could be the difference in half time with initial TTC and OTC

loads or heterogeneity (macropores and micropores) of the surface sites of Al-WTR

(Makris et al., 2004) which may result in rapid-stage TTC and OTC sorption followed by

the relatively slow phase. Also, Kulshrestha et al. (2004), Bao et al. (2009), and Wang

and Yates (2008) observed similar biphasic (two distinct stages with very rapid phase and

relatively slow phase) TTC and OTC adsorption by clays, soils (Alfisols and Ultisols),

manure, and manure amended soil (sandy loam), respectively. Reaction rate kinetics of

25 and 50 g kg^{-1} Al-WTR application rates on manure, soils, and manure amended soils

showed significant difference ($p < 0.001$) when compared to control (no Al-WTR

application) (Table 4-2). However, there was no significant effect ($p > 0.05$) of initial

TTC and OTC concentration on the kinetic rate reaction at different Al-WTR application

rates.). But with the increase in initial TTC/OTC load, there was a decrease in TTC/OTC

sorption/immobilization (%) contributed by limited potential binding sites. Also, there

was no significant ($p > 0.1$) difference between sorption/immobilization of TTC and

OTC in manure, soils, and TTC/OTC rich manure amended soils mixed with Al-WTR at

all the concentrations and SSRs tested, showing species independent sorption behavior (because OTC and TTC have similar functional groups).

In Al-WTR applied manure the time to reach equilibrium was 7 d for both 25 and 50 g kg^{-1} Al-WTR application rates at initial concentration of 2.25 mg kg^{-1} TTC and OTC (Figure 4-1 and 4-2). In case of control (no AL-WTR application) the time to reach equilibrium was 21 d. However, significant difference ($p < 0.001$) was observed in sorption and immobilization of TTC and OTC between different Al-WTR application rates; 0 g kg^{-1} (28-30%) 25 g kg^{-1} (80-82%) and 50 g kg^{-1} (> 95%), respectively. The kinetics behavior of Al-WTR applied manure amended with 22.5 mg kg^{-1} was similar to that of 2.25 mg kg^{-1} TTC and OTC. But at higher initial concentration (22.5 mg kg^{-1}), as expected, the time to reach relative equilibrium was slightly higher, 0 g kg^{-1} (28 d with 26-28% sorption/immobilization) 25 g kg^{-1} (10 d with 78-80 % sorption/immobilization) and 50 g kg^{-1} (> 95%), respectively. However, there was no effect of initial TTC and OTC concentration observed on sorption/immobilization in Al-WTR amended manure.

In Al-WTR applied soils a different sorption kinetics behavior was observed compared to Al-WTR applied manure. The sorption kinetics for Immokalee and Belleglade was dependent on their physico-chemical properties. In Al-WTR applied Immokalee soil the time to reach equilibrium was 14 and 10 d for 25 and 50 g kg^{-1} Al-WTR application rates respectively, where 79-82% (25 g kg^{-1}) and 89-92% (50 g kg^{-1}) of initially added TTC and OTC were adsorbed/immobilized at initial concentration of 2.25 mg kg^{-1} TTC and OTC (Figure 4-3 and 4-4). The kinetics behavior of Al-WTR applied

Table 4-2: Pseudo- second order reaction rate constants in Al-WTR-amended manure, soils, and soil amended manure (50 g kg^{-1} application rate) at different initial TTC and OTC loads. Contact time ranged from 1 to 90 days.

Soil/Manure	TCs	1st order rate fit (R2)	2nd order rate fit (R2)
Immokalee	TTC (1.0mM)	0.84	0.98
	OTC (1.0mM)	0.83	0.99
Immokalee + Manure	TTC (1.0mM)	0.86	0.98
	OTC (1.0mM)	0.85	0.99
Belleglade	TTC (1.0mM)	0.83	0.98
	OTC (1.0mM)	0.85	0.98
Belleglade + Manure	TTC (1.0mM)	0.87	0.99
	OTC (1.0mM)	0.82	0.99
Manure	TTC (1.0mM)	0.79	0.98
	OTC (1.0mM)	0.78	0.99

The data for 10mM TTC and OTC also best fits second order pseudo kinetics model ($R^2 = >0.98$)

manure amended with 22.5 concentration mg kg^{-1} was different from that of 2.25 mg kg^{-1} TTC and OTC. At higher initial (22.5 mg kg^{-1}), as the time to reach relative equilibrium was higher, 0 g kg^{-1} (60 d with 18-20% sorption/immobilization) 25 g kg^{-1} (21 d with 75-77 % sorption/immobilization) and 50 g kg^{-1} (> 90%), respectively. In case of control (no AL-WTR application) the time to reach equilibrium was 60 d. The observed kinetics behavior in Immokalee soil can be explained based on the physico-chemical properties. As discussed above, Immokalee soil is a sandy in nature with low content of Fe/Al, Ca/Mg, and P (Datta and Sarkar, 2005). Further, due to sandy nature and lacking positively charged surfaces (e.g., low content of amorphous Fe/Al oxides) (Datta and Sarkar, 2005), the Immokalee soil is likely to have low retaining capacity thereby possibly releasing high amount of TCs spiked and greater time to reach equilibrium(Datta and Sarkar, 2005). Significant difference ($p < 0.001$) was observed between Al-WTR applied Immokalee soil and control (no Al-WTR application) to immobilize and stabilize TTC and OTC. There was some effect, but not significant ($p > 0.15$), of initial TTC and OTC concentration observed on sorption/immobilization in Immokalee soils.

The kinetics sorption behavior of Belleglade soil amended with Al-WTR was different compared to Al-WTR applied Immokalee soil and manure (Figure 4-5 and 4-6). In Al-WTR applied Belleglade soil, the time to reach equilibrium was 14 d for both 25 and 50 g kg^{-1} Al-WTR application rates, where 82-88% (25 g kg^{-1}) and >92% (50 g kg^{-1}) of initially added TTC and OTC were adsorbed/immobilized at initial concentration of 2.25 mg kg^{-1} TTC and OTC (Figure 4-5 and 4-6). The kinetics behavior of Al-WTR applied manure amended with 22.5 mg kg^{-1} was significantly different ($p < 0.05$) from

that of 2.25 mg kg^{-1} TTC and OTC. At higher initial concentration (22.5 mg kg^{-1}), the time to reach relative equilibrium was higher, 0 g kg^{-1} (21 d with 26-29% sorption/immobilization) 25 g kg^{-1} (14 d with 83-88 % sorption/immobilization) and 50 g kg^{-1} (> 90%), respectively. In case of control (no AL-WTR application) the time to reach equilibrium was 21 d. The observed kinetics behavior in Belleglade soil without Al-WTR application can be explained based on the physico-chemical properties. Belleglade soil has high concentrations of Fe, Al, Ca, and Mg compared to Immokalee soil indicating higher TCs retention potential (Gu and Karthikeyan, 2005; Figueroa and MacKay, 2005). Also, Belleglade soil has high soil organic matter compared to Immokalee soil resulting in higher adsorption (Wang and Yates, 2008). Significant difference ($p < 0.001$) was observed between Al-WTR applied Belleglade soil and control (no Al-WTR application) to immobilize and stabilize TTC and OTC at all the concentration tested.

A diverse kinetics sorption behavior was observed in manure-amended soils (Figure 4-2 and 4-3). The application rates of Al-WTR were chosen based on practical application rates of biosolids in agricultural fields (Elliott et al. 2002), manure at a rate of 11.2 Mg ha^{-1}. In Al-WTR applied manure amended Immokalee and Belleglade soils the time to reach equilibrium was 14 (82-85% sorption) and 10 d (90-92% sorption) and 10 (87-89% sorption) and 7d (92-95%) for 25 and 50 g kg^{-1} Al-WTR application rates, respectively, at initial concentration of 2.25 mg kg^{-1} TTC and OTC (Figure 4-2 to 4-6). The kinetics behavior of Al-WTR applied manure amended soils with 22.5 mg kg^{-1} was different from that of 2.25 mg kg^{-1} TTC and OTC. However, no significant effect ($p >$
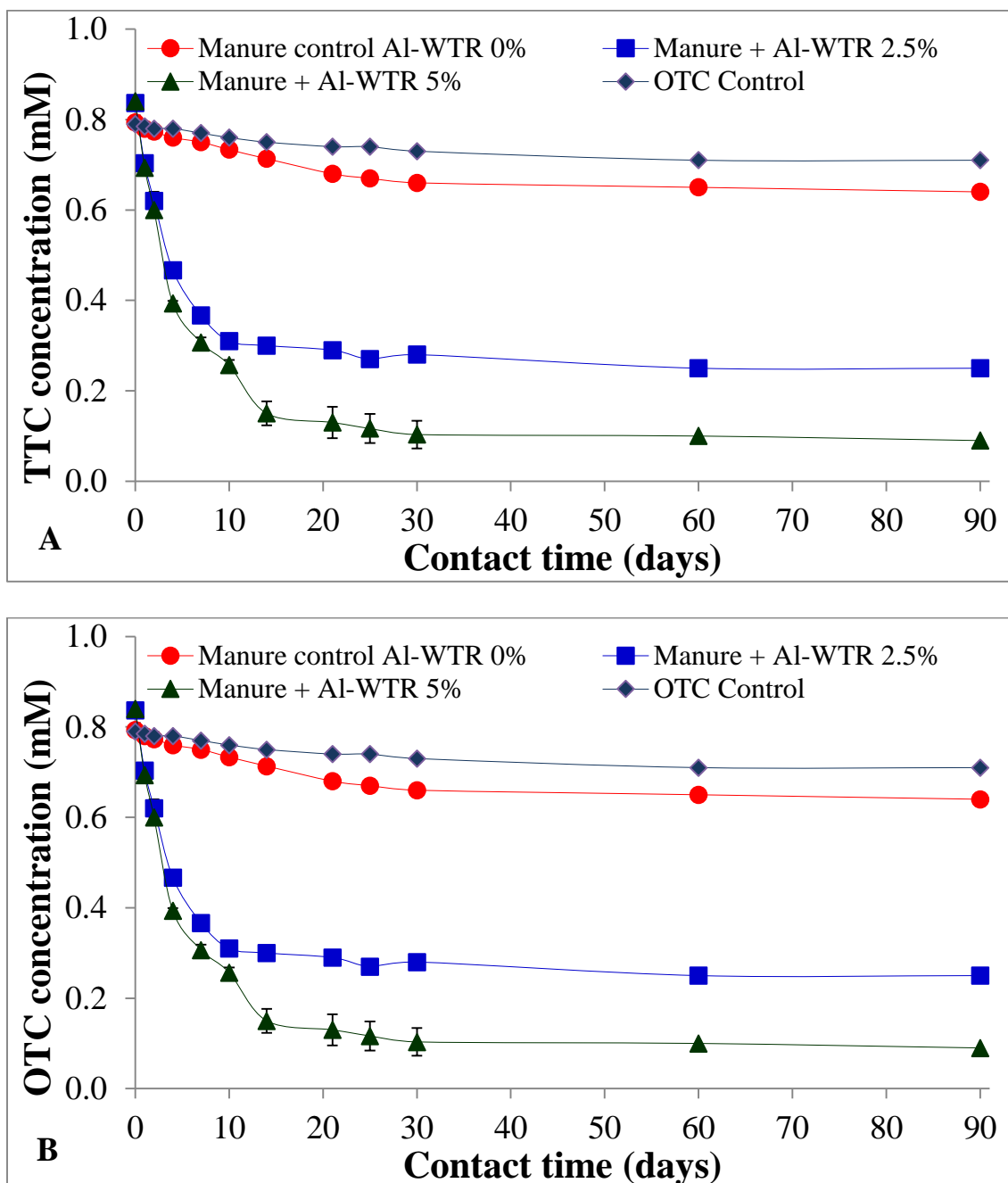
Figure 4-1: TTC (A) and OTC (B) sorption by Al-WTR amended manure at 25 and 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads (2.25 mg kg^{-1}). Data are expressed as mean of three replicates ± one standard deviation.
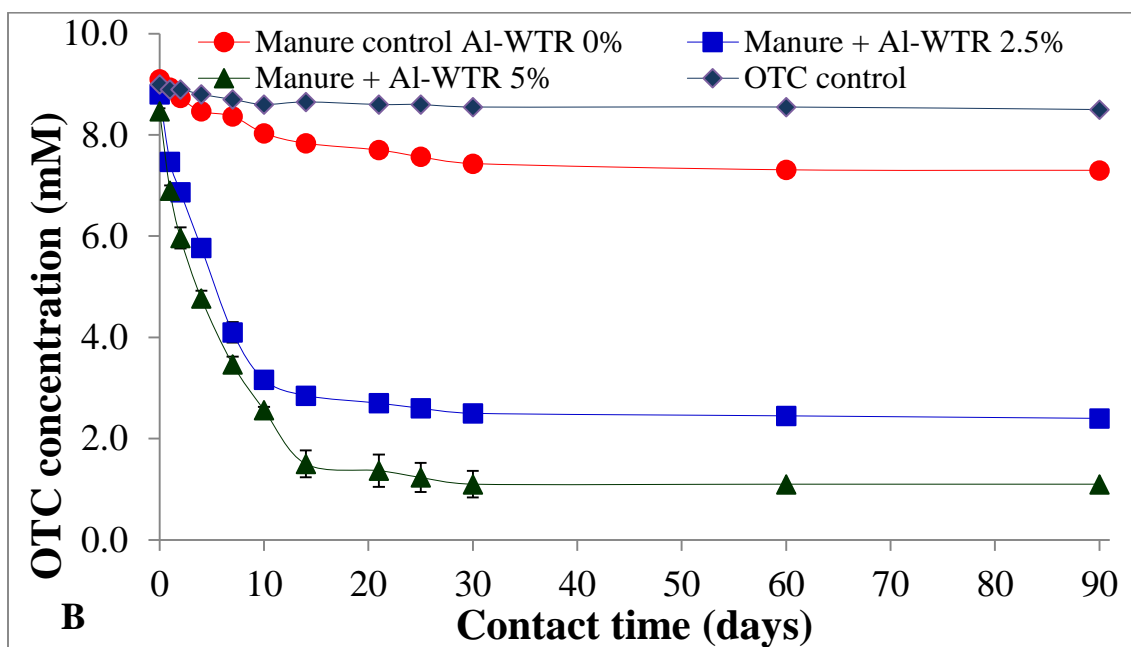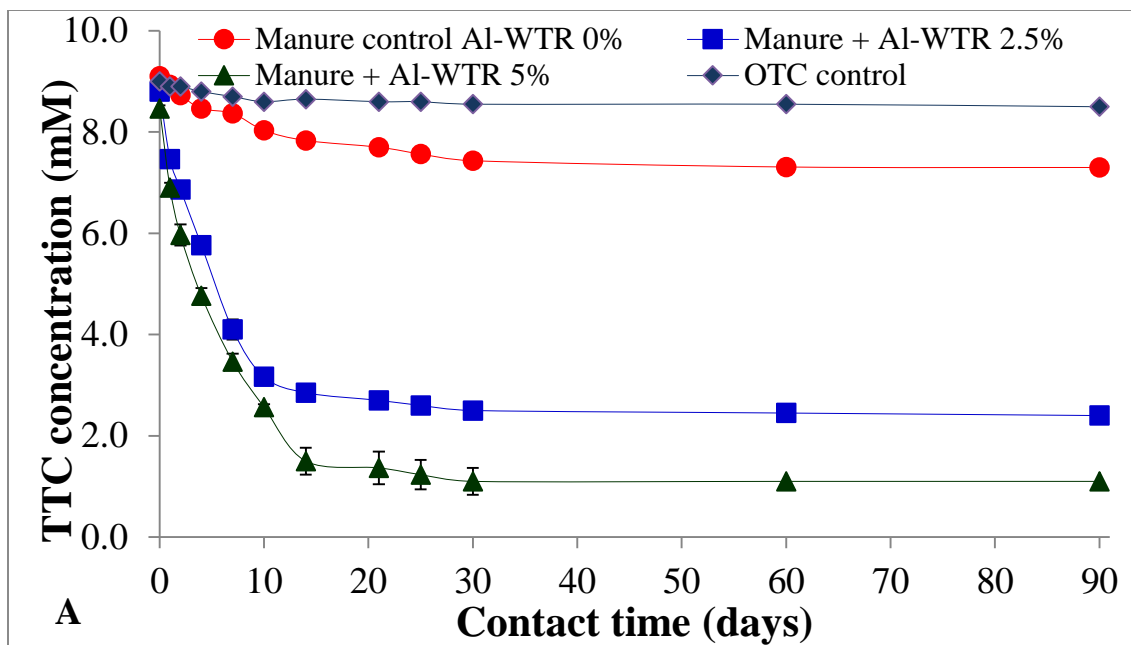
Figure 4-2: TTC (A) and OTC (B) sorption by Al-WTR amended manure at 25 and 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads (22.5 mg kg^{-1}). Data are expressed as mean of three replicates ± one standard deviation.
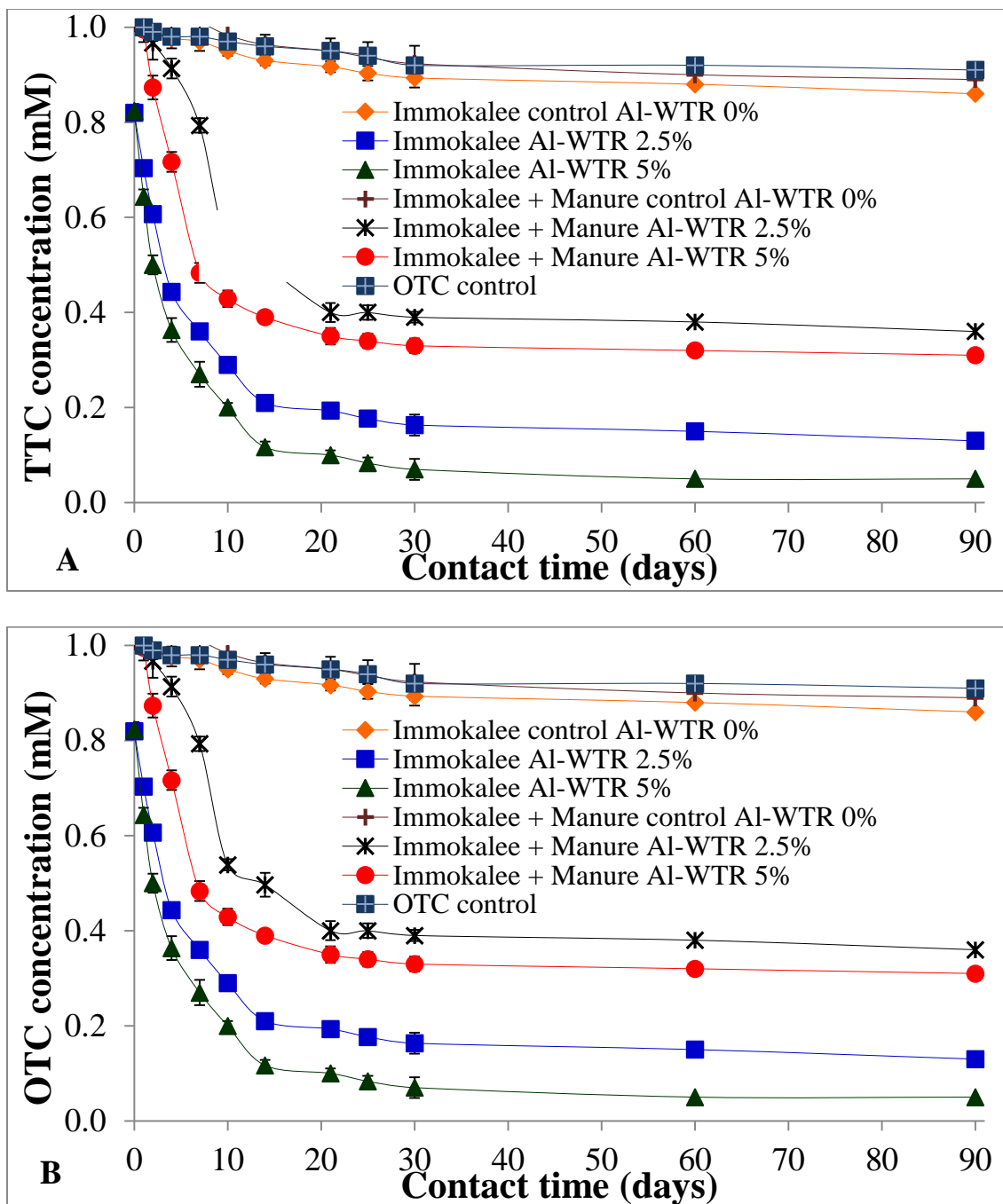
Figure 4-3: TTC (A) and OTC (B) sorption by Al-WTR amended Immokalee soil and manure amended Immokalee soils at 25 and 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads (2.25 mg kg^{-1}). Data are expressed as mean of three replicates ± one standard deviation.
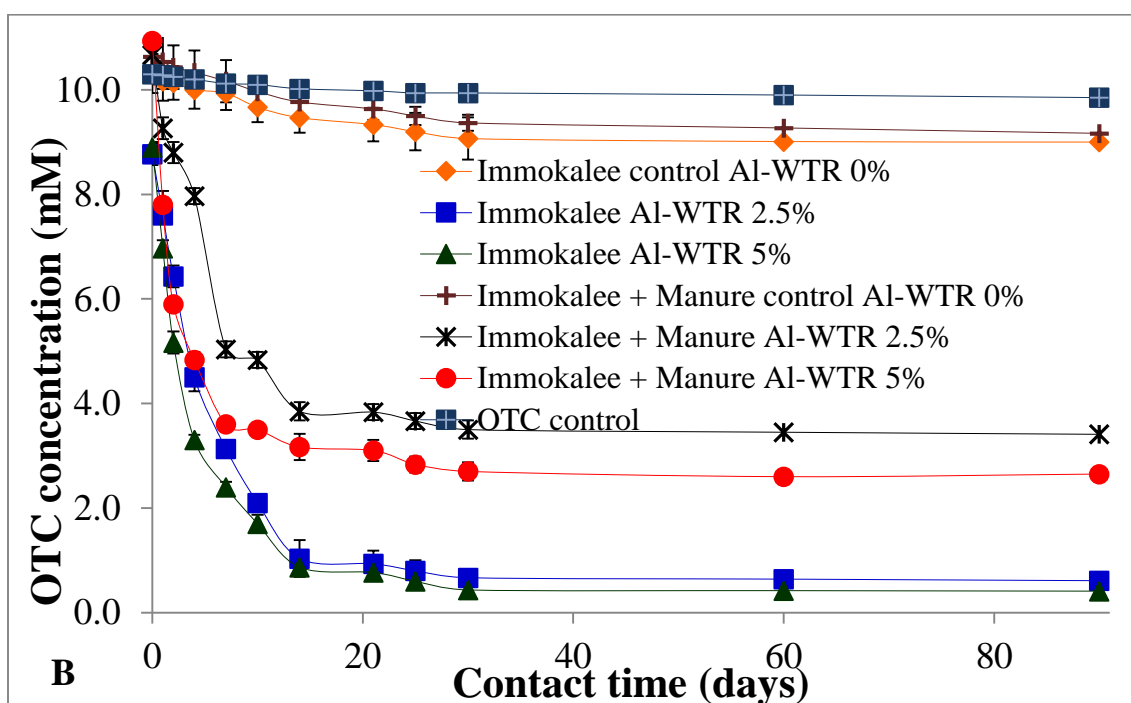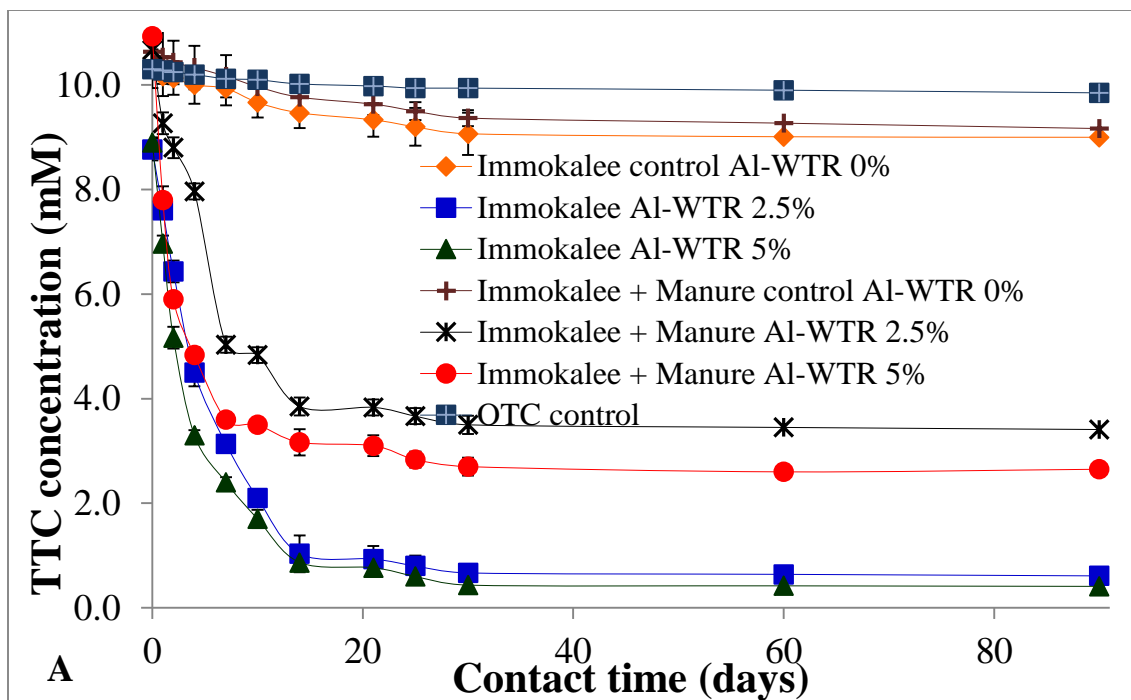
Figure 4-4: TTC (A) and OTC (B) sorption by Al-WTR amended Immokalee soil and manure amended Immokalee soils at 25 and 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads (22.5 mg kg^{-1}). Data are expressed as mean of three replicates ± one standard deviation.
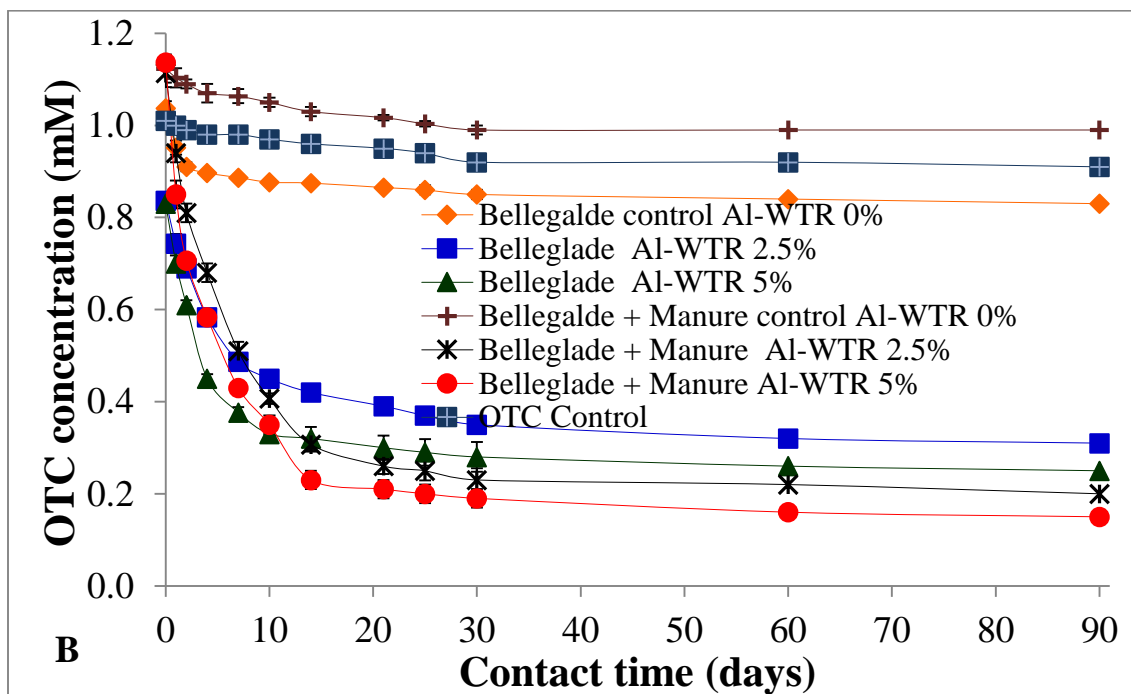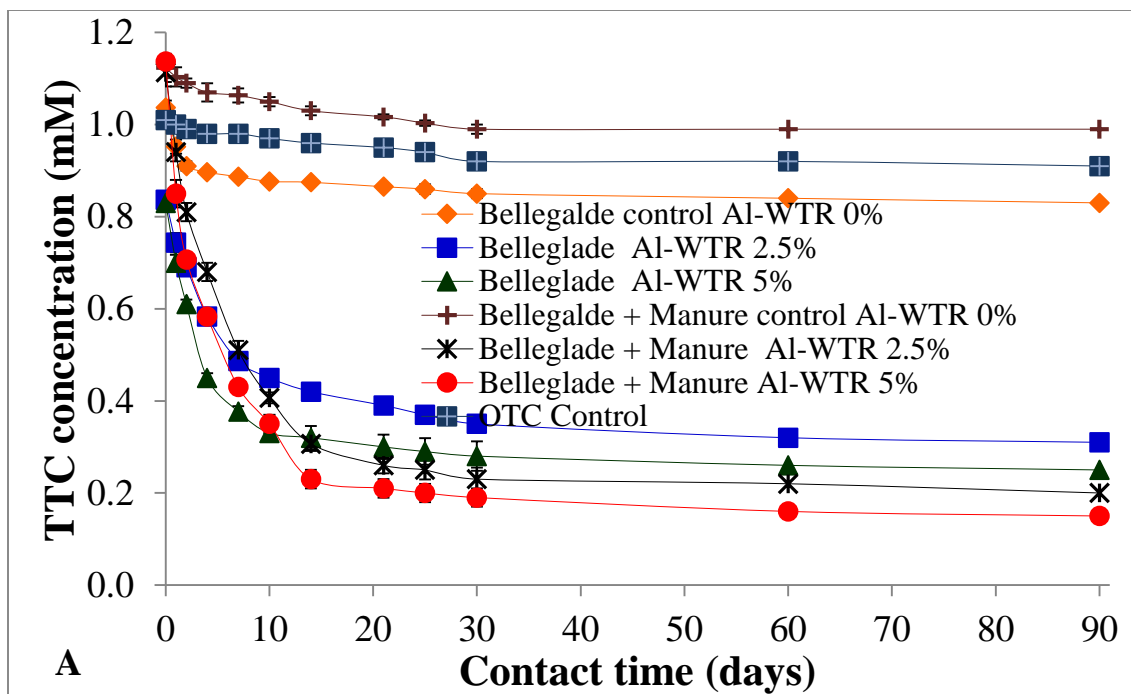
Figure 4-5: TTC (A) and OTC (B) sorption by Al-WTR amended Belleglade soil and manure amended Belleglade soils at 25 and 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads (2.25 mg kg^{-1}). Data are expressed as mean of three replicates ± one standard deviation.
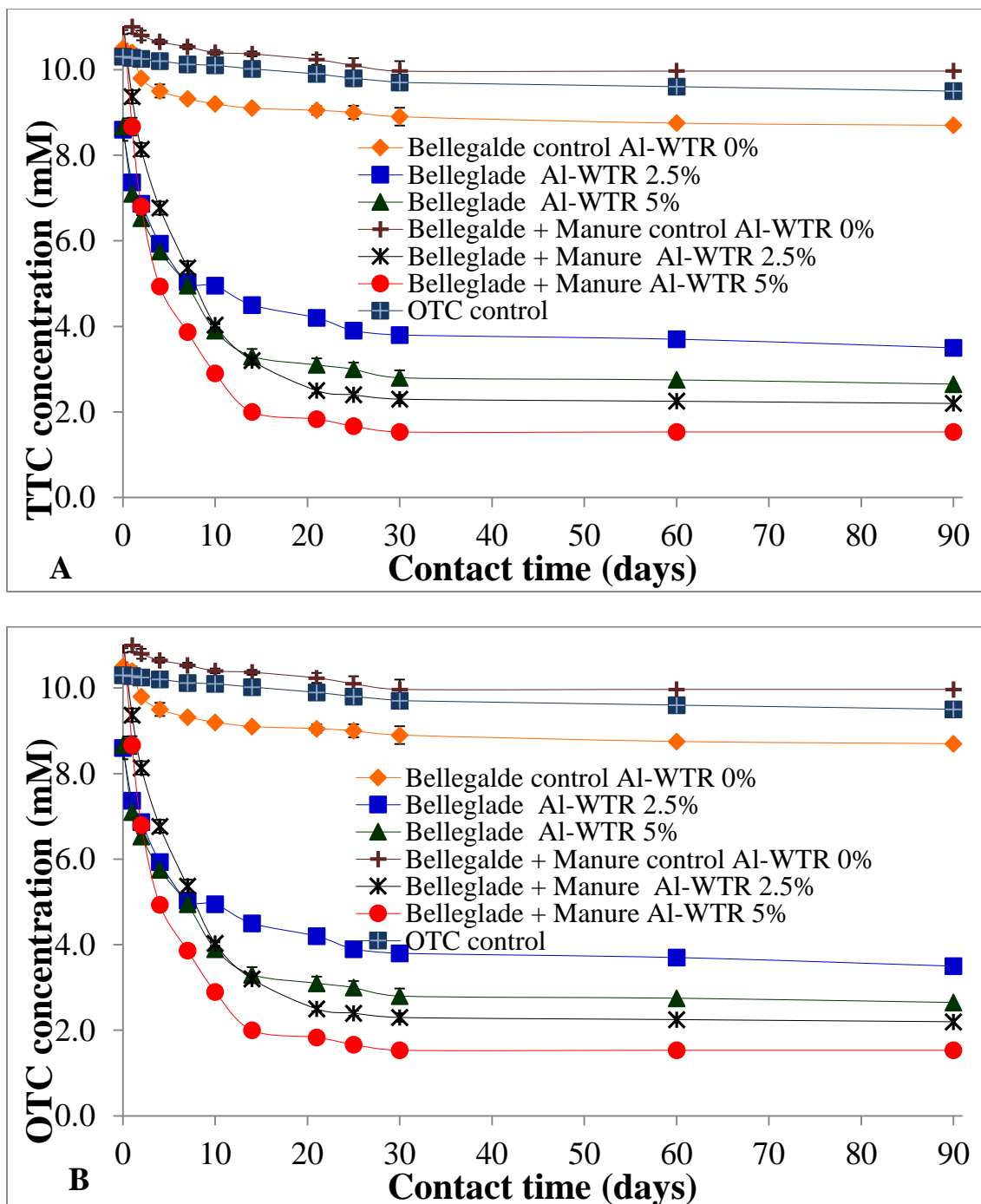
Figure 4-6: TTC (A) and OTC(B) sorption by Al-WTR amended Belleglade soil and manure amended Belleglade soils at 25 and 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads (22.5 mg kg^{-1}). Data are expressed as mean of three replicates ± one standard deviation.

0.12) of initial concentration on sorption was seen. At higher initial concentration of

TTC/OTC, the time to reach relative equilibrium was higher in some instances and

similar in other cases. In case of Immokalee soil amended with TTC/OTC rich manure

and amended with Al-WTR 0, 25, and 50 g kg^{-1} the time to reach equilibrium was 30, 14,

and 10 d with 20-22 %, 80-82 %, and 90-93% sorption/immobilization, respectively;

whereas, for Belleglade Immokalee soil amended with TTC/OTC rich manure and

amended with Al-WTR 0, 25, and 50 g kg^{-1} the time to reach equilibrium was 21, 14, and

7d with 23-25 %, 89-92 %, and 93-95% sorption/immobilization, respectively. The

equilibration time for manure amended soils was lower than non-amended soils. The

observed kinetics rate was faster than non-amended soils but slower that manure applied

Al-WTR and spiked TTC and OTC. Wang and Yates (2008) observed similar behavior

for OTC in non-amended and manure amended soil (Sandy loam). Further, the observed

behavior can be explained by increase in the organic matter content of the soil by manure

application and possible change in the pH and CEC of the soils (Wang and Yates, 2008;

Ter Laak et al., 2006).

4.2.3 Sorption kinetics in presence of competing agricultural species (phosphate and sulfate)

Adsorption kinetics studies were performed to evaluate the adsorption affinity of

Al-WTR applied at two different rates (0 and 50 g kg^{-1}) with fixed initial TTC and OTC

concentrations (2.5 mg kg^{-1}) for soils at room temperature (25 ± 2 °C) under aerobic

conditions in presence of competing agricultural species; phosphate and sulfate at three
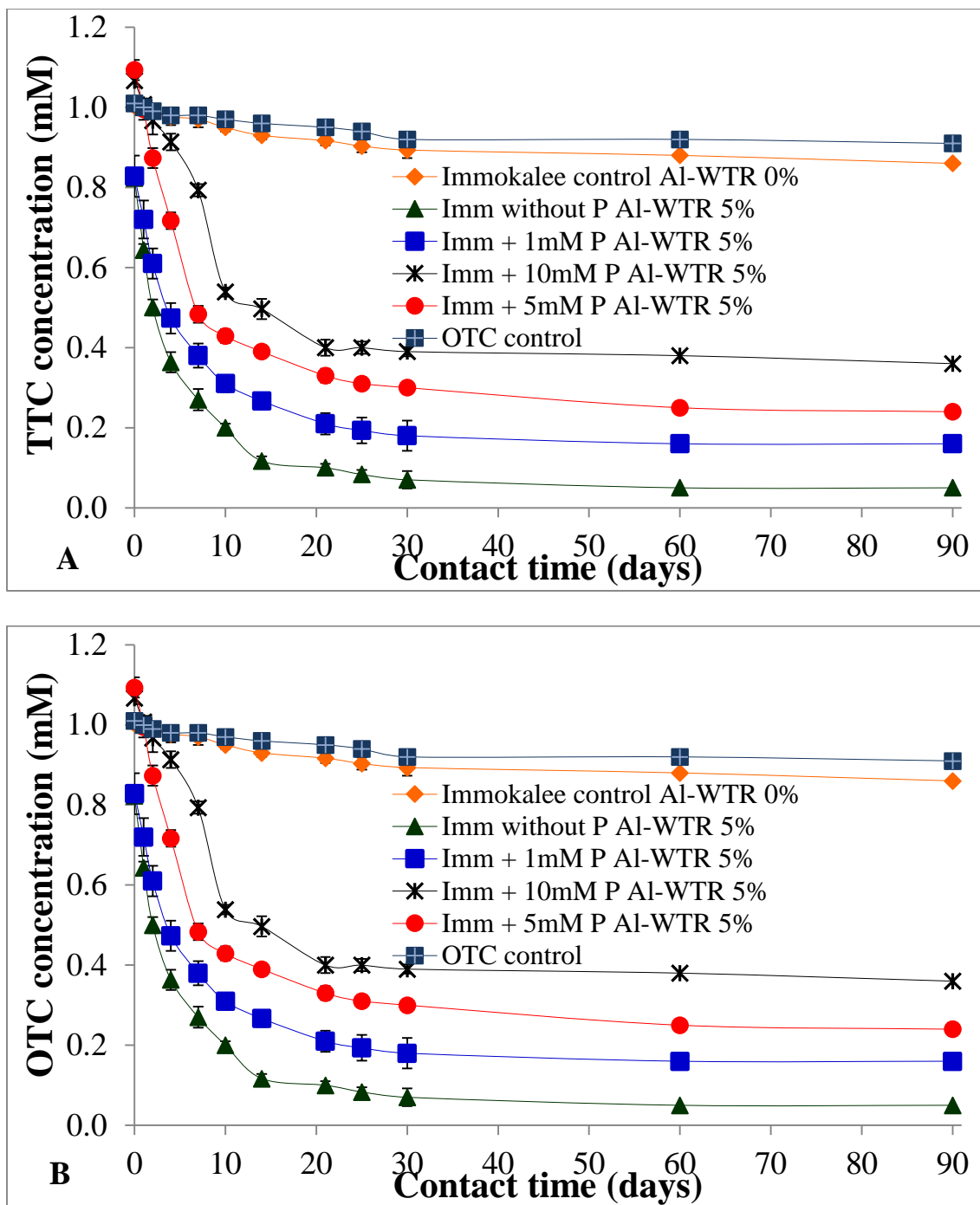
Figure 4-7: Effect of competing agricultural species (phosphate) on TTC (A) and OTC (B) sorption by Al-WTR amended Immokalee soil and manure amended Immokalee soils at 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads. TTC/OTC:P molar ratios were- 1:1, 1: and 1:10. Control was without phosphate. Data are expressed as mean of three replicates ± one standard deviation.
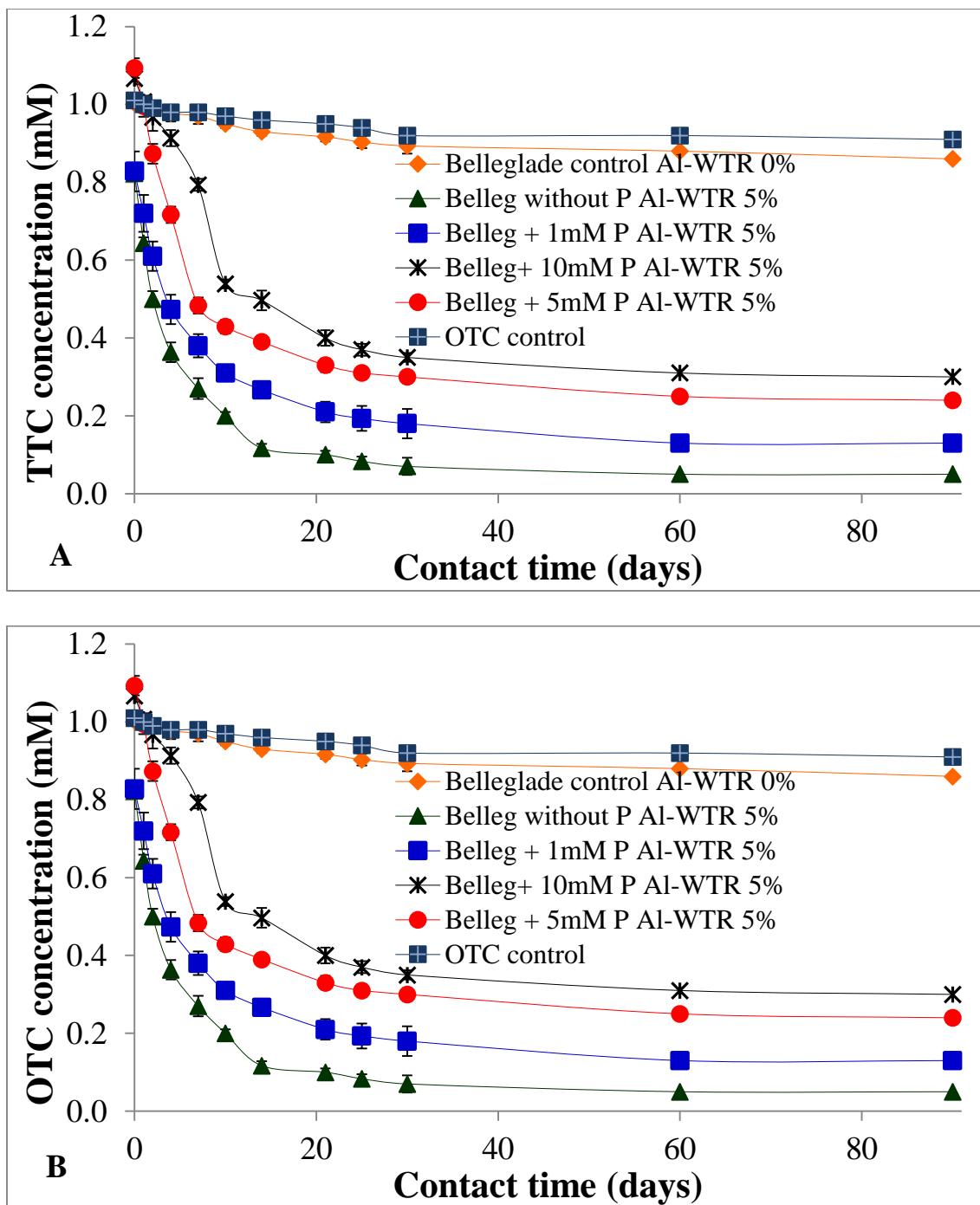
Figure 4-8: Effect of competing agricultural species (phosphate) on TTC (A) and OTC (B) sorption by Al-WTR amended Belleglade soil and manure amended Belleglade soils at 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads. TTC/OTC:P molar ratios were- 1:1, 1: and 1:10. Control was without phosphate. Data are expressed as mean of three replicates ± one standard deviation

different ratios (Figure 4-7 to 4-10). Competitive sorption study for TTC and OTC and P(V) added simultaneously to soils was conducted at pH values between 5.9-7.1 at 50 g L^{-1} Al-WTR application rate to study kinetics sorption in the presence of competing agricultural species. Different initial molar ratios of TTC/OTC to competing agricultural species (1:1, 1:5, and 1:10) were chosen, and the sorption was measured during different time intervals. The total concentration was chosen to provide near maximum surface coverage, according to previous specific surface area measurements for Al-WTR (Makris et al., 2004). Visual MINTEQ version 4.0.3 (chemical equilibrium model) calculations showed no secondary precipitation and/or formation of any mineral at the highest P(V) concentration used (data not shown) under the experimental conditions used in the study. The presence of phosphate in Immokalee and Belleglade soils resulted in the significant ($p < 0.005$) decrease in TTC and OTC sorption/immobilization by Al-WTR at higher ratios of TTC/OTC:P (1:5 and 1:10) (Figure 4-7 and 4-8). However, no significant effect ($p > 0.2$) of phosphate was seen on the reaction rate. The relative equilibrium was reached in 21 and 14 d for Immokalee and Belleglade, respectively with for all the TTC/OTC:P ratios tested. However, the rate of sorption/immobilization varied significantly ($p < 0.005$) at the different TTC/OTC:P ratios tested. In case of Immokalee amended soil with Al-WTR simultaneously mixed with phosphate at TTC/OTC:P ratios 1:0, 1:1, 1:5, and 1:10; the rate of sorption/immobilization decreased from >90% to 81-83%, 75-77% , and 66-68%, whereas in Belleglade soil, it decreased from >90% to 85-86%, 75-78%, and 67-69%, respectively. The kinetics behavior observed is similar with our previous kinetic batch sorption study on TTC and OTC sorption by Al-WTR in
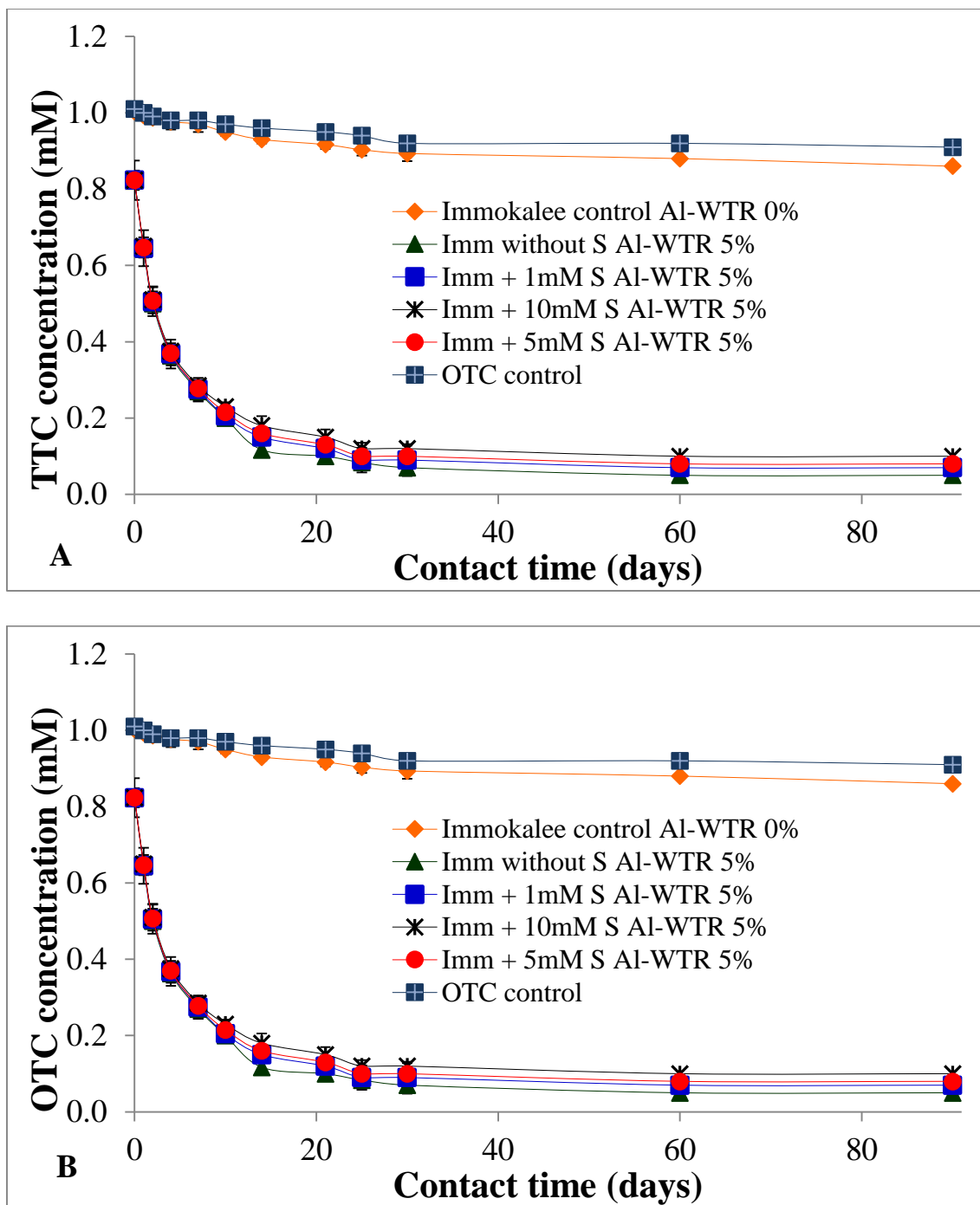
Figure 4-9: Effect of competing agricultural species (sulfate) on TTC (A) and OTC (B) sorption by Al-WTR amended Immokalee soil and manure amended Immokalee soils at 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads. TTC/OTC:Sulfate molar ratios were- 1:1, 1: and 1:10. Control was without sulfate. Data are expressed as mean of three replicates ± one standard deviation
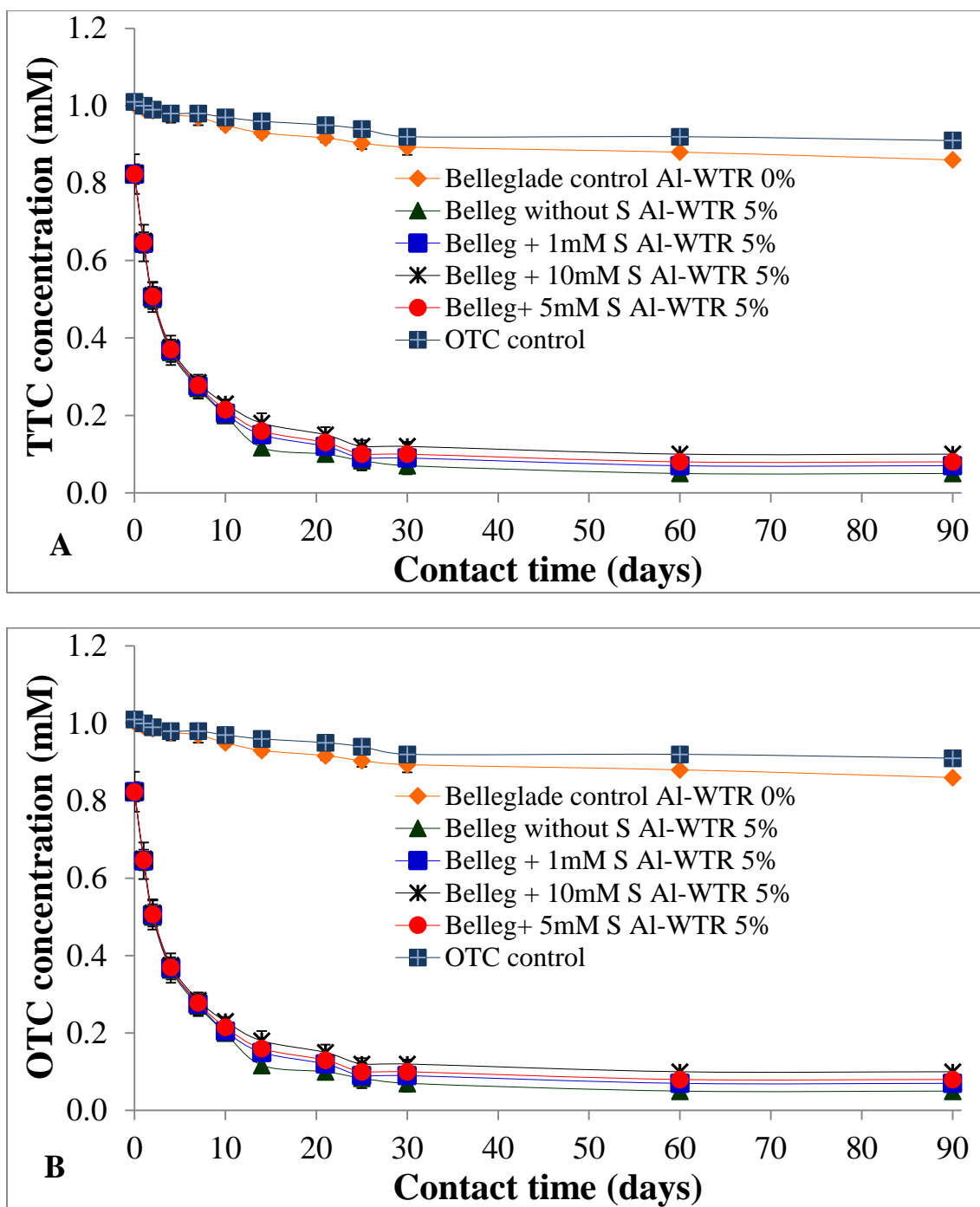
Figure 4-10: Effect of competing agricultural species (sulfate) on TTC (A) and OTC (B) sorption by Al-WTR amended Belleglade soil and manure amended Belleglade soils at 50 g kg^{-1} as a function of contact time and initial TTC and OTC loads. TTC/OTC:Sulfate molar ratios were- 1:1, 1: and 1:10. Control was without sulfate. Data are expressed as mean of three replicates ± one standard deviation

presence of phosphate at solid-water interface (Punamiya et al., 2013b). Similarly, studies

have shown a decrease in the sorption (pH-dependent) of antibiotics in the presence of

various competing ligands on clays, sediments, soils, oxides, and various sorbents (Zhang

et al., 2011; Ji et al., 2009; Kang et al., 2010; Wang et al., 2008;) under circum-neutral

pH. Reports from Wang et al. (2008b), Ji et al. (2009), Kang et al. (2010), and Zhang et

al. (2011) showed that the presence of competing anion can increase as well as decrease

the adsorption of tetracycline on montmorillonite, carbon nanotubes and chitosan,

respectively by competing for sorption sites or by surface charge reversal.

A competitive sorption study for TTC and OTC and sulfate added simultaneously

to soils was conducted at pH range of 5.9 – 6.8 at 50 g L^{-1} Al-WTR application rate to

study sorption kinetics in the presence of sulfate. Different initial molar ratios of

TTC/OTC to

sulfate (1:1, 1:5, and 1:10) were chosen, and the sorption kinetics were measured during

different time intervals. Presence of sulfate had no significant (*p > 0.1*) effect on TTC

and OTC sorption on the Al-WTR amended soils (Figure 4-9 and 4-10). The relative

equilibrium was reached in 14 and 10 d for Immokalee and Belleglade, respectively with

all the TTC/OTC: sulfate ratios tested. In case of Immokalee amended soil with 50 g L^{-1}

Al-WTR simultaneously mixed with sulfate at TTC/OTC: sulfate ratios 1:0, 1:1, 1:5, and

1:10; the rate of sorption/immobilization were >90% to 88-89%, 86-88% , and 85-86%,

whereas in Belleglade soil it decreased from >92% to 89-90%, 87-89%, and 87-88%,

respectively. The kinetics behavior observed is similar with our previous kinetic batch

sorption study on TTC and OTC sorption by Al-WTR in presence of sulfate in absence of

soils (Punamiya et al., 2013b). Bui and Choi (2010) studied influence of anion (sulfate) on the adsorption of pharmaceuticals to porous silica and reported similar behavior of sorption. The presence of sulfate did not cause any effect on the adsorption of pharmaceuticals to porous silica. However, the impact was dependent on various other parameters (ionic strength, natural organic matter, cations, etc.).

4.2.4 FTIR analysis

The ATR-FTIR spectra of crystalline TTC, untreated: Al-WTR, Immokalee, Belleglade, and manure, TTC treated: Immokalee, Belleglade, manure, manure applied Immokalee and Belleglade soils amended with Al-WTR are shown in figure 4-11A, B, C and spectra of crystalline OTC, untreated: Al-WTR, Immokalee, Belleglade, and manure, OTC

treated: Immokalee, Belleglade, manure, manure applied Immokalee and Belleglade soils amended with Al-WTR are shown in figures 4-11 D, E, F. Characteristic IR bands associated with the functional groups of crystalline OTC/TTC molecule resulted peaks near 1678 cm^{-1} for amide I (the C=O group of the –CONH$_2$ moiety), 1618 & 1584 cm^{-1} for amide II (two N-H bonds of –CONH$_2$), 1518 & 1447 cm^{-1} for skeletal C=C vibrations (aromatic ring), 1280, 1247, and 1228 cm^{-1} for C-N bonds, and 1177 and 1117 for phenolic –OH (Rakshit et al., 2013b). Any IR band shift in the OTC/ TTC treated spectra compared to that of crystalline TTC and OTC would indicate potential interactions between OTC / TTC with soils, manure, and manure applied soils amended with Al-WTR (Kang et al., 2011; Zhao et al., 2012, Rakshit et al., 2013b) (Fig.4-11A- F).

For Immokalee, characteristic IR bands were discernible in the OTC / TTC treated Immokalee + Al-WTR experiments (Figure 4-11 A and D). Low wave number shifts in the OTC / TTC treated Immokalee + Al-WTR spectra compared to that of crystalline OTC/ TTC were observed for the peaks at 1684/1678, 1618, 1584, 1518, 1247, 1228, and 1176 cm^{-1}. These shifts pertain to strong inner-sphere type interaction of OTC/ TTC molecule with the Immokalee + Al-WTR matrix via $-CONH_2$ and phenolic $-OH$ groups (Punamiya et al., 2013; Rakshit et al., 2013b). The IR bands of OTC/ TTC treated manure +Immokalee + Al-WTR experiment indicated very noisy spectra probably due to the presence of manure (Figure 4-11 A and D). However, some peaks are identifiable. The IR bands of crystalline OTC/TTC spectra at 1248, 1227, and 1176 cm^{-1} were shifted to lower wave numbers in the OTC/ TTC treated manure +Immokalee + Al-WTR experiment, indicating strong inner-sphere type bonding via the $-CONH_2$ and phenolic-OH groups.

For Belleglade, the IR bands in the OTC/TTC treated Belleglade + Al-WTR indicated a downward shift near 1281, 1248, and 1176 cm^{-1} (Figure 4-11 B and E). The IR band shifts in these regions revealed that OTC/TTC bound with Belleglade + Al-WTR matrix via $-CONH_2$ & phenolic-OH groups. Similar observation could be made for the manure + Belleglade + Al-WTR experiment (Figure 4-11 B and E).

The IR bands of OTC/ TTC treated manure amended with Al-WTR experiment indicated noisy spectra (Figure 4-11 C and F) probably due to the presence of manure as seen for manure amended Immokalee and Belleglade soils applied with Al-WTR (Figure
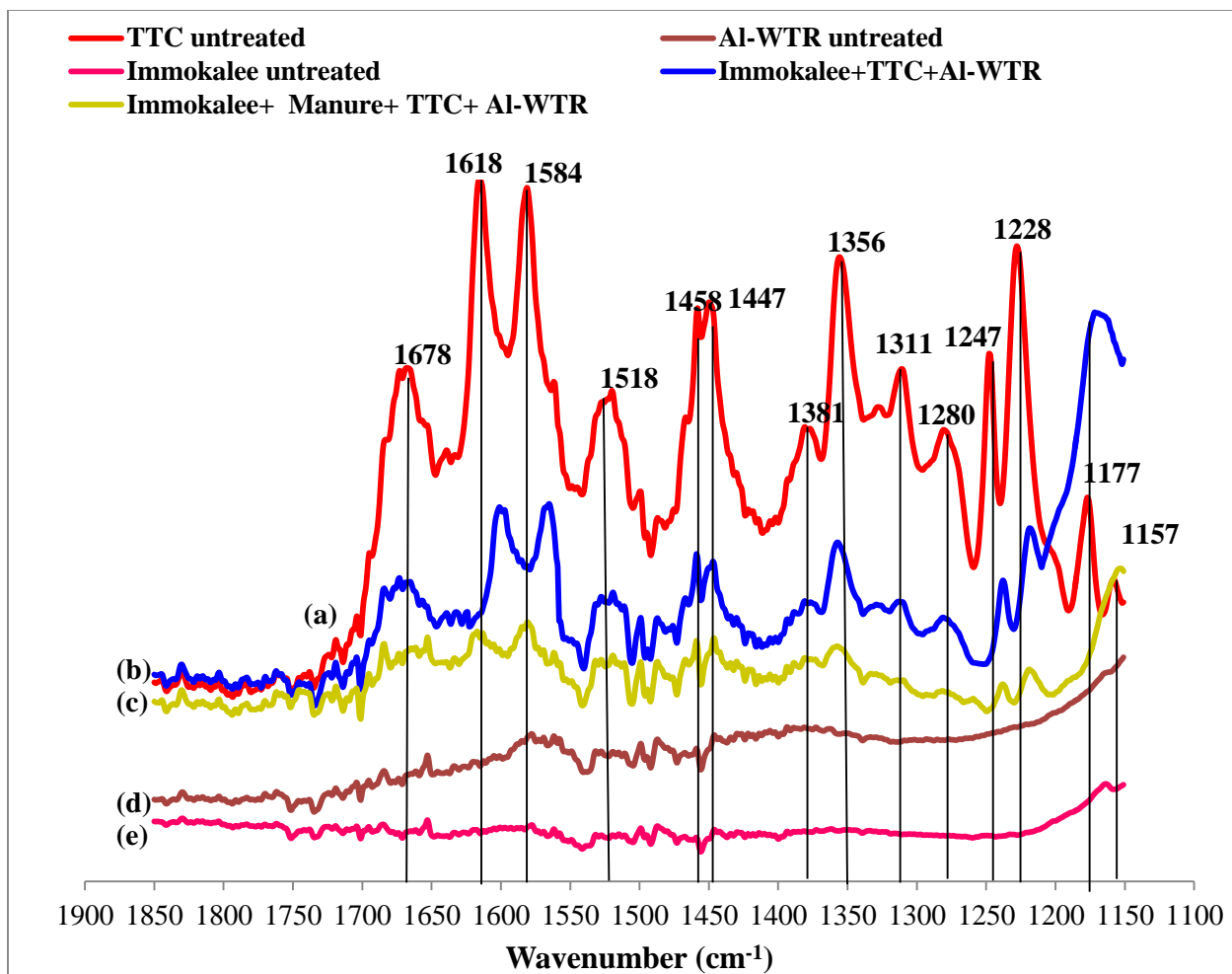
Figure 4-11A: ATR-FTIR spectra: (a) crystalline untreated TTC, (b)Immokalee treated with TTC amended with Al-WTR, (c)Immokalee treated with TTC applied with manure and amended with Al-WTR, (d)Al-WTR untreated, and (e)Immokalee untreated. All spectra are normalized with the highest peak observed.
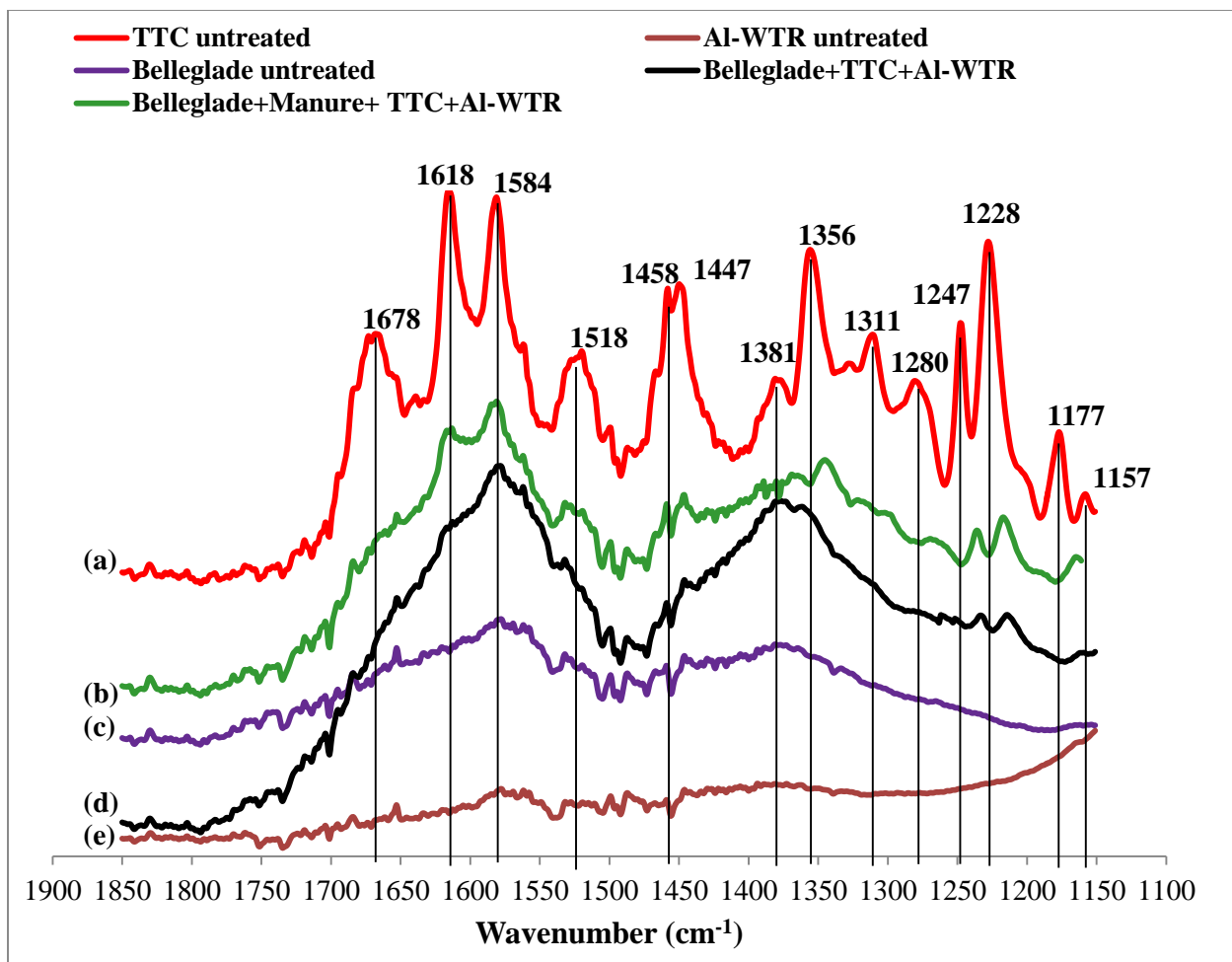
Figure 4-11B: ATR-FTIR spectra: (a) crystalline untreated TTC, (b) Belleglade treated with TTC applied with manure and amended with Al-WTR, (c) Belleglade untreated, (d) Belleglade treated with TTC amended with Al-WTR, and (e) Al-WTR untreated. All spectra are normalized with the highest peak observed.
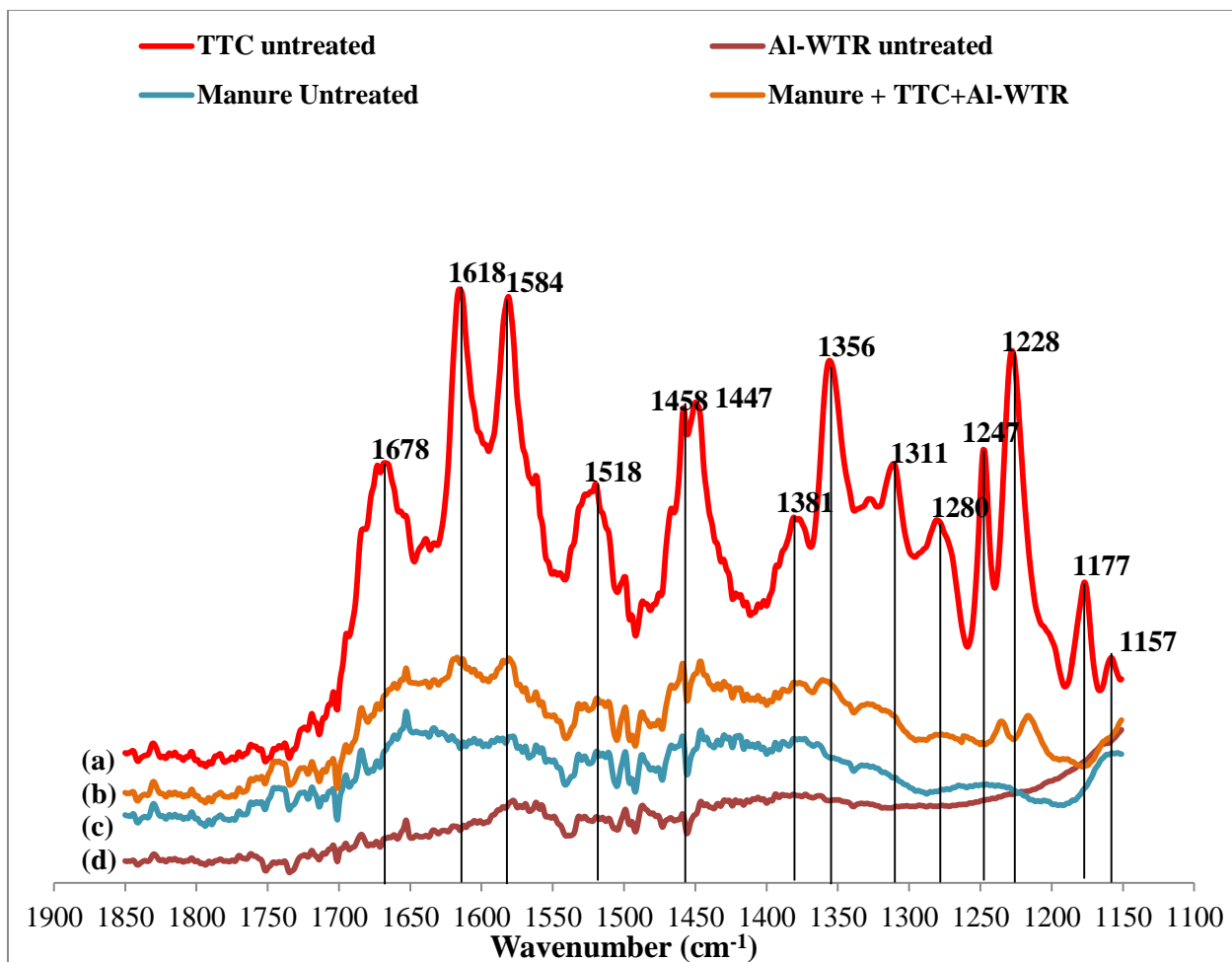
Figure 4-11C: ATR-FTIR spectra: (a) crystalline untreated TTC, (b) Manure treated with TTC amended with Al-WTR, (c) Manure untreated, and (e) Al-WTR untreated. All spectra are normalized with the highest peak observed.
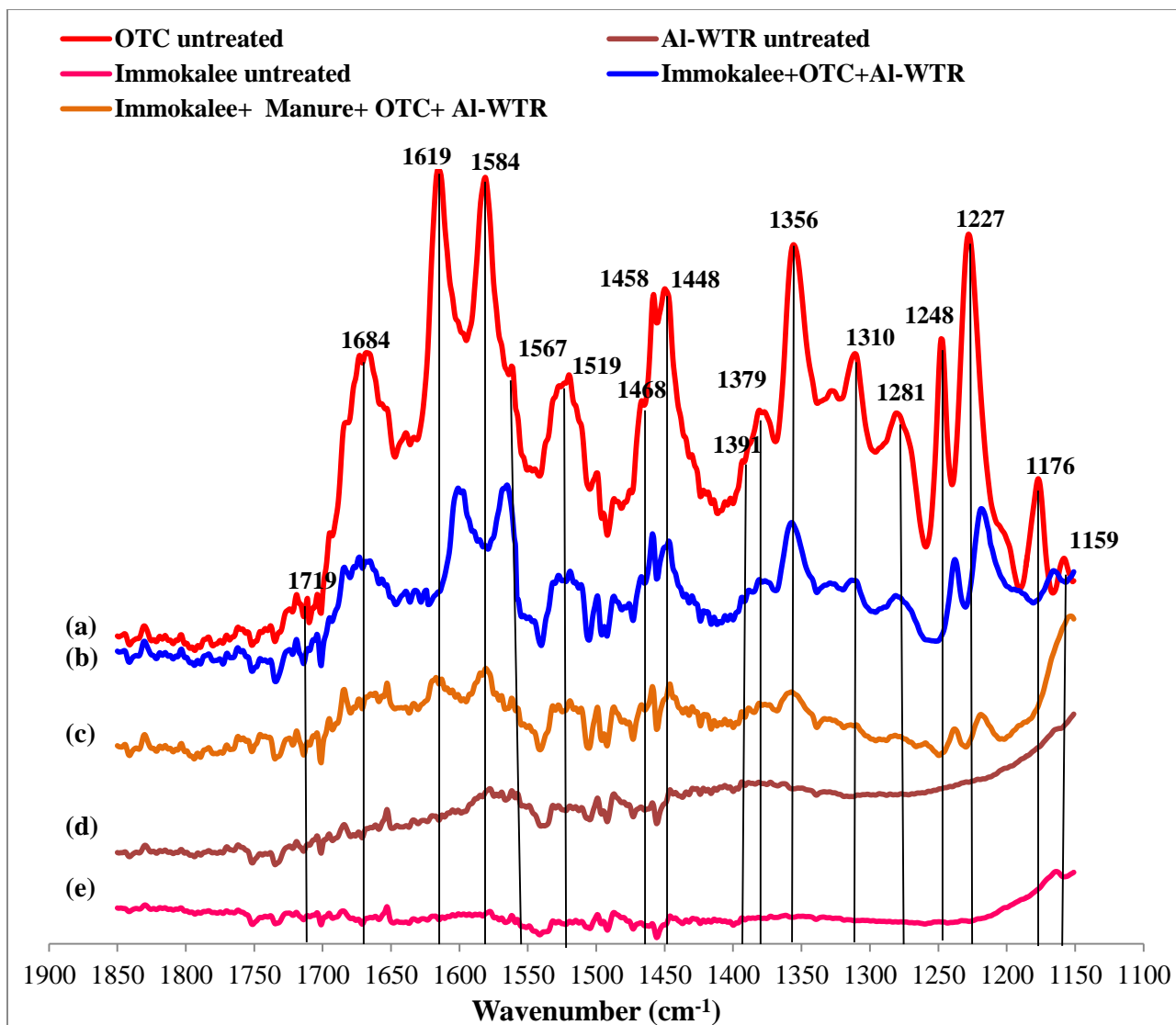
Figure 4-11D: ATR-FTIR spectra: (a) crystalline untreated OTC, (b)Immokalee treated with OTC amended with Al-WTR, (c)Immokalee treated with OTC applied with manure and amended with Al-WTR, (d)Al-WTR untreated, and (e)Immokalee untreated. All spectra are normalized with the highest peak observed.
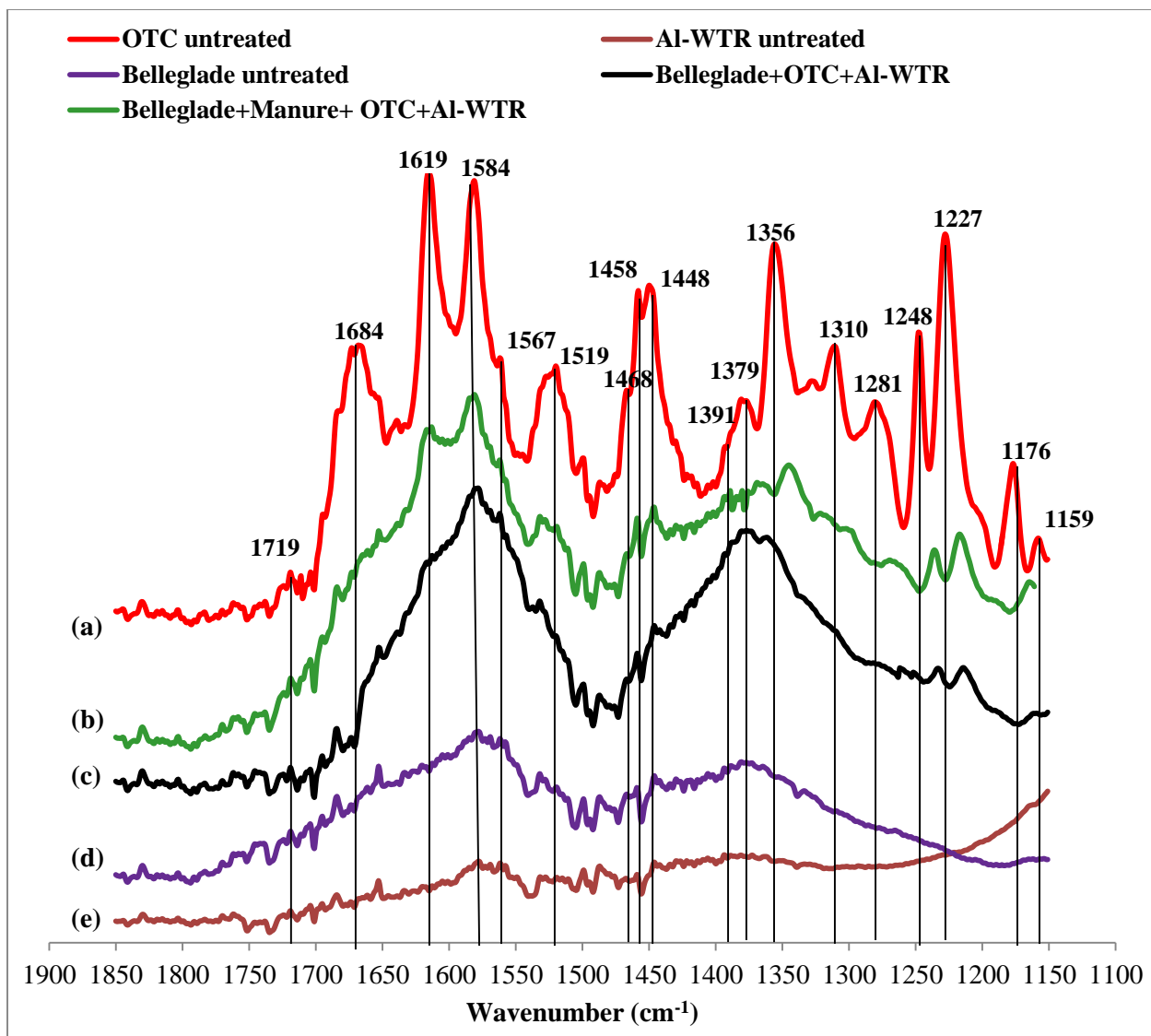
Figure 4-11E: ATR-FTIR spectra: (a) crystalline untreated OTC, (b) Belleglade treated with OTC applied with manure and amended with Al-WTR, (c) Belleglade treated with OTC amended with Al-WTR, (d) Belleglade untreated, and (e) Al-WTR untreated. All spectra are normalized with the highest peak observed.
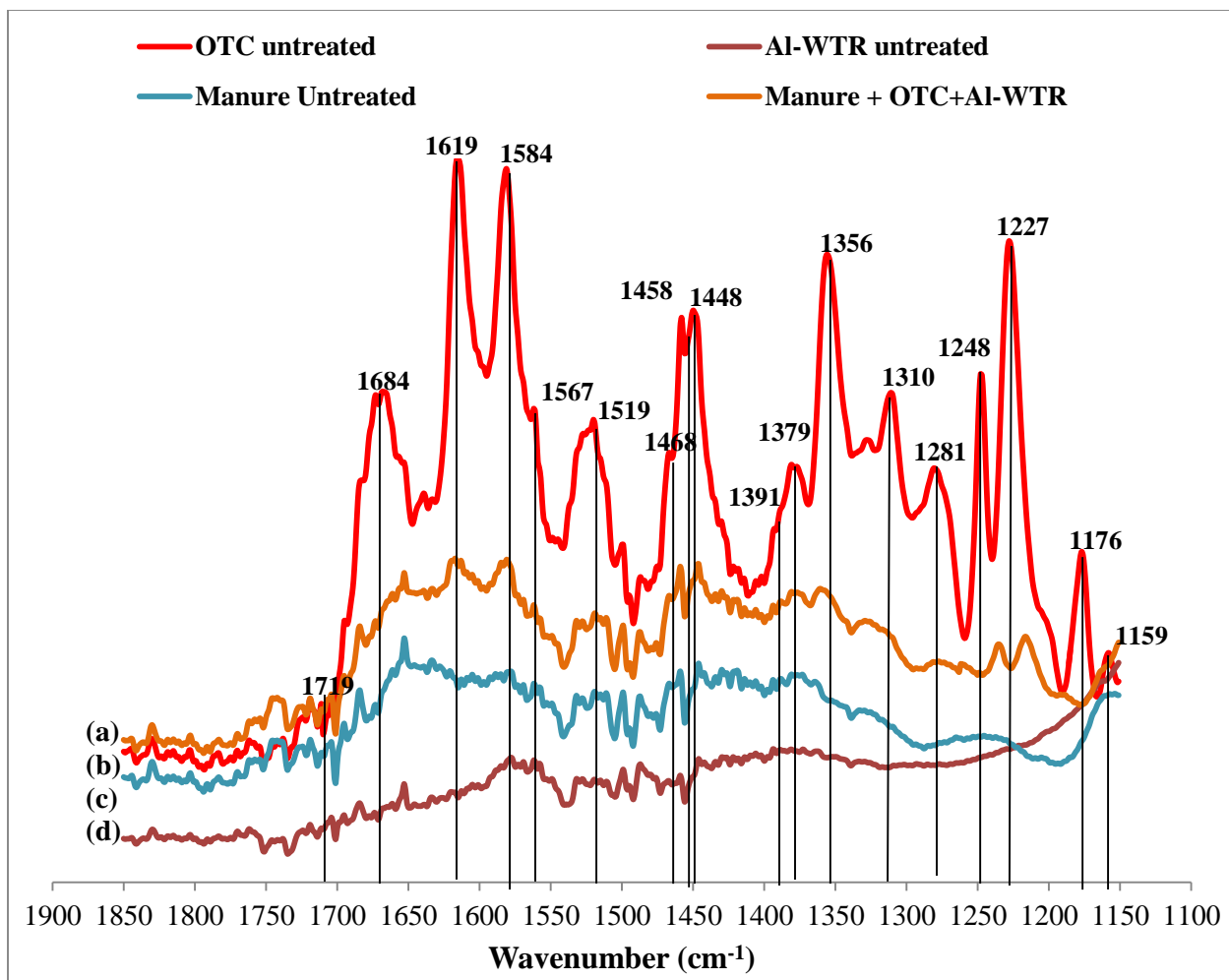
Figure 4-11F: ATR-FTIR spectra: (a) crystalline untreated OTC, (b) Manure treated with OTC amended with Al-WTR, (c) Manure untreated, and (e) Al-WTR untreated. All spectra are normalized with the highest peak observed.

4-11 A, B, D, and E) . However, some identifiable peaks showed shift to lower wave number compared to untreated TTC/OTC spectra. Changes in peak intensities and energy positions can be seen for the C–N peaks located near 1247 and 1228 cm^{-1} when comparing the OTC and TTC crystalline spectra to the spectra of treatments (1240 and 1222 cm−1, Fig. 4-11 C and F) (Kulshrestha et al., 2004; Aristilde et al., 2010). This possibly reflects an interaction of phenolic OH with the surfaces.

Overall, IR spectra indicated both for Immokalee and Belleglade soil, addition of Al-WTR influenced strong inner-sphere type adsorption of OTC/ TTC molecule in the mixed matrix. The IR band shift observed were similar to that of Punamiya et al. (2013), Chapter 2, and Chapter 3 in which the authors conducted batch sorption study with OTC/TTC on Al-WTR.

4.2.5 TCLP analysis of soils, manure, and manure applied soils amended with Al-WTR

The TCLP of soils, manure, and soils applied manure amended with different rates of Al-WTR were determined using USEPA SW-846 Method 1311 to ensure that the toxicity

characteristics concentration of several metals and metalloids were below the EPA threshold limit for their safe use and land application of Al-WTR in arable land. The TCLP values (Table 4-3) for all the metals and metalloids tested were well below the hazardous waste toxicity characteristic criterion as defined in Title 40 of the Code of Federal Regulations (CFR), Part 261.24 and below the USEPA threshold values of biosolids, thus, permitting safe land application of Al-WTR. Al and Fe does not fall under USEPA hazardous waste criteria therefore the limits of Al and Fe were compared

Table 4-3: Toxicity characteristics values of several metals and metalloids measured in soils, manure, and soils amended with manure and Al-WTR at different rates using the EPA 1311 TCLP extraction method. The results are expressed in mg L^{-1}.

Sample ID	Al*	Cr	Fe*	Ni	Cu	Zn	As	Ag	Cd	Pb
	---------------------- mg L^{-1} ----------------------									
Imm control	0.07	0.01	0.14	0.01	0.01	0.06	0.01	0.01	0.01	>MDL
Imm + 2.5% Al-WTR	57.73	0.05	0.53	0.01	0.01	0.07	0.01	0.01	0.01	>MDL
Imm + 5% Al-WTR	107.70	0.09	0.63	0.01	0.02	0.10	0.01	0.01	0.01	>MDL
Belle control	1.11	0.01	1.63	0.02	0.01	0.01	0.02	0.01	0.02	>MDL
Belle + 2.5% Al-WTR	2.85	0.01	1.37	0.02	0.01	0.01	0.01	0.01	0.02	>MDL
Belle + 5% Al-WTR	3.25	0.01	1.31	0.02	0.01	0.01	0.01	0.01	0.02	>MDL
Manure control	0.99	0.01	1.69	0.02	0.09	1.53	0.00	0.01	0.02	0.01
Manure + 2.5% Al-WTR	4.48	0.03	3.94	0.03	0.20	1.90	>MDL†	>MDL	>MDL	0.01
Manure + 5% Al-WTR	4.50	0.03	2.66	0.09	0.26	2.87	>MDL	>MDL	0.01	0.02
Imm + Manure control	0.15	0.01	0.19	0.01	0.01	0.10	>MDL	>MDL	>MDL	>MDL
Imm + Manure + 2.5% Al-WTR	33.15	0.03	0.22	0.01	0.01	0.31	>MDL	>MDL	>MDL	>MDL
Imm + Manure + 5% Al-WTR	22.35	0.02	0.16	0.01	0.01	0.21	>MDL	>MDL	>MDL	>MDL
Belle + Manure control	40.67	0.04	0.59	0.01	0.01	0.08	>MDL	>MDL	>MDL	>MDL
Belle + Manure + 2.5% Al-WTR	0.59	0.00	0.81	0.01	0.01	0.01	0.01	0.01	>MDL	>MDL
Belle + Manure + 5% Al-WTR	2.05	0.01	0.77	0.01	0.01	0.01	0.01	0.01	>MDL	>MDL

†<MDL † (Below Method Detection Limit for As, Ag, Cd, and Pb 10 mg L^{-1} using ICP-MS)

*NR: Not regulated under the Toxicity Characteristics Leaching Protocol by USEPA

to the solid industrial waste. The values for Al and Fe were also well below the threshold values of safe solid industrial waste disposal.

4.3 Conclusions

Since the beginning of the last decade, extensive research has been carried out looking into the fate and transport of VAs in the water-soil environment. Despite widespread research, reports on TCs rich manure, TCs rich manure amended soils, and finding novel remediation alternatives for immobilization of TCs have been rare. Our previous studies (Punamiya et al., 2013a and 2013b) showed promising results in removing TTC and OTC from aqueous medium with varying solution chemistry. The reported study demonstrated the effectiveness of Al-WTR at all the tested application rates in immobilizing and stabilizing TTC and OTC in manure, soils with varying physico-chemical properties, and manure amended soils under short term incubation study settings. The relative time to reach equilibrium followed the sequence: manure < Belleglade < manure amended soils < Immokalee. The presence of phosphate resulted in significant ($p<0.01$) decrease in TTC/OTC sorption by Al-WTR, with a strong dependence on initial phosphate concentration in manure, soils, and TCs rich manure amended soils. However, the rate of sorption and immobilization of TTC and OTC in manure, soils, and TCs rich manure amended soils was unaffected by the presence of sulfate. . Fourier transform infrared spectroscopy analysis indicated the possibility of TTC and OTC forming surface complex through strong innersphere-type bonds on soils, manure, and manure applied soils amended with Al-WTR. The present study provides

understanding of TTC and OTC sorption/immobilization by manure, soils, and manure amended soils with Al-WTR under different conditions, which is necessary to demonstrate the effectiveness of Al-WTR in manure-soil environment. Further, long-term green house and simulated field based studies under dynamic system with manure, soil, water, and plants are required to validate the current findings of Al-WTR in immobilizing and stabilizing TCs in manure and soils.

4.4 References

- AHI. Animal Health Institute News Release, 2011 AHI: Washington, DC, USA.
- Aga, D. S., O'Connor, S., Ensley, S., Payero, J. O., Snow, D., Tarkalson, D., 2005. Determination of the persistence of tetracycline antibiotics and their degradates in manure-amended soil using enzyme-linked immunosorbent assay and liquid chromatography-mass spectrometry. J. Agric. Food Chem., 53(18), 7165-7171.
- Arikan, O. A., Sikora, L. J., Mulbry, W., Khan, S. U., Foster, G. D., 2007. Composting rapidly reduces levels of extractable oxytetracycline in manure from therapeutically treated beef calves. Bioresource technology, 98(1), 169-176.
- Bao, Y., Zhou, Q., Wang, Y., 2009. Adsorption characteristics of tetracycline by two soils: assessing role of soil organic matter. Soil Research 47(3), 286-295.
- Bao, Y., Zhou, Q., Wan, Y., Yu, Q., Xie, X., 2010. Effects of soil/solution ratios and cation types on adsorption and desorption of tetracycline in soils. Soil Science Society of America Journal 74(5), 1553-1561.
- Bassil, R. J., Bashour, I. I., Sleiman F. T., Abou-Jawdeh, Y. A., 2013.Antibiotic uptake by plants from manure-amended soils, J Environ Sci Health B., 48 (7), 570-574
- Ben-Dor E., Banin. A., 1989. Determination of organic matter content in arid-zone soils using a simple "loss-on-ignition" method. Commun. Soil Sci. Plant Anal. 20:1675–1695
- Boxall, A.B.A., Johnson, P., Smith, E.J., Sinclair,C.J., Stutt, E., Levy, L., 2006. Uptake of veterinary medicines from soils into plants J. Agr. Food Chem., 54, 2288–2297
- Bui, T. X., Choi, H., 2010. Influence of ionic strength, anions, cations, and natural organic matter on the adsorption of pharmaceuticals to silica. Chemosphere, 80, 681-686.
- Chander, Y., Kumar, K., Goyal, S. M., & Gupta, S. C., 2005. Antibacterial activity of soil-bound antibiotics. J. Environ. Qual., 34(6), 1952-1957.
- Chee-Sanford, J. C., Aminov, R. I., Krapac, I. J., Garrigues-Jeanjean, N., Mackie, R. I., 2007. Occurrence and diversity of tetracycline resistance genes in lagoons and groundwater underlying two swine production facilities Appl. Environ. Microbiol 67, 1494-1502.
- Datta, R., Sarkar, D., 2005. Consideration of soil properties in assessment of human health risk from exposure to arsenic-enriched soils Int. Environ. Asses. Manage., 1, 55–59
- Dolliver, H., Gupta, S., and Noll, S., 2008. Antibiotic degradation during manure composting. J. Environ. Qual., 37(3), 1245-1253.
- Elliot, H.A., O'Connor, G.A. Lu, P., Brinton, S., 2002. Influence of water treatment residuals on phosphorus solubility and leaching. J. Environ. Qual. 31:1362-1369.

- Elliott, H.A., Dempsey, B.A., 1991. Agronomic effects of land application of water treatment sludges. J. AWWA, 83 126–131.
- Figueroa, R.A., Leonard, A., MacKay, A.A., 2004. Modeling Tetracycline Antibiotic Sorption to Clays. Environmental Science & Technology 38, 476-483.
- Figueroa, R.A., MacKay, A.A., 2005. Sorption of Oxytetracycline to Iron Oxides and Iron Oxide-Rich Soils. Environmental Science & Technology 39, 6664-6671.
- Fritz, J.W., Zuo, Y., 2007. Simultaneous determination of tetracycline, oxytetracycline, and 4-epitetracycline in milk by high-performance liquid chromatography. Food Chemistry 105, 1297-1301.
- Gee, G. W., Bauder, J. W., Klute, A., 1986. Particle-size analysis. Methods of soil analysis. Part 1. Physical and mineralogical methods, 383-412.
- Gu, C., Karthikeyan, K., 2005. Interaction of tetracycline with aluminum and iron hydrous oxides. Environmental Science & Technology 39, 2660-2667.
- Hakk, H., Millner, P, Larsen, G., 2005. Decrease in water-soluble 17β-estradiol and testosterone in composted poultry manure with time J. Environ. Qual. 34, 943-950.
- Hamscher, G., Sczesny, S., Höper, H., Nau, H., 2002. Determination of persistent tetracycline residues in soil fertilised with liquid manure by high-performance liquid chromatography with electrospray ionisation tandem mass spectrometry, Anal. Chem.74, 1509–1518.
- Hamscher, G., Pawelzick, H.T., Hoper, H., Nau, H., 2005. Different behaviour of tetracyclines and sulfonamides in sandy soils after repeated fertilisation with liquid manure, Environ. Toxicol. Chem. 24, 861–868.
- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997a. Soil pH (1:2v/v). IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL) chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p15.
- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997b. Electrical Conductivity. IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL) chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p24.
- Ippolito, J., Barbarick, K., Elliott, H., 2011. Drinking Water Treatment Residuals: A Review of Recent Uses. Journal of Environmental Quality 40, 1-12.
- Ji, L., Chen, W., Duan, L., Zhu, D., 2009. Mechanisms for strong adsorption of tetracycline to carbon nanotubes: A comparative study using activated carbon and graphite as adsorbents. Environmental Science & Technology 43, 2322-2327.
- Kang, J., Liu, H.J., Zheng, Y.M., Qu, J.H., Chen, J.P., 2011. Application of nuclear magnetic resonance spectroscopy, Fourier transform infrared spectroscopy, UV-Visible spectroscopy and kinetic modeling for elucidation of adsorption chemistry in uptake of tetracycline by zeolite beta. Journal of Colloid and Interface Science 354, 261-267.
- Kay, P., Blackwell, P. A., Boxall, A. B., 2004. A. Fate of veterinary antibiotics in a macroporous tile drained clay soil Environ. Toxicol. Chem. 23, 1136-1144.

- Klute, A., 1986. Methods of soil analysis. Part 1. Physical and mineralogical methods. SSSA Publications, Madison, WI.
- Kulshrestha, P., Giese, R.F., Aga, D.S., 2004. Investigating the Molecular Interactions of Oxytetracycline in Clay and Organic Matter: Insights on Factors Affecting Its Mobility in Soil. Environmental Science & Technology 38, 4097-4105.
- Kumar, K., Gupta, S.C., Chander, Y., Singh. A.K., 2005. Antibiotic use in agriculture and its impact on the terrestrial environment. Adv. Agron 87, 1–54.
- Makris, K., Harris, W., O'Connor, G., Obreza, T., 2004. Phosphorus immobilization in micropores of drinking-water treatment residuals: Implications for long-term stability. Environmental Science & Technology 38, 6590-6596.
- Makris, K., Harris, W., O'Connor, G., Obreza, T., Elliott, H., 2005. Physicochemical properties related to long-term phosphorus retention by drinking-water treatment residuals. Environmental Science & Technology 39, 4280-4289.
- McClellan, K., Halden, R. U., 2010. Pharmaceuticals and personal care products in archived U.S. biosolids from the 2001 EPA National Sewage Sludge Survey. Water Res. 44 (2), 658−668.
- Muller, A. K., Westergaard, K., Christensen, S., Soresen, S. J., 2002. The diversity and function of soil microbial communities exposed to different disturbances Microb. Ecol 44, 49-58.
- Munir, M., Xagoraraki, I., 2011. Levels of Antibiotic Resistance Genes in Manure, Biosolids, and Fertilized Soil. J. Environ. Qual. 40 (1) 248-255.
- Oberle, K., Capdeville, M.J., Berthe, T., Budzinski, H., Petit, F., 2012. Evidence for a Complex Relationship between Antibiotics and Antibiotic-Resistant Escherichia Coli: From Medical Center Patients to a Receiving Environment. Environmental Science & Technology 46, 1859-1868.
- Ostermann, A., Siemens, J., Welp, G., Xue, Q., Lin, X., Liu, X., Amelung, W., 2013. Leaching of veterinary antibiotics in calcareous Chinese croplands. Chemosphere, 91 (7) 928-934.
- O'Connor, G.A., Elliott, H.A., Lu, R., 2002. Characterizing water treatment residuals phosphorus retention. Soil and Crop Science Society of Florida Proceedings 61, 67-73.
- Pils, J., Laird, D., 2007. Sorption of tetracycline and chlortetracycline on K- and Ca-saturated soil clays, humic substances, and clay-humic complexes. Environmental Science & Technology 41, 1928-1933.
- Prakash, P., Sengupta, A.K., 2003. Selective coagulant recovery from water treatment plant residuals using donnan membrane process. Environmental Science & Technology 37, 4468-4474.
- Punamiya, P., Sarkar, D., Rakshit, S., Datta, R., 2013a. Effectiveness of Al- based drinking water treatment residuals as a novel sorbent to remove tetracyclines from aqueous medium. J. Environ. Qual. *doi:10.2134/jeq2013.03.0082*.

- Punamiya, P., Sarkar, D., Rakshit, S., Datta, R., 2013b. Effect of solution properties, competing ligands, and complexing metal on sorption of tetracyclines on Al-based drinking water treatment residuals, J. Hazard. Mater. (To be submitted).
- Sall, J., Creighton, L., Lehman, A., 2005. JMP start statistics, 8th edition. Cary, NC, SAS Institute.
- Sarmah, A. K., Meyer, M. T.; Boxall, A. B. A., 2006. A global perspective on the use, sale, exposure pathways, occurrence, fate and effects of veterinary antibiotics (VAs) in the environment Chemosphere 65, 725-759.
- Sassman, S. A., Lee, L. S., 2005. Sorption of three tetracyclines by several soils: assessing the role of pH and cation exchange. Environmental science & technology 39(19), 7452-7459.
- Sengelov, G., Agerso, Y., Halling-Sorensen, B., Baloda, S. B., Anderson, J. S., Jensen, L. B., 2003. Bacterial antibiotic resistance levels in Danish farmland as a result of treatment with pig manure slurry Environ. Int 28. 587-595.
- Stoob, K., Singer, H., Mueller, S.R., R.P. Schwarzenbach, R.P., C.H. Stamm, C.H., 2007 Dissipation and transport of veterinary sulfonamide antibiotics after manure application to grassland in a small catchment Environ. Sci. Technol 41, 7349–7355
- Teixido, M., Hurtado, C., Pignatello, J.J., Beltran, J.L., Grenados, M., and Peccia, J., 2013. Predicting contaminant adsorption in black carbon (biochar)-amended soil for the veterinary antimicrobial sulfamethazine. Environ. Sci. Technol. 47, 6197.
- Ter Laak, T.L.,Wouter, A.G., Tolls, J., 2006. The effect of pH and ionic strength on the sorption of sulfachloropyridazine, tylosin, and oxytetracycline to soil. Environ. Toxicol. Chem. 25, 904–911.
- Tolls, J., 2001. Sorption of veterinary pharmaceuticals in soils: a review, Environ. Sci. Technol. **35**, pp. 3397–3406.
- Topp, E., Monteiro, S.C., Beck, A.., Ball Coelho, B., Boxall, P.W. Duenk, A.B.A., Kleywegt, S., Lapen, D.R., Payne, M., Sabourin, L., Metcalfe, C.D., 2008. Runoff of pharmaceuticals and personal care products following application of biosolids to an agricultural field Sci. Total Environ., 396, 52–59
- U.S. Department of Agriculture, Economic Research Service (USDA/ERS), 2005. Confined animal and manure nutrient data system. http://www.ers.usda.gov/Data/manure/default
- Van Dijk, J., & Keukens, H. J., 2000. The stability of some veterinary drugs and coccidiostats during composting and storage of laying hen and broiler faeces. In Residues of veterinary drugs in food. Proc. of the Euroresidue IV Conf., Veldhoven, Netherlands (pp. 8-10).
- Wang, Y. J., Jia, D. A., Sun, R. J., Zhu, H. W., Zhou, D. M., 2008a. Adsorption and cosorption of tetracycline and copper (II) on montmorillonite as affected by solution pH. Environmental Science and Technology 42(9), 3254-3259.

- Wang, Q., Yates, S.R., 2008b. Laboratory study of oxytetracycline degradation kinetics in animal manure and soil. J. Agric. Food Chem. 56, 1683-1688.
- Wang, Y. J., Sun,R. J., Xiao, A. Y., Wang, S. Q., and Zhou, D. M., 2010. Phosphate affects the adsorption of tetracycline on two soils with different characteristics. Geoderma, 156, 237-242.
- Watanabe, F. S., Olsen, S. R., 1965. Test of an ascorbic acid method for determining phosphorus in water and $NaHCO_3$ extracts from soil. Soil Science Society of America Journal. 29(6), 677-678.
- Westergaard, K., Muller, A. K., Christensen, S., Bloem, J., Sorensen, S. J., 2001. Effects of tylosin as a disturbance on the soil microbial community Soil Biol. Biochem 33, 2061-2071.
- Winckler, C. and A. Grafe, A., 2001. Use of veterinary drugs in intensive animal production Evidence for persistence of tetracycline in pig slurry. J Soil Sediment 1, 67-70.
- Zhang, D., Pan, B., Wu, M., Wang, B., Zhang, H., Peng, H. B., Wu, D., Ning P., 2011. Adsorption of sulfamethoxazole on functionalized carbon nanotubes as affected by cations and anions. Environ. Pollut., 159, 2616–2621
- Zhang, Y., Zhang, C., Parker, D. B., Snow, D. D., Zhou, Z., Li, X., 2013. Occurrence of antimicrobials and antimicrobial resistance genes in beef cattle storage ponds and swine treatment lagoons. Sci Total Environ.
- Zhao, Y., Gu, X., Gao, S., Geng, J., and Wang, X., 2012. Adsorption of tetracycline (TC) onto montmorillonite: Cations and humic acid effects. Geoderma 183-184, 12-18.

Chapter 5

Immobilization and Stabilization of Tetracyclines in Soils and Manure Amended Soils: A Long-Term Greenhouse Column Study

[Portion of this chapter has been submitted to a Journal for Publication]

Abstract

The presence of tetracyclines (TCs) in manure, soil, sediments, sewage, surface water, and groundwater has been reported by several recent studies. Our earlier batch sorption and incubation studies revealed high adsorption affinity of Al-based drinking water treatment residuals (Al-WTR) for tetracycline (TTC) and oxytetracycline (OTC). Based on the successful results from the sorption studies, we hypothesized that Al-WTR could be a promising "green" sorbent for TTC and OTC rich soils and manure amended soils. In the current one year greenhouse column study, we evaluated the effectiveness of Al-WTR to immobilize and stabilize TTC and OTC in soils and manure amended soils. Two physico-chemically variant soil types (Immokalee Spodosol series and Belleglade Pahokee muck series) were chosen based on their potential differences with regard to TCs reactivity. Bermuda grass (*Cynodon dactylon*) and corn (*Zea mays L.)* were used as control and test plants, respectively. Manure and soil samples were spiked with various concentrations of TTC/OTC (0, 2.25, and 22.5 mg kg^{-1}) and amended at three rates (0, 25, and 50 g kg^{-1}) of Al-WTR. Soils, manure amended soils, plants, and leachate samples were collected periodically for one year. Soil and manure amended soil samples were subjected to different treatments followed by solid phase extraction to understand retention and release mechanisms. Results showed that, compared to the unamended (no

Al-WTR) soils and manure amended soils, leaching and mobility of TTC/OTC significantly ($p<0.001$) decreased by 44-68% within 12 months across all the treatments tested. Leaching of TTC and OTC reduced significantly ($p< 0.05$) from soils and manure amended soils amended with 50 g kg^{-1} Al-WTR as compared to those with 25 g kg^{-1} Al-WTR. Presence of plant cover in the form of bermuda grass and corn reduced leaching of TTC/OTC by 6-9% compared to columns with no plant cover. Highest total leaching (time zero to 12 months) was observed in Immokalee soil, followed by Belleglade soil and manure amended soils, showing physico-chemically dependent leaching behavior. Data from SPE showed less than 12% release of the initial TTC/OTC concentration in both soils and manure amended soils in different phases tested, indicating strong binding of TCs on Al-WTR. Results obtained from the current greenhouse column study are encouraging and will potentially help to develop an optimal low-cost remediation technique for TCs and other veterinary antibiotics using a waste by-product.

Keywords: Tetracyclines, Soil, Remediation, Drinking Water Treatment Residuals, Greenhouse column study.

5. Introduction

Veterinary antibiotics (VAs) are biologically active compounds and primarily used to treat infections, to protect animals from infectious diseases, or to promote growth at concentrated animal feeding operations (CAFOs) (Boxall et al., 2003). Since early 2000, VAs has been submitted to an environmental risk assessment. VAs get partially metabolized in the gut of the animal and considerable amount is excreted either unchanged, as conjugate, or as active metabolites in urine and manure (Winckler and Grafe, 2001; Kumar et al., 2005a). Additional, degradation of VAs can occur during various processes at CAFO's (storage ponds, treatment lagoon, etc.), but large amount of VAs can still reach the water and soil environment through accidental leakage or leaching from CAFOs waste storage and land application of manure as a rich source of nutrients (Ostermann et al., 2013; Stoob et al., 2007; Topp et al., 2008). As a result, several VAs have been found in aquatic (ground water and surface water) and terrestrial (agricultural soils, waste storage at CAFOs, etc.) environments.

Tetracyclines (TCs) are broad-spectrum antibiotics, which are extensively used for therapeutic purposes in the livestock industry. Studies have shown that tetracyclines can enter the environment in significant concentrations via use of reclaimed waste water for irrigation (Tanoue et al., 2012; Shenker et al., 2011) or by repeated land application of manure/biosolids (Bassil et al., 2013; McClellan and Halden, 2010. Further they tend to accumulate in soils (Kay et al., 2004; Hamscher et al., 2002 and 2005), generating potential environmental and human health risks (Oberle et al., 2012; Munir and Xagoraraki**,** 2011; Daughton and Ternes, 1999). The presence of TCs in the environment

is of great concern because even at ng L^{-1} levels, these molecules are biologically active

and can affect critical development stages and endocrine systems of aquatic and

terrestrial organisms (Zhang et al., 2013; Allen et al., 2010; Sarmah et al., 2006).

TCs have been found in the ground water, surface water, surface soils, and soils

amended with TCs rich manure in concentration ranging from few micrograms to

milligrams per kilogram (Aga et al., 2005, Hamscher et al., 2002). Kumar et al. (2005a)

reported trace levels to > 200 mg kg^{-1} or L^{-1} antibiotic concentration in manure with

typical concentration in the range of 1 to 10 mg kg^{-1} or L^{-1}. Hamscher et al. (2005)

reported concentrations of up to 270 μg kg^{-1} (OTC), 443 μg kg^{-1} (TTC), and 93 μg kg^{-1}

(CTC) in manure impacted surface soils. Winckler and Grafe (2001) found TCs to persist

in agricultural soils at concentrations of 450–900 μg kg^{-1}, and Hamscher et al. (2002)

detected high levels (20 mg kg^{-1}) of TTC in the top soil fertilized with liquid manure.

Further, Kay et al. (2004) observed that after manure was applied to a field in 2

consecutive years, OTC concentration in the soil was 1691 μg kg^{-1}, and concentrations as

high as 613.2 μg L^{-1} were observed in drain flows, which end up in aquatic systems

through agricultural effluent discharges. Sarmah et al. (2006) also reported high

possibility of TCs release from soil surface to mineral horizon to the aquifer, causing

surface and groundwater contamination. Further, a bioaccumulation study carried out by

Kumar et al. (2005b) showed uptake of TCs (0.002 to 0.017 μg kg^{-1} fresh weight) by

plant grown in soils amended with VAs-rich manure. Boxall et al. (2006) showed uptake

of various VAs (3 to 38 μg kg^{-1} fresh weight) in plant tissue and leaves in plants grown in

a sandy soil. Similarly, various recent studies have shown uptake of VAs in plant tissues,

leaves, fruit, etc. on plants grown on soils spiked with TCs or on TCs rich manure amended soils (Bassil et al., 2013; Chitescu et al., 2013; Tanoue et ., 2012; Wu et al., 2010). However, the associated health risks and health implications of VAs residue in plants is unknown, but several potential adverse impacts include allergic/toxic reactions, chronic toxic effects due to prolonged low-level intake, development of antibiotic resistant bacteria, and disruption of digestive system (Kumar et al., 2005b). Thus, there is a great need to develop remediation techniques to immobilize and stabilize TCs from soils and manure amended soils to avoid their transportation, leaching, and release in to the aqueous environment, uptake by plants and animal, and thereby prevent associated human health risks.

Our previous research have demonstrated high affinity of Al-WTR for TCs in aqueous medium under different environmental conditions (Punamiya et al., 2013a and 2013b) and in manure, soils, and manure amended soil under short term incubation study settings. In the current study, we evaluated the effectiveness of Al-WTR to immobilize and stabilize TTC and OTC in soils and manure amended soils under a dynamic system with soils, manure, water, and plants in one year column greenhouse study. The objectives of the study were; i) to assess the long term (one year) effect of Al-WTR on TTC and OTC immobilization and stabilization in two soils and manure amended soils with varying physico-chemical properties, and ii) to evaluate the effect of Al-WTR on TTC and OTC uptake by plants and TCs concentration in leachates over that period of time.

5.1 Materials and Methods

5.1.1 Soils, manure, and Al-WTR collecting, sampling, preparation, and

characterization

Two types of soils with varying physico-chemical properties (the Immokalee

Spodosols series and Belleglade Pahokee Muck series), cattle manure, plants (Bermuda

grass (*Cynodon dactylon*) and Corn (*Zea mays*)) and Al-WTR were used in this study.

The Immokalee series Spodosols soils were collected from surface horizons in the

Southwest Florida Research and Education Center, Immokalee, Florida and the

Belleglade Pahokee Muck series soils were collected from the surface horizons from

Everglades Research and Education Center at Belle Glade, Florida. The Al-based WTR

were obtained from the drinking-water treatment plant in Bradenton, FL, USA. Corn and

Bermuda grass seeds were obtained from U.S. Department of Agriculture (USDA) with

>90% germination rate. Manure, WTR, and Soil samples were air-dried and then sieved

with a 2-mm sieve before the Manure, WTR, and Soil being subjected to characterization

and greenhouse experiments. Fresh cattle manure samples were systematically collected

from surface, adjacent corners, and bottom of the manure pit at Rutgers Cook Campus,

NJ, USA. The fresh manure samples were thoroughly mixed into a single batch and

refrigerator at 4°C until further use.

Manure, soils, and Al-WTR were characterized for selected physicochemical

properties before the initiation of the study. The pH, electrical conductivity, water

content, and texture were measured using standard protocols (Hanlon et al., 2002).

Organic matter was measured using the loss-on-ignition method (Ben-Dor and Banin,

1989; Klute 1996). Total C and N were determined by combustion at 1100°C using an

Elementar Vario EL CHNS/O analyzer (Elementar, NJ USA). Concentrations of Oxalate-

extractable Al and Fe of the soils and WTRs samples were determined using Tamm's

reagent (Klute 1996). Acid digestion was used for total-recoverable Fe and Al

concentrations following the USEPA 3050B method (USEPA, 2000). Toxicity

Characteristic Leaching Procedure (TCLP) using SW-846 Method 1311 was used for the

WTRs to assess their potential for any waste leaching in a landfill environment (EPA

method 131). The concentrations of RCRA metals and metalloids in the extract of the

TCLP method were then assessed against regulatory levels to check whether WTRs

exhibits the Toxicity Characteristic (TC). Phosphorus concentration was colorimetrically

measured with a UV/vis-spectrophotometer, using the molybdate–ascorbic acid method

(Watanabe and Olsen, 1965). Texture of the soils was determined by pipette method and

Malvern particle size analyzer (Gee and Bauder, 1986).

5.1.2 Study design and soil amendments

A one-year greenhouse study was initiated by preparing 80 columns. The detailed

column design is illustrated in Figure 5-1. In brief, the columns were made of PVC pipes

(0.38 m high × 0.15 m internal diameter). An outlet nozzle was fitted at the bottom and

connected with a tube to collect the leachate in a clean, 1 L Nalgene polyethylene bottle.

To prevent soil leaching from the column, a fine nylon mesh (8 μm mesh size) was

placed at the bottom of the column above the outlet nozzle lined with non-reactive clear

glass marbles. Each column was filled with 0.18 m height of play sand, followed by

0.15 m of soil/soil amended with manure.

Immokalee and Belleglade soils were spiked with TTC or OTC in form of

tetracycline hydrochloride and oxytetracycline hydrochloride (USP grade, ≥99%)

obtained from Sigma-Aldrich chemical (St. Louis MO) to achieve final TTC/OTC load of

2.25 and 22.5 mg kg^{-1}, respectively. In another set of treatment, soils were amended with

TTC/OTC rich manure at a rate of 11.2 Mg ha^{-1} to simulate a realistic field loading rate to

achieve the final required concentration of TTC/OTC in the soils. Further after

equilibration the soils and manure amended soils were mixed with Al- WTR at two rates

25 and 50 g kg^{-1}, respectively. Soils and manure amended soils spiked with and without

TTC/OTC but not WTR, with/without plants, were included as a control. Columns were

filled with the soil/manure amended soils-TTC/OTC-Al-WTR mixture after thorough

mixing.
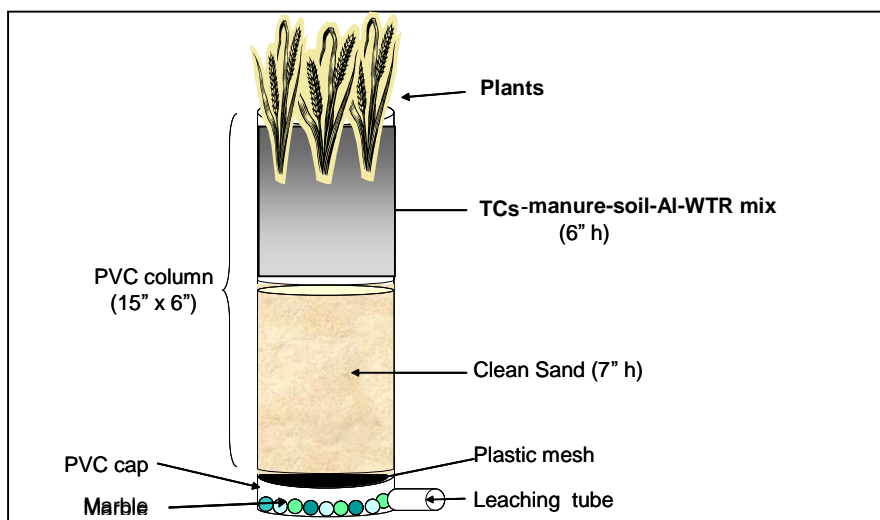
Figure 5-1: Design of the PVC column for the green house study (modified from Andra, 2008)

For samples spiked with TTC/OTC, there were a total of 32 pots with Al-WTR treatments (two types of TCs × two concentrations × two soils × two rates of Al-WTR × two replicates). In addition, 16 control columns without Al-WTR, with and without plants (two soils × two TCs × two plants × two replicates) were also prepared. The columns were maintained at optimum soil moisture conditions (70% of water holding capacity). The pots were arranged in a randomized block design and were rotated periodically to account for variances in temperature and sunlight within the greenhouse. Bermuda grass was used a control plant, whereas corn was used as the test crop.

Soil samples were collected by composite sampling from the top 10-cm surface. The first soil sampling was done immediately after spiking (time zero), and then after 0.25 and 0.5 year of equilibration time. The columns were over-watered twice (after 0.25 and 0.5 year) to induce leaching. The leachate was collected for each column and analyzed for soluble TTC and OTC. Plant samples were harvested after time of maturity i.e. 0.5 year. These TTC and OTC extractions in the plant samples were done according to Boxall et al., 2006. Columns were retained for the remaining time without plant cover to understand the effect of soil/Al-WTR aging on TTC and OTC fraction and mobility. After 0.5 year, soils were sampled at the end of 1 year. After each sampling, soil samples were extracted using method described in Wang and Yates, 2008 and analyzed (Fritz and Zuo, 2007) for total TTC and OTC using HPLC (Finnigan surveyor plus, Thermo Scientific).

5.1.3 Sample Extraction and TCs Analysis

Plant samples were extracted using citric acid and methanol (Boxall et al., 2006). Further the samples were loaded on Oasis HLB cartridge and elution was done using HPLC grade methanol. Soil samples were extracted using citric acid, oxalic acid, and methanol/water mixture for total TTC and OTC (Wang and Yates, 2008). In brief, 5 g of soil sample was extracted using mixture of 2.5 g of citric acid, 1.5 g of oxalic acid, and 15 mL of methanol/water mixture (9:1 volume by volume) by shaking for 30 min, followed by centrifugation and filtration of the supernatant.

Further, four different extractions were conducted on the soil samples. Four different treatments, DI water, 1 mol L^{-1} KCl, methanol, and 0.25 mol L^{-1} EDTA were used. The methanol treatment was used to check the role of hydrophobicity on sorption, KCl was used to see the effect of competing ion on sorption, and EDTA was used as a chelating agent to evaluate competitively displaced sorbed TTC and OTC from Al-WTR. The sample solutions were shaken at 250 rpm on a reciprocating shaker for 1 h. The samples were withdrawn, centrifuged, and filtered.

Detailed HPLC of TCs analysis has been discussed in details elsewhere (Punamiya et al., 2013). In brief, Finnigan surveyor plus HPLC system (Thermo Scientific) with surveyor PDA plus detector (photodiode array) and a surveyor plus auto-sampler were used for all analyses. A hypersil gold C_{18} column (150×4.6 mm, 5 μm) (Thermo Scientific) with a corresponding hypersil gold guard column (10×4 mm, 5 μm) (Thermo Scientific) at room temperature was used for all separations. Samples were eluted isocratically with a mobile phase consisting of 0.01 mol L^{-1} aqueous oxalic acid: acetonitrile: methanol (150:20:20 by volume) (Fritz and Zuo, 2007). All the elemental

analyses were performed using Thermo X-series Inductively Coupled Plasma Mass

Spectrometer (ICP-MS) (Thermo Electron).

5.1.4 Statistical Analysis

Statistical analysis was performed using JMP IN version pro 10 (Sall et al. 2005).

Two/Three-way analysis of variance (ANOVA) was performed to examine the effect of

Al-WTR on TTC and OTC immobilization in soils and manure amended soils as a

function of contact time, initial TTC and OTC concentration, presence/absence of plants,

and different rates of Al-WTR. To evaluate differences among treatment means for the

amount of TTC/OTC in leachates and extent of immobilization, Tukey–Kramer honest

significant difference (HSD) test was used. Treatment differences were deemed

significant at $\alpha = \leq 0.05$ or 0.01 at 95 and 99 % confidence interval, respectively. All data

were expressed as mean ($n = 2$) along with one standard deviation.

5.2 Results and Discussion

5.2.1 Soils and manure amended soils TTC/OTC Immobilization: Effect of Al-WTR

Immokalee spodosol series and Belleglade Pahokee muck series soils were used

in the current greenhouse study based on their distinct physico-chemical properties. Table

5-1 shows the selected general physico-chemical properties of soils, Al-WTR, and

manure. The general physico-chemical properties of soils, Al-WTR, and manure have

been discussed in the previous chapter. In brief, being sandy and lacking positively

charged surfaces (e.g., amorphous Fe/Al oxides), the Immokalee soil is likely to have

minimal retention capacity (Datta and Sarkar,2005; Figueroa and MacKay, 2005; Gu and

Karthikeyan 2005), thereby potentially releasing high content of TCs spiked into the environment. The Belleglade soil was slightly acidic-neutral (pH 6.45) with high SOM (85%) and much higher concentrations of Fe, Al, Ca, and Mg compared to Immokalee soil indicating higher TCs retention potential (Gu and Karthikeyan, 2005; Figueroa and MacKay, 2005;).The soils also varied widely in their salinity (measured as electrical conductivity [EC]) and the CEC, both likely to affect TCs sorption (Sassman and Lee, 2005; Ter Laak et al., 2006; Bao et al., 2010). General physicochemical properties of the Al-WTR are given in Table 5-1 and have been discussed in details in the previous chapter. In brief, Al-WTR was acidic (6.1) and amorphous (about 75% of total Al) in nature. The total C values (185 g kg^{-1}) for Al-WTR are in the range of organic C found in various WTRs nationwide (Ippolito et al., 2011). The organic matter for Al- WTR was 240 g kg^{-1} higher than those of both soils. Cattle manure used in the study was acidic (pH 6.1) in nature with high organic matter (250.5 ± 2.5 kg^{-1}) and total P content (4.5 ± 2.1 kg^{-1}). Total Ca+Mg ranged between 0.11 to 0.15 g kg^{-1}. No detectable background levels (method detection limit 1×10^{-3} mmol L^{-1}) of TTC and OTC were found in the soils, manure, and Al-WTR used in the greenhouse study.

5.2.2 Effect of Al-WTR in Immokalee and manure amended Immokalee soil

At time zero, no significant ($p > 0.05$) difference was found in TTC/OTC immobilization and stabilization at both the concentrations tested (2.25 and 22.5 mg kg^{-1}) in Immokalee soil and manure amended Immokalee soil between the unamended control) and the Al-WTR-amended soil (treatment) at two different rates (25 and 50 g kg^{-1}). However, the trend changed after 0.25 year of the equilibration time for both the

Table 5-1: Selected general physico-chemical properties of Immokalee and Belleglade soils, cattle manure, and Al-WTR used in the greenhouse study. Data are expressed as mean of three replicates ± one standard deviation.

	Al-WTR	Immokalee	Belleglade	Manure
pH	5.1 ± 0.34	5.9 ± 0.42	7.85 ± 0.12	6.2 ± 0.1
EC^a (μs/cm)	363 ± 12.3	59.5 ± 4.5	503 ± 13	240 ± 5.5
CEC^b (C mol/kg)	Nd	$774 \pm 28^§$	$18,908 \pm 1204^§$	Nd
OM^c (g/kg)	240 ± 8.78	$8.40 \pm 0.2^§$	80.0 ± 1.5	250.5 ± 2.5
Sand (%)	65 ± 6	99 ± 0.02	84.2 ± 1.5	Nd
Clay (%)	15 ± 3	0.57 ± 0.01	4.58 ± 2.2	Nd
Silt (%)	13 ± 3	0.35 ± 0.05	7.02 ± 1.4	Nd
$(Al+Fe)_{Total}$ (g/kg)	122.2 ± 8.5	0.08 ± 0.001	5.42 ± 0.46	<MDL
$(Al+Fe)_{ox}$ (g/kg)	92.3 ± 4.5	0.02 ± 0.001	1.20 ± 0.005	<MDL
Total P g kg^{-1}	2.5 ± 0.3	0.23 ± 0.006	6.8 ± 0.058	4.5 ± 2.1
Total Ca + Mg (g kg^{-1})	12 ± 4.2	$1.178 \pm 0.1^§$	$40.8 \pm 2.3^§$	0.13 ± 0.02
TCs (mM)	<MDL†	<MDL	<MDL	<MDL

Nd; Not determined
[a] EC = Electrical conductivity
[b] CEC = Cation exchange capacity
[c] OM = Organic matter
†= Below Method detection limit
[§] Datta and Sarkar, 2005

TTC/OTC concentrations and Al-WTR rates tested (Figure 5-2 and 5-3). After 0.25 year

of equilibration time, the effect of Al-WTR amendment on soil and manure amended

soils, TTC and OTC immobilization became significant ($p < 0.001$), with increase in

extent of immobilization compared to the unamended control at both the Al-WTR

application rates and TTC/OTC concentrations tested (Figure 5-1). Further with increase

in the equilibration time from 0.25 to 0.5 year, the effect of Al-WTR amended soil

became pronounced on TTC and OTC immobilization. The trend remained unchanged

until 1 year of equilibration period. At the end of 1 year equilibration period the TTC

immobilization rate increased by 58-64 % and 72-79% in Immokalee soil, 63-69% and

79-84% in manure amended Immokalee soils applied with 25 and 50 g kg^{-1} Al-WTR,

respectively compared to unamended control at both the TTC concentrations tested

(Figure 5-2). Similar immobilization behavior was seen for OTC. However, the rate of

OTC immobilization by Al-WTR was slightly lower compared to TTC. After 1 year of

equilibration period, the OTC immobilization rate increased by 54-60 % and 69-74% in

Immokalee soil, 60-66% and 75-80% in manure amended Immokalee soils applied with

25 and 50 g kg^{-1} Al-WTR, respectively compared to unamended control at OTC

concentrations tested (Figure 5-3). Immokalee is a sandy soil with very low organic

matter (8.40 ± 0.2 g kg^{-1}); whereas, manure has very high organic matter (250.5 ± 2.5 kg^{-1}) (Table 5-1). The relatively high rate of TTC and OTC immobilization seen in the

manure amended Immokalee soil compared to spiked Immokalee soil can be explained

by contribution of high organic matter from the manure in the soil, resulting in change of

soil properties via increase in organic matter and thereby enhancing the binding of TTC

and OTC. Similar sorption behavior was observed by Wang and Yates (2008) in TCs

kinetics and degradation study in a sandy loam soil amended with fresh animal manure.

They attributed the observed behavior to the higher moisture content in the manure and

introduction of organic content from manure in to the soil. Further, Wang et al. (2006)

observed similar behavior in kinetics and degradation study of sulfadimethoxine in

manure amended soils. There was no significant effect ($p > 0.05$) of initial TTC and OTC

concentrations observed on the rate of immobilization by Al-WTR in Immokalee and

manure amended Immokalee soil. However, Al-WTR application rate had a significant

positive effect ($p < 0.001$) on the rate of TTC and OTC immobilization in both the

TTC/OTC concentrations tested and Immokalee and manure amended Immokalee soil.

The observed results are in agreement with the previous short time incubation study

(chapter 4) conducted with Immokalee and TTC/OTC rich manure amended Immokalee

soil in absence of plants with application of Al-WTR with different initial concentrations

of TTC and OTC.

5.2.3 Effect of Al-WTR in Belleglade and manure amended Belleglade soil

Belleglade and manure amended Belleglade soil control without Al-WTR

application showed a significant difference ($p < 0.005$) in TTC and OTC immobilization

compared to Immokalee and manure amended Immokalee soil control. This observed

behavior can be explained by the high SOM (85%) and higher concentrations of Fe, Al,

165
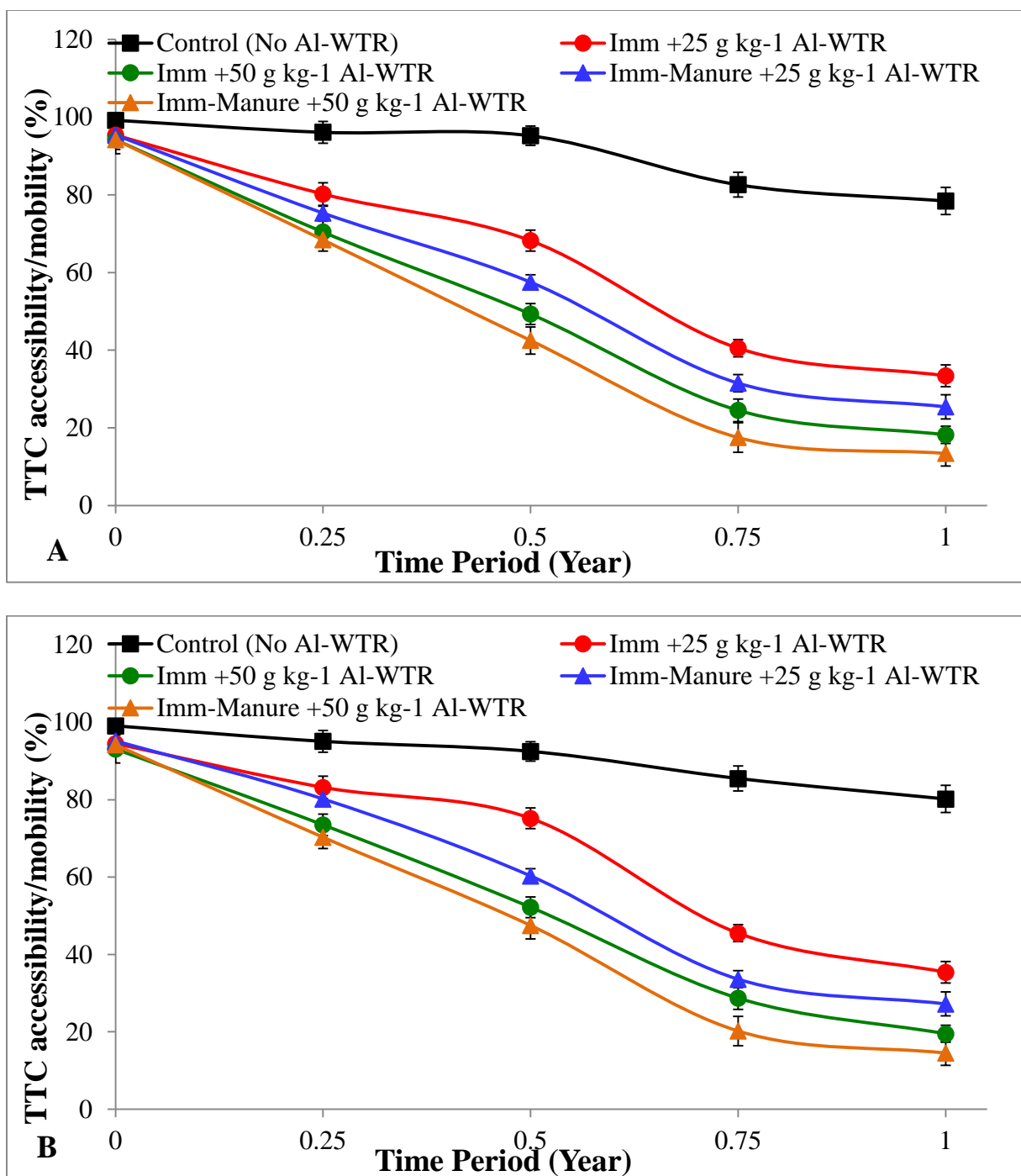
Figure 5-2: Effect of Al-WTR application rates (0, 25, and 50 g kg^{-1}) on immobilization of TTC at two different initial concentrations, 2.25 (A) and 22.5 (B) mg kg^{-1}, respectively as a function of equilibration time in Immokalee soil and Immokalee soil amended with TTC rich manure. Immokalee soil spiked with TTC without Al-WTR amendment was used as a control. Data are expressed as mean of two replicates ± one standard deviation.
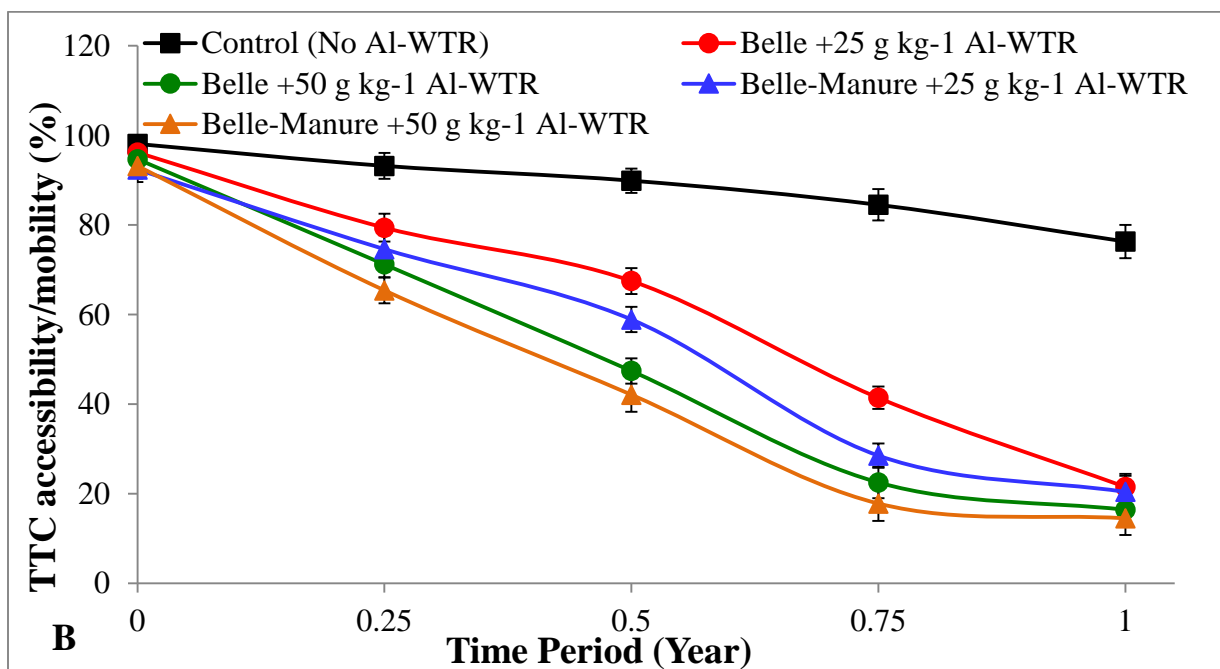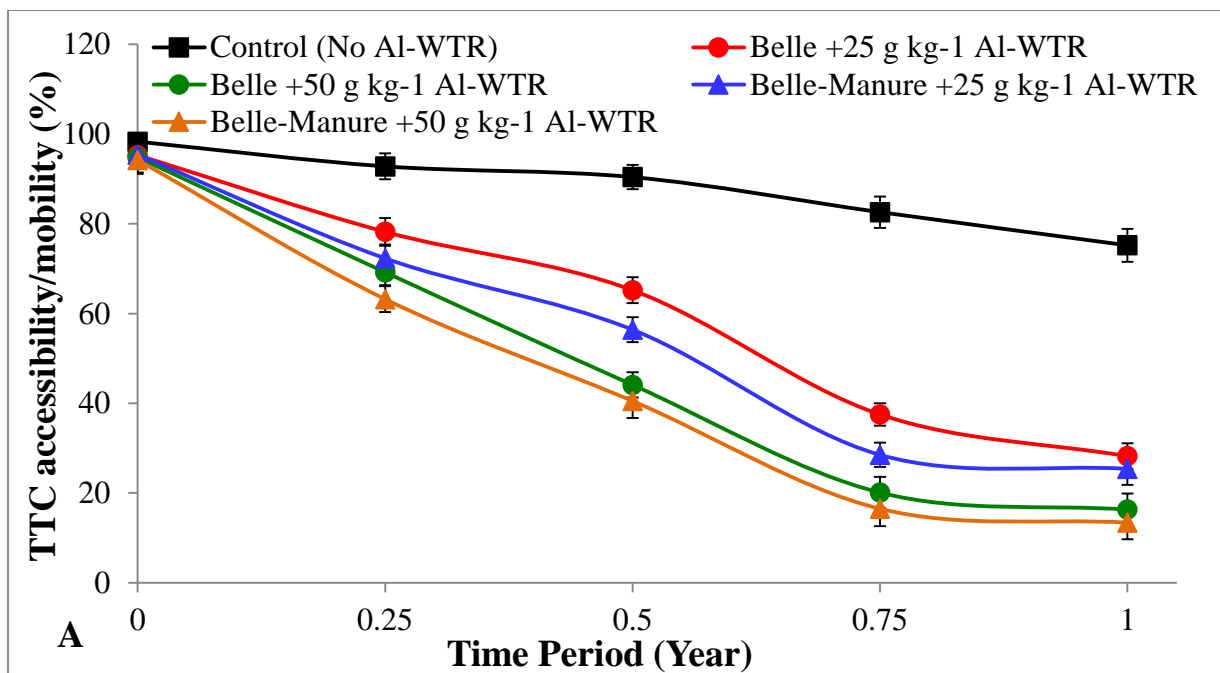
Figure 5-3: Effect of Al-WTR application rates (0, 25, and 50 g kg^{-1}) on immobilization of TTC at two different initial concentrations, 2.25 (A) and 22.5 (B) mg kg^{-1}, respectively as a function of equilibration time in Belleglade soil and Belleglade soil amended with TTC rich manure. Belleglade soil spiked with TTC without Al-WTR amendment was used as a control. Data are expressed as mean of two replicates ± one standard deviation.

Ca, and Mg compared to Immokalee soil indicating higher TCs retention potential (Gu and Karthikeyan, 2005; Figueroa and MacKay, 2005). However, similar to Immokalee soil treatments no significant difference ($p > 0.11$) in Al-WTR amended Belleglade soil and unamended control was seen at time zero (Figure 5-4 and 5-5). At the end of 1 year equilibration period, the TTC immobilization rate increased by 67-71 % and 75-81% in Belleglade soil, 72-75% and 82-87% in manure amended Belleglade soils applied with 25 and 50 g kg^{-1} Al-WTR, respectively compared to unamended control at both the TTC concentrations tested (Figure 5-4). In case of OTC the immobilization behavior was similar to TTC. However, as seen in Immokalee soil, the rate of OTC immobilization by Al-WTR was slightly lower compared to TTC. After 1 year of equilibration period, the OTC immobilization rate increased by 64-69 % and 72-76% in Belleglade soil, 70-74% and 80-85% in manure amended Belleglade soils applied with 25 and 50 g kg^{-1} Al-WTR, respectively compared to unamended control at OTC concentrations tested (Figure 5-5). As compared to Immokalee soil treatments the difference between rate of TTC and OTC immobilization in manure amended to unamended Belleglade soil was relatively low. This is mainly due to the physico-chemical properties of Belleglade. Further, application of manure would not significantly change the soil properties in terms of organic matter content, total P, and total Ca+Mg, which may contribute to increase in the rate of immobilization. There was no significant effect ($p > 0.05$) of initial TTC and OTC concentrations observed on the rate of immobilization by Al-WTR in Belleglade and manure amended Belleglade soil. However, as observed in case of Immokalee soils, Al-
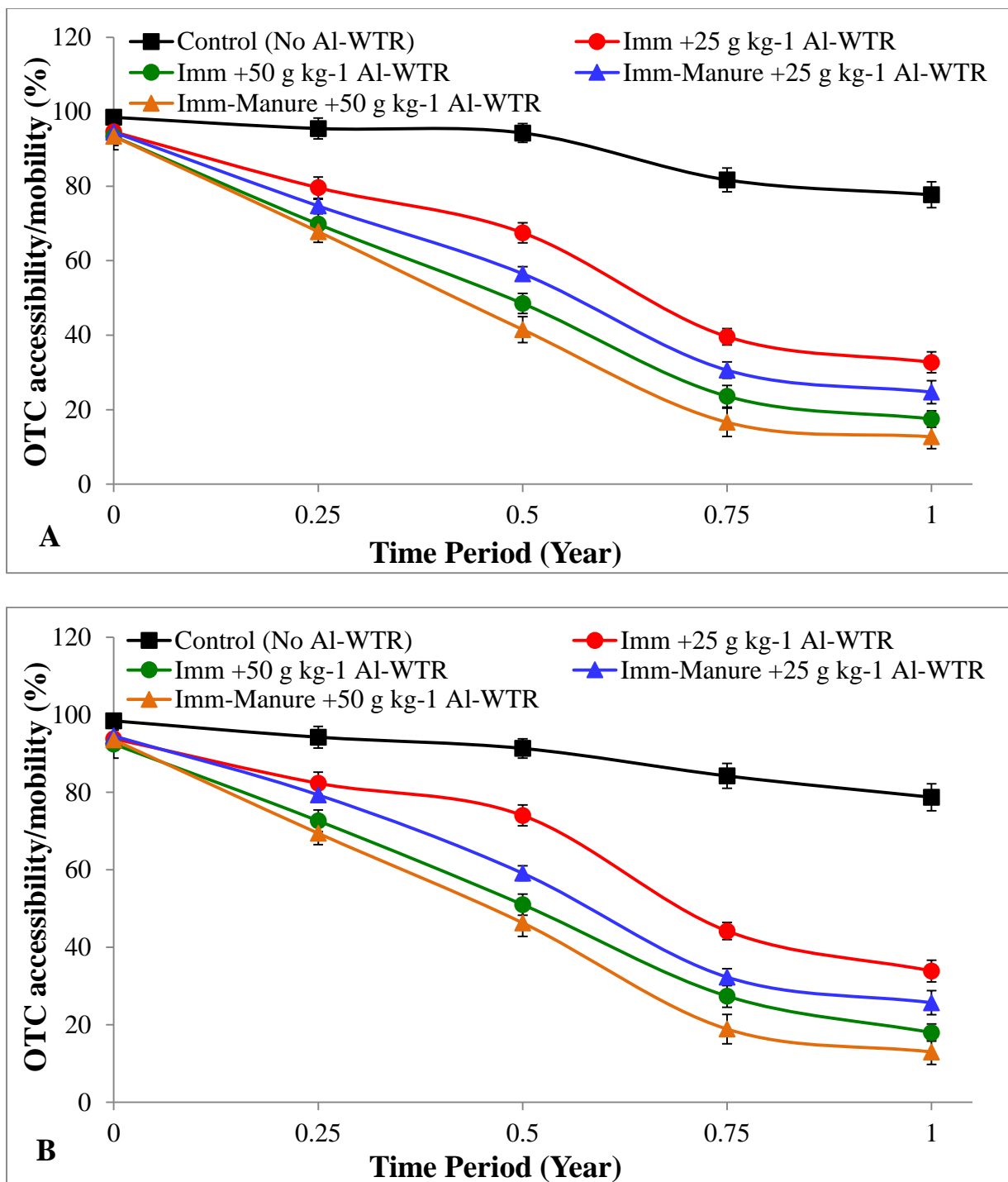
Figure 5-4: Effect of Al-WTR application rates (0, 25, and 50 g kg^{-1}) on immobilization of OTC at two different initial concentrations, 2.25 (A) and 22.5 (B) mg kg^{-1} respectively as a function of equilibration time in Immokalee soil and Immokalee soil amended with OTC rich manure. Immokalee soil spiked with OTC without Al-WTR amendment was used as a control. Data are expressed as mean of two replicates ± one standard deviation.
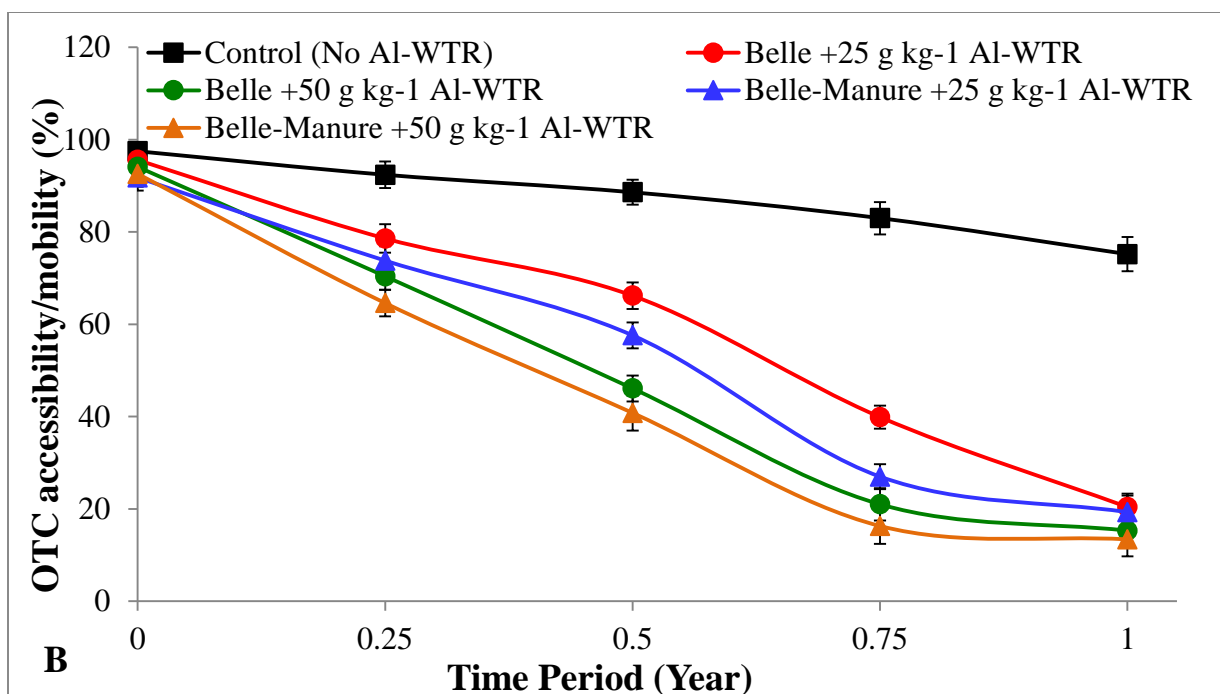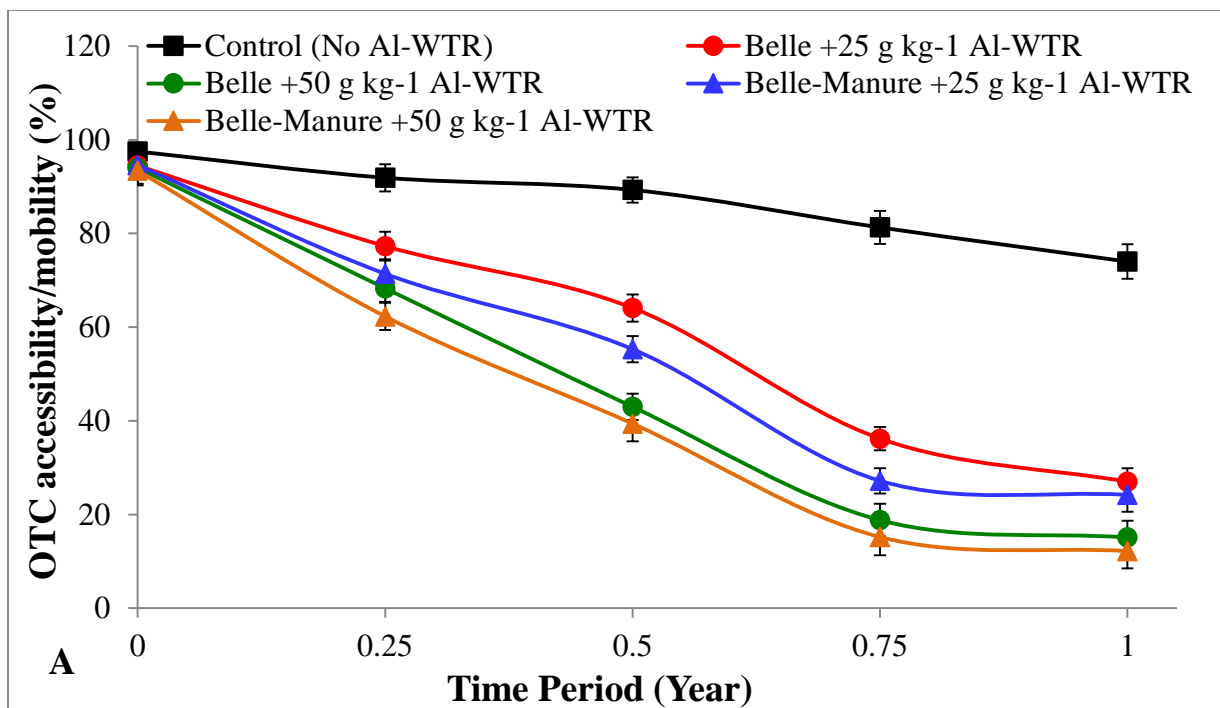
Figure 5-5: Effect of Al-WTR application rates (0, 25, and 50 g kg^{-1}) on immobilization of OTC at two different initial concentrations, 2.25 (A) and 22.5 (B) mg kg^{-1} respectively as a function of equilibration time in Belleglade soil and Belleglade soil amended with OTC rich manure. Belleglade soil spiked with OTC without Al-WTR amendment was used as a control. Data are expressed as mean of two replicates ± one standard deviation.

WTR application rate had a significant positive ($p < 0.001$) effect on the rate of TTC and OTC immobilization in both the TTC/OTC concentrations tested in manure amended and unamended Belleglade soil. The observed results are in agreement with the previous short time incubation study (chapter 4) conducted with Belleglade and TTC/OTC rich manure amended Belleglade soil in the absence of plants with application of Al-WTR with different initial concentrations of TTC and OTC. The effect of Al-WTR in immobilization and stabilization of TTC and OTC in Belleglade soil unamended and amended with manure was significantly ($p > 0.05$) higher compared to Immokalee soil unamended and amended with manure (due to their physico-chemical properties; Table 5-1). Studies have demonstrated that physico-chemical properties such as pH, CEC, clay content, (Kulshreshta et al., 2004; Sassman and Lee, 2005; Ter Laak et al., 2006; Bao et al., 2010), Al and Fe hydrous oxide (Figueroa et al., 2004; Figueroa and MacKay, 2005; Gu and Karthikeyan 2005), soil organic matter (Bao et al., 2009), humic materials (Gu et al., 2007; Pils and Laird, 2007;) presence of competing ligands and complexing metals (Jia et al ., 2009; Wang et al., 2010; Zhang et al., 2011; Zhao et al., 2012) etc. of manure, soil, and sorbent may highly influence the fate and transport of TCs in the soil system.

5.2.4 TTC/OTC in leachates and plants: Effect of Al-WTR

The effect of different Al-WTR application rates (0, 25, and 50 g kg^{-1}) on leaching of TTC (Figure 5-6A and 5-7A) and OTC (Figure 5-6B and 5-7B) was investigated as a function of initial TTC and OTC concentrations (2.25 and 22.5 mg kg^{-1}) after 0.25 and 0.5 year of equilibration time in Immokalee and Belleglade soils
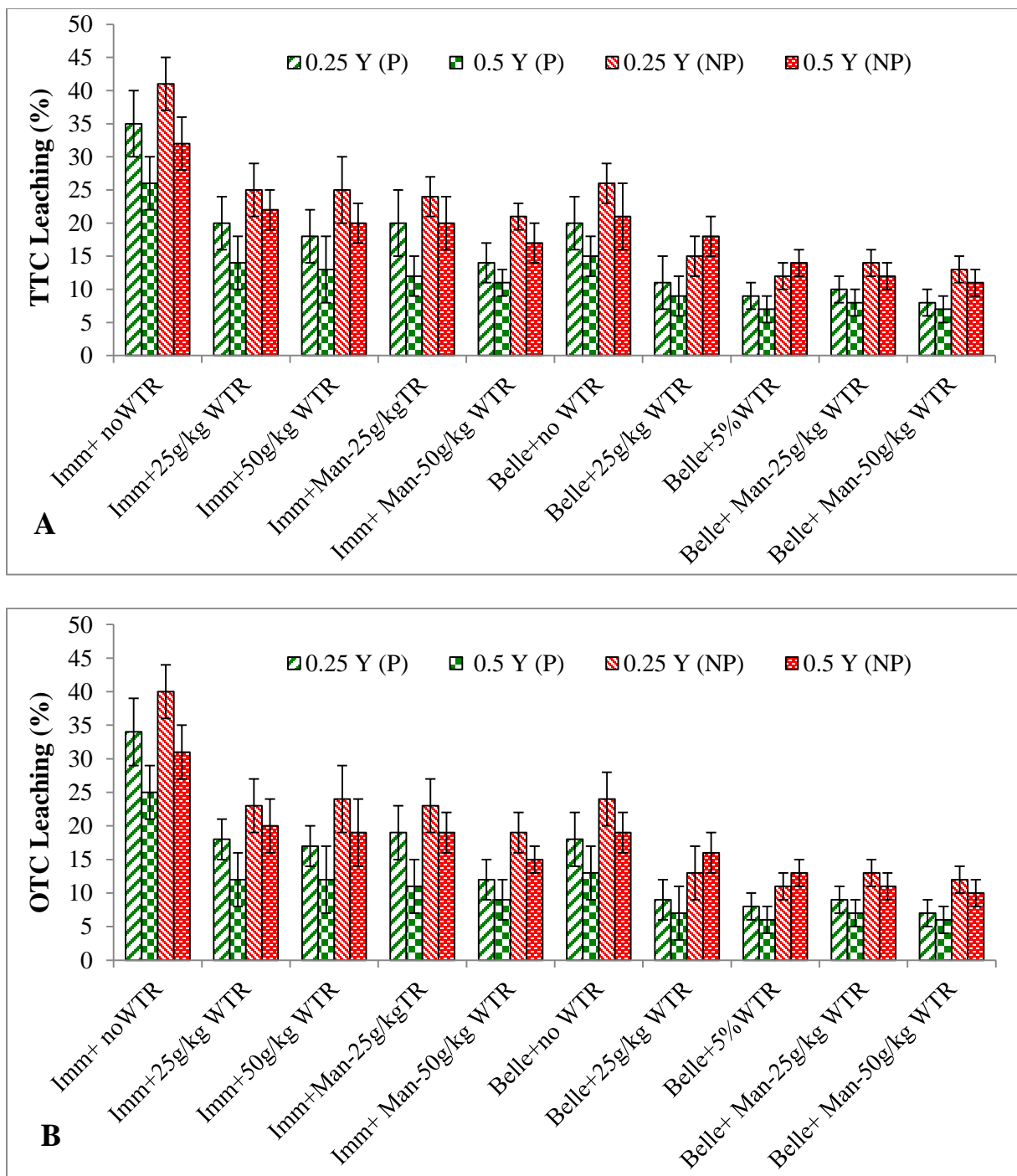
Figure 5-6: Effect of Al-WTR application rates (0, 25, and 50 g kg^{-1}) on leaching of TTC (A) and OTC (B) as a function of initial concentration (2.25 mg kg^{-1}) after 0.25 and 0.5 year of equilibration in Immokalee and Belleglade soils and soils amended with TTC/OTC rich manure in presence (P) and absence (NP) of plants. Soils spiked with TTC/OTC without Al-WTR amendment were used as control. Data are expressed as mean of two replicates ± one standard deviation.
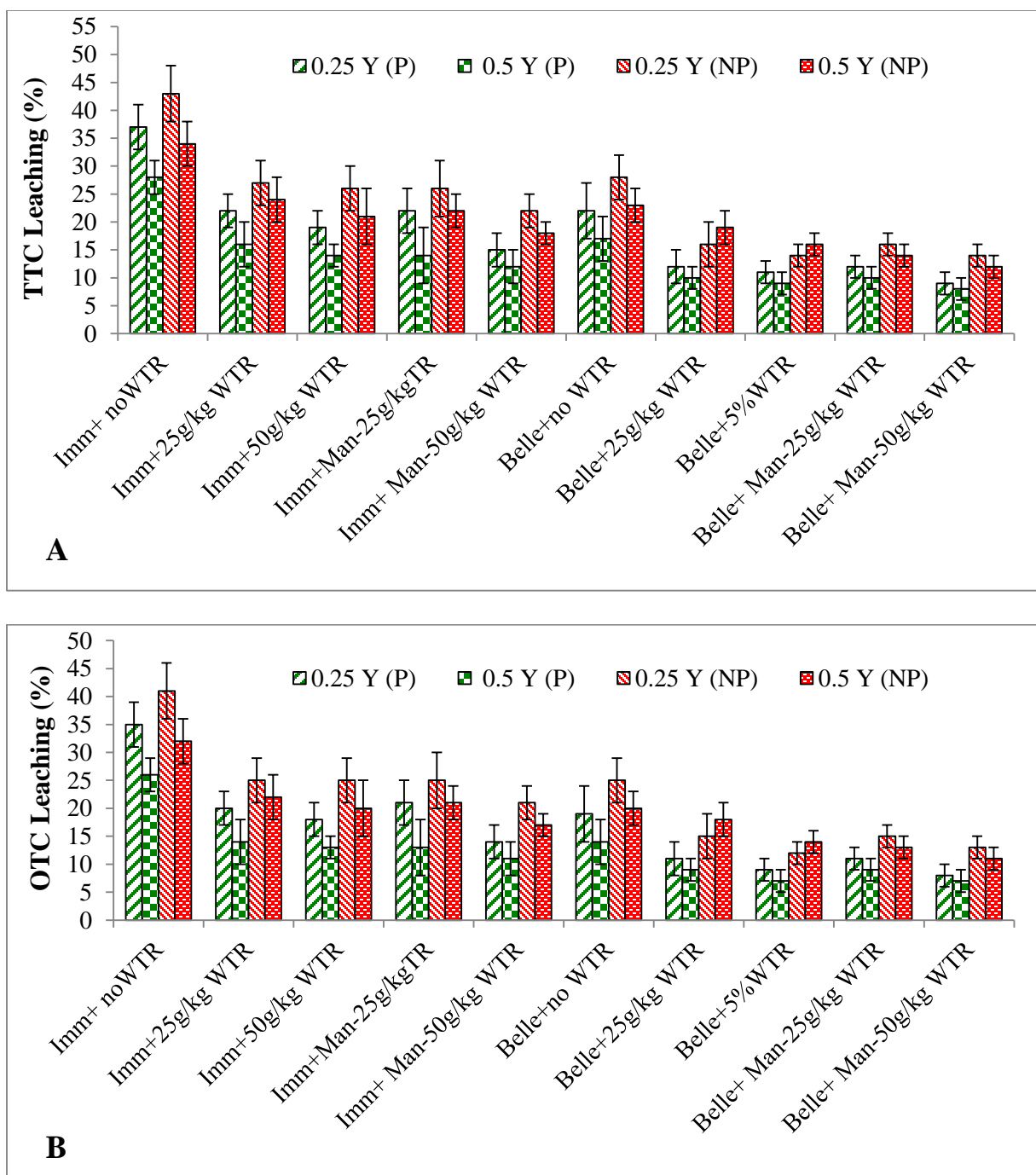
Figure 5-7: Effect of Al-WTR application rates (0, 25, and 50 g kg^{-1}) on leaching of TTC (A) and OTC (B) as a function of initial concentration (22.5 mg kg^{-1}) after 0.25 and 0.5 year of equilibration in Immokalee and Belleglade soils and soils amended with TTC/OTC rich manure in presence (P) and absence (NP) of plants. Soils spiked with TTC/OTC without Al-WTR amendment were used as control. Data are expressed as mean of two replicates ± one standard deviation.

unamended and amended with TTC/OTC rich manure in presence and absence of

Bermuda grass (control plant) and corn (test crop). Soils amended with manure and

spiked with TTC/OTC without Al-WTR amendment were used as control. In the

unamended Immokalee soil (no Al-WTR), the

highest amount of leaching was observed; due to sandy nature, low organic matter, and

very low content of Fe, Al, Ca, and Mg. In presence and absence of plant cover 25-32%

and 38-42% , TTC and OTC leached from Immokalee soil after equilibration period of

0.25 and 0.5 year. However, in unamended Belleglade soil relatively low amount of

leaching was observed; 15-20% and 20-25% in presence and absence of plants after

equilibration period of 0.25 and 0.5 year. Application of Al-WTR in Immokalee and

Belleglade soils significantly ($p < 0.01$) decreased the downward movement of TTC and

OTC via leachate, by immobilization and stabilization. Similar trend was observed for

manure amended Immokalee and Belleglade soil; However, the amount of leaching was

lower than soils unamended with manure; due to increase in the organic matter content

and increase in the TTC and OTC binding extent in manure amended soils (Wang and

Yates, 2008).

Further, the effect of Al-WTR was investigated on uptake of TTC and OTC by corn (test

crop). After harvesting (maturity) corn kernel and leaves were analyzed for TTC/OTC. In

soils amended with Al-WTR there was no uptake of TTC/OTC in corn kernel and leaves

samples (Table 5-2 and 5-3). Also, the uptake of TTC/OTC in control soils (no Al-WTR)

was minimal (< 0.1 % of the initially added TTC/OTC concentration). Kumar et al.

(2005b) studied the uptake of chlortetracycline in cabbage, corn, and green onion from

manure-amended soil with antibiotic ranging from 25 to 125 mg kg^{-1} in manure. The

study reported uptake of chlortetracycline was between 0.002 and 0.017 mg kg^{-1} fresh

tissue weight. Other studies have also reported minimal uptake of VAs in plant tissue in

manure-amended soils (Boxall et al., 2006; Dolliver et al., 2008). However, the uptake is

compound-plant specific, dependent on plant physiology and growth stage, based on soil

properties, and environmental conditions.   Furthermore, LC/MS/MS analyses were

performed of one sample each for TTC and OTC to confirm if there is any transformation

or degradation of TTC and OTC in plant tissue. No known daughter compounds or

metabolites of TTC and OTC were observed in the corn kernel and leaves extract. The

percent recoveries (calculated from the sum total of TTC/OTC in the soils, lost in the

leachate, and uptake in the plants) of TTC (Table 5-2) and OTC (Table 5-3) in the current

greenhouse study ranged between 85 to 91% for Immokalee soil and 86 to 92% for

Belleglade soil (Table 5-2 and 5-3). The residual 12-18% of TTC and OTC can be

attributed to loss by photodegradation, removal by microbes, and unextractable fraction

in manure, soils, and Al-WTR.

175

A

B
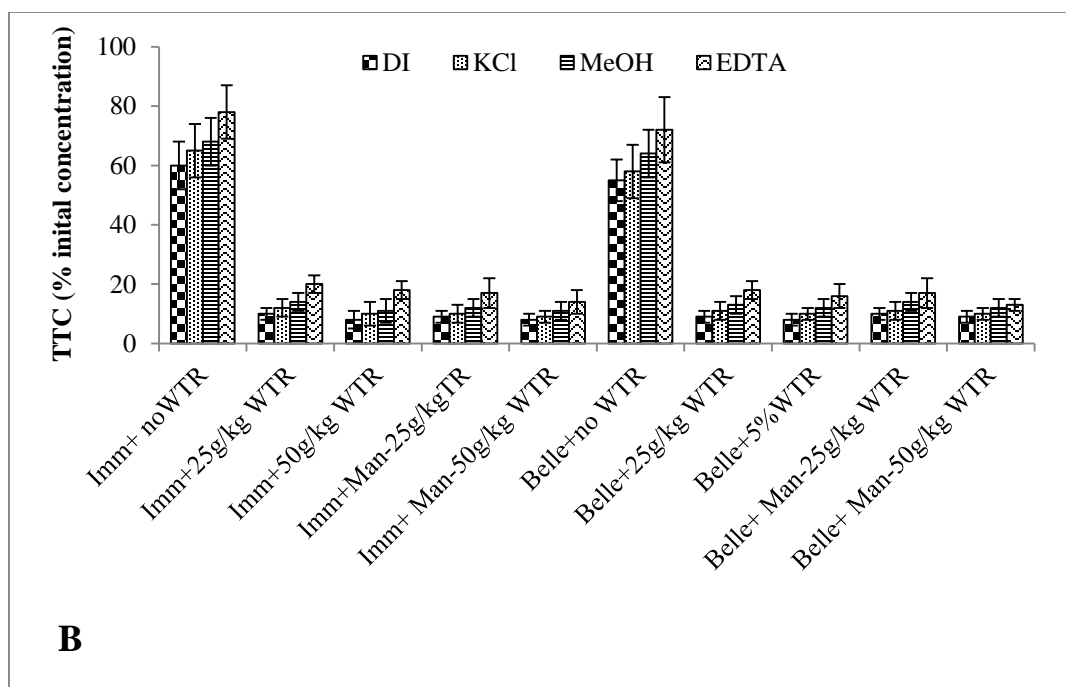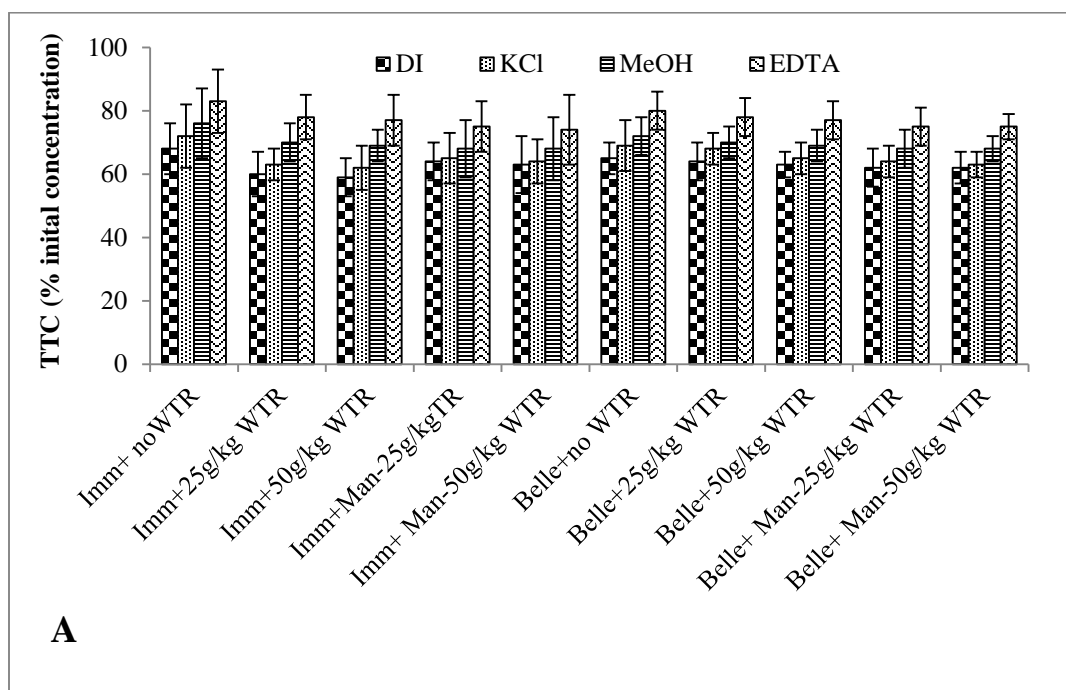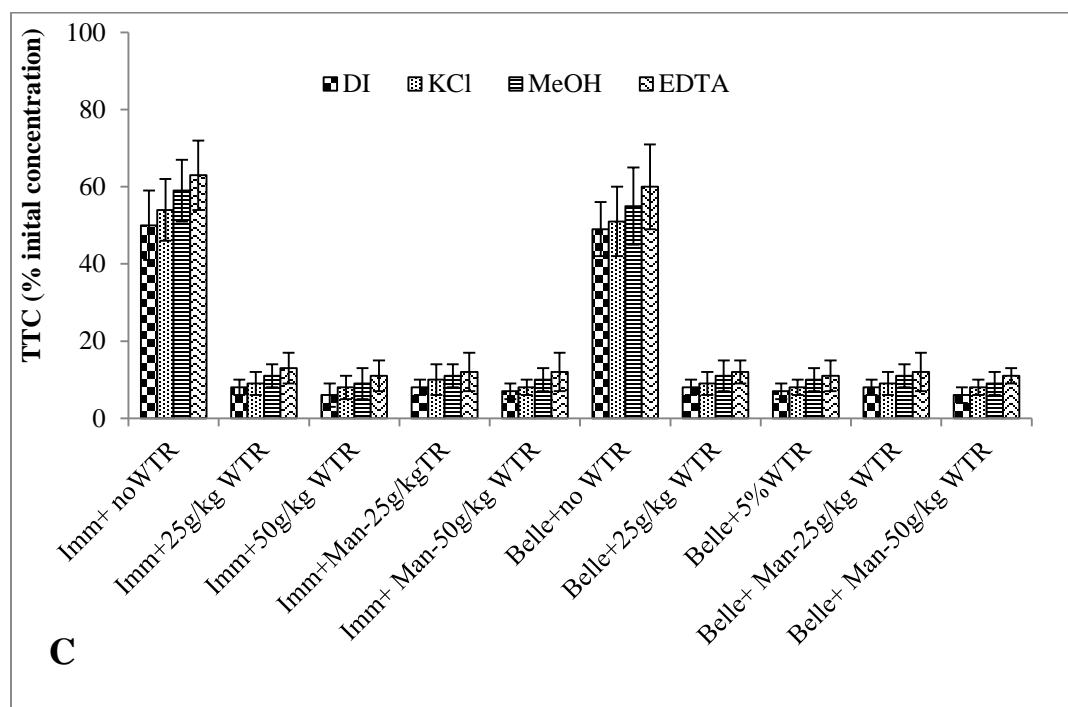
Figure 5-8: Different extractions (DI water, 1 mol L^{-1} KCl, methanol (MeOH), and 0.25 mol L^{-1} EDTA) conducted on Immokalee and Belleglade soils and soils amended with TTC rich manure amended with Al-WTR (0, 25, and 50 g kg^{-1}) with initial concentration (22.5 mg kg^{-1}) after 0 (A) 0.5 (B) and 1 (C) year of equilibration. Soils spiked with TTC without Al-WTR amendment were used as control. Data are expressed as mean of two replicates ± one standard deviation.

A

B
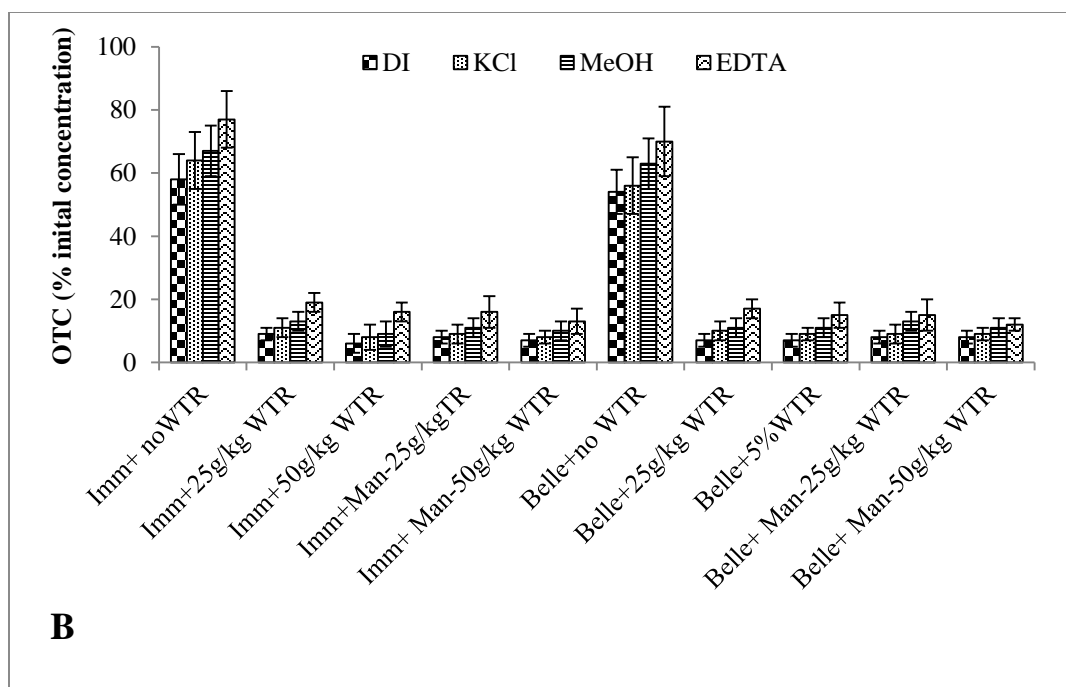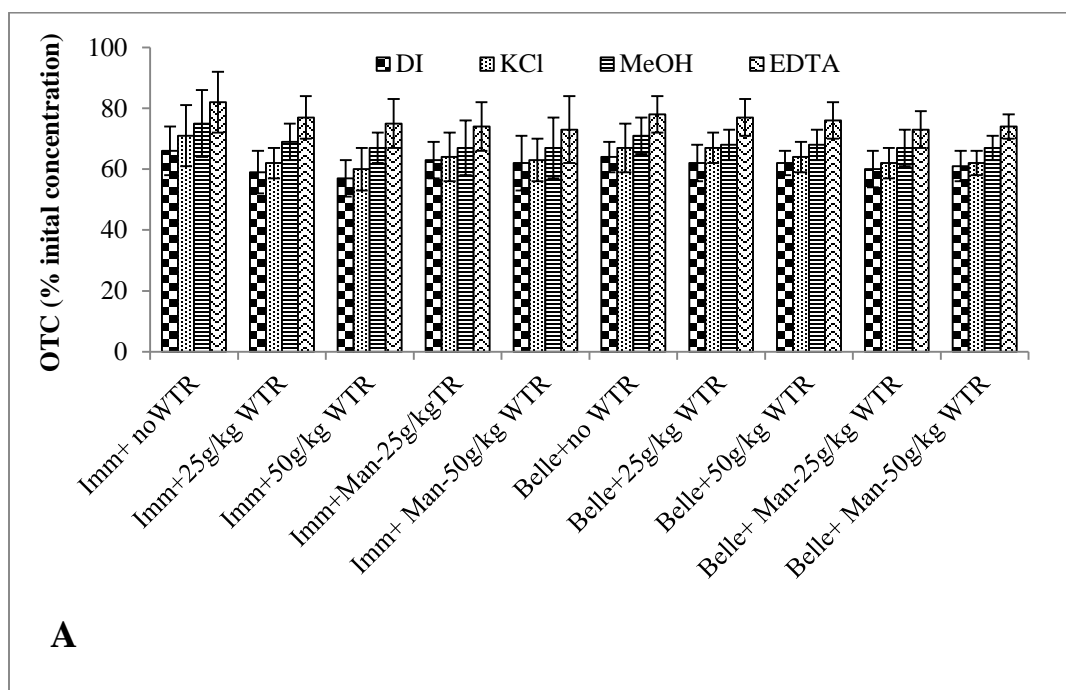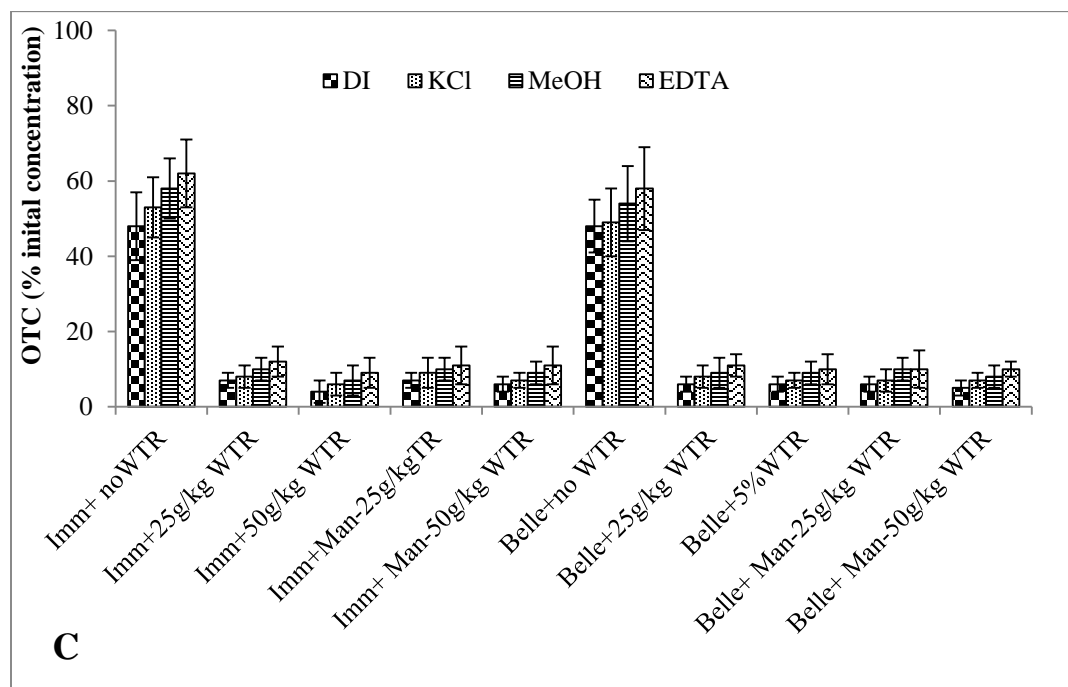
Figure 5-9: Different extractions (DI water, 1 mol L^{-1} KCl, methanol (MeOH), and 0.25 mol L^{-1} EDTA) conducted on Immokalee and Belleglade soils and soils amended with OTC rich manure amended with Al-WTR (0, 25, and 50 g kg^{-1}) with initial concentration (22.5 mg kg^{-1}) after 0 (A) 0.5 (B) and 1 (C) year of equilibration. Soils spiked with OTC without Al-WTR amendment were used as control. Data are expressed as mean of two replicates ± one standard deviation.

5.2.5 Extraction/Treatments: Effect of Al-WTR

Experiments were performed to determine the potential release of TTC and OTC from Immokalee and Belleglade soil amended and unamended with manure and treated with different Al-WTR rates at high initial TTC (Figure 5-8) and OTC concentration (Figure 5-9) as a function equilibration time. Four different treatments namely; DI water, 1 mol L^{-1} KCl, methanol, and 0.25 mol L^{-1} EDTA were used. DI water extraction was used to determine soluble-water extractable phase, methanol treatment was used to check the role of hydrophobicity, KCl was used to see the effect of competing ion, and EDTA was used as a chelating agent to evaluate competitively displaced bound TTC and OTC from Soil-Manure-Al-WTR mixture at time zero, 0.5, and 1 year equilibration time. At time zero, no significant difference ($p > 0.05$) was observed in the amount of TTC and OTC released/extracted between the Al-WTR applied Immokalee and Belleglade soil amended and unamended with manure compared to control (no Al-WTR application) in all the treatments tested (Figure 5-8A and 5-9A). The DI water extraction ranged between 64-75% and 52-60 % of the initial TTC and OTC concentration (22.5 mg kg^{-1}) for Immokalee and Belleglade soil amended and unamended with manure, respectively. The soluble form likely represents the TTC and OTC fraction that can be lost through runoffs and leaching or is available for plants to uptake. In case of unamended Immokalee soil (no manure and Al-WTR) nearly 75% of the TTC/OTC was in soluble form, which was expected due to the sandy nature and low Fe, Al, Ca, and Mg content of the Immokalee soil. The application of manure and Al-WTR in Immokalee soil at time zero did not exert any significant effect on lowering the TTC and OTC soluble fraction. With the treatment

of 1 mol L^{-1} KCl and methanol, the extraction was in the range of 58-73% and 60-75%, respectively. As expected, highest extraction was attained via treatment of EDTA, 75-83% of initially added TTC/OTC in Immokalee soil amended with manure and applied with AL-WTR (Figure 5-7A and 5-8A). However, the addition of Al-WTR at both the rates tested significantly decreased ($p < 0.01$) extraction by all treatments at 0.5 and 1 year of equilibration time for TTC (Figure 5-8B and 5-8C) and OTC (Figure 5-9B and 5-9C) indicating immobilization of TCs. After 1 year of equilibration time, KCl, methanol, and EDTA extraction reduced to 8-10%, 11-13%, 14-16%, and 17-20% of initial TTC (Figure 5-8C) and OTC (Figure 5-9C) concentration, respectively; whereas, control unamended with Al-WTR extraction were 48-55% (DI water), 54-57% (KCl), 59-62% (Methanol), and 63-72% (EDTA). In case of unamended Belleglade soil (no manure and Al-WTR) 65-69% of the total TTC/OTC was in soluble form, which was relatively lower than Immokalee soil (Figure 5-8A and 5-9A). The application of manure and Al-WTR in Belleglade soil at time zero did not exert any significant effect ($p > 0.05$) on lowering the TTC and OTC soluble fraction as seen for Immokalee soil treatments. The extractions of TTC and OTC by KCl, methanol, and EDTA at time zero were 59-70% and 62-72% and 72-78%, respectively.

Table 5-2: Mass balance of TTC in Immokalee and Belleglade soils, and soil amended with manure at high initial TTC concentration (22.5 mg kg^{-1}) rate. The percent recoveries are calculated from TTC remaining in the soils, manure amended soils, TTC in leachates, and TTC accumulation in plants.

Soil Type	Soil Treatment	Time Zero		Time-Final (1 Year)				
		Initial TTC –theoretical (mg)	Initial TTC – experimental (mg)	TTC remaining in the soil[a] (mg)	TTC in the plant (mg)[b]	TTC in the leachates (mg)	Sum (mg)	Percent recovery
Immokalee	No Al-WTR	90	91.4 ± 1.1	26.5 ± 2.8	0.4 ± 0.1	51.5 ± 3.1	76.5 ±6.0	85 ± 6.6
	25 g kg^{-1} Al-WTR	90	91.5 ± 1.2	50.1 ± 4.7	<MDL†	31.2 ± 3.3	81.3 ± 8	90.3 ± 7.2
	50 g kg^{-1} Al-WTR	90	92.3 ± 1.5	58.3 ± 3.5	<MDL†	24.4 ± 2.4	82.7 ± 5.9	91.8 ± 6.5
Immokalee + Manure	25 g kg^{-1} Al-WTR	90	92.5 ± 1.6	52.3 ± 2.9	<MDL†	27.5 ± 1.7	79.8 ± 5.9	88.6 ± 6.6
	50 g kg^{-1} Al-WTR	90	91.5 ± 1.3	62.9 ± 1.8	<MDL†	18.2 ± 1.4	81.1± 3.2	90.1± 3.5
Belleglade	No Al-WTR	90	91.7 ± 1.2	43.5 ± 3.8	0.34 ± 0.08	34.2 ± 2.6	77.7 ± 6.4	86.3 ± 7.1
	25 g kg^{-1} Al-WTR	90	90.9 ± 1.1	58.3 ± 2.9	<MDL†	23.4 ± 3.3	81.7 ± 6.2	90.7 ± 6.8
	50 g kg^{-1} Al-WTR	90	91.3 ± 1.2	64.3 ± 3.7	<MDL†	14.4 ± 2.1	79.1 ± 5.8	87.4 ± 6.4
Belleglade + Manure	25 g kg^{-1} Al-WTR	90	91.5 ± 1.3	59.4 ± 3.2	<MDL†	22.5 ± 1.8	81.9 ± 5	91 ± 5.5
	50 g kg^{-1} Al-WTR	90	91.6 ± 1.2	65.9 ± 1.9	<MDL†	14.2 ± 1.6	80.1± 3.5	89 ± 3.5

[a] Citric acid, oxalic acid, and methanol/water mixture extraction (Wang and Yates, 2008)

[b] Plant samples were extracted using citric acid and methanol (Boxall et al., 2006), † Below Method detection limit

Table 5-3: Mass balance of OTC in Immokalee and Belleglade soils, and soil amended with manure at high initial OTC concentration (22.5 mg kg^{-1}) rate. The percent recoveries are calculated from OTC remaining in the soils, manure amended soils, OTC in leachates, and OTC accumulation in plants.

Soil Type	Soil Treatment	Time Zero		Time-Final (1 Year)				
		Initial OTC theoretical (mg)	Initial OTC experimental (mg)	OTC remaining in the soil[a] (mg)	OTC in the plant (mg)[b]	OTC in the leachates (mg)	Sum (mg)	Percent recovery
Immokalee	No Al-WTR	90	91.5 ± 0.9	26.8 ± 2.5	0.51 ± 0.1	50.6 ± 2.7	77.6 ± 5.3	86.2 ± 5.8
	25 g kg^{-1} Al-WTR	90	91.8 ± 1.1	51.3 ± 3.2	<MDL†	30.5 ± 2.4	81.8 ± 8	90.8 ± 6.2
	50 g kg^{-1} Al-WTR	90	91.3 ± 1.2	58.9 ± 2.8	<MDL†	22.3 ± 2.5	81.2 ± 5.4	90.2 ± 4.8
Immokalee + Manure	25 g kg^{-1} Al-WTR	90	91.5 ± 1.2	53.4 ± 2.4	<MDL†	25.9 ± 1.8	79.3 ± 5.9	88.1 ± 3.8
	50 g kg^{-1} Al-WTR	90	91.5 ± 1.3	63.4 ± 1.9	<MDL†	17.9 ± 1.5	81.3± 3.2	90.3 ± 3.5
Belleglade	No Al-WTR	90	91.4 ± 1.4	46.7 ± 3.6	0.38 ± 0.02	33.6 ± 2.5	80.7 ± 6.5	89.6 ± 7.2
	25 g kg^{-1} Al-WTR	90	90.8 ± 0.9	59.6 ± 3.1	<MDL†	22.7 ± 3.1	82.3 ± 6.2	91.4 ± 6.8
	50 g kg^{-1} Al-WTR	90	91.2 ± 1.2	65.4 ± 3.2	<MDL†	13.9 ± 2.2	79.3 ± 5.4	88.1 ± 6
Belleglade + Manure	25 g kg^{-1} Al-WTR	90	91.3 ± 1.1	60.5 ± 3.2	<MDL†	21.4 ± 2	81.9 ± 5	91 ± 5.5
	50 g kg^{-1} Al-WTR	90	91 ± 1.2	66.8 ± 1.9	<MDL†	14.1 ± 1.5	80.9± 3.4	88.9 ± 3.7

[a] Citric acid, oxalic acid, and methanol/water mixture extraction (Wang and Yates, 2008)
[b] Plant samples extracted using citric acid and methanol (Boxall et al., 2006), † Below Method detection limit

However, extraction of TTC and OTC in manure amended and unamended Belleglade soil by all the four treatment tested significantly reduced ($p < 0.05$) in Al-WTR applied Belleglade soil compared to the control unamended (no Al-WTR) soil after 0.5 and 1 year of equilibration time. After 1 year of equilibration time, KCl, methanol, and EDTA extraction reduced to 7-9%, 9-11%, 12-14%, and 16-19% of initial TTC (Figure 5-8C) and OTC (Figure 5-9C) concentration, respectively; whereas, control unamended with Al-WTR extraction were 42-45% (DI water), 49-55% (KCl), 52-58% (Methanol), and 63-66% (EDTA). Al-WTR application rates did not have a significant effect ($p > 0.1$) in decreasing the release of TTC and OTC by different extractions tested in both Immokalee and Belleglade soils manure amended and unamended. Al-WTR was highly effective in decreasing the release of TTC and OTC from Immokalee and Belleglade soils manure amended and unamended and thereby increasing the immobilization and stabilization under dynamic system with soil, water, manure, and plants.

5.2.6 TCLP analysis of soils and manure applied soils amended with Al-WTR

The TCLP of soils and soils applied manure amended with different rates of Al-WTR in presence of plants were determined using USEPA SW-846 Method 1311 to ensure that the toxicity characteristics concentration of several metals and metalloids were below the EPA threshold limit for their safe use and land application on arable land as well as there is no toxic effects on plants. The TCLP values (Table 5-4) for all the metals and metalloids tested were well below the hazardous waste toxicity characteristic criterion as defined in Title 40 of the Code of Federal Regulations (CFR), Part 261.24 and below the USEPA threshold values of biosolids, thus, permitting safe land

Table 5-4: Toxicity characteristics values of several metals and metalloids measured for soils and soils applied with manure amended with Al-WTR at different rates using the EPA 1311 TCLP extraction method. The results are expressed as mg L^{-1}.

Sample ID	Al*	Cr	Fe*	Ni	Cu	Zn	As	Ag	Cd	Pb
Immokalee control	0.65	0.01	0.18	0.01	0.06	0.53	0.01	0.01	0.01	>MDL
Imm + 2.5% Al-WTR	14.63	0.05	0.75	0.01	0.11	1.16	0.01	0.01	0.01	>MDL
Imm + 5% Al-WTR	24.30	0.10	0.80	0.01	0.11	0.78	0.01	0.01	0.01	>MDL
Belleglade control	1.11	0.01	1.63	0.02	0.01	0.25	0.02	0.01	0.02	>MDL
Belle + 2.5% Al-WTR	1.59	0.01	1.87	0.02	0.01	0.28	0.01	0.01	0.02	>MDL
Belle + 5% Al-WTR	1.85	0.01	1.95	0.02	0.01	0.05	0.01	0.01	0.02	>MDL
Imm + Manure control	0.15	0.01	0.19	0.01	0.01	0.02	>MDL†	>MDL	>MDL	>MDL
Imm + Manure+ 2.5% Al-WTR	26.15	0.03	0.22	0.01	0.01	0.02	>MDL	>MDL	>MDL	>MDL
Imm + Manure+ 5% Al-WTR	32.35	0.04	0.16	0.01	0.01	0.01	>MDL	>MDL	>MDL	>MDL
Belle + Manure control	1.45	0.04	1.25	0.01	0.01	0.07	>MDL	>MDL	>MDL	>MDL
Belle + Manure+ 2.5% Al-WTR	1.70	0.01	1.89	0.01	0.01	0.01	0.01	0.01	>MDL	>MDL
Belle + Manure+ 5% Al-WTR	2.07	0.01	1.95	0.01	0.01	0.01	0.01	0.01	>MDL	>MDL

†<MDL † (Below Method Detection Limit for As, Ag, Cd, and Pb 10 mg L^{-1} using ICP-MS)
*NR: Not regulated under the Toxicity Characteristics Leaching Protocol by USEPA

Table 5-5: Dry plant biomass of corn plants (without corn cob and kernel/seeds) harvested after full maturation period grown in Immokalee and Belleglade soils applied with manure and amended with different rates of Al-WTR spiked with high TTC/OTC (22.5 mg kg^{-1}) concentration. Data are expressed as mean of three replicates (n=3) with one standard deviation.

Sample ID	TTC	OTC
Treatment with presence corn plants	------g----	
Immokalee Control*	189.4 ± 13.6	179.1 ± 15.2
Immokalee + 2.5% Al-WTR	192.8 ± 17.1	182 ± 17.8
Immokalee + 5% Al-WTR	179 ± 15.4	194 ± 13.1
Belleglade Control	242.5 ± 18.9	258 ± 22.4
Belleglade + 2.5% Al-WTR	235.6 ± 21.5	243.2 ± 21.5
Belleglade + 5% Al-WTR	228.1 ± 20.3	245.9 ± 11.2
Immokalee + Manure Control	212.5 ± 20.5	208 ± 21.5
Immokalee + Manure + 2.5% Al-WTR	202.5 ± 19.2	219 ± 15.9
Immokalee + Manure + 5% Al-WTR	207.8 ± 24.2	218.5 ± 16.2
Belleglade + Manure Control	268.5 ± 15.4	259 ± 25.9
Belleglade + Manure + 2.5% Al-WTR	255.2 ± 23.2	253.6 ± 27.4
Belleglade + Manure + 5% Al-WTR	268.1 ± 28.1	255.9 ± 13.2

*Control indicates absence of Al-WTR application

†Manure applied at recommended agricultural application rates

application of Al-WTR. Al and Fe does not fall under USEPA hazardous waste criteria

therefore the limits of Al and Fe were compared to the solid industrial waste. The values

for Al and Fe were also well below the threshold values of safe solid industrial waste

disposal. One of the issues associated with Al-WTR land application may be the Al-

toxicity to plants and restriction of root growth. The plant dry biomass of corn plants

(without corn cob and kernel/seeds) harvested after full maturation period grown in

Immokalee and Belleglade soils applied with manure and amended with different rates of

Al-WTR spiked with high TTC/OTC (22.5 mg kg^{-1}) concentration were recorded (Table

5-5). The data from plant dry biomass shows there was no significant ($p > 0.2$) effect of

Al-WTR application rates on the plant biomass compared to control (no Al-WTR

application). Several long term field and greenhouse studies have shown similar results,

where no Al toxicity, no reduced yields nor increased plant Al phytoavailability have

been observed on plants (bahiagrass (*Paspalum notatum* Fluggae), ryegrass (*Lolium

perenne* L.), etc.) at similar application rates use in the current study (Silveria et al.,2013;

Oladeji et al., 2009). In addition, several states use WTRs as soil amendments or as a soil

conditioner and to prevent excess P from leaching into surface water bodies. The benefits

associated with use of WTRs as a soil amendment include improved soil structure (El-

Swaify and Emerson, 1975), increased moisture-holding capacity (Bugbee and Frink,

1985), and increased availability of nutrients for various plants (Heil and Barbarick,

1989). Further, several studies have reported field application of the WTRs for agronomic

and ecological benefits (e.g., Jacobs and Teppen, 2000; Agyin-Birikorang et al., 2007).

The mean plant biomass (without corn cob and kernels/seeds) values ranged from 179 to

268 g for TTC/OTC treatment. The plant biomass was higher for Belleglade soil when compared to Immokalee soil as expected based on the physico-chemical properties of both the soils. Also, application of manure to the soils increased the mean plant biomass.

5.3 Conclusions

Considerable efforts have been made to understand the fate and transport of veterinary antibiotics in the soil and water environment and very few attempts to develop novel remediation strategy to overcome the problem. To our knowledge, the present study is one of the very few attempts where an industrial waste by-product in form of Al-WTR has been evaluated for its effectiveness to immobilize TCs in a dynamic system with soil, water, plants, and manure in a one year greenhouse column study. Previous studies in batch sorption and incubation settings demonstrated that Al-WTR was an effective sorbent for TTC and OTC. The current study further documented the effectiveness of Al-WTR to immobilize and stabilize TTC and OTC in soils and manure amended soils in a dynamic greenhouse column setting for one year, thereby reducing the mobility of TTC and OTC and thus potentially decreasing the risk associated with TCs getting in surface runoff, leachate, and been available for plants to uptake and get to higher levels in the food chain. Further, long-term (≥ 3 years) simulated field based studies under a dynamic system with repetitive application of TCs rich manure (due to persistent nature of TCs), cyclic crop rotation, and uncontrolled natural condition are required to validate the current findings of Al-WTR in immobilizing and stabilizing TCs in soil system.

5.4 References

- Aga, D. S., O'Connor, S., Ensley, S., Payero, J. O., Snow, D., Tarkalson, D., 2005. Determination of the persistence of tetracycline antibiotics and their degradates in manure-amended soil using enzyme-linked immunosorbent assay and liquid chromatography-mass spectrometry. J. Agric. Food Chem., 53(18), 7165-7171.
- Agyin-Birikorang, S., O'Connor, G., Jacobs, L., Makris, K., Brinton, S., 2007. Long-term phosphorus immobilization by a drinking water treatment residual. Journal of Environmental Quality 36, 316-323.
- Andra, S. S. P., 2008. Phytoremediation of lead contaminated soils. Dissertation: Thesis (Ph. D.)--University of Texas at San Antonio.
- Bao, Y., Zhou, Q., Wan, Y., Yu, Q., Xie, X., 2010. Effects of soil/solution ratios and cation types on adsorption and desorption of tetracycline in soils. Soil Science Society of America Journal 74(5), 1553-1561.
- Bao, Y., Zhou, Q., Wang, Y., 2009. Adsorption characteristics of tetracycline by two soils: assessing role of soil organic matter. Soil Research 47(3), 286-295.
- Bassil, R. J., Bashour, I. I., Sleiman F. T., Abou-Jawdeh, Y. A., 2013.Antibiotic uptake by plants from manure-amended soils, J Environ Sci Health B., 48 (7), 570-574
- Ben-Dor E., Banin. A., 1989. Determination of organic matter content in arid-zone soils using a simple "loss-on-ignition" method. Commun. Soil Sci. Plant Anal. 20:1675–1695
- Boxall, A.B.A., Johnson, P., Smith, E.J., Sinclair,C.J., Stutt, E., Levy, L., 2006. Uptake of veterinary medicines from soils into plants J. Agr. Food Chem., 54, 2288–2297
- Boxall, A.B.A., Kolpin, D.W., Halling-Sørensen, B., Tolls, J., 2003. Peer Reviewed: Are Veterinary Medicines Causing Environmental Risks? Environmental Science & Technology 37, 286A-294A.
- Bugbee, G.J., Frink, C.R., 1985. Alum sludge as soil amendment: Effects on soil properties and plant growth. New Haven Connecticut Agric. Exp. Stn. Bull. 827.
- Chitescu, C, L., Nicolau, A. I., Stolker, A. A., 2013. Uptake of oxytetracycline, sulfamethoxazole and ketoconazole from fertilised soils by plants. Food Addit Contam Part A Chem Anal Control Expo Risk Assess., 30 (6) 1138-46
- Datta, R., Sarkar, D., 2005. Consideration of soil properties in assessment of human health risk from exposure to arsenic-enriched soils Int. Environ. Asses. Manage., 1, 55–59
- Daughton, C., Ternes, T., 1999. Pharmaceuticals and personal care product in environment: agents of ssubtle change? Environ. Health Perspect. 107, 907-937
- Dolliver, H., Gupta, S., and Noll, S., 2008. Antibiotic degradation during manure composting. J. Environ. Qual., 37(3), 1245-1253.

- El-Swaify, S.A., Emerson, W. W., 1975. Changes in the physical properties of soil clays due to precipitated aluminum and iron hydroxides: I. Swelling and aggregate stability after drying. Soil Sci. Soc. Am. J., 39, 1056-1063.
- Figueroa, R.A., Leonard, A., MacKay, A.A., 2004. Modeling Tetracycline Antibiotic Sorption to Clays. Environmental Science & Technology 38, 476-483.
- Figueroa, R.A., MacKay, A.A., 2005. Sorption of Oxytetracycline to Iron Oxides and Iron Oxide-Rich Soils. Environmental Science & Technology 39, 6664-6671.
- Fritz, J.W., Zuo, Y., 2007. Simultaneous determination of tetracycline, oxytetracycline, and 4-epitetracycline in milk by high-performance liquid chromatography. Food Chemistry 105, 1297-1301.
- Gee, G. W., Bauder, J. W., Klute, A., 1986. Particle-size analysis. Methods of soil analysis. Part 1. Physical and mineralogical methods, 383-412.
- Gu, C., Karthikeyan, K., 2005. Interaction of tetracycline with aluminum and iron hydrous oxides. Environmental Science & Technology 39, 2660-2667.
- Hamscher, G., Pawelzick, H.T., Hoper, H., Nau, H., 2005. Different behaviour of tetracyclines and sulfonamides in sandy soils after repeated fertilisation with liquid manure, Environ. Toxicol. Chem. 24, 861–868.
- Hamscher, G., Sczesny, S., Höper, H., Nau, H., 2002. Determination of persistent tetracycline residues in soil fertilised with liquid manure by high-performance liquid chromatography with electrospray ionisation tandem mass spectrometry, Anal. Chem.74, 1509–1518.
- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997a. Soil pH (1:2v/v). IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL) chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p15.
- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997b. Electrical Conductivity. IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL) chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p24.
- Heil, D.M., Barbarick, K.A., 1989. Water treatment sludge influence on the growth of sorghum-sudangrass. J. Environ. Qual., 18, 292-298.
- Ippolito, J., Barbarick, K., Elliott, H., 2011. Drinking Water Treatment Residuals: A Review of Recent Uses. Journal of Environmental Quality 40, 1-12.
- Jacobs, L.W., Teppen, B.J., 2000. WTR as a soil amendment to reduce nonpoint source pollution from phosphorus-enriched soils. 9 p. *In* Proc. 14th Annual Residuals and Biosolids Management Conference, 27–29 Feb. 2000, MA. CD-ROM, Water Environment Federation, Alexandria, VA.
- Jia, D.A., Zhou, D.M., Wang, Y.J., Zhu, H.W., and Chen, J.L. 2008. Adsorption and cosorption of Cu(II) and tetracycline on two soils with different characteristics. Geoderma 146 (1–2), 224–230.
- Kay, P., Blackwell, P. A., Boxall, A. B., 2004. A. Fate of veterinary antibiotics in a macroporous tile drained clay soil Environ. Toxicol. Chem. 23, 1136-1144.

- Klute, A., 1986. Methods of soil analysis. Part 1. Physical and mineralogical methods. SSSA Publications, Madison, WI.
- Kulshrestha, P., Giese, R.F., Aga, D.S., 2004. Investigating the Molecular Interactions of Oxytetracycline in Clay and Organic Matter: Insights on Factors Affecting Its Mobility in Soil. Environmental Science & Technology 38, 4097-4105.
- Kumar, K., Gupta S. C., Baidoo S. K., Chander, Y., Rosen C. J., 2005b. Antibiotic uptake by plants from soil fertilized with animal manure. J. Environ Qual 34:2082–2085.
- Kumar, K., Gupta, S.C., Chander, Y., Singh. A.K., 2005a. Antibiotic use in agriculture and its impact on the terrestrial environment. Adv. Agron 87, 1–54.
- McClellan, K., Halden, R. U., 2010. Pharmaceuticals and personal care products in archived U.S. biosolids from the 2001 EPA National Sewage Sludge Survey. Water Res. 44 (2), 658−668.
- Munir, M., Xagoraraki, I., 2011. Levels of Antibiotic Resistance Genes in Manure, Biosolids, and Fertilized Soil. J. Environ. Qual. 40 (1) 248-255.
- Oberle, K., Capdeville, M.J., Berthe, T., Budzinski, H., Petit, F., 2012. Evidence for a Complex Relationship between Antibiotics and Antibiotic-Resistant Escherichia Coli: From Medical Center Patients to a Receiving Environment. Environmental Science & Technology 46, 1859-1868.
- Oladeji, O. O., Sartain, B. J., O'Connor, G.A.,2009. Land Application of Aluminum Water Treatment Residual: Aluminum Phytoavailability and Forage Yield. Communications in Soil Science and Plant Analysis, 40, 9-10, 1483-1498.
- Ostermann, A., Siemens, J., Welp, G., Xue, Q., Lin, X., Liu, X., Amelung, W., 2013. Leaching of veterinary antibiotics in calcareous Chinese croplands. Chemosphere, 91 (7) 928-934.
- Pils, J., Laird, D., 2007. Sorption of tetracycline and chlortetracycline on K- and Ca-saturated soil clays, humic substances, and clay-humic complexes. Environmental Science & Technology 41, 1928-1933.
- Punamiya, P., Sarkar, D., Rakshit, S., Datta, R., 2013a. Effectiveness of Al- based drinking water treatment residuals as a novel sorbent to remove tetracyclines from aqueous medium. J. Environ. Qual. *doi:10.2134/jeq2013.03.0082*.
- Punamiya, P., Sarkar, D., Rakshit, S., Datta, R., 2013b. Effect of solution properties, competing ligands, and complexing metal on sorption of tetracyclines on Al-based drinking water treatment residuals, J. Hazard. Mater. (To be submitted).
- Sall, J., Creighton, L., Lehman, A., 2005. JMP start statistics, 8th edition. Cary, NC, SAS Institute.
- Sarmah, A. K., Meyer, M. T.; Boxall, A. B. A., 2006. A global perspective on the use, sale, exposure pathways, occurrence, fate and effects of veterinary antibiotics (VAs) in the environment Chemosphere 65, 725-759.

- Sassman, S. A., Lee, L. S., 2005. Sorption of three tetracyclines by several soils: assessing the role of pH and cation exchange. Environmental science & technology 39(19), 7452-7459.
- Sengelov, G., Agerso, Y., Halling-Sorensen, B., Baloda, S. B., Anderson, J. S., Jensen, L. B., 2003. Bacterial antibiotic resistance levels in Danish farmland as a result of treatment with pig manure slurry Environ. Int 28. 587-595.
- Shenker, M., Harush, D., Ben-Ari, J., Chefetz, B., 2011. Uptake of carbamazepine by cucumber plants - A case study related to irrigation with reclaimed wastewater. Chemosphere 82, 905-910.
- Silveira, M. L., Driscoll, J. L., Silveira, C. P., Graetz, D. A., Sollenberger, L. E., Vendramini J. M. B., 2013.Land Application of Aluminum Water Treatment Residual to Bahiagrass Pastures: Soil and Forage Responses. Agron. J. 105, 3, 796-802.
- Stoob, K., Singer, H., Mueller, S.R., R.P. Schwarzenbach, R.P., C.H. Stamm, C.H., 2007 Dissipation and transport of veterinary sulfonamide antibiotics after manure application to grassland in a small catchment Environ. Sci. Technol 41, 7349–7355
- Tanoue, R., Sato, Y., Motoyama, M., Nakagawa, S., Shinohara, R., Nomiyama, K., 2012. Plant Uptake of Pharmaceutical Chemicals Detected in Recycled Organic Manure and Reclaimed Wastewater. J. Agric. Food Chem. 60, 10203−10211
- Teixido, M., Hurtado, C., Pignatello, J.J., Beltran, J.L., Grenados, M., and Peccia, J., 2013. Predicting contaminant adsorption in black carbon (biochar)-amended soil for the veterinary antimicrobial sulfamethazine. Environ. Sci. Technol. 47, 6197.
- Ter Laak, T.L.,Wouter, A.G., Tolls, J., 2006. The effect of pH and ionic strength on the sorption of sulfachloropyridazine, tylosin, and oxytetracycline to soil. Environ. Toxicol. Chem. 25, 904–911.
- Tolls, J., 2001. Sorption of veterinary pharmaceuticals in soils: a review, Environ. Sci. Technol. 35, pp. 3397–3406.
- Topp, E., Monteiro, S.C., Beck, A.., Ball Coelho, B., Boxall, P.W. Duenk, A.B.A., Kleywegt, S., Lapen, D.R., Payne, M., Sabourin, L., Metcalfe, C.D., 2008. Runoff of pharmaceuticals and personal care products following application of biosolids to an agricultural field Sci. Total Environ., 396, 52–59
- U.S. Department of Agriculture, Economic Research Service (USDA/ERS), 2005. Confined animal and manure nutrient data system. http://www.ers.usda.gov/Data/manure/default
- Van Dijk, J., & Keukens, H. J., 2000. The stability of some veterinary drugs and coccidiostats during composting and storage of laying hen and broiler faeces. In Residues of veterinary drugs in food. Proc. of the Euroresidue IV Conf., Veldhoven, Netherlands (pp. 8-10).
- Wang, Q., Yates, S.R., 2008b. Laboratory study of oxytetracycline degradation kinetics in animal manure and soil. J. Agric. Food Chem. 56, 1683-1688.

- Wang, Y. J., Jia, D. A., Sun, R. J., Zhu, H. W., Zhou, D. M., 2008a. Adsorption and cosorption of tetracycline and copper (II) on montmorillonite as affected by solution pH. Environmental Science and Technology 42(9), 3254-3259.
- Wang, Y. J., Sun,R. J., Xiao, A. Y., Wang, S. Q., and Zhou, D. M., 2010. Phosphate affects the adsorption of tetracycline on two soils with different characteristics. Geoderma, 156, 237-242.
- Watanabe, F. S., Olsen, S. R., 1965. Test of an ascorbic acid method for determining phosphorus in water and $NaHCO_3$ extracts from soil. Soil Science Society of America Journal. 29(6), 677-678.
- Westergaard, K., Muller, A. K., Christensen, S., Bloem, J., Sorensen, S. J., 2001. Effects of tylosin as a disturbance on the soil microbial community Soil Biol. Biochem 33, 2061-2071.
- Winckler, C. and A. Grafe, A., 2001. Use of veterinary drugs in intensive animal production Evidence for persistence of tetracycline in pig slurry. J Soil Sediment 1, 67-70.
- Wu, C., Spongberg, A. L., Witter, J. D., Fang, M., Czajkowski, K. P.,2010 Uptake of Pharmaceutical and Personal Care Products by Soybean Plants from Soils Applied with Biosolids and Irrigated with Contaminated Water. Environ. Sci. Technol. 44 (16), 6157-6161
- Zhang, D., Pan, B., Wu, M., Wang, B., Zhang, H., Peng, H. B., Wu, D., Ning P., 2011. Adsorption of sulfamethoxazole on functionalized carbon nanotubes as affected by cations and anions. Environ. Pollut., 159, 2616–2621
- Zhang, Y., Zhang, C., Parker, D. B., Snow, D. D., Zhou, Z., Li, X., 2013. Occurrence of antimicrobials and antimicrobial resistance genes in beef cattle storage ponds and swine treatment lagoons. Sci Total Environ.
- Zhao, Y., Gu, X., Gao, S., Geng, J., and Wang, X., 2012. Adsorption of tetracycline (TC) onto montmorillonite: Cations and humic acid effects. Geoderma 183-184, 12-18.

REFERENCES

- Aga, D. S. 2008. Fate of pharmaceuticals in the environment and in water treatment systems. CRC Press: Boca Raton, FL.

- Aga, D. S., O'Connor, S., Ensley, S., Payero, J. O., Snow, D., Tarkalson, D., 2005. Determination of the persistence of tetracycline antibiotics and their degradates in manure-amended soil using enzyme-linked immunosorbent assay and liquid chromatography-mass spectrometry. J. Agric. Food Chem., 53(18), 7165-7171.

- Aga, D. S., O'Connor, S., Ensley, S., Payero, J. O., Snow, D., Tarkalson, D., 2005. Determination of the persistence of tetracycline antibiotics and their degradates in manure-amended soil using enzyme-linked immunosorbent assay and liquid chromatography-mass spectrometry. J. Agric. Food Chem., 53(18), 7165-7171.

- Agerso, Y., Wulff, G., Vaclavik, E., Halling-Sorensen, B., Jensen, L., 2006. Effect of tetracycline residues in pig manure slurry on tetracycline-resistant bacteria and resistance gene tet(M) in soil microcosms. Environment International 32, 876-882.

- Agyin-Birikorang, S., O'Connor, G., Jacobs, L., Makris, K., Brinton, S., 2007. Long-term phosphorus immobilization by a drinking water treatment residual. Journal of Environmental Quality 36, 316-323.

- AHI. Animal Health Institute News Release, 2011 AHI: Washington, DC, USA.

- Allen, H.K., Donato, J., Wang, H.H., Cloud-Hansen, K.A., Davies, J., Handelsman, J., 2010. Call of the wild: antibiotic resistance genes in natural environments. Nature Reviews Microbiology 8, 251-259.

- Arikan, O. A., Sikora, L. J., Mulbry, W., Khan, S. U., Foster, G. D., 2007. Composting rapidly reduces levels of extractable oxytetracycline in manure from therapeutically treated beef calves. Bioresource technology, 98(1), 169-176.

- Aristilde, L., Marichal, C., Miehe-Brendle, J., Lanson, B., Charlet, L., 2010. Interactions of Oxytetracycline with a Smectite Clay: A Spectroscopic Study with Molecular Simulations. Environmental Science & Technology 44.

- Baguer, A.J., Jensen, J., Krogh, P.H., 2000. Effects of the antibiotics oxytetracycline and tylosin on soil fauna. Chemosphere 40, 751-757.

- Bao, Y., Zhou, Q., Wan, Y., Yu, Q., Xie, X., 2010. Effects of soil/solution ratios and cation types on adsorption and desorption of tetracycline in soils. Soil Science Society of America Journal 74(5), 1553-1561.

- Bao, Y., Zhou, Q., Wang, Y., 2009. Adsorption characteristics of tetracycline by two soils: assessing role of soil organic matter. Soil Research 47(3), 286-295.

- Bassil, R. J., Bashour, I. I., Sleiman F. T., Abou-Jawdeh, Y. A., 2013.Antibiotic uptake by plants from manure-amended soils, J Environ Sci Health B., 48 (7), 570-574

- Ben-Dor E., Banin. A., 1989. Determination of organic matter content in arid-zone soils using a simple "loss-on-ignition" method. Commun. Soil Sci. Plant Anal. 20:1675–1695

- Blackwell, P.A., Kay, P., Boxall, A.B.A., 2007. The dissipation and transport of veterinary antibiotics in a sandy loam soil. Chemosphere 67, 292-299.

- Boxall, A.B.A., Johnson, P., Smith, E.J., Sinclair,C.J., Stutt, E., Levy, L., 2006. Uptake of veterinary medicines from soils into plants J. Agr. Food Chem., 54, 2288–2297

- Boxall, A.B.A., Kolpin, D.W., Halling-Sørensen, B., Tolls, J., 2003. Peer Reviewed: Are Veterinary Medicines Causing Environmental Risks? Environmental Science & Technology 37, 286A-294A.

- Boyd, S.L., Boyd, R.J., Bossonnette, P.W., Kerdaron, D.I., Aucoin, N.T., 1995. A theorrtical study of the effects of protonation and deprotonation on bond dissociation energies. J. Am. Chem. Soc. 117, 8816-8822.

- Bugbee, G.J., Frink, C.R., 1985. Alum sludge as soil amendment: Effects on soil properties and plant growth. New Haven Connecticut Agric. Exp. Stn. Bull. 827.

- Bui, T. X., Choi, H., 2010. Influence of ionic strength, anions, cations, and natural organic matter on the adsorption of pharmaceuticals to silica. Chemosphere, 80, 681-686.

- Caporale, A. G., Punamiya, P., Pigna, M., Violante, A., Sarkar, D., 2013. Effect of particle size of drinking-water treatment residuals on the sorption of arsenic in the presence of competing ions. J. Hazard Mater.260, 644-651

- Chander, Y., Kumar, K., Goyal, S. M., & Gupta, S. C., 2005. Antibacterial activity of soil-bound antibiotics. J. Environ. Qual., 34(6), 1952-1957.

- Chee-Sanford, J. C., Aminov, R. I., Krapac, I. J., Garrigues-Jeanjean, N., Mackie, R. I., 2007. Occurrence and diversity of tetracycline resistance genes in lagoons and groundwater underlying two swine production facilities Appl. Environ. Microbiol 67, 1494-1502.

- Chen, W.-R., Huang, C.-H., 2010. Adsorption and transformation of tetracycline antibiotics with aluminum oxide. Chemosphere 79, 779-785.

- Chitescu, C, L., Nicolau, A. I., Stolker, A. A., 2013. Uptake of oxytetracycline, sulfamethoxazole and ketoconazole from fertilised soils by plants. Food Addit Contam Part A Chem Anal Control Expo Risk Assess., 30 (6) 1138-46

- Datta, R., Das, P., Smith, S., Punamiya, P., Ramanathan, D.M., Reddy, R., Sarkar, D., 2013. Phytoremediation potential of vetiver grass [Chrysopogon zizanioides (L.)] for tetracycline. *Internatl.* J. Phytorem. 15:343-351

- Datta, R., Sarkar, D., 2005. Consideration of soil properties in assessment of human health risk from exposure to arsenic-enriched soils Int. Environ. Asses. Manage., 1, 55–59

- Daughton, C., Ternes, T., 1999. Pharmaceuticals and personal care products in the environment: Agents of subtle change? Environmental Health Perspectives 107, 907-938.

- Dayton, E.A., Basta, N.T., 2005. A method for determining the phosphorus sorption capacity and amorphous aluminum of aluminum-based water treatment residuals. J. Environ. Qual.

- De Liguoro, M., Cibin, V., Capolongo, F., Halling-Sørensen, B., Montesissa, C., 2003. Use of oxytetracycline and tylosin in intensive calf farming: evaluation of transfer to manure and soil. Chemosphere 52, 203-212.

- Dolliver, H., Gupta, S., and Noll, S., 2008. Antibiotic degradation during manure composting. J. Environ. Qual., 37(3), 1245-1253.

- Dos Santos, H.F., Xavier, E.S., Zerner, M.C., De Almeida, W.B., 2000. Spectroscopic investigation of the Al(III)-anhydrotetracycline complexation process. Journal of Molecular Structure-Theochem 527, 193-202.

- Elliott, H.A., Dempsey, B.A., 1991. Agronomic effects of land application of water treatment sludges. J. AWWA, 83 126–131.

- Elliott, H.A., O'Connor, G.A., Lu, P., Brinton, S., 2002. Influence of water treatment residuals on phosphorus solubility and leaching. Journal of Environmental Quality 31, 1362-1369.

- El-Swaify, S.A., Emerson, W. W., 1975. Changes in the physical properties of soil clays due to precipitated aluminum and iron hydroxides: I. Swelling and aggregate stability after drying. Soil Sci. Soc. Am. J., 39, 1056-1063.

- Essington, M. E., 2004. Soil and Water Chemistry; CRC Press: Boca Raton, FL.

- Evanko, C.R., Dzombak. D.A., 1999. Surface Complexation Modeling of Organic Acid Sorption to Goethite. J. Colloid Interf. Sci. 214:189-206.

- Figueroa, R.A., Leonard, A., MacKay, A.A., 2004. Modeling Tetracycline Antibiotic Sorption to Clays. Environmental Science & Technology 38, 476-483.

- Figueroa, R.A., MacKay, A.A., 2005. Sorption of Oxytetracycline to Iron Oxides and Iron Oxide-Rich Soils. Environmental Science & Technology 39, 6664-6671.

- Fritz, J.W., Zuo, Y., 2007. Simultaneous determination of tetracycline, oxytetracycline, and 4-epitetracycline in milk by high-performance liquid chromatography. Food Chemistry 105, 1297-1301.

- Gee, G. W., Bauder, J. W., Klute, A., 1986. Particle-size analysis. Methods of soil analysis. Part 1. Physical and mineralogical methods, 383-412.

- Goldberg, S., Johnston. C., 2001. Mechanisms of arsenic adsorption on amorphous oxides evaluated using macroscopic measurements, vibrational spectroscopy, and surface complexation modeling. J. Colloid Interf. Sci. 234:204-216.

- Gonzalez-Pleiter, M., Gonzalo, S., Rodea-Palomares, I., Leganes, F., Rosal, R., Boltes, K., Marco, E., Fernandez-Pinas, F., 2013. Toxicity of five antibiotics and their mixtures towards photosynthetic aquatic organisms: Implications for environmental risk assessment. Water Research 47, 2050-2064.

- Gu, C., Karthikeyan, K., 2005. Interaction of tetracycline with aluminum and iron hydrous oxides. Environmental Science & Technology 39, 2660-2667.

- Hakk, H., Millner, P, Larsen, G., 2005. Decrease in water-soluble 17β-estradiol and testosterone in composted poultry manure with time J. Environ. Qual. 34, 943-950.

- Halling-Sørensen, B., Nielsen, S.N., Lanzky, P.F., Ingerslev, F., Lutzhoft, H.C.H., Jorgensen. S.E., 1998. Occurrence, fate and effects of pharmaceutical substances in the environment − a review. Chemosphere **36,** 357–393.

- Hamscher, G., Pawelzick, H.T., Hoper, H., Nau, H., 2005. Different behaviour of tetracyclines and sulfonamides in sandy soils after repeated fertilisation with liquid manure, Environ. Toxicol. Chem. 24, 861–868.

- Hamscher, G., Sczesny, S., Höper, H., Nau, H., 2002. Determination of persistent tetracycline residues in soil fertilised with liquid manure by high-performance liquid chromatography with electrospray ionisation tandem mass spectrometry, Anal. Chem.74, 1509–1518.

- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997a. Soil pH (1:2v/v). IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL) chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p15.

- Hanlon, E.A.,. Gonzalez, J.S., Bartos, J.M., 1997b. Electrical Conductivity. IFAS extension soil testing laboratory (ESTL) and analytical research laboratory (ARL) chemical procedures and training manual. Fl. Coop. Ext. Ser. Cir. 812. Univ. of Florida, Gainesville, p24.

- Heil, D.M., Barbarick, K.A., 1989. Water treatment sludge influence on the growth of sorghum-sudangrass. J. Environ. Qual., 18, 292-298.

- Herbelin, A.L., Westall, J.C., 1999. FITEQL 4.0: A computer program for determination of chemical equilibrium constants from experimental data. Department of Chemistry, Oregon State University: Corvallis, Oregon.

- Hovsepyan, A., Bonzongo, J.C.J., 2009. Aluminum drinking water treatment residuals (Al-WTRs) as sorbent for mercury: Implications for soil remediation. J. Hazard Mater. 164:73-80.

- Ippolito, J., Barbarick, K., Elliott, H., 2011. Drinking Water Treatment Residuals: A Review of Recent Uses. Journal of Environmental Quality 40, 1-12.

- Ippolito, J.A., Scheckel, K.G., Barbarick, K.A., 2009. Selenium adsorption to aluminum-based water treatment residuals. Journal of Colloid and Interface Science 338, 48-55.

- Jacobs, L.W., Teppen, B.J., 2000. WTR as a soil amendment to reduce nonpoint source pollution from phosphorus-enriched soils. 9 p. *In* Proc. 14th Annual Residuals and Biosolids Management Conference, 27–29 Feb. 2000, MA. CD-ROM, Water Environment Federation, Alexandria, VA.

- Janos, P., Sypecká, J., Mlckovská, P., Kurán, P., Pilarová, V., 2007. Removal of metal ions from aqueous solutions by sorption onto untreated low-rank coal (oxihumolite). Separation and Purification Technology 53, 322-329.

- Ji, L., Chen, W., Duan, L., Zhu, D., 2009. Mechanisms for strong adsorption of tetracycline to carbon nanotubes: A comparative study using activated carbon and graphite as adsorbents. Environmental Science & Technology 43, 2322-2327.

- Kang, J., Liu, H.J., Zheng, Y.M., Qu, J.H., Chen, J.P., 2011. Application of nuclear magnetic resonance spectroscopy, Fourier transform infrared spectroscopy, UV-Visible spectroscopy and kinetic modeling for elucidation of adsorption chemistry in uptake of tetracycline by zeolite beta. Journal of Colloid and Interface Science 354, 261-267.

- Kay, P., Blackwell, P. A., Boxall, A. B., 2004. A. Fate of veterinary antibiotics in a macroporous tile drained clay soil Environ. Toxicol. Chem. 23, 1136-1144.

- Kay, P., Blackwell, P., Boxall, A., 2004. Fate of veterinary antibiotics in a macroporous tile drained clay soil. Environmental Toxicology and Chemistry 23, 1136-1144.

- Kemper, N. 2008. Veterinary antibiotics in the aquatic and terrestrial environment.Ecol. Indicators 8:1-13.

- Klute, A., 1986. Methods of soil analysis. Part 1. Physical and mineralogical methods. SSSA Publications, Madison, WI.

- Kolpin, D., Furlong, E., Meyer, M., Thurman, E., Zaugg, S., Barber, L., Buxton, H., 2002. Pharmaceuticals, hormones, and other organic wastewater contaminants in US streams, 1999-2000: A national reconnaissance. Environmental Science & Technology 36, 1202-1211.

- Kulshrestha, P., Giese, R.F., Aga, D.S., 2004. Investigating the Molecular Interactions of Oxytetracycline in Clay and Organic Matter: Insights on Factors Affecting Its Mobility in Soil. Environmental Science & Technology 38, 4097-4105.

- Kumar, K., Gupta S. C., Baidoo S. K., Chander, Y., Rosen C. J., 2005b. Antibiotic uptake by plants from soil fertilized with animal manure. J. Environ Qual 34:2082–2085.

- Kumar, K., Gupta, S.C., Chander, Y., Singh. A.K., 2005a. Antibiotic use in agriculture and its impact on the terrestrial environment. Adv. Agron 87, 1–54.

- Kümmerer, K. 2009. Antibiotics in the aquatic environment – A review – Part I.Chemosphere 75:417-434.

- Levy, S. B., 1998. The challenge of antibiotic resistance. Sci. Am., 278:46-53.

- Levy, S. B., 2002. The Antibiotic Paradox; Perseus Publishing: Cambridge,MA.

- Loeppert, R.H., Inskeep, W.P., 1996. Iron. In: Sparks, D.L., Page, A.L., Helmke, P.A., Loeppert, R.H., Soltanpour, P.N., Tabatabai, M.A., Johnston, C.T., Summer, M.E. (Eds.), Method of Soil Analysis Part 3 Chemical methods, SSSA Book Series, pp. 639–664.

- Loffler, D., Rombke, J., Meller, M., Ternes, T.A., 2005. Environmental fate of pharmaceuticals in water/sediment systems. Environmental Science & Technology 39, 5209-5218.

- Makris, K., Harris, W., O'Connor, G., Obreza, T., 2004. Phosphorus immobilization in micropores of drinking-water treatment residuals: Implications for long-term stability. Environmental Science & Technology 38, 6590-6596.

- Makris, K., Harris, W., O'Connor, G., Obreza, T., Elliott, H., 2005. Physicochemical properties related to long-term phosphorus retention by

drinking-water treatment residuals. Environmental Science & Technology 39, 4280-4289.

- Makris, K., Sarkar, D., Datta, R., 2006a. Aluminum-based drinking-water treatment residuals: A novel sorbent for perchlorate removal. Environmental Pollution 140, 9-12.

- Makris, K., Sarkar, D., Datta, R., 2006b. Evaluating a drinking-water waste by-product as a novel sorbent for arsenic. Chemosphere 64, 730-741.

- McClellan, K., Halden, R. U., 2010. Pharmaceuticals and personal care products in archived U.S. biosolids from the 2001 EPA National Sewage Sludge Survey. Water Res. 44 (2), 658−668.

- Miller, M.L., Bhadha, J.H., O'Connor, G.A., Jawitz, J.W., Mitchell, J., 2011. Aluminum water treatment residuals as permeable reactive barrier sorbents to reduce phosphorus losses. Chemosphere 83, 978-983.

- Moore, P.A. Jr., Daniel, T.C., Sharpley, A.N., Wood. C.W., 1995. Poultry manure management − Environmentally sound options. J. Soil Water Conserv. 50, 321-327.

- Muller, A. K., Westergaard, K., Christensen, S., Soresen, S. J., 2002. The diversity and function of soil microbial communities exposed to different disturbances Microb. Ecol 44, 49-58.

- Munir, M., Xagoraraki, I., 2011. Levels of Antibiotic Resistance Genes in Manure, Biosolids, and Fertilized Soil. J. Environ. Qual. 40 (1) 248-255.

- Nagar, R., Sarkar, D., Makris, K., Datta, R., 2010. Effect of solution chemistry on arsenic sorption by Fe- and Al-based drinking-water treatment residuals. Chemosphere 78, 1028-1035.

- Netthisinghe, A. M. P., Cook, K. L., Rice, C., Gilfillen, R. A., Sistani, K. R., 2013. Soil Nutrients, Bacteria Populations, and Veterinary Pharmaceuticals across a Backgrounding Beef Feedlot. J. Environ. Qual. 42, 532–544

- Nowack, B., Lützenkirchen, J., Behra, P., Sigg, L., 1996. Modeling the adsorption of metal-EDTA complexes onto oxides. Environmental Science and Technology 30, 2397- 2405.

- Oberle, K., Capdeville, M.J., Berthe, T., Budzinski, H., Petit, F., 2012. Evidence for a Complex Relationship between Antibiotics and Antibiotic-Resistant Escherichia Coli: From Medical Center Patients to a Receiving Environment. Environmental Science & Technology 46, 1859-1868.

- O'Connor, G.A., Elliott, H.A., Lu, R., 2002. Characterizing water treatment residuals phosphorus retention. Soil and Crop Science Society of Florida Proceedings 61, 67-73.

- Ostermann, A., Siemens, J., Welp, G., Xue, Q., Lin, X., Liu, X., and Amelung, W.2013. Leaching of veterinary antibiotics in calcareous Chinese croplands, Chemosphere, 91 (7) 928-934.

- Park, S., Choi, K., 2008. Hazard assessment of commonly used agricultural antibiotics on aquatic ecosystems. Ecotoxicology 17, 526-538.

- Pei, Z.G., Shan, X.Q., Zhang, S.Z., Kong, J.J., Wen, B., Zhang, J., Zheng, L.R., Xie, Y.N., Janssens, K., 2011. Insight to ternary complexes of co-adsorption of norfloxacin and Cu(II) onto montmorillonite at different ph using EXAFS. Journal of Hazardous Materials 186, 842-848.

- Pils, J., Laird, D., 2007. Sorption of tetracycline and chlortetracycline on K- and Ca-saturated soil clays, humic substances, and clay-humic complexes. Environmental Science & Technology 41, 1928-1933.

- Prakash, P., Sengupta, A.K., 2003. Selective coagulant recovery from water treatment plant residuals using donnan membrane process. Environmental Science & Technology 37, 4468-4474.

- Punamiya, P., Sarkar, D., Rakshit, S., Datta, R., 2013a. Effectiveness of Al- based drinking water treatment residuals as a novel sorbent to remove tetracyclines from aqueous medium. J. Environ. Qual. *doi:10.2134/jeq2013.03.0082*.

- Punamiya, P., Sarkar, D., Rakshit, S., Datta, R., 2013b. Effect of solution properties, competing ligands, and complexing metal on sorption of tetracyclines on Al-based drinking water treatment residuals, J. Hazard. Mater. (To be submitted).

- Rakshit, S., Punamiya, P., Datta, R., Sarkar, D., 2010. Sorption of oxytetracycline on magnetite-water interface. Geochimica Et Cosmochimica Acta 74, A846-A846.

- Rakshit, S., Punamiya, P., Datta, R., Sarkar, D., 2010. Sorption of oxytetracycline on magnetite-water interface. Geochimica Et Cosmochimica Acta 74, A846-A846.

- Rakshit, S., Sarkar, D., Elzinga, E.J., Datta, R.., 2013b. In Situ ATR-FTIR study of Oxytetracycline Sorption on Magnetite . J. Environ. Qual. 42:822-827.

- Rakshit, S., Sarkar, D., Elzinga, E.J., Punamiya, P., Datta, R., 2013a. Mechanisms of ciprofloxacin removal by nano-sized magnetite. J. Hazard Mater. 246-247:221-226.

- Sall, J., Creighton, L., Lehman, A., 2005. JMP start statistics, 8th edition. Cary, NC, SAS Institute.

- Sarkar, D., Makris, K., Vandanapu, V., Datta, R., 2007. Arsenic immobilization in soils amended with drinking-water treatment residuals. Environmental Pollution 146, 414-419.

- Sarmah, A. K., Meyer, M. T.; Boxall, A. B. A., 2006. A global perspective on the use, sale, exposure pathways, occurrence, fate and effects of veterinary antibiotics (VAs) in the environment Chemosphere 65, 725-759.

- Sassman, S. A., Lee, L. S., 2005. Sorption of three tetracyclines by several soils: assessing the role of pH and cation exchange. Environmental science & technology 39(19), 7452-7459.

- Schmitt, M.O., Schneider, S., 2000. Spectroscopic investigation of complexation between various tetracyclines and Mg^{2+} or Ca^{2+}. Phys. Chem. Comm. 9, 1–14.

- Sengelov, G., Agerso, Y., Halling-Sorensen, B., Baloda, S.B., Andersen, J.S., Jensen, L.B., 2003. Bacterial antibiotic resistance levels in Danish farmland as a result of treatment with pig manure slurry. Environment International 28, 587-595.

- Shenker, M., Harush, D., Ben-Ari, J., Chefetz, B., 2011. Uptake of carbamazepine by cucumber plants - A case study related to irrigation with reclaimed wastewater. Chemosphere 82, 905-910.

- Sparks, D. L. 1995. Environmental soil chemistry, 2nd ed. Academic Press, San Diego, CA.

- Stoob, K., Singer, H.P., Mueller, S.R., Schwarzenbach, R.P., Stamm, C.H., 2007. Dissipation and transport of veterinary sulfonamide antibiotics after manure application to grassland in a small catchment. Environmental Science & Technology 41, 7349-7355.

- Tanoue, R., Sato, Y., Motoyama, M., Nakagawa, S., Shinohara, R., Nomiyama, K., 2012. Plant Uptake of Pharmaceutical Chemicals Detected in Recycled Organic Manure and Reclaimed Wastewater. J. Agric. Food Chem. 60, 10203−10211

- Teixido, M., Hurtado, C., Pignatello, J.J., Beltran, J.L., Grenados, M., and Peccia, J., 2013. Predicting contaminant adsorption in black carbon (biochar)-amended soil for the veterinary antimicrobial sulfamethazine. Environ. Sci. Technol. 47, 6197.

- Ter Laak, T.L.,Wouter, A.G., Tolls, J., 2006. The effect of pH and ionic strength on the sorption of sulfachloropyridazine, tylosin, and oxytetracycline to soil. Environ. Toxicol. Chem. 25, 904–911.

- Thiele-Bruhn, S., 2003. Pharmaceutical antibiotic compounds in soils - a review. Journal of Plant Nutrition and Soil Science-Zeitschrift Fur Pflanzenernahrung Und Bodenkunde 166, 145-167.

- Tolls, J., 2001. Sorption of Veterinary Pharmaceuticals in Soils: A Review. Environmental Science & Technology 35, 3397-3406.

- Topp, E., Monteiro, S.C., Beck, A.., Ball Coelho, B., Boxall, P.W. Duenk, A.B.A., Kleywegt, S., Lapen, D.R., Payne, M., Sabourin, L., Metcalfe, C.D., 2008. Runoff of pharmaceuticals and personal care products following application of biosolids to an agricultural field Sci. Total Environ., 396, 52–59

- U.S. Department of Agriculture, Economic Research Service (USDA/ERS), 2005. Confined animal and manure nutrient data system. http://www.ers.usda.gov/Data/manure/default

- Van Dijk, J., & Keukens, H. J., 2000. The stability of some veterinary drugs and coccidiostats during composting and storage of laying hen and broiler faeces. In Residues of veterinary drugs in food. Proc. of the Euroresidue IV Conf., Veldhoven, Netherlands (pp. 8-10).

- Van Riemsdijk, W.H., Lyklema, J., 1980. Reaction of phosphate with gibbsite (AI(OH)$_3$) beyond the adsorption maximum. J. Colloid Interf. Sci. 76:55-66.

- Wang, Q., Yates, S.R., 2008b. Laboratory study of oxytetracycline degradation kinetics in animal manure and soil. J. Agric. Food Chem. 56, 1683-1688.

- Wang, Y. J., Jia, D. A., Sun, R. J., Zhu, H. W., Zhou, D. M., 2008a. Adsorption and cosorption of tetracycline and copper (II) on montmorillonite as affected by solution pH. Environmental Science and Technology 42(9), 3254-3259.

- Wang, Y. J., Sun,R. J., Xiao, A. Y., Wang, S. Q., and Zhou, D. M., 2010. Phosphate affects the adsorption of tetracycline on two soils with different characteristics. Geoderma, 156, 237-242.

- Watanabe, F. S., Olsen, S. R., 1965. Test of an ascorbic acid method for determining phosphorus in water and $NaHCO_3$ extracts from soil. Soil Science Society of America Journal. 29(6), 677-678.

- Watkinson, A. J., Murby, E. J., Kolpin, D. W., Costanzo, S. D., 2009. The occurrence of antibiotics in an urban watershed: From wastewater to drinking water. Sci. Total Environ. 407 (8), 2711−2723.

- Wessels, J.M., Ford, W.E., Szymczak, W., Schneider, S., 1998. The complexation of tetracycline and anhydrotetracycline with Mg2+ and Ca2+: A spectroscopic study. Journal of Physical Chemistry B 102, 9323-9331.

- Westergaard, K., Muller, A. K., Christensen, S., Bloem, J., Sorensen, S. J., 2001. Effects of tylosin as a disturbance on the soil microbial community Soil Biol. Biochem 33, 2061-2071.

- Winckler, C., Grafe, A., 2001. Use of veterinary drugs in intensive animal production Evidence for persistence of tetracycline in pig slurry. Journal of Soil Sediment - 1, 67 - 70.

- Wollenberger, L., Halling-Sorensen, B., Kusk, K.O., 2000. Acute and chronic toxicity of veterinary antibiotics to Daphnia magna. Chemosphere 40, 723-730.

- Wu, C., Spongberg, A. L., Witter, J. D., Fang, M., Czajkowski, K. P.,2010 Uptake of Pharmaceutical and Personal Care Products by Soybean Plants from Soils Applied with Biosolids and Irrigated with Contaminated Water. Environ. Sci. Technol. 44 (16), 6157-6161

- Yang, Y., Zhao, Y.Q., Babatunde, A.O., Wang, L., Ren, Y.X., Han, Y., 2006. Characteristics and mechanisms of phosphate adsorption on dewatered alum sludge. Sep. Purif. Technol. 51, 193-200.

- Zaimes, G. N., Schultz, R, C., 2002. Phosphorus in Agricultural Watersheds: A Literature Review. Iowa State University, Ames, Department of Forestry

- Zhang, D., Pan, B., Wu, M., Wang, B., Zhang, H., Peng, H., Wu, D., Ning, P., 2011a. Adsorption of sulfamethoxazole on functionalized carbon nanotubes as affected by cations and anions. Environmental Pollution 159, 2616-2621.

- Zhang, L., Song, X.Y., Liu, X.Y., Yang, L.J., Pan, F., Lv, J.N., 2011. Studies on the removal of tetracycline by multi-walled carbon nanotubes. Chemical Engineering Journal 178, 26-33.

- Zhang, Y., Cai, X.Y., Lang, X.M., Qiao, X.L., Li, X.H., Chen, J.W., 2012. Insights into aquatic toxicities of the antibiotics oxytetracycline and ciprofloxacin

in the presence of metal: Complexation versus mixture. Environmental Pollution 166, 48-56.

- Zhang, Y., Zhang, C., Parker, D. B., Snow, D. D., Zhou, Z., Li, X., 2013. Occurrence of antimicrobials and antimicrobial resistance genes in beef cattle storage ponds and swine treatment lagoons. Sci Total Environ. 6,463-464C:631-638

- Zhao, Y., Gu, X., Gao, S., Geng, J., and Wang, X., 2012. Adsorption of tetracycline (TC) onto montmorillonite: Cations and humic acid effects. Geoderma 183-184, 12-18.

LIST OF APPENDICIES

Appendix A: Experimental data for Chapter 2

Appendix B: Experimental data for Chapter 3

Appendix C: Experimental data for Chapter 4

Appendix D: Experimental data for Chapter 5

Appendix E: Pictures from Incubation and Greenhouse studies

Appendix F: Preface

Appendix A

A.1: Effect of SSR on amount of TTC and OTC sorption by Al-WTR. Experimental parameters: reaction time 24 h, ionic strength 0.01 M KCl, and pH 6.0 ± 0.1. Data are expressed as mean of three replicates ± one standard deviation.

	% Mean Sorption					
SSR	Initial TTC concentration			Standard Deviation		
g L^{-1}	0.1 mM	0.5 mM	1 mM	0.1 mM	0.5 mM	1 mM
1	15.0	10.0	8.0	0.88	0.78	1.73
5	40.2	33.0	29.7	0.98	0.78	1.15
10	65.1	60.0	57.0	1.53	1.50	1.73
15	82.2	80.1	77.7	2.30	0.89	2.31
20	99.0	98.0	97.7	3.36	2.10	0.58
40	99.0	99.0	99.0	1.53	0.70	0.51
75	99.1	99.0	99.0	1.50	1.30	0.42
100	99.2	99.1	99.0	2.10	2.30	0.45

	% Mean Sorption					
SSR	Initial OTC concentration			Standard Deviation		
g L^{-1}	0.1 mM	0.5 mM	1 mM	0.1 mM	0.5 mM	1 mM
1	15.3	10.0	8.0	0.88	0.78	0.85
5	37.1	33.3	29.7	0.98	0.78	0.75
10	63.0	60.5	57.2	1.53	1.50	1.20
15	82.0	80.3	77.7	2.30	0.89	1.50
20	99.0	98.2	97.7	3.36	2.10	2.20
40	99.0	99.0	99.0	1.53	0.70	0.92
75	99.0	99.0	99.0	1.50	1.30	1.40
100	99.0	99.0	99.0	2.10	2.30	1.20

A.2: Sorption edge of TTC and OTC sorption by Al-WTR as a function of different pH and initial TTC and OTC concentrations. Experimental parameters: reaction time 24 h, ionic strength 0.01 M KCl, and SSR 20 g L^{-1}. Data are expressed as mean of three replicates ± one standard deviation.

	% Mean Sorption (Experimental)			% Sorption (FITEQL Fit)			Standard Deviation (Experimental)		
	Initial TTC concentration			Initial TTC concentration			Initial TTC concentration		
Final pH	0.1 mM	0.5 mM	1.0 mM	0.1 mM	0.5 mM	1.0 mM	0.1mM	0.5mM	1.0 mM
3.05	93.50	90.60	88.50	91.00	90.00	89.00	0.58	2.65	1.50
4.09	95.50	91.33	90.20	95.00	94.00	92.00	0.00	1.15	0.00
4.96	96.50	95.33	93.66	100.00	100.00	100.00	0.58	0.58	0.87
6.05	96.17	97.00	95.00	100.00	100.00	100.00	0.00	0.00	1.44
7.08	96.17	95.00	94.00	100.00	100.00	100.00	0.00	0.00	1.44
7.96	93.83	92.67	90.66	100.00	100.00	100.00	1.15	1.15	0.29
8.92	93.17	90.00	87.33	98.00	97.00	96.00	0.58	3.46	1.44

	% Mean Sorption (Experimental)			% Sorption (FITEQL Fit)			Standard Deviation (Experimental)		
	Initial TTC concentration			Initial TTC concentration			Initial TTC concentration		
Final pH	0.1 mM	0.5 mM	1.0 mM	0.1 mM	0.5 mM	1.0 mM	0.1mM	0.5mM	1.0 mM
3.07	91.50	89.50	87.50	91.00	90.00	89.00	1.13	1.18	0.28
4.02	92.97	91.16	90.30	95.00	94.00	92.00	0.57	1.18	0.57
5.01	94.44	93.23	92.44	100.00	100.00	100.00	1.13	0.59	0.57
6.04	96.44	97.28	95.04	100.00	100.00	100.00	0.00	0.59	0.57
6.96	95.75	94.97	94.10	100.00	100.00	100.00	0.00	0.12	0.57
7.94	93.30	92.52	91.81	100.00	100.00	100.00	0.57	1.18	0.28
8.97	91.34	89.80	89.54	98.00	97.00	96.00	0.57	3.53	0.28

A.3: Effect of ionic strength on the amount of TTC and OTC sorbed by Al-WTR as a function of different initial TTC and OTC concentrations. Experimental parameters: reaction time 24 h, pH 6.0 ± 0.1, and SSR 20 g L-1. Data are expressed as mean of three replicates ± one standard deviation.

Initial Conc.	Mean TTC Conc. Solution	Mean TTC Conc. In solid	Mean TTC Sorption (%)	S.D
IS = 0.01 M KCl				
mM	mM	mM		
0.050	0.0001	0.050	99.893	0.012
0.100	0.0001	0.100	99.851	0.010
0.200	0.001	0.199	99.500	0.020
0.500	0.021	0.479	95.884	0.014
1.000	0.107	0.894	89.444	1.503
2.000	0.253	1.752	87.611	0.788
IS = 0.1 M KCl				
Initial Conc.	Mean TTC Conc. Solution	Mean TTC Conc. In solid	Mean TTC Sorption (%)	S.D
mM	mM	mM		
0.050	0.0001	0.050	99.893	0.012
0.100	0.0001	0.100	99.851	0.010
0.200	0.001	0.199	99.500	0.030
0.500	0.051	0.448	89.689	0.619
1.000	0.153	0.849	84.889	0.192
2.000	0.350	1.650	82.500	1.000
IS = 0.5 M KCl				
Initial Conc.	Mean TTC Conc. Solution	Mean TTC Conc. In solid	Mean TTC Sorption (%)	S.D
mM	mM	mM		
0.050	0.0001	0.050	99.893	0.012
0.100	0.0001	0.100	99.851	0.010
0.200	0.001	0.199	99.500	0.050
0.500	0.040	0.460	92.089	0.102
1.000	0.193	0.809	80.889	0.192
2.000	0.390	1.603	80.167	0.577

IS = 0.01 M KCl				
Initial Conc.	Mean OTC Conc. Solution	Mean OTC Conc. In solid	Mean OTC Sorption (%)	S.D
mM	mM	mM		
0.050	0.0001	0.050	99.889	0.010
0.100	0.0002	0.100	99.849	0.010
0.200	0.0010	0.199	99.500	0.030
0.500	0.0213	0.479	95.711	0.102
1.000	0.1100	0.890	89.000	1.000
2.000	0.2467	1.751	87.556	0.509
IS = 0.1 M KCl				
Initial Conc.	Mean OTC Conc. Solution	Mean OTC Conc. In solid	Mean OTC Sorption (%)	S.D
mM	mM	mM		
0.050	0.0001	0.050	99.889	0.010
0.100	0.0002	0.100	99.849	0.010
0.200	0.0010	0.199	99.500	0.020
0.500	0.0510	0.449	89.733	0.702
1.000	0.1633	0.842	84.222	0.694
2.000	0.3533	1.649	82.444	1.005
IS = 0.5 M KCl				
Initial Conc.	Mean OTC Conc. Solution	Mean OTC Conc. In solid	Mean OTC Sorption (%)	S.D
mM	mM	mM		
0.050	0.0001	0.050	99.889	0.010
0.100	0.0002	0.100	99.849	0.010
0.200	0.0010	0.199	99.500	0.030
0.500	0.0443	0.458	91.644	0.454
1.000	0.1967	0.808	80.778	0.385
2.000	0.4200	1.567	78.333	1.155

A.4: TTC and OTC sorption isotherm with simulated Freundlich fit for Al-WTR at different ionic strength. Experimental parameters: reaction time 24 h, pH 6.0 ± 0.1, and SSR 20 g L^{-1}. Data are expressed as mean of three replicates ± one standard deviation.

Initial Conc.	Mean Conc. In soln (Ce)	Mean Conc. In solid	Mean Conc. In solid (qe)	log (Ce)	log(qe)
IS = 0.01 M KCl					
TTC mM	TTC mM	TTC mM	TTC mmol/g		
0.050	0.0001	0.0500	0.0025	-4.3010	-2.6025
0.100	0.0001	0.0999	0.0050	-3.8337	-2.3017
0.200	0.0010	0.1990	0.0100	-3.0000	-2.0022
0.500	0.0207	0.4793	0.0240	-1.6847	-1.6204
1.000	0.1067	0.8933	0.0447	-0.9720	-1.3500
2.000	0.2533	1.7467	0.0873	-0.5963	-1.0588
IS = 0.1 M KCl					
TTC mM	TTC mM	TTC mM	TTC mmol/g		
0.050	0.0001	0.0500	0.0025	-4.3010	-2.6025
0.100	0.0001	0.0999	0.0050	-3.8337	-2.3017
0.200	0.0010	0.1990	0.0100	-3.0000	-2.0022
0.500	0.0507	0.4493	0.0225	-1.2953	-1.6485
1.000	0.1533	0.8467	0.0423	-0.8144	-1.3733
2.000	0.3500	1.6500	0.0825	-0.4559	-1.0835
IS = 0.5 M KCl					
TTC mM	TTC mM	TTC mM	TTC mmol/g		
0.050	0.0001	0.0500	0.0025	-4.3010	-2.6025
0.100	0.0001	0.0999	0.0050	-3.8337	-2.3017
0.200	0.0010	0.1990	0.0100	-3.0000	-2.0022
0.500	0.0397	0.4603	0.0230	-1.4016	-1.6380
1.000	0.1933	0.8067	0.0403	-0.7137	-1.3943
2.000	0.3900	1.6100	0.0805	-0.4089	-1.0942

IS = 0.01 M KCl					
Initial Conc	**Mean conc in soln(Ce)**	**Mean conc in solid**	**Mean conc in solid (qe)**	log (Ce)	log(qe)
OTC mM	OTC mM	OTC mM	OTC mmol/g		
0.050	0.0001	0.0499	0.0025	-4.2467	-2.6026
0.100	0.0002	0.0998	0.0050	-3.8144	-2.3017
0.200	0.0010	0.1990	0.0100	-3.0000	-2.0022
0.500	0.0213	0.4787	0.0239	-1.6709	-1.6210
1.000	0.1100	0.8900	0.0445	-0.9586	-1.3516
2.000	0.2467	1.7533	0.0877	-0.6079	-1.0572
IS = 0.1 M KCl					
Initial Conc	**Mean conc in soln(Ce)**	**Mean conc in solid**	**Mean conc in solid (qe)**	log (Ce)	log(qe)
OTC mM	OTC mM	OTC mM	OTC mmol/g		
0.050	0.0001	0.0499	0.0025	-4.2467	-2.6026
0.100	0.0002	0.0998	0.0050	-3.8144	-2.3017
0.200	0.0010	0.1990	0.0100	-3.0000	-2.0022
0.500	0.0510	0.4490	0.0225	-1.2924	-1.6488
1.000	0.1633	0.8367	0.0418	-0.7869	-1.3785
2.000	0.3533	1.6467	0.0823	-0.4518	-1.0844
IS = 0.5 M KCl					
Initial Conc	**Mean conc in soln(Ce)**	**Mean conc in solid**	**Mean conc in solid (qe)**	log (Ce)	log(qe)
OTC mM	OTC mM	OTC mM	OTC mmol/g		
0.050	0.0001	0.0499	0.0025	-4.2467	-2.6026
0.100	0.0002	0.0998	0.0050	-3.8144	-2.3017
0.200	0.0010	0.1990	0.0100	-3.0000	-2.0022
0.500	0.0443	0.4557	0.0228	-1.3533	-1.6424
1.000	0.1967	0.8033	0.0402	-0.7063	-1.3961
2.000	0.4200	1.5800	0.0790	-0.3768	-1.1024

A.5: Effect of reaction time on the amount of TTC (7A) and OTC (7B) sorbed by Al-WTR as a function of different initial TTC and OTC concentrations (0.5 and 1 mM). Experimental parameters: SSR 20 g L^{-1}, pH 6.0 ± 0.1, and ionic strength 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.

Reaction time		Initial TTC (0.5mM)		Initial TTC (1.0 mM)		Initial Conc.	Initial Conc.
		Solid	Solution	Solid	Solution	0.5mM	1mM
h	min	mM	mM	mM	mM		
		Mean Conc.	Mean Conc.	Mean Conc.	Mean Conc.	Standard deviation	
0.0	0	0	0.52	0	1.002	0	0
0.167	10	0.37	0.13	0.802	0.2	0.02	0.04
0.333	20	0.39	0.11	0.792	0.21	0.01	0.05
0.5	30	0.41	0.09	0.902	0.1	0.02	0.04
1	60	0.44	0.06	0.912	0.09	0.01	0.03
2	120	0.44	0.06	0.932	0.07	0.02	0.05
5	300	0.45	0.05	0.947	0.055	0.03	0.04
10	600	0.46	0.04	0.97	0.032	0.03	0.03
24	1440	0.49	0.01	0.992	0.01	0.03	0.05
48	2880	0.499	0.001	0.999	0.001	0.04	0.05
72	4320	0.499	0.001	0.999	0.001	0.04	0.04

Reaction time		Initial OTC (0.5mM)		Initial OTC (1mM)		Initial Conc.	Initial Conc.
		Solid	Solution	Solid	Solution	0.5mM	1mM
h	min	mM	mM	mM	mM		
		Mean Conc.	Mean Conc.	Mean Conc.	Mean Conc.	Standard deviation	
0.0	0	0	0.52	0	1.002	0	0
0.167	10	0.38	0.14	0.812	0.19	0.01	0.03
0.333	20	0.4	0.12	0.802	0.2	0.02	0.05
0.5	30	0.43	0.09	0.892	0.11	0.02	0.04
1	60	0.46	0.06	0.912	0.09	0.03	0.03
2	120	0.46	0.06	0.922	0.08	0.02	0.05
5	300	0.47	0.05	0.945	0.057	0.02	0.04
10	600	0.48	0.04	0.972	0.03	0.03	0.03
24	1440	0.5	0.02	0.992	0.01	0.02	0.03
48	2880	0.5	0.02	0.998	0.002	0.04	0.04
72	4320	0.5	0.02	0.998	0.002	0.03	0.04

A.6: ATR-FTIR spectra. All spectra are normalized with the highest peak observed.

Wavenumber	TTC	Al-WTR	OTC	TTC+ Al-WTR	OTC+ Al-WTR
1301.74	0.16953	0.03401	0.19953	0.08931	0.06931
1303.66	0.17121	0.03462	0.20121	0.08951	0.06951
1305.59	0.17174	0.03441	0.20174	0.08974	0.06974
1307.52	0.17119	0.03452	0.20119	0.09002	0.07002
1309.45	0.16951	0.03489	0.19951	0.09013	0.07013
1311.38	0.16707	0.03529	0.19707	0.09016	0.07016
1313.31	0.16446	0.03595	0.19446	0.09040	0.07040
1315.24	0.16220	0.03628	0.19220	0.09073	0.07073
1317.16	0.16078	0.03663	0.19078	0.09091	0.07091
1319.09	0.16036	0.03692	0.19036	0.09100	0.07100
1321.02	0.16124	0.03736	0.19124	0.09121	0.07121
1322.95	0.16343	0.03825	0.19343	0.09153	0.07153
1324.88	0.16664	0.03881	0.19664	0.09175	0.07175
1326.81	0.16991	0.03949	0.19991	0.09201	0.07201
1328.74	0.17224	0.03992	0.20224	0.09239	0.07239
1330.66	0.17285	0.04084	0.20285	0.09274	0.07274
1332.59	0.17133	0.04197	0.20133	0.09302	0.07302
1334.52	0.16765	0.04284	0.19765	0.09351	0.07351
1336.45	0.16260	0.04388	0.19260	0.09400	0.07400
1338.38	0.15718	0.04465	0.18718	0.09445	0.07445
1340.31	0.15235	0.04580	0.18235	0.09480	0.07480
1342.23	0.14896	0.04666	0.17896	0.09521	0.07521
1344.16	0.14704	0.04782	0.17704	0.09576	0.07576
1346.09	0.14627	0.04890	0.17627	0.09631	0.07631
1348.02	0.14634	0.04994	0.17634	0.09675	0.07675
1349.95	0.14685	0.05077	0.17685	0.09718	0.07718
1351.88	0.14776	0.05203	0.17776	0.09768	0.07768
1353.81	0.14880	0.05309	0.17880	0.09809	0.07809
1355.73	0.14973	0.05448	0.17973	0.09786	0.07846
1357.66	0.15050	0.05524	0.18050	0.09762	0.07890
1359.59	0.15104	0.05628	0.18104	0.09566	0.07950
1361.52	0.15118	0.05820	0.18118	0.09558	0.07994
1363.45	0.15086	0.05967	0.18086	0.09705	0.08021
1365.38	0.15019	0.06002	0.18019	0.09740	0.08068
1367.31	0.14929	0.05998	0.17929	0.09736	0.08089
1369.23	0.14843	0.06082	0.17843	0.09820	0.08098

1371.16	0.14805	0.06145	0.17805	0.09883	0.08129
1373.09	0.14811	0.06216	0.17811	0.09954	0.08158
1375.02	0.14860	0.06231	0.17860	0.09969	0.08156
1376.95	0.14949	0.06307	0.17949	0.10045	0.08169
1378.88	0.15034	0.06266	0.18034	0.10004	0.08173
1380.80	0.15051	0.06301	0.18051	0.10039	0.08155
1382.73	0.14975	0.06348	0.17975	0.10086	0.08134
1384.66	0.14864	0.06366	0.17864	0.10104	0.08117
1386.59	0.14738	0.06348	0.17738	0.10086	0.08081
1388.52	0.14575	0.06362	0.17575	0.10100	0.08025
1390.45	0.14386	0.06373	0.17386	0.10111	0.08014
1392.38	0.14170	0.06373	0.17170	0.10111	0.07978
1394.30	0.13924	0.06405	0.16924	0.10070	0.07970
1396.23	0.13713	0.06439	0.16713	0.10076	0.07976
1398.16	0.13555	0.06276	0.16555	0.10114	0.08014
1400.09	0.13423	0.06249	0.16423	0.10206	0.08106
1402.02	0.13358	0.06195	0.16358	0.10304	0.08204
1403.95	0.13311	0.06161	0.16311	0.10334	0.08234
1405.88	0.13278	0.06103	0.16278	0.10247	0.08147
1407.80	0.13270	0.06039	0.16270	0.10164	0.08064
1409.73	0.13276	0.05983	0.16276	0.10146	0.08046
1411.66	0.13314	0.05956	0.16314	0.10185	0.08085
1413.59	0.13406	0.05925	0.16406	0.10289	0.08189
1415.52	0.13504	0.05825	0.16504	0.10442	0.08342
1417.45	0.13534	0.05761	0.16534	0.10668	0.08568
1419.37	0.13447	0.05700	0.16447	0.10974	0.08874
1421.30	0.13364	0.05675	0.16364	0.11356	0.09256
1423.23	0.13346	0.05658	0.16346	0.11770	0.09670
1425.16	0.13385	0.05585	0.16385	0.12226	0.10126
1427.09	0.13489	0.05548	0.16489	0.12701	0.10601
1429.02	0.13642	0.05487	0.16642	0.13214	0.11114
1430.95	0.13868	0.05480	0.16868	0.13264	0.11264
1432.87	0.14174	0.05588	0.17174	0.13207	0.11107
1434.80	0.14556	0.05342	0.17556	0.13262	0.11062
1436.73	0.14970	0.05288	0.17970	0.13193	0.11093
1438.66	0.15426	0.05218	0.18426	0.12980	0.10880
1440.59	0.15901	0.05223	0.18901	0.12883	0.10783
1442.52	0.16414	0.05107	0.19414	0.12607	0.10507
1444.44	0.16864	0.05027	0.19864	0.11986	0.09886

1446.37	0.17007	0.04977	0.20007	0.11299	0.09199
1448.30	0.16762	0.04919	0.19762	0.10674	0.08574
1450.23	0.16393	0.04805	0.19393	0.09302	0.08290
1452.16	0.16180	0.04752	0.19180	0.09249	0.08253
1454.09	0.16083	0.04699	0.19083	0.09196	0.08246
1456.02	0.15807	0.04690	0.18807	0.09187	0.08179
1457.94	0.15186	0.04604	0.18186	0.09101	0.08019
1459.87	0.14499	0.04609	0.17499	0.09106	0.07990
1461.80	0.13874	0.04629	0.16874	0.09126	0.07989
1463.73	0.13474	0.04628	0.16474	0.09125	0.07945
1465.66	0.13305	0.04581	0.16305	0.09078	0.07865
1467.59	0.13254	0.04579	0.16254	0.09076	0.07834
1469.52	0.13138	0.04607	0.16138	0.09104	0.07825
1471.44	0.12926	0.04597	0.15926	0.09094	0.07778
1473.37	0.12666	0.04663	0.15666	0.09160	0.07694
1475.30	0.12458	0.04703	0.15458	0.09200	0.07677
1477.23	0.12251	0.04755	0.15251	0.09252	0.07664
1479.16	0.12026	0.04929	0.15026	0.09426	0.07679
1481.09	0.11734	0.05053	0.14734	0.09550	0.07696
1483.01	0.11432	0.04997	0.14432	0.09494	0.07721
1484.94	0.11177	0.05074	0.14177	0.09571	0.07759
1486.87	0.11016	0.05177	0.14016	0.09674	0.07786
1488.80	0.10954	0.05259	0.13954	0.09756	0.07807
1490.73	0.10975	0.05420	0.13975	0.09917	0.07823
1492.66	0.11064	0.05454	0.14064	0.09951	0.07896
1494.59	0.11193	0.05683	0.14193	0.10180	0.07992
1496.51	0.11337	0.05691	0.14337	0.10188	0.08036
1498.44	0.11527	0.05879	0.14527	0.10376	0.08121
1500.37	0.11741	0.05979	0.14741	0.10476	0.08265
1502.30	0.11975	0.06034	0.14975	0.10531	0.08431
1504.23	0.12204	0.06204	0.15204	0.10769	0.08669
1506.16	0.12407	0.06428	0.15407	0.11900	0.09800
1508.09	0.12647	0.06458	0.15647	0.12021	0.09921
1510.01	0.12931	0.06623	0.15931	0.12045	0.09945
1511.94	0.13206	0.06920	0.16206	0.12214	0.10114
1513.87	0.13480	0.06911	0.16480	0.12525	0.10425
1515.80	0.13732	0.07060	0.16732	0.1270333	0.10603
1517.73	0.13969	0.07328	0.16969	0.1280333	0.10703
1519.66	0.15100	0.07413	0.18100	0.1335986	0.11160

1521.58	0.15221	0.07609	0.18221	0.134002	0.11002
1523.51	0.15245	0.07773	0.18245	0.1346776	0.11168
1525.44	0.15414	0.08031	0.18414	0.1337931	0.11179
1527.37	0.15725	0.08113	0.18725	0.1316416	0.11064
1529.30	0.1590333	0.0842721	0.1890333	0.13055	0.10955
1531.23	0.1600333	0.0851051	0.1900333	0.12757	0.10657
1533.16	0.1655986	0.0848403	0.1955986	0.12441	0.10341
1535.08	0.169002	0.088436	0.199002	0.12230	0.10130
1537.01	0.1696776	0.0903961	0.1996776	0.12080	0.09980
1538.94	0.1657931	0.0917052	0.1957931	0.12021	0.09921
1540.87	0.1636416	0.0937542	0.1936416	0.12021	0.09921
1542.80	0.16255	0.09418	0.19255	0.12021	0.09921
1544.73	0.15957	0.09534	0.18957	0.12745	0.10645
1546.66	0.15641	0.09701	0.18641	0.12748	0.10648
1548.58	0.15430	0.09556	0.18430	0.12917	0.10817
1550.51	0.15280	0.09781	0.18280	0.13082	0.10982
1552.44	0.15221	0.09812	0.18221	0.13221	0.11121
1554.37	0.15221	0.09817	0.18221	0.13377	0.11277
1556.30	0.15221	0.09830	0.18221	0.13579	0.11479
1558.23	0.15945	0.09762	0.18945	0.13937	0.11837
1560.15	0.15948	0.09738	0.18948	0.14236	0.12336
1562.08	0.16117	0.09649	0.19117	0.14371	0.12471
1564.01	0.16282	0.09466	0.19282	0.14379	0.12579
1565.94	0.16421	0.09256	0.19421	0.14237	0.12437
1567.87	0.16577	0.09001	0.19577	0.14236	0.12336
1569.80	0.16779	0.08641	0.19779	0.14168	0.12368
1571.73	0.17137	0.08272	0.20137	0.14025	0.12225
1573.65	0.17636	0.08027	0.20636	0.13700	0.11600
1575.58	0.17871	0.07993	0.20871	0.13165	0.11065
1577.51	0.17879	0.08142	0.20879	0.12757	0.10657
1579.44	0.17837	0.08290	0.20837	0.12441	0.10341
1581.37	0.17736	0.08347	0.20736	0.12230	0.10130
1583.30	0.17668	0.08334	0.20668	0.12080	0.10195
1585.23	0.17525	0.08320	0.20525	0.12021	0.10121
1587.15	0.16900	0.08373	0.19900	0.12045	0.09945
1589.08	0.16365	0.08114	0.19365	0.12014	0.10114
1591.01	0.15957	0.07891	0.18957	0.12525	0.10425
1592.94	0.15641	0.07792	0.18641	0.13021	0.10921
1594.87	0.15430	0.07471	0.18430	0.13603	0.11503

1596.80	0.15280	0.07271	0.18280	0.13760	0.11660
1598.72	0.15221	0.07048	0.18221	0.13800	0.11600
1600.65	0.15245	0.07026	0.18245	0.13978	0.11678
1602.58	0.15414	0.07108	0.18414	0.14079	0.11793
1604.51	0.15725	0.07438	0.18725	0.13854	0.11554
1606.44	0.16221	0.07368	0.19221	0.13502	0.11402
1608.37	0.16803	0.07386	0.19803	0.12757	0.10657
1610.30	0.17360	0.07428	0.20360	0.12441	0.10341
1612.22	0.17700	0.07410	0.20700	0.12230	0.10130
1614.15	0.17768	0.07355	0.20768	0.12080	0.09980
1616.08	0.17479	0.07296	0.20479	0.12021	0.09921
1618.01	0.17054	0.07147	0.20054	0.12021	0.09921
1619.94	0.16702	0.07112	0.19702	0.12021	0.09921
1621.87	0.15957	0.07132	0.18957	0.12072	0.10172
1623.80	0.15641	0.06923	0.18641	0.12312	0.10212
1625.72	0.15430	0.06821	0.18430	0.12472	0.10312
1627.65	0.15280	0.06747	0.18280	0.12572	0.10316
1629.58	0.15221	0.06593	0.18221	0.12572	0.10352
1631.51	0.15221	0.06517	0.18221	0.12572	0.10362
1633.44	0.15221	0.06406	0.18221	0.12572	0.10372
1635.37	0.15672	0.06325	0.18672	0.12487	0.10387
1637.29	0.15512	0.06313	0.18512	0.12557	0.10457
1639.22	0.15672	0.06104	0.18672	0.12637	0.10537
1641.15	0.15772	0.06041	0.18772	0.12680	0.10580
1643.08	0.15772	0.06040	0.18772	0.12748	0.10641
1645.01	0.15772	0.05872	0.18772	0.12745	0.10645
1646.94	0.15772	0.05809	0.18772	0.12748	0.10648
1648.87	0.15687	0.05671	0.18687	0.12917	0.10817
1650.79	0.15757	0.05666	0.18757	0.13082	0.10982
1652.72	0.15837	0.05685	0.18837	0.13221	0.11121
1654.65	0.15880	0.05416	0.18880	0.13377	0.11277
1656.58	0.15948	0.05364	0.18948	0.13579	0.11479
1658.51	0.15945	0.05276	0.18945	0.13937	0.11837
1660.44	0.15948	0.05216	0.18948	0.14436	0.12336
1662.37	0.16117	0.05256	0.19117	0.14971	0.12771
1664.29	0.16282	0.05021	0.19282	0.14889	0.12779
1666.22	0.16421	0.05063	0.19421	0.14837	0.12737
1668.15	0.16577	0.04906	0.19577	0.14736	0.12636
1670.08	0.16779	0.04882	0.19779	0.14597	0.12497

1672.01	0.17137	0.04816	0.20137	0.14325	0.12225
1673.94	0.17636	0.04710	0.20636	0.13700	0.11600
1675.86	0.18171	0.04661	0.21171	0.13165	0.11065
1677.79	0.18679	0.04592	0.21679	0.12518	0.10418
1679.72	0.18937	0.04533	0.21937	0.12035	0.09935
1681.65	0.18736	0.04511	0.21736	0.11696	0.09596
1683.58	0.18197	0.04545	0.21197	0.11504	0.09404
1685.51	0.17525	0.04357	0.20525	0.11032	0.09327
1687.44	0.16900	0.04318	0.19900	0.10993	0.08997
1689.36	0.16365	0.04238	0.19365	0.10913	0.08960
1691.29	0.15718	0.04181	0.18718	0.10856	0.08920
1693.22	0.15235	0.04090	0.18235	0.10765	0.08922
1695.15	0.14896	0.04026	0.17896	0.10701	0.08850
1697.08	0.14704	0.03980	0.17704	0.10655	0.08775
1699.01	0.14627	0.04057	0.17627	0.10732	0.08787
1700.94	0.14634	0.03918	0.17634	0.10593	0.08661
1702.86	0.14685	0.03887	0.17685	0.10562	0.08687
1704.79	0.14776	0.03875	0.17776	0.10550	0.08679
1706.72	0.14880	0.03810	0.17880	0.10485	0.08639
1708.65	0.14973	0.03812	0.17973	0.10487	0.08636
1710.58	0.15050	0.03761	0.18050	0.10436	0.08635
1712.51	0.15104	0.03738	0.18104	0.10413	0.08634
1714.43	0.15118	0.03681	0.18118	0.10356	0.08626
1716.36	0.15086	0.03740	0.18086	0.10415	0.08587
1718.29	0.15019	0.03638	0.18019	0.10313	0.08490
1720.22	0.14929	0.03616	0.17929	0.10291	0.08462
1722.15	0.14843	0.03574	0.17843	0.10249	0.08466
1724.08	0.14805	0.03550	0.17805	0.10225	0.08430
1726.01	0.14811	0.03544	0.17811	0.10219	0.08404
1727.93	0.14860	0.03473	0.17860	0.10148	0.08387
1729.86	0.14949	0.03436	0.17949	0.10111	0.08359
1731.79	0.15034	0.03812	0.18034	0.10487	0.08344
1733.72	0.15051	0.03761	0.18051	0.10436	0.08282
1735.65	0.14975	0.03738	0.17975	0.10413	0.08225
1737.58	0.14864	0.03681	0.17864	0.10356	0.08255

Appendix B
B.1: The effect of pH on the amount of TTC sorbed by Al-WTR at different SSR's: Data
are expressed as mean of three replicates ± one standard deviation.

| pH | % TTC Mean Sorption (SSR 5 g/L) | | | | Standard Deviation | | | |
	0.1 mM	0.5 mM	1.0 mM	5.0 mM	0.1 mM	0.5 mM	1.0 mM	5.0 mM
3.21	68.50	66.50	66.00	61	2.40	2.60	2.40	1.90
4.08	71.50	67.50	66.50	61.5	2.80	1.80	1.90	1.80
5.16	72.50	71.50	67.50	64	2.70	1.60	2.50	1.80
6.03	76.00	73.50	71.50	67.50	2.40	2.50	1.60	2.70
7.1	75.00	72.50	69.50	65.00	1.80	2.40	1.80	2.90
8.08	72.50	71.50	66.50	61.50	1.90	1.80	1.50	2.10
9.04	63.50	61.50	53.50	46.50	2.50	2.80	1.80	2.50
10.02	58.50	56.50	48.50	40.00	1.80	2.50	1.80	2.50
11.06	54.50	51.50	40.50	32.50	2.50	1.80	1.90	2.50
pH	%TTC Mean Sorption (SSR 10 g/L)				Standard Deviation			
	0.1 mM	0.5 mM	1.0 mM	5.0 mM	0.1 mM	0.5 mM	1.0 mM	5.0 mM
3.21	78.50	78.40	75.00	71.5	2.20	1.50	1.80	1.50
4.08	80.00	80.20	77.00	72.5	2.40	2.10	1.70	1.40
5.16	83.55	82.50	79.00	75	2.10	2.10	1.40	1.85
6.03	85.50	86.50	83.50	78.50	1.80	1.50	1.50	1.90
7.1	85.50	85.50	82.50	79.00	1.90	1.50	1.44	2.80
8.08	83.50	84.50	81.50	76.50	2.10	1.80	0.29	2.20
9.04	82.00	82.00	78.00	71.50	1.50	2.90	1.80	2.60
10.02	78.00	78.50	75.00	66.00	1.20	3.10	1.50	2.50
11.06	73.00	72.10	68.00	61.00	2.30	2.50	1.15	2.40
pH	%TTC Mean Sorption (SSR 20 g/L)				Standard Deviation			
	0.1 mM	0.5 mM	1.0 mM	5.0 mM	0.1 mM	0.5 mM	1.0 mM	5.0 mM
3.21	89.50	87.50	88.50	84.5	2.50	2.50	1.80	1.80
4.08	91.00	89.50	89.50	84.5	2.30	3.10	1.90	2.10
5.16	94.00	93.50	90.50	87	2.50	2.80	2.10	2.50
6.03	96.50	95.50	91.00	87.50	2.80	2.40	2.50	1.50
7.1	94.50	93.50	91.50	83.50	2.50	3.50	2.20	2.60
8.08	92.50	91.50	88.50	82.00	3.10	2.20	1.90	2.20
9.04	88.00	88.50	86.00	81.00	1.50	1.50	1.60	2.60
10.02	87.50	88.00	83.00	76.00	1.70	2.40	1.50	2.20
11.06	77.50	75.00	76.50	68.50	2.31	2.50	1.50	2.50

B.2: The effect of pH on the amount of OTC sorbed by Al-WTR at different SSR's: Data are expressed as mean of three replicates ± one standard deviation.

	% OTC Mean Sorption (SSR 5 g/L)				Standard Deviation			
pH	0.1 mM	0.5 mM	1.0 mM	5.0 mM	0.1 mM	0.5 mM	1.0 mM	5.0 mM
3.21	67.00	65.50	64.50	61.50	2.30	1.50	2.10	2.20
4.08	70.50	67.00	65.50	62.00	1.90	1.80	2.20	2.10
5.16	72.00	71.00	66.00	64.50	2.40	2.60	1.70	1.50
6.03	75.50	72.50	71.00	66.50	1.20	2.90	1.50	1.70
7.1	75.00	71.50	70.00	64.00	1.60	1.90	2.10	1.80
8.08	72.00	71.00	66.00	61.00	2.10	2.30	2.70	2.50
9.04	63.00	60.50	54.00	47.50	1.90	1.60	1.80	1.30
10.02	58.00	56.00	49.00	41.00	1.80	1.90	2.20	2.6
11.06	54.50	52.00	41.50	31.50	1.50	1.80	2.50	2.30
	%OTC Mean Sorption (SSR 10 g/L)				Standard Deviation			
pH	0.1 mM	0.5 mM	1.0 mM	5.0 mM	0.1 mM	0.5 mM	1.0 mM	5.0 mM
3.21	77.00	77.40	73.50	72.00	2.80	3.10	1.90	2.10
4.08	79.00	79.70	76.00	73.00	2.10	2.10	3.10	1.40
5.16	83.05	82.00	77.50	74.50	2.40	2.10	2.80	1.85
6.03	85.00	85.50	83.00	77.50	1.80	1.50	2.40	1.90
7.1	85.50	85.00	83.00	78.00	1.90	2.40	1.44	1.50
8.08	83.00	84.00	81.00	76.00	2.50	2.10	0.29	2.60
9.04	81.50	81.00	78.50	71.00	2.80	2.90	2.40	2.20
10.02	77.50	78.00	75.50	67.50	2.50	3.10	1.50	2.50
11.06	72.50	72.60	69.00	60.00	2.30	2.40	1.15	2.10
	%OTC Mean Sorption (SSR 20 g/L)				Standard Deviation			
pH	0.1 mM	0.5 mM	1.0 mM	5.0 mM	0.1 mM	0.5 mM	1.0 mM	5.0 mM
3.21	88.50	86.50	87.00	85.00	2.70	2.50	1.70	1.90
4.08	90.00	89.00	88.00	85.00	1.50	3.10	2.20	2.10
5.16	93.50	93.00	89.00	86.50	1.80	2.80	2.10	2.50
6.03	96.00	94.50	90.50	86.00	2.80	2.40	2.50	1.50
7.1	94.50	93.00	91.00	82.00	2.40	3.50	2.20	2.60
8.08	92.50	91.00	88.00	81.50	3.50	1.80	1.90	2.20
9.04	87.50	87.50	86.50	80.50	1.50	1.40	1.50	2.60
10.02	87.00	87.50	83.50	77.50	1.70	2.10	2.20	2.20
11.06	77.00	75.50	74.50	67.50	2.31	1.90	1.50	2.50

B.3: The effect of phosphate on the amount of TTC and OTC sorbed by Al-WTR as a function of pH and different TTC/OTC:P ratios. Initial TTC concentration 1.0 mM L^{-1}, reaction time 24 h, and ionic strength buffer 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.

1:0 (TTC:P)			
pH	TTC in Solution	TTC in Solid	% TTC Sorption
	mM	mM	
3.21	0.062	0.938	93.8
5.15	0.024	0.976	97.6
7.09	0.04	0.96	96
8.98	0.066	0.934	93.4
10.93	0.158	0.842	84.2
1:1 (TTC:P)			
pH	TTC in Solution	TTC in Solid	% TTC Sorption
	mM	mM	
3.21	0.058	0.942	94.2
5.15	0.036	0.964	96.4
7.09	0.038	0.962	96.2
8.98	0.078	0.922	92.2
10.93	0.182	0.818	81.8
1:10 (TTC:P)			
pH	TTC in Solution	TTC in Solid	% TTC Sorption
	mM	mM	
3.21	0.18	0.82	82
5.15	0.15	0.85	85
7.09	0.178	0.822	82.2
8.98	0.198	0.802	80.2
10.93	0.214	0.786	78.6
1:20 (TTC:P)			
pH	TTC in Solution	TTC in Solid	% TTC Sorption
	mM	mM	
3.21	0.24	0.76	76
5.15	0.2	0.8	80.1
7.09	0.28	0.72	72
8.98	0.36	0.64	64.3
10.93	0.38	0.62	62.5

1:0 (OTC:P)			
pH	OTC in Solution	OTC in Solid	% OTC Sorption
	mM	mM	
3.07	0.098	0.922	92.2
5.02	0.072	0.948	94.8
6.96	0.054	0.966	96.6
8.97	0.092	0.928	92.8
11.02	0.24	0.78	78

1:1 (0TC:P)			
pH	OTC in Solution	OTC in Solid	% OTC Sorption
	mM	mM	
3.07	0.118	0.902	90.2
5.02	0.096	0.924	92.4
6.96	0.058	0.962	96.2
8.97	0.098	0.922	92.2
11.02	0.26	0.76	76

1:10 (OTC:P)			
pH	OTC in Solution	OTC in Solid	% OTC Sorption
	mM	mM	
3.07	0.15	0.87	87
5.02	0.138	0.882	88.2
6.96	0.138	0.882	88.2
8.97	0.198	0.822	82.2
11.02	0.28	0.74	74

1:20 (0TC:P)			
pH	OTC in Solution	OTC in Solid	% OTC Sorption
	mM	mM	
3.07	0.26	0.76	76
5.02	0.22	0.8	80
6.96	0.22	0.8	80
8.97	0.38	0.64	64
11.02	0.4	0.62	62

B.4: The effect of sulfate on the amount of TTC and OTC sorbed by Al-WTR as a function of pH and different TTC/OTC: sulfate ratios. Initial TTC concentration 1.0 mM L^{-1}, reaction time 24 h, and ionic strength buffer 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.

1:0 (TTC:S)			
pH	TTC in Solution (mM)	TTC in Solid (mM)	% TTC Sorption
3.21	0.062	0.938	93.8
5.15	0.024	0.976	97.6
7.09	0.04	0.96	96
8.98	0.066	0.934	93.4
10.93	0.158	0.842	84.2
1:1(TTC:S)			
pH	TTC in Solution (mM)	TTC in Solid (mM)	% TTC Sorption
3.21	0.064	0.936	93.6
5.15	0.026	0.974	97.4
7.09	0.042	0.958	95.8
8.98	0.068	0.932	93.2
10.93	0.16	0.84	84
1:5 (TTC:S)			
pH	TTC in Solution (mM)	TTC in Solid (mM)	% TTC Sorption
3.21	0.068	0.932	93.2
5.15	0.028	0.972	97.2
7.09	0.044	0.956	95.6
8.98	0.072	0.928	92.8
10.93	0.162	0.838	83.8
1:10 (TTC:S)			
pH	TTC in Solution (mM)	TTC in Solid (mM)	% TTC Sorption
3.21	0.072	0.928	92.8
5.15	0.034	0.966	96.6
7.09	0.048	0.952	95.2
8.98	0.076	0.924	92.4
10.93	0.17	0.83	83
1:20 (TTC:S)			
pH	TTC in Solution (mM)	TTC in Solid (mM)	% TTC Sorption
3.21	0.078	0.922	92.2
5.15	0.038	0.962	96.2
7.09	0.054	0.946	94.6
8.98	0.082	0.918	91.8
10.93	0.178	0.822	82.2

1:0 (OTC:S)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.098	0.922	92.2
5.02	0.072	0.948	94.8
6.96	0.054	0.966	96.6
8.97	0.092	0.928	92.8
11.02	0.24	0.78	78
1:1(OTC:S)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.106	0.914	91.4
5.02	0.086	0.934	93.4
6.96	0.064	0.956	95.6
8.97	0.074	0.946	94.6
11.02	0.26	0.76	76
1:5 (OTC:S)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.108	0.912	91.2
5.02	0.092	0.928	92.8
6.96	0.068	0.952	95.2
8.97	0.078	0.942	94.2
11.02	0.264	0.756	75.6
1:10 (OTC:S)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.112	0.908	90.8
5.02	0.096	0.924	92.4
6.96	0.072	0.948	94.8
8.97	0.078	0.942	94.2
11.02	0.268	0.752	75.2
1:20 (OTC:S)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.118	0.902	90.2
5.02	0.1	0.92	92
6.96	0.074	0.946	94.6
8.97	0.084	0.936	93.6
11.02	0.28	0.74	74

B.5: The effect of calcium on the amount of TTC and OTC sorbed by Al-WTR as a function of pH and different TTC/OTC: calcium ratios. Initial TTC/OTC concentration 1.0 mM L^{-1}, reaction time 24 h, and ionic strength buffer 0.01 M KCl. Data are expressed as mean of three replicates ± one standard deviation.

pH	TTC in Solution (mM)	TTC in Solid (mM)	% TTC Sorption
1:0 (TTC:Ca)			
3.21	0.062	0.938	93.8
5.15	0.024	0.976	97.6
7.09	0.04	0.96	96
8.98	0.066	0.934	93.4
10.93	0.158	0.842	84.2
1:1 (TTC:Ca)			
3.21	0.198	0.802	80.2
5.15	0.122	0.878	87.8
7.09	0.158	0.842	84.2
8.98	0.118	0.882	88.2
10.93	0.176	0.824	82.4
1:5 (TTC:Ca)			
3.21	0.228	0.772	77.2
5.15	0.188	0.812	81.2
7.09	0.204	0.796	79.6
8.98	0.14	0.86	86
10.93	0.184	0.816	81.6
1:10 (TTC:Ca)			
3.21	0.272	0.728	72.8
5.15	0.194	0.806	80.6
7.09	0.208	0.792	79.2
8.98	0.136	0.864	86.4
10.93	0.17	0.83	83
1:20 (TTC:Ca)			
3.21	0.298	0.702	70.2
5.15	0.198	0.802	80.2
7.09	0.234	0.766	76.6
8.98	0.142	0.858	85.8
10.93	0.136	0.864	86.4

1:0 (OTC:Ca)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.098	0.922	92.2
5.02	0.072	0.948	94.8
6.96	0.054	0.966	96.6
8.97	0.092	0.928	92.8
11.02	0.24	0.78	78
1:1 (OTC:Ca)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.19	0.83	83
5.02	0.156	0.864	86.4
6.96	0.198	0.822	82.2
8.97	0.156	0.864	86.4
11.02	0.198	0.822	82.2
1:5 (OTC:Ca)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.21	0.81	81
5.02	0.168	0.852	85.2
6.96	0.266	0.754	75.4
8.97	0.134	0.886	88.6
11.02	1.88	-0.86	-86
1:10 (OTC:Ca)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.22	0.8	80
5.02	0.182	0.838	83.8
6.96	0.27	0.75	75
8.97	0.132	0.888	88.8
11.02	0.1732	0.8468	84.68
1:20 (OTC:Ca)			
pH	OTC in Solution (mM)	OTC in Solid (mM)	% OTC Sorption
3.07	0.258	0.762	76.2
5.02	0.186	0.834	83.4
6.96	0.272	0.748	74.8
8.97	0.152	0.868	86.8
11.02	0.156	0.864	86.4

B.6: ATR-FTIR spectra. All spectra are normalized with the highest peak observed.

Wavenumber	TTC	Al-WTR	TTC+Al-WTR (pH 4.12)	TTC+Al-WTR (pH 6.3)	TTC+Al-WTR (pH 9.11)
1301.74	0.1695	0.0270	0.0893	0.0693	0.0493
1303.66	0.1712	0.0276	0.0895	0.0695	0.0495
1305.59	0.1717	0.0274	0.0897	0.0697	0.0497
1307.52	0.1712	0.0275	0.0900	0.0700	0.0500
1309.45	0.1695	0.0279	0.0901	0.0701	0.0501
1311.38	0.1671	0.0283	0.0902	0.0702	0.0502
1313.31	0.1645	0.0289	0.0904	0.0704	0.0504
1315.24	0.1622	0.0293	0.0907	0.0707	0.0507
1317.16	0.1608	0.0296	0.0909	0.0709	0.0509
1319.09	0.1604	0.0299	0.0910	0.0710	0.0510
1321.02	0.1612	0.0304	0.0912	0.0712	0.0512
1322.95	0.1634	0.0312	0.0915	0.0715	0.0515
1324.88	0.1666	0.0318	0.0918	0.0718	0.0518
1326.81	0.1699	0.0325	0.0920	0.0720	0.0520
1328.74	0.1722	0.0329	0.0924	0.0724	0.0524
1330.66	0.1729	0.0338	0.0927	0.0727	0.0527
1332.59	0.1713	0.0350	0.0930	0.0730	0.0530
1334.52	0.1677	0.0358	0.0935	0.0735	0.0535
1336.45	0.1626	0.0369	0.0940	0.0740	0.0540
1338.38	0.1572	0.0377	0.0945	0.0745	0.0545
1340.31	0.1523	0.0388	0.0948	0.0748	0.0548
1342.23	0.1490	0.0397	0.0952	0.0752	0.0552
1344.16	0.1470	0.0408	0.0958	0.0758	0.0558
1346.09	0.1463	0.0419	0.0963	0.0763	0.0563
1348.02	0.1463	0.0421	0.0968	0.0768	0.0568
1349.95	0.1468	0.0423	0.0972	0.0772	0.0572
1351.88	0.1478	0.0425	0.0977	0.0777	0.0577
1353.81	0.1488	0.0422	0.0981	0.0781	0.0581
1355.73	0.1497	0.0426	0.0979	0.0785	0.0585
1357.66	0.1505	0.0429	0.0976	0.0789	0.0589
1359.59	0.1510	0.0429	0.0957	0.0795	0.0595
1361.52	0.1512	0.0432	0.0956	0.0799	0.0599
1363.45	0.1509	0.0434	0.0970	0.0802	0.0602
1365.38	0.1502	0.0436	0.0974	0.0807	0.0607
1367.31	0.1493	0.0439	0.0974	0.0809	0.0609

1369.23	0.1484	0.0438	0.0982	0.0810	0.0610
1371.16	0.1481	0.0446	0.0988	0.0813	0.0613
1373.09	0.1481	0.0449	0.0995	0.0816	0.0616
1375.02	0.1486	0.0451	0.0997	0.0816	0.0616
1376.95	0.1495	0.0459	0.1005	0.0817	0.0617
1378.88	0.1503	0.0449	0.1000	0.0817	0.0617
1380.80	0.1505	0.0448	0.1004	0.0816	0.0616
1382.73	0.1498	0.0449	0.1009	0.0813	0.0613
1384.66	0.1486	0.0451	0.1010	0.0812	0.0612
1386.59	0.1474	0.0459	0.1009	0.0808	0.0608
1388.52	0.1458	0.0462	0.1010	0.0803	0.0603
1390.45	0.1439	0.0470	0.1011	0.0801	0.0605
1392.38	0.1417	0.0449	0.1011	0.0798	0.0610
1394.30	0.1392	0.0451	0.1007	0.0797	0.0613
1396.23	0.1371	0.0455	0.1008	0.0798	0.0619
1398.16	0.1356	0.0465	0.1011	0.0801	0.0620
1400.09	0.1342	0.0468	0.1021	0.0811	0.0620
1402.02	0.1336	0.0470	0.1030	0.0820	0.0621
1403.95	0.1331	0.0475	0.1033	0.0823	0.0621
1405.88	0.1328	0.0481	0.1025	0.0815	0.0620
1407.80	0.1327	0.0483	0.1016	0.0806	0.0613
1409.73	0.1328	0.0485	0.1015	0.0805	0.0613
1411.66	0.1331	0.0489	0.1018	0.0808	0.0618
1413.59	0.1341	0.0491	0.1029	0.0819	0.0628
1415.52	0.1350	0.0493	0.1044	0.0834	0.0638
1417.45	0.1353	0.0495	0.1067	0.0857	0.0647
1419.37	0.1345	0.0496	0.1097	0.0887	0.0655
1421.30	0.1336	0.0498	0.1136	0.0926	0.0660
1423.23	0.1335	0.0496	0.1177	0.0967	0.0662
1425.16	0.1338	0.0488	0.1223	0.1013	0.0659
1427.09	0.1349	0.0485	0.1270	0.1060	0.0652
1429.02	0.1364	0.0479	0.1321	0.1111	0.0643
1430.95	0.1387	0.0478	0.1326	0.1126	0.0634
1432.87	0.1417	0.0489	0.1321	0.1111	0.0631
1434.80	0.1456	0.0464	0.1326	0.1106	0.0631
1436.73	0.1497	0.0459	0.1319	0.1109	0.0636
1438.66	0.1543	0.0452	0.1298	0.1088	0.0645
1440.59	0.1590	0.0452	0.1288	0.1078	0.0653

1442.52	0.1641	0.0441	0.1261	0.1051	0.0655
1444.44	0.1686	0.0433	0.1199	0.0989	0.0648
1446.37	0.1701	0.0428	0.1130	0.0920	0.0636
1448.30	0.1676	0.0422	0.1067	0.0857	0.0624
1450.23	0.1639	0.0410	0.0930	0.0829	0.0608
1452.16	0.1618	0.0405	0.0925	0.0825	0.0605
1454.09	0.1608	0.0400	0.0920	0.0825	0.0601
1456.02	0.1581	0.0399	0.0919	0.0818	0.0598
1457.94	0.1519	0.0390	0.0910	0.0802	0.0593
1459.87	0.1450	0.0391	0.0911	0.0799	0.0590
1461.80	0.1387	0.0393	0.0913	0.0799	0.0585
1463.73	0.1347	0.0393	0.0913	0.0795	0.0580
1465.66	0.1330	0.0388	0.0908	0.0787	0.0558
1467.59	0.1325	0.0388	0.0908	0.0783	0.0554
1469.52	0.1314	0.0391	0.0910	0.0782	0.0550
1471.44	0.1293	0.0390	0.0909	0.0778	0.0559
1473.37	0.1267	0.0396	0.0916	0.0769	0.0561
1475.30	0.1246	0.0400	0.0920	0.0768	0.0568
1477.23	0.1225	0.0405	0.0925	0.0766	0.0566
1479.16	0.1203	0.0423	0.0943	0.0768	0.0568
1481.09	0.1173	0.0435	0.0955	0.0770	0.0570
1483.01	0.1143	0.0430	0.0949	0.0772	0.0572
1484.94	0.1118	0.0437	0.0957	0.0776	0.0576
1486.87	0.1102	0.0448	0.0967	0.0779	0.0579
1488.80	0.1095	0.0456	0.0976	0.0781	0.0581
1490.73	0.1097	0.0472	0.0992	0.0782	0.0582
1492.66	0.1106	0.0475	0.0995	0.0790	0.0590
1494.59	0.1119	0.0498	0.1018	0.0799	0.0599
1496.51	0.1134	0.0499	0.1019	0.0804	0.0604
1498.44	0.1153	0.0518	0.1038	0.0812	0.0612
1500.37	0.1174	0.0528	0.1048	0.0826	0.0626
1502.30	0.1198	0.0533	0.1053	0.0843	0.0643
1504.23	0.1220	0.0550	0.1077	0.0867	0.0667
1506.16	0.1241	0.0573	0.1190	0.0980	0.0780
1508.09	0.1265	0.0576	0.1202	0.0992	0.0792
1510.01	0.1293	0.0578	0.1205	0.0995	0.0795
1511.94	0.1321	0.0579	0.1221	0.1011	0.0811
1513.87	0.1348	0.0581	0.1252	0.1042	0.0842

1515.80	0.1373	0.0583	0.1270	0.1060	0.0860
1517.73	0.1397	0.0589	0.1280	0.1070	0.0870
1519.66	0.1510	0.0595	0.1336	0.1103	0.0880
1521.58	0.1522	0.0600	0.1340	0.1100	0.0891
1523.51	0.1525	0.0601	0.1347	0.1117	0.0892
1525.44	0.1541	0.0602	0.1338	0.1118	0.0900
1527.37	0.1572	0.0601	0.1316	0.1106	0.0906
1529.30	0.1590	0.0602	0.1306	0.1096	0.0896
1531.23	0.1600	0.0603	0.1276	0.1066	0.0866
1533.16	0.1656	0.0605	0.1244	0.1034	0.0834
1535.08	0.1690	0.0606	0.1223	0.1013	0.0813
1537.01	0.1697	0.0607	0.1208	0.0998	0.0798
1538.94	0.1658	0.0605	0.1202	0.0992	0.0792
1540.87	0.1636	0.0606	0.1202	0.0992	0.0792
1542.80	0.1626	0.0607	0.1202	0.0992	0.0792
1544.73	0.1596	0.0609	0.1275	0.1065	0.0865
1546.66	0.1564	0.0612	0.1275	0.1065	0.0865
1548.58	0.1543	0.0615	0.1292	0.1082	0.0882
1550.51	0.1528	0.0625	0.1308	0.1098	0.0898
1552.44	0.1522	0.0628	0.1322	0.1112	0.0912
1554.37	0.1522	0.0632	0.1338	0.1128	0.0928
1556.30	0.1522	0.0600	0.1358	0.1148	0.0948
1558.23	0.1595	0.0601	0.1394	0.1184	0.0950
1560.15	0.1595	0.0602	0.1424	0.1234	0.0953
1562.08	0.1612	0.0601	0.1437	0.1247	0.0963
1564.01	0.1628	0.0602	0.1438	0.1258	0.0971
1565.94	0.1642	0.0603	0.1424	0.1244	0.0972
1567.87	0.1658	0.0605	0.1424	0.1234	0.0974
1569.80	0.1678	0.0606	0.1417	0.1237	0.0984
1571.73	0.1714	0.0607	0.1402	0.1222	0.0973
1573.65	0.1764	0.0605	0.1370	0.1160	0.0960
1575.58	0.1787	0.0606	0.1317	0.1107	0.0907
1577.51	0.1788	0.0607	0.1276	0.1066	0.0866
1579.44	0.1784	0.0609	0.1244	0.1034	0.0834
1581.37	0.1774	0.0612	0.1223	0.1013	0.0813
1583.30	0.1767	0.0615	0.1208	0.1020	0.0820
1585.23	0.1752	0.0625	0.1202	0.1012	0.0812
1587.15	0.1690	0.0628	0.1205	0.0995	0.0795

1589.08	0.1637	0.0632	0.1201	0.1011	0.0811
1591.01	0.1596	0.0638	0.1252	0.1042	0.0842
1592.94	0.1564	0.0643	0.1302	0.1092	0.0892
1594.87	0.1543	0.0648	0.1360	0.1150	0.0950
1596.80	0.1528	0.0652	0.1376	0.1166	0.0966
1598.72	0.1522	0.0659	0.1380	0.1160	0.0960
1600.65	0.1525	0.0662	0.1398	0.1168	0.0968
1602.58	0.1541	0.0665	0.1408	0.1179	0.0979
1604.51	0.1572	0.0667	0.1385	0.1155	0.0955
1606.44	0.1622	0.0669	0.1350	0.1140	0.0940
1608.37	0.1680	0.0670	0.1276	0.1066	0.0866
1610.30	0.1736	0.0671	0.1244	0.1034	0.0834
1612.22	0.1770	0.0671	0.1223	0.1013	0.0813
1614.15	0.1777	0.0665	0.1208	0.0998	0.0798
1616.08	0.1748	0.0660	0.1202	0.0992	0.0792
1618.01	0.1705	0.0645	0.1202	0.0992	0.0792
1619.94	0.1670	0.0641	0.1202	0.0992	0.0792
1621.87	0.1596	0.0643	0.1207	0.1017	0.0817
1623.80	0.1564	0.0622	0.1231	0.1021	0.0821
1625.72	0.1543	0.0612	0.1247	0.1031	0.0831
1627.65	0.1528	0.0605	0.1257	0.1032	0.0832
1629.58	0.1522	0.0589	0.1257	0.1035	0.0835
1631.51	0.1522	0.0582	0.1257	0.1036	0.0836
1633.44	0.1522	0.0571	0.1257	0.1037	0.0837
1635.37	0.1567	0.0562	0.1249	0.1039	0.0839
1637.29	0.1551	0.0561	0.1256	0.1046	0.0846
1639.22	0.1567	0.0540	0.1264	0.1054	0.0842
1641.15	0.1577	0.0534	0.1268	0.1058	0.0836
1643.08	0.1577	0.0534	0.1275	0.1064	0.0825
1645.01	0.1577	0.0517	0.1275	0.1065	0.0815
1646.94	0.1577	0.0511	0.1275	0.1065	0.0810
1648.87	0.1569	0.0497	0.1292	0.1082	0.0802
1650.79	0.1576	0.0497	0.1308	0.1098	0.0818
1652.72	0.1584	0.0499	0.1322	0.1112	0.0832
1654.65	0.1588	0.0472	0.1338	0.1128	0.0848
1656.58	0.1595	0.0466	0.1358	0.1148	0.0868
1658.51	0.1595	0.0458	0.1394	0.1184	0.0904
1660.44	0.1595	0.0452	0.1444	0.1234	0.0954

1662.37	0.1612	0.0456	0.1497	0.1277	0.0997
1664.29	0.1628	0.0432	0.1489	0.1278	0.0998
1666.22	0.1642	0.0436	0.1484	0.1274	0.0994
1668.15	0.1658	0.0421	0.1474	0.1264	0.0984
1670.08	0.1678	0.0418	0.1460	0.1250	0.0970
1672.01	0.1714	0.0412	0.1432	0.1222	0.0942
1673.94	0.1764	0.0401	0.1370	0.1160	0.0880
1675.86	0.1817	0.0396	0.1317	0.1107	0.0875
1677.79	0.1868	0.0389	0.1252	0.1042	0.0842
1679.72	0.1894	0.0383	0.1203	0.0993	0.0793
1681.65	0.1874	0.0381	0.1170	0.0960	0.0760
1683.58	0.1820	0.0385	0.1150	0.0940	0.0740
1685.51	0.1752	0.0366	0.1103	0.0933	0.0733
1687.44	0.1690	0.0362	0.1099	0.0900	0.0700
1689.36	0.1637	0.0354	0.1091	0.0896	0.0696
1691.29	0.1572	0.0348	0.1086	0.0892	0.0692
1693.22	0.1523	0.0339	0.1077	0.0892	0.0692
1695.15	0.1490	0.0333	0.1070	0.0885	0.0685
1697.08	0.1470	0.0328	0.1065	0.0877	0.0677
1699.01	0.1463	0.0336	0.1073	0.0879	0.0679
1700.94	0.1463	0.0322	0.1059	0.0866	0.0666
1702.86	0.1468	0.0319	0.1056	0.0869	0.0669
1704.79	0.1478	0.0317	0.1055	0.0868	0.0668
1706.72	0.1488	0.0311	0.1048	0.0864	0.0664
1708.65	0.1497	0.0311	0.1049	0.0864	0.0664
1710.58	0.1505	0.0306	0.1044	0.0864	0.0664
1712.51	0.1510	0.0304	0.1041	0.0863	0.0663
1714.43	0.1512	0.0298	0.1036	0.0863	0.0663
1716.36	0.1509	0.0304	0.1041	0.0859	0.0659
1718.29	0.1502	0.0294	0.1031	0.0849	0.0649
1720.22	0.1493	0.0292	0.1029	0.0846	0.0646
1722.15	0.1484	0.0287	0.1025	0.0847	0.0647
1724.08	0.1481	0.0285	0.1022	0.0843	0.0643
1726.01	0.1481	0.0284	0.1022	0.0840	0.0640
1727.93	0.1486	0.0277	0.1015	0.0839	0.0639
1729.86	0.1495	0.0274	0.1011	0.0836	0.0636
1731.79	0.1503	0.0311	0.1049	0.0834	0.0634
1733.72	0.1505	0.0306	0.1044	0.0828	0.0628

1735.65	0.1498	0.0304	0.1041	0.0823	0.0623
1737.58	0.1486	0.0298	0.1036	0.0826	0.0626

Wavenumber	TTC	Al-WTR	OTC+Al-WTR (pH 4.15)	OTC+Al-WTR (pH 6.32)	OTC+Al-WTR (pH 9.13)
1301.74	0.16953	0.0270	0.0893	0.0693	0.0493
1303.66	0.17121	0.0276	0.0895	0.0695	0.0495
1305.59	0.17174	0.0274	0.0897	0.0697	0.0497
1307.52	0.17119	0.0275	0.0900	0.0700	0.0500
1309.45	0.16951	0.0279	0.0901	0.0701	0.0501
1311.38	0.16707	0.0283	0.0902	0.0702	0.0502
1313.31	0.16446	0.0289	0.0904	0.0704	0.0504
1315.24	0.16220	0.0293	0.0907	0.0707	0.0507
1317.16	0.16078	0.0296	0.0909	0.0709	0.0509
1319.09	0.16036	0.0299	0.0910	0.0710	0.0510
1321.02	0.16124	0.0304	0.0912	0.0712	0.0512
1322.95	0.16343	0.0312	0.0915	0.0715	0.0515
1324.88	0.16664	0.0318	0.0918	0.0718	0.0518
1326.81	0.16991	0.0325	0.0920	0.0720	0.0520
1328.74	0.17224	0.0329	0.0924	0.0724	0.0524
1330.66	0.17285	0.0338	0.0927	0.0727	0.0527
1332.59	0.17133	0.0350	0.0930	0.0730	0.0530
1334.52	0.16765	0.0358	0.0935	0.0735	0.0535
1336.45	0.16260	0.0369	0.0940	0.0740	0.0540
1338.38	0.15718	0.0377	0.0945	0.0745	0.0545
1340.31	0.15235	0.0388	0.0948	0.0748	0.0548
1342.23	0.14896	0.0397	0.0952	0.0752	0.0552
1344.16	0.14704	0.0408	0.0958	0.0758	0.0558
1346.09	0.14627	0.0419	0.0963	0.0763	0.0563
1348.02	0.14634	0.0421	0.0968	0.0768	0.0568
1349.95	0.14685	0.0423	0.0972	0.0772	0.0572
1351.88	0.14776	0.0425	0.0977	0.0777	0.0577
1353.81	0.14880	0.0422	0.0981	0.0781	0.0581
1355.73	0.14973	0.0426	0.0979	0.0785	0.0585
1357.66	0.15050	0.0429	0.0976	0.0789	0.0589
1359.59	0.15104	0.0429	0.0957	0.0795	0.0595
1361.52	0.15118	0.0432	0.0956	0.0799	0.0599
1363.45	0.15086	0.0434	0.0970	0.0802	0.0602
1365.38	0.15019	0.0436	0.0974	0.0807	0.0607
1367.31	0.14929	0.0439	0.0974	0.0809	0.0609

1369.23	0.14843	0.0438	0.0982	0.0810	0.0610
1371.16	0.14805	0.0446	0.0988	0.0813	0.0613
1373.09	0.14811	0.0449	0.0995	0.0816	0.0616
1375.02	0.14860	0.0451	0.0997	0.0816	0.0616
1376.95	0.14949	0.0459	0.1005	0.0817	0.0617
1378.88	0.15034	0.0449	0.1000	0.0817	0.0617
1380.80	0.15051	0.0448	0.1004	0.0816	0.0616
1382.73	0.14975	0.0449	0.1009	0.0813	0.0613
1384.66	0.14864	0.0451	0.1010	0.0812	0.0612
1386.59	0.14738	0.0459	0.1009	0.0808	0.0608
1388.52	0.14575	0.0462	0.1010	0.0803	0.0603
1390.45	0.14386	0.0470	0.1011	0.0801	0.0605
1392.38	0.14170	0.0449	0.1011	0.0798	0.0610
1394.30	0.13924	0.0451	0.1007	0.0797	0.0613
1396.23	0.13713	0.0455	0.1008	0.0798	0.0619
1398.16	0.13555	0.0465	0.1011	0.0801	0.0620
1400.09	0.13423	0.0468	0.1021	0.0811	0.0620
1402.02	0.13358	0.0470	0.1030	0.0820	0.0621
1403.95	0.13311	0.0475	0.1033	0.0823	0.0621
1405.88	0.13278	0.0481	0.1025	0.0815	0.0620
1407.80	0.13270	0.0483	0.1016	0.0806	0.0613
1409.73	0.13276	0.0485	0.1015	0.0805	0.0613
1411.66	0.13314	0.0489	0.1018	0.0808	0.0618
1413.59	0.13406	0.0491	0.1029	0.0819	0.0628
1415.52	0.13504	0.0493	0.1044	0.0834	0.0638
1417.45	0.13534	0.0495	0.1067	0.0857	0.0647
1419.37	0.13447	0.0496	0.1097	0.0887	0.0655
1421.30	0.13364	0.0498	0.1136	0.0926	0.0660
1423.23	0.13346	0.0496	0.1177	0.0967	0.0662
1425.16	0.13385	0.0488	0.1223	0.1013	0.0659
1427.09	0.13489	0.0485	0.1270	0.1060	0.0652
1429.02	0.13642	0.0479	0.1321	0.1111	0.0643
1430.95	0.13868	0.0478	0.1326	0.1126	0.0634
1432.87	0.14174	0.0489	0.1321	0.1111	0.0631
1434.80	0.14556	0.0464	0.1326	0.1106	0.0631
1436.73	0.14970	0.0459	0.1319	0.1109	0.0636
1438.66	0.15426	0.0452	0.1298	0.1088	0.0645

1440.59	0.15901	0.0452	0.1288	0.1078	0.0653
1442.52	0.16414	0.0441	0.1261	0.1051	0.0655
1444.44	0.16864	0.0433	0.1199	0.0989	0.0648
1446.37	0.17007	0.0428	0.1130	0.0920	0.0636
1448.30	0.16762	0.0422	0.1067	0.0857	0.0624
1450.23	0.16393	0.0410	0.0930	0.0829	0.0608
1452.16	0.16180	0.0405	0.0925	0.0825	0.0605
1454.09	0.16083	0.0400	0.0920	0.0825	0.0601
1456.02	0.15807	0.0399	0.0919	0.0818	0.0598
1457.94	0.15186	0.0390	0.0910	0.0802	0.0593
1459.87	0.14499	0.0391	0.0911	0.0799	0.0590
1461.80	0.13874	0.0393	0.0913	0.0799	0.0585
1463.73	0.13474	0.0393	0.0913	0.0795	0.0580
1465.66	0.13305	0.0388	0.0908	0.0787	0.0558
1467.59	0.13254	0.0388	0.0908	0.0783	0.0554
1469.52	0.13138	0.0391	0.0910	0.0782	0.0550
1471.44	0.12926	0.0390	0.0909	0.0778	0.0559
1473.37	0.12666	0.0396	0.0916	0.0769	0.0561
1475.30	0.12458	0.0400	0.0920	0.0768	0.0568
1477.23	0.12251	0.0405	0.0925	0.0766	0.0566
1479.16	0.12026	0.0423	0.0943	0.0768	0.0568
1481.09	0.11734	0.0435	0.0955	0.0770	0.0570
1483.01	0.11432	0.0430	0.0949	0.0772	0.0572
1484.94	0.11177	0.0437	0.0957	0.0776	0.0576
1486.87	0.11016	0.0448	0.0967	0.0779	0.0579
1488.80	0.10954	0.0456	0.0976	0.0781	0.0581
1490.73	0.10975	0.0472	0.0992	0.0782	0.0582
1492.66	0.11064	0.0475	0.0995	0.0790	0.0590
1494.59	0.11193	0.0498	0.1018	0.0799	0.0599
1496.51	0.11337	0.0499	0.1019	0.0804	0.0604
1498.44	0.11527	0.0518	0.1038	0.0812	0.0612
1500.37	0.11741	0.0528	0.1048	0.0826	0.0626
1502.30	0.11975	0.0533	0.1053	0.0843	0.0643
1504.23	0.12204	0.0550	0.1077	0.0867	0.0667
1506.16	0.12407	0.0573	0.1190	0.0980	0.0780
1508.09	0.12647	0.0576	0.1202	0.0992	0.0792
1510.01	0.12931	0.0578	0.1205	0.0995	0.0795

1511.94	0.13206	0.0579	0.1221	0.1011	0.0811
1513.87	0.13480	0.0581	0.1252	0.1042	0.0842
1515.80	0.13732	0.0583	0.1270	0.1060	0.0860
1517.73	0.13969	0.0589	0.1280	0.1070	0.0870
1519.66	0.15100	0.0595	0.1336	0.1103	0.0880
1521.58	0.15221	0.0600	0.1340	0.1100	0.0891
1523.51	0.15245	0.0601	0.1347	0.1117	0.0892
1525.44	0.15414	0.0602	0.1338	0.1118	0.0900
1527.37	0.15725	0.0601	0.1316	0.1106	0.0906
1529.30	0.1590333	0.0602	0.1306	0.1096	0.0896
1531.23	0.1600333	0.0603	0.1276	0.1066	0.0866
1533.16	0.1655986	0.0605	0.1244	0.1034	0.0834
1535.08	0.169002	0.0606	0.1223	0.1013	0.0813
1537.01	0.1696776	0.0607	0.1208	0.0998	0.0798
1538.94	0.1657931	0.0605	0.1202	0.0992	0.0792
1540.87	0.1636416	0.0606	0.1202	0.0992	0.0792
1542.80	0.16255	0.0607	0.1202	0.0992	0.0792
1544.73	0.15957	0.0609	0.1275	0.1065	0.0865
1546.66	0.15641	0.0612	0.1275	0.1065	0.0865
1548.58	0.15430	0.0615	0.1292	0.1082	0.0882
1550.51	0.15280	0.0625	0.1308	0.1098	0.0898
1552.44	0.15221	0.0628	0.1322	0.1112	0.0912
1554.37	0.15221	0.0632	0.1338	0.1128	0.0928
1556.30	0.15221	0.0600	0.1358	0.1148	0.0948
1558.23	0.15945	0.0601	0.1394	0.1184	0.0950
1560.15	0.15948	0.0602	0.1424	0.1234	0.0953
1562.08	0.16117	0.0601	0.1437	0.1247	0.0963
1564.01	0.16282	0.0602	0.1438	0.1258	0.0971
1565.94	0.16421	0.0603	0.1424	0.1244	0.0972
1567.87	0.16577	0.0605	0.1424	0.1234	0.0974
1569.80	0.16779	0.0606	0.1417	0.1237	0.0984
1571.73	0.17137	0.0607	0.1402	0.1222	0.0973
1573.65	0.17636	0.0605	0.1370	0.1160	0.0960
1575.58	0.17871	0.0606	0.1317	0.1107	0.0907
1577.51	0.17879	0.0607	0.1276	0.1066	0.0866
1579.44	0.17837	0.0609	0.1244	0.1034	0.0834
1581.37	0.17736	0.0612	0.1223	0.1013	0.0813

1583.30	0.17668	0.0615	0.1208	0.1020	0.0820
1585.23	0.17525	0.0625	0.1202	0.1012	0.0812
1587.15	0.16900	0.0628	0.1205	0.0995	0.0795
1589.08	0.16365	0.0632	0.1201	0.1011	0.0811
1591.01	0.15957	0.0638	0.1252	0.1042	0.0842
1592.94	0.15641	0.0643	0.1302	0.1092	0.0892
1594.87	0.15430	0.0648	0.1360	0.1150	0.0950
1596.80	0.15280	0.0652	0.1376	0.1166	0.0966
1598.72	0.15221	0.0659	0.1380	0.1160	0.0960
1600.65	0.15245	0.0662	0.1398	0.1168	0.0968
1602.58	0.15414	0.0665	0.1408	0.1179	0.0979
1604.51	0.15725	0.0667	0.1385	0.1155	0.0955
1606.44	0.16221	0.0669	0.1350	0.1140	0.0940
1608.37	0.16803	0.0670	0.1276	0.1066	0.0866
1610.30	0.17360	0.0671	0.1244	0.1034	0.0834
1612.22	0.17700	0.0671	0.1223	0.1013	0.0813
1614.15	0.17768	0.0665	0.1208	0.0998	0.0798
1616.08	0.17479	0.0660	0.1202	0.0992	0.0792
1618.01	0.17054	0.0645	0.1202	0.0992	0.0792
1619.94	0.16702	0.0641	0.1202	0.0992	0.0792
1621.87	0.15957	0.0643	0.1207	0.1017	0.0817
1623.80	0.15641	0.0622	0.1231	0.1021	0.0821
1625.72	0.15430	0.0612	0.1247	0.1031	0.0831
1627.65	0.15280	0.0605	0.1257	0.1032	0.0832
1629.58	0.15221	0.0589	0.1257	0.1035	0.0835
1631.51	0.15221	0.0582	0.1257	0.1036	0.0836
1633.44	0.15221	0.0571	0.1257	0.1037	0.0837
1635.37	0.15672	0.0562	0.1249	0.1039	0.0839
1637.29	0.15512	0.0561	0.1256	0.1046	0.0846
1639.22	0.15672	0.0540	0.1264	0.1054	0.0842
1641.15	0.15772	0.0534	0.1268	0.1058	0.0836
1643.08	0.15772	0.0534	0.1275	0.1064	0.0825
1645.01	0.15772	0.0517	0.1275	0.1065	0.0815
1646.94	0.15772	0.0511	0.1275	0.1065	0.0810
1648.87	0.15687	0.0497	0.1292	0.1082	0.0802
1650.79	0.15757	0.0497	0.1308	0.1098	0.0818
1652.72	0.15837	0.0499	0.1322	0.1112	0.0832

1654.65	0.15880	0.0472	0.1338	0.1128	0.0848
1656.58	0.15948	0.0466	0.1358	0.1148	0.0868
1658.51	0.15945	0.0458	0.1394	0.1184	0.0904
1660.44	0.15948	0.0452	0.1444	0.1234	0.0954
1662.37	0.16117	0.0456	0.1497	0.1277	0.0997
1664.29	0.16282	0.0432	0.1489	0.1278	0.0998
1666.22	0.16421	0.0436	0.1484	0.1274	0.0994
1668.15	0.16577	0.0421	0.1474	0.1264	0.0984
1670.08	0.16779	0.0418	0.1460	0.1250	0.0970
1672.01	0.17137	0.0412	0.1432	0.1222	0.0942
1673.94	0.17636	0.0401	0.1370	0.1160	0.0880
1675.86	0.18171	0.0396	0.1317	0.1107	0.0875
1677.79	0.18679	0.0389	0.1252	0.1042	0.0842
1679.72	0.18937	0.0383	0.1203	0.0993	0.0793
1681.65	0.18736	0.0381	0.1170	0.0960	0.0760
1683.58	0.18197	0.0385	0.1150	0.0940	0.0740
1685.51	0.17525	0.0366	0.1103	0.0933	0.0733
1687.44	0.16900	0.0362	0.1099	0.0900	0.0700
1689.36	0.16365	0.0354	0.1091	0.0896	0.0696
1691.29	0.15718	0.0348	0.1086	0.0892	0.0692
1693.22	0.15235	0.0339	0.1077	0.0892	0.0692
1695.15	0.14896	0.0333	0.1070	0.0885	0.0685
1697.08	0.14704	0.0328	0.1065	0.0877	0.0677
1699.01	0.14627	0.0336	0.1073	0.0879	0.0679
1700.94	0.14634	0.0322	0.1059	0.0866	0.0666
1702.86	0.14685	0.0319	0.1056	0.0869	0.0669
1704.79	0.14776	0.0317	0.1055	0.0868	0.0668
1706.72	0.14880	0.0311	0.1048	0.0864	0.0664
1708.65	0.14973	0.0311	0.1049	0.0864	0.0664
1710.58	0.15050	0.0306	0.1044	0.0864	0.0664
1712.51	0.15104	0.0304	0.1041	0.0863	0.0663
1714.43	0.15118	0.0298	0.1036	0.0863	0.0663
1716.36	0.15086	0.0304	0.1041	0.0859	0.0659
1718.29	0.15019	0.0294	0.1031	0.0849	0.0649
1720.22	0.14929	0.0292	0.1029	0.0846	0.0646
1722.15	0.14843	0.0287	0.1025	0.0847	0.0647
1724.08	0.14805	0.0285	0.1022	0.0843	0.0643

1726.01	0.14811	0.0284	0.1022	0.0840	0.0640
1727.93	0.14860	0.0277	0.1015	0.0839	0.0639
1729.86	0.14949	0.0274	0.1011	0.0836	0.0636
1731.79	0.15034	0.0311	0.1049	0.0834	0.0634
1733.72	0.15051	0.0306	0.1044	0.0828	0.0628
1735.65	0.14975	0.0304	0.1041	0.0823	0.0623

Appendix C

C.1: TTC and OTC sorption by Al-WTR amended manure at different Al-WTR rates as a function of contact time and initial TTC and OTC loads.

Time (D)	Initial TTC concentration (2.25mg kg^{-1})			Initial TTC concentration (22.5mg kg^{-1})		
	Manure control Al-WTR 0%	Manure + Al-WTR 2.5%	Manure + Al-WTR 5%	Manure control Al-WTR 0%	Manure +Al-WTR 2.5%	Manure +Al-WTR 5%
	TTC concentration (mM)					
0	0.741	0.782	0.787	8.94	8.63	8.337
1	0.753	0.68	0.673	8.683	7.197	6.7
2	0.753	0.599	0.582	8.51	6.617	5.777
4	0.742	0.447	0.378	8.237	5.527	4.577
7	0.734	0.348	0.291	8.346	4.38	3.297
10	0.713	0.29	0.247	7.83	2.94	2.39
14	0.691	0.211	0.139	7.65	2.06	1.34
21	0.68	0.193	0.118	7.5	1.84	1.22
25	0.671	0.175	0.105	7.38	1.69	1.08
30	0.642	0.162	0.089	7.27	1.55	0.98
60	0.636	0.151	0.08	7.23	1.49	0.92
90	0.629	0.145	0.06	7.24	1.44	0.88

Time (D)	Initial OTC concentration (2.25mg kg^{-1})			Initial OTC concentration (22.5mg kg^{-1})		
	Manure control Al-WTR 0%	Manure + Al-WTR 2.5%	Manure + Al-WTR 5%	Manure control Al-WTR 0%	Manure +Al-WTR 2.5%	Manure +Al-WTR 5%
	OTC concentration (mM)					
0	0.793	0.837	0.840	9.100	8.800	8.467
1	0.780	0.703	0.693	8.933	7.467	6.900
2	0.773	0.620	0.600	8.730	6.867	5.967
4	0.760	0.467	0.393	8.467	5.767	4.767
7	0.750	0.367	0.307	8.367	4.600	3.467
10	0.73	0.31	0.26	8.03	3.17	2.57
14	0.71	0.23	0.15	7.83	2.27	1.50
21	0.70	0.21	0.13	7.70	2.10	1.37
25	0.69	0.19	0.12	7.57	1.93	1.23
30	0.67	0.18	0.10	7.43	1.77	1.10
60	0.65	0.17	0.09	7.38	1.70	1.02
90	0.64	0.16	0.07	7.35	1.62	0.98

C.2: TTC and OTC sorption by Al-WTR amended Immokalee soil and manure amended Immokalee soil at different Al-WTR rates as a function of contact time and initial TTC and OTC loads.

Time (D)	TTC Control	Initial TTC concentration (2.25mg kg^{-1})					
		Immokalee control Al-WTR 0%	Immokalee Al-WTR 2.5%	Immokalee Al-WTR 5%	Immokalee + Manure control Al-WTR 0%	Immokalee + Manure Al-WTR 2.5%	Immokalee + Manure Al-WTR 5%
		TTC concentration (mM)					
0	1.06	1.053	0.875	0.875	1.116	1.117	1.042
1	1.04	1.01	0.726	0.67	1.065	1.028	1.017
2	1.02	1.008	0.628	0.52	1.052	0.987	0.894
4	1.00	0.995	0.463	0.381	1.043	0.932	0.735
7	1.00	0.986	0.379	0.286	1.023	0.813	0.499
10	0.98	0.963	0.31	0.217	0.991	0.559	0.447
14	0.97	0.941	0.229	0.139	0.972	0.515	0.409
21	0.96	0.932	0.207	0.12	0.961	0.413	0.37
25	0.95	0.915	0.195	0.099	0.95	0.412	0.359
30	0.93	0.901	0.178	0.088	1.02	0.401	0.345
60	0.93	0.89	0.169	0.064	0.91	0.39	0.333
90	0.92	0.87	0.145	0.061	0.9	0.37	0.32

Time (D)	OTC Control	Initial OTC concentration (2.25mg kg^{-1})					
		Immokalee control Al-WTR 0%	Immokalee Al-WTR 2.5%	Immokalee Al-WTR 5%	Immokalee + Manure control Al-WTR 0%	Immokalee + Manure Al-WTR 2.5%	Immokalee + Manure Al-WTR 5%
		OTC concentration (mM)					
0	1.01	1.00	0.820	0.823	1.063	1.067	1.093
1	1.00	0.99	0.703	0.643	1.047	1.007	0.995
2	0.99	0.99	0.607	0.500	1.037	0.967	0.873
4	0.98	0.98	0.443	0.363	1.027	0.913	0.717
7	0.98	0.97	0.360	0.270	1.010	0.793	0.483
10	0.97	0.95	0.29	0.20	0.98	0.54	0.43
14	0.96	0.93	0.21	0.12	0.96	0.50	0.39
21	0.95	0.92	0.19	0.10	0.95	0.40	0.35
25	0.94	0.90	0.18	0.08	0.94	0.40	0.34
30	0.92	0.89	0.16	0.07	0.92	0.39	0.33
60	0.92	0.88	0.15	0.05	0.90	0.38	0.32
90	0.91	0.86	0.13	0.05	0.89	0.36	0.31

C.3: TTC and OTC sorption by Al-WTR amended Immokalee soil and manure amended Immokalee soil at different Al-WTR rates as a function of contact time and initial TTC and OTC

Time (D)	Initial TTC concentration (22.5mg kg^{-1})					
	Immokalee control Al-WTR 0%	Immokalee Al-WTR 2.5%	Immokalee Al-WTR 5%	Immokalee + Manure control Al-WTR 0%	Immokalee + Manure Al-WTR 2.5%	Immokalee + Manure Al-WTR 5%
	TTC concentration (mM)					
0	10.43	8.937	9.06	10.843	10.624	10.181
1	10.37	7.87	7.217	10.793	9.457	8.04
2	10.32	6.683	5.387	10.673	8.99	6.12
4	10.19	4.74	3.53	10.553	8.137	5.063
7	10.1	3.353	2.421	10.377	5.213	3.79
10	9.85	2.33	1.9	10.15	5.02	3.66
14	9.63	1.24	1.05	9.95	4.01	3.32
21	9.48	1.19	0.97	9.83	3.98	3.21
25	9.35	1.04	0.79	9.69	3.78	2.94
30	9.19	0.89	0.59	9.53	3.62	2.82
60	9.11	0.85	0.57	9.42	3.46	2.61
90	9.1	0.79	0.52	9.28	3.42	2.66

Time (D)	Initial OTC concentration (22.5mg kg^{-1})					
	Immokalee control Al-WTR 0%	Immokalee Al-WTR 2.5%	Immokalee Al-WTR 5%	Immokalee + Manure control Al-WTR 0%	Immokalee + Manure Al-WTR 2.5%	Immokalee + Manure Al-WTR 5%
	OTC concentration (mM)					
0	10.30	8.767	8.900	10.633	10.667	10.933
1	10.17	7.600	6.967	10.533	9.267	7.800
2	10.13	6.433	5.167	10.433	8.800	5.900
4	10.00	4.500	3.300	10.333	7.967	4.833
7	9.93	3.133	2.400	10.167	5.033	3.600
10	9.67	2.10	1.70	9.97	4.83	3.50
14	9.47	1.03	0.87	9.77	3.85	3.17
21	9.33	0.93	0.77	9.63	3.83	3.10
25	9.20	0.80	0.60	9.50	3.67	2.83
30	9.07	0.67	0.43	9.37	3.50	2.70
60	9.01	0.64	0.42	9.27	3.45	2.60
90	9.00	0.61	0.41	9.17	3.41	2.65

C.4: TTC and OTC sorption by Al-WTR amended Belleglade soil and manure amended Belleglade soil at different Al-WTR rates as a function of time and initial load.

Time (D)	Initial TTC concentration (2.25mg kg⁻¹)					
	Belleglade control Al-WTR 0%	Belleglade Al-WTR 2.5%	Belleglade Al-WTR 5%	Belleglade + Manure control Al-WTR 0%	Belleglade + Manure Al-WTR 2.5%	Belleglade + Manure Al-WTR 5%
	TTC concentration (mM)					
0	1.06	0.859	0.85	1.145	1.134	1.118
1	0.972	0.764	0.718	1.124	0.958	0.87
2	0.929	0.71	0.625	1.108	0.825	0.726
4	0.917	0.602	0.466	1.086	0.696	0.603
7	0.906	0.507	0.39	1.08	0.523	0.449
10	0.895	0.469	0.321	1.069	0.421	0.366
14	0.883	0.437	0.222	1.05	0.322	0.284
21	0.882	0.405	0.205	1.039	0.305	0.262
25	0.871	0.383	0.181	1.015	0.281	0.241
30	0.861	0.362	0.17	1.003	0.273	0.23
60	0.85	0.331	0.17	1.001	0.27	0.22
90	0.84	0.321	0.17	1	0.269	0.21
Time (D)	Initial OTC concentration (2.25mg kg⁻¹)					
	Belleglade control Al-WTR 0%	Belleglade Al-WTR 2.5%	Belleglade Al-WTR 5%	Belleglade + Manure control Al-WTR 0%	Belleglade + Manure Al-WTR 2.5%	Belleglade + Manure Al-WTR 5%
	OTC concentration (mM)					
0	1.037	0.837	0.830	1.123	1.113	1.137
1	0.952	0.743	0.700	1.103	0.940	0.850
2	0.910	0.690	0.610	1.090	0.810	0.707
4	0.897	0.583	0.450	1.070	0.680	0.583
7	0.887	0.487	0.377	1.063	0.510	0.430
10	0.88	0.45	0.31	1.05	0.41	0.35
14	0.87	0.42	0.21	1.03	0.31	0.27
21	0.87	0.39	0.19	1.02	0.29	0.25
25	0.86	0.37	0.17	1.00	0.27	0.23
30	0.85	0.35	0.16	0.99	0.26	0.21
60	0.84	0.32	0.16	0.99	0.26	0.21
90	0.83	0.31	0.16	0.99	0.26	0.21

The initial TTC/OTC concentration superscript is 2.25mg kg^{-1}.

C.5: TTC and OTC sorption by Al-WTR amended Belleglade soil and manure amended Belleglade soil at different Al-WTR rates as a function of contact time and initial TTC and OTC loads.

Time (D)	Initial TTC concentration (22.5mg kg^{-1})					
	Belleglade control Al-WTR 0%	Belleglade Al-WTR 2.5%	Belleglade Al-WTR 5%	Belleglade + Manure control Al-WTR 0%	Belleglade + Manure Al-WTR 2.5%	Belleglade + Manure Al-WTR 5%
	TTC concentration (mM)					
0	8.554	8.652	8.716	11.286	11.188	11.019
1	8.42	7.389	6.922	11.087	9.39	8.694
2	8.385	6.888	6.054	10.951	8.154	6.82
4	8.249	5.952	4.918	10.882	6.787	4.951
7	8.046	5.051	3.783	10.749	5.386	3.883
10	7.881	4.086	2.847	10.583	4.05	2.917
14	7.682	3.245	2.016	10.381	3.219	2.019
21	7.511	3.083	1.85	10.242	3.047	1.848
25	7.34	2.912	1.689	10.115	2.885	1.689
30	7.27	2.741	1.515	9.981	2.718	1.548
60	7.18	2.74	1.513	9.98	2.719	1.544
90	7.18	2.74	1.51	9.98	2.715	1.541

Time (D)	Initial OTC concentration (22.5mg kg^{-1})					
	Belleglade control Al-WTR 0%	Belleglade Al-WTR 2.5%	Belleglade Al-WTR 5%	Belleglade + Manure control Al-WTR 0%	Belleglade + Manure Al-WTR 2.5%	Belleglade + Manure Al-WTR 5%
	OTC concentration (mM)					
0	8.500	8.600	8.667	11.233	11.133	11.367
1	8.400	7.367	6.900	11.067	9.367	8.667
2	8.367	6.867	6.033	10.933	8.133	6.800
4	8.233	5.933	4.900	10.867	6.767	4.933
7	8.033	5.033	3.767	10.733	5.367	3.867
10	7.87	4.07	2.83	10.57	4.03	2.90
14	7.67	3.23	2.00	10.37	3.20	2.00
21	7.50	3.07	1.83	10.23	3.03	1.83
25	7.33	2.90	1.67	10.10	2.87	1.67
30	7.17	2.73	1.50	9.97	2.70	1.53
60	7.17	2.73	1.50	9.97	2.70	1.53
90	7.17	2.73	1.50	9.97	2.70	1.53

C.6: Effect of competing agricultural species (phosphate) on TTC and OTC sorption by Al-WTR amended Immokalee soil as a function of contact time different TTC/OTC:P molar ratios

Time (D)	TTC Control	Immokalee control Al-WTR 0%	Imm without P Al-WTR 5%	Imm + 1mM P Al-WTR 5%	Imm + 5mM P Al-WTR 5%	Imm + 10mM P Al-WTR 5%
TTC sorption/immobilization (% on initial concentration)						
0	1.07	0.52	2.25	3.50	3.30	3.70
1	1.05	1.45	23.25	12.85	9.25	6.35
2	1.02	2.20	41.42	25.45	22.35	10.98
4	1.00	2.85	56.85	43.26	36.20	15.57
7	0.99	3.29	68.44	55.76	56.89	26.30
10	0.98	5.84	76.29	63.27	62.50	51.20
14	0.98	8.23	86.70	65.62	65.90	55.40
21	0.96	9.15	88.20	75.10	71.24	63.80
25	0.94	10.38	89.95	76.90	72.20	63.96
30	0.91	11.55	91.65	79.20	72.26	64.22
60	0.89	12.49	92.70	80.70	76.30	65.08
90	0.89	15.20	93.20	80.78	78.20	66.42

Time (D)	OTC Control	Immokalee control Al-WTR 0%	Imm without P Al-WTR 5%	Imm + 1mM P Al-WTR 5%	Imm + 5mM P Al-WTR 5%	Imm + 10mM P Al-WTR 5%
OTC sorption/immobilization (% on initial concentration)						
0	1.01	0.58	2.35	3.30	2.90	3.55
1	1.00	1.00	21.86	13.04	8.99	5.59
2	0.99	1.66	39.27	26.33	20.12	9.34
4	0.98	2.66	55.87	42.83	34.45	14.38
7	0.98	3.32	67.21	54.11	55.79	25.63
10	0.97	5.32	75.71	62.56	60.76	49.50
14	0.96	7.31	85.83	67.79	64.33	53.44
21	0.95	8.64	87.85	74.64	69.82	62.50
25	0.94	9.97	89.88	76.65	71.65	62.50
30	0.92	10.96	91.50	78.26	72.56	63.44
60	0.92	12.29	93.93	80.68	77.13	64.38
90	0.91	14.30	93.93	80.68	78.05	66.25

C.7: Effect of competing agricultural species (phosphate) on TTC and OTC sorption by Al-WTR amended Belleglade soil as a function of contact time different TTC/OTC:P molar ratios

Time (D)	TTC Control	Belleglade control Al-WTR 0%	Belleg without P Al-WTR 5%	Belleg + 1mM P Al-WTR 5%	Belleg + 5mM P Al-WTR 5%	Belleg+ 10mM P Al-WTR 5%
0	1.07	0.78	4.28	4.08	3.76	3.65
1	1.05	1.52	22.58	14.3	9.55	7.2
2	1.02	2.32	40.45	27.55	19.53	12.62
4	1.00	3.35	56.82	43.82	33.55	18.4
7	0.99	4.5	68.5	55.55	52.45	28.46
10	0.98	6.23	76.45	64.35	60.53	38.5
14	0.98	8.45	86.2	68.3	64.35	45.8
21	0.96	10.3	87.9	75.45	68.5	55.9
25	0.94	12.58	89.94	77.35	70.25	62.55
30	0.91	13.7	90.55	79.76	71.55	65.8
60	0.89	14.38	92.7	86.2	71.92	69.41
90	0.89	15.24	93.73	85.83	75.45	70.2

Time (D)	OTC Control	Belleglade control Al-WTR 0%	Belleg without P Al-WTR 5%	Belleg + 1mM P Al-WTR 5%	Belleg + 5mM P Al-WTR 5%	Belleg+ 10mM P Al-WTR 5%
0	1.01	0.85	3.95	3.80	2.72	2.60
1	1.00	1.88	22.50	14.58	12.40	7.35
2	0.99	2.45	42.30	28.47	23.55	10.55
4	0.98	4.52	58.74	42.89	37.25	15.62
7	0.98	6.53	69.50	55.75	48.20	27.54
10	0.97	8.55	78.30	64.32	58.65	51.30
14	0.96	10.20	87.25	68.72	63.22	54.70
21	0.95	12.65	89.32	75.30	69.75	62.50
25	0.94	15.75	90.55	76.70	72.30	66.35
30	0.92	16.20	92.50	79.45	72.95	68.05
60	0.92	16.80	94.20	85.32	74.60	69.85
90	0.91	17.50	94.55	86.40	75.90	70.50

C.8: Effect of competing agricultural species (sulfate) on TTC and OTC sorption by Al-WTR amended Immokalee soil as a function of contact time different TTC/OTC: sulfate molar ratios

Time (D)	TTC Control	Immokalee control Al-WTR 0%	Imm without S Al-WTR 5%	Imm + 1mM S Al-WTR 5%	Imm + 5mM S Al-WTR 5%	Imm + 10mM S Al-WTR 5%
0	1.07	0.52	2.25	2.04	2.1	1.96
1	1.05	1.45	23.25	21.8	20.7	18.5
2	1.02	2.2	41.42	39.55	38.4	36.2
4	1.00	2.85	56.85	55.75	54.75	52.8
7	0.99	3.29	68.44	66.9	65.5	64.45
10	0.98	5.84	76.29	74.85	73.8	72.6
14	0.98	8.23	86.70	74.55	72.4	71.85
21	0.96	9.15	88.20	86.23	85.4	85.2
25	0.94	10.38	89.95	87.56	86.2	86.5
30	0.91	11.55	91.65	89.5	88.5	87.2
60	0.89	12.49	92.70	89.95	89.28	88.69
90	0.89	15.2	93.20	90.6	90.2	89.2

Time (D)	OTC Control	Immokalee control Al-WTR 0%	Imm without S Al-WTR 5%	Imm + 1mM S Al-WTR 5%	Imm + 5mM S Al-WTR 5%	Imm + 10mM S Al-WTR 5%
0	1.01	0.58	2.35	2.35	2.15	2.06
1	1.00	1.00	21.86	21.66	21.42	21.05
2	0.99	1.66	39.27	38.66	38.42	38.06
4	0.98	2.66	55.87	55.30	55.06	54.45
7	0.98	3.32	67.21	66.60	66.23	65.38
10	0.97	5.32	75.71	75.10	73.89	72.06
14	0.96	7.31	85.83	81.78	80.57	78.14
21	0.95	8.64	87.85	85.43	84.21	81.78
25	0.94	9.97	89.88	89.07	87.85	85.43
30	0.92	10.96	91.50	89.07	87.85	85.43
60	0.92	12.29	93.93	91.50	90.28	87.85
90	0.91	14.30	93.93	91.50	90.28	87.95

C.9: Effect of competing agricultural species (sulfate) on TTC and OTC sorption by Al-WTR amended Belleglade soil as a function of contact time different TTC/OTC: sulfate molar ratios

Time (D)	TTC Control	Belleglade control Al-WTR 0%	Belleg without S Al-WTR 5%	Belleg + 1mM S Al-WTR 5%	Belleg+ 5mM S Al-WTR 5%	Belleg + 10mM S Al-WTR 5%
0	1.07	0.78	4.28	3.78	3.55	3.45
1	1.05	1.52	22.58	21.85	20.8	20.4
2	1.02	2.32	40.45	38.79	38.45	37.58
4	1.00	3.35	56.82	55.45	54.95	54.6
7	0.99	4.5	68.5	67.6	66.8	65.5
10	0.98	6.23	76.45	75.4	73.25	72.5
14	0.98	8.45	86.2	85.7	84.2	82.25
21	0.96	10.3	87.9	86.2	85.5	83.75
25	0.94	12.58	89.94	87.8	86.4	84.9
30	0.91	13.7	90.55	88.7	87.5	86.1
60	0.89	14.38	92.7	89.95	88.76	88.55
90	0.89	15.24	93.73	91.2	89.6	89.6

Time (D)	OTC Control	Belleglade control Al-WTR 0%	Belleg without S Al-WTR 5%	Belleg + 1mM S Al-WTR 5%	Belleg+ 5mM S Al-WTR 5%	Belleg + 10mM S Al-WTR 5%
0	1.01	0.85	3.95	3.800	3.720	3.500
1	1.00	1.88	22.50	21.75	20.59	19.89
2	0.99	2.45	42.30	39.60	38.59	37.65
4	0.98	4.52	58.74	56.75	54.96	54.20
7	0.98	6.53	69.50	67.20	66.75	64.98
10	0.97	8.55	78.30	76.50	74.60	72.50
14	0.96	10.20	87.25	83.80	81.30	79.65
21	0.95	12.65	89.32	86.40	84.68	81.30
25	0.94	15.75	90.55	88.32	86.95	84.90
30	0.92	16.20	92.50	90.35	88.20	86.20
60	0.92	16.80	94.20	92.40	89.50	87.80
90	0.91	17.50	94.55	93.20	90.20	87.90

Appendix D

D.1: Effect of Al-WTR application rates on immobilization of TTC at two different initial concentrations as a function of equilibration time in Immokalee soil and Immokalee soil amended with TTC rich manure.

TTC 2.25 mg/kg	TTC mobility/accessibility (% initial concentration)				
Time Period (Year)	Control (No Al-WTR)	Imm +25 g kg^{-1} Al-WTR	Imm +50 g kg^{-1} Al-WTR	Imm-Manure +25 g kg^{-1} Al-WTR	Imm-Manure +50 g kg^{-1} Al-WTR
0	99.2	95.4	94.2	95.4	94.2
0.25	96.1	80.2	70.4	75.3	68.4
0.5	95.2	68.2	49.3	57.5	42.5
0.75	82.6	40.5	24.5	31.5	17.5
1	78.4	33.4	18.2	25.4	13.4
TTC 22.5 mg/kg	TTC mobility/accessibility (% initial concentration)				
Time Period (Year)	Control (No Al-WTR)	Imm +25 g kg^{-1} Al-WTR	Imm +50 g kg^{-1} Al-WTR	Imm-Manure +25 g kg^{-1} Al-WTR	Imm-Manure +50 g kg^{-1} Al-WTR
0	99.1	94.5	93.1	95.2	94.2
0.25	95.1	83.2	73.5	80.2	70.3
0.5	92.5	75.2	52.2	60.3	47.5
0.75	85.5	45.5	28.7	33.6	20.2
1	80.2	35.4	19.5	27.2	14.5

258

D.2: Effect of Al-WTR application rates on immobilization of TTC at two different initial concentrations as a function of equilibration time in Belleglade soil and Belleglade soil amended with TTC rich manure.

TTC 2.25 mg/kg	TTC mobility/accessibility (% initial concentration)				
Time Period (Year)	Control (No Al-WTR)	Belle +25 g kg^{-1} Al-WTR	Belle +50 g kg^{-1} Al-WTR	Belle-Manure +25 g kg^{-1} Al-WTR	Belle-Manure +50 g kg^{-1} Al-WTR
0	98.3	95.3	94.8	95.4	94.2
0.25	92.8	78.2	69.2	72.3	63.2
0.5	90.4	65.2	44.1	56.4	40.5
0.75	82.6	37.5	20.1	28.5	16.5
1	75.2	28.2	16.3	25.4	13.4
TTC 22.5 mg/kg	TTC mobility/accessibility (% initial concentration)				
Time Period (Year)	Control (No Al-WTR)	Belle +25 g kg^{-1} Al-WTR	Belle +50 g kg^{-1} Al-WTR	Belle-Manure +25 g kg^{-1} Al-WTR	Belle-Manure +50 g kg^{-1} Al-WTR
0	98.1	96.2	94.7	92.4	93.2
0.25	93.2	79.4	71.2	74.6	65.4
0.5	89.9	67.5	47.4	58.9	42.1
0.75	84.5	41.4	22.5	28.5	17.8
1	76.3	21.5	16.4	20.4	14.5

D.3: Effect of Al-WTR application rates on immobilization of OTC at two different initial concentrations as a function of equilibration time in Immokalee soil and Immokalee soil amended with OTC rich manure

OTC 2.25 mg/kg	OTC mobility/accessibility (% initial concentration)				
Time Period (Year)	Control (No Al-WTR)	Imm +25 g kg^{-1} Al-WTR	Imm +50 g kg^{-1} Al-WTR	Imm-Manure +25 g kg^{-1} Al-WTR	Imm-Manure +50 g kg^{-1} Al-WTR
0	98.5	94.6	93.4	94.5	93.4
0.25	95.5	79.6	69.8	74.7	67.8
0.5	94.3	67.5	48.5	56.5	41.5
0.75	81.7	39.6	23.6	30.6	16.6
1	77.7	32.7	17.5	24.7	12.7
OTC 22.5 mg/kg	OTC mobility/accessibility (% initial concentration)				
Time Period (Year)	Control (No Al-WTR)	Imm +25 g kg^{-1} Al-WTR	Imm +50 g kg^{-1} Al-WTR	Imm-Manure +25 g kg^{-1} Al-WTR	Imm-Manure +50 g kg^{-1} Al-WTR
0	98.4	93.8	92.4	94.5	93.5
0.25	94.2	82.3	72.6	79.3	69.4
0.5	91.3	74	51	59.1	46.3
0.75	84.2	44.2	27.4	32.3	18.9
1	78.7	33.9	18	25.7	13

D.4: Effect of Al-WTR application rates on immobilization of OTC at two different initial concentrations as a function of equilibration time in Belleglade soil and Belleglade soil amended with OTC rich manure.

OTC 2.25 mg/kg	OTC mobility/accessibility (% initial concentration)				
Time Period (Year)	Control (No Al-WTR)	Belle +25 g kg^{-1} Al-WTR	Belle +50 g kg^{-1} Al-WTR	Belle-Manure +25 g kg^{-1} Al-WTR	Belle-Manure +50 g kg^{-1} Al-WTR
0	97.5	94.5	94	94.6	93.4
0.25	91.9	77.3	68.3	71.4	62.3
0.5	89.3	64.1	43	55.3	39.4
0.75	81.3	36.2	18.8	27.2	15.2
1	74	27	15.1	24.2	12.2
OTC 22.5 mg/kg	OTC mobility/accessibility (% initial concentration)				
Time Period (Year)	Control (No Al-WTR)	Belle +25 g kg^{-1} Al-WTR	Belle +50 g kg^{-1} Al-WTR	Belle-Manure +25 g kg^{-1} Al-WTR	Belle-Manure +50 g kg^{-1} Al-WTR
0	97.5	95.6	94.1	91.8	92.6
0.25	92.4	78.6	70.4	73.8	64.6
0.5	88.6	66.2	46.1	57.6	40.8
0.75	83	39.9	21	27	16.3
1	75.2	20.4	15.3	19.3	13.4

D.5: Effect of Al-WTR application rates on leaching of TTC and OTC as a function of initial concentration (2.25 mg kg^{-1}) after 0.25 and 0.5 year of equilibration in Immokalee and Belleglade soils and soils amended with TTC/OTC rich manure in presence (P) and absence (NP) of plants.

	TTC leaching (%)			
Treatment (2.25 mg/kg TTC)	0.25 Y (P)	0.5 Y (P)	0.25 Y (NP)	0.5 Y (NP)
Imm+ noWTR	35	26	41	32
Imm+25g/kg WTR	20	14	25	22
Imm+50g/kg WTR	18	13	25	20
Imm+Man-25g/kgTR	20	12	24	20
Imm+ Man-50g/kg WTR	14	11	21	17
Belle+no WTR	20	15	26	21
Belle+25g/kg WTR	11	9	15	18
Belle+5%WTR	9	7	12	14
Belle+ Man-25g/kg WTR	10	8	14	12
Belle+ Man-50g/kg WTR	8	7	13	11
	OTC leaching (%)			
Treatment (2.25 mg/kg OTC)	0.25 Y (P)	0.5 Y (P)	0.25 Y (NP)	0.5 Y (NP)
Imm+ noWTR	34	25	40	31
Imm+25g/kg WTR	18	12	23	20
Imm+50g/kg WTR	17	12	24	19
Imm+Man-25g/kgTR	19	11	23	19
Imm+ Man-50g/kg WTR	12	9	19	15
Belle+no WTR	18	13	24	19
Belle+25g/kg WTR	9	7	13	16
Belle+5%WTR	8	6	11	13
Belle+ Man-25g/kg WTR	9	7	13	11
Belle+ Man-50g/kg WTR	7	6	12	10

D.6: Effect of Al-WTR application rates on leaching of TTC and OTC as a function of initial concentration (22.5 mg kg^{-1}) after 0.25 and 0.5 year of equilibration in Immokalee and Belleglade soils and soils amended with TTC/OTC rich manure in presence (P) and absence (NP) of plants.

Treatment (22.5 mg/kg TTC)	TTC leaching (%)			
	0.25 Y (P)	0.5 Y (P)	0.25 Y (NP)	0.5 Y (NP)
Imm+ noWTR	37	28	43	34
Imm+25g/kg WTR	22	16	27	24
Imm+50g/kg WTR	19	14	26	21
Imm+Man-25g/kgTR	22	14	26	22
Imm+ Man-50g/kg WTR	15	12	22	18
Belle+no WTR	22	17	28	23
Belle+25g/kg WTR	12	10	16	19
Belle+5%WTR	11	9	14	16
Belle+ Man-25g/kg WTR	12	10	16	14
Belle+ Man-50g/kg WTR	9	8	14	12
Treatment (22.5 mg/kg OTC)	OTC leaching (%)			
	0.25 Y (P)	0.5 Y (P)	0.25 Y (NP)	0.5 Y (NP)
Imm+ noWTR	35	26	41	32
Imm+25g/kg WTR	20	14	25	22
Imm+50g/kg WTR	18	13	25	20
Imm+Man-25g/kgTR	21	13	25	21
Imm+ Man-50g/kg WTR	14	11	21	17
Belle+no WTR	19	14	25	20
Belle+25g/kg WTR	11	9	15	18
Belle+5%WTR	9	7	12	14
Belle+ Man-25g/kg WTR	11	9	15	13
Belle+ Man-50g/kg WTR	8	7	13	11

D.7: Different extractions conducted on soils and soils amended with TTC rich manure

Time Zero	Initial Concentration 22.5 mg/kg			
	TTC leaching (%)			
Treatment (22.5 mg/kg TTC)	DI	KCl	MeOH	EDTA
Imm+ noWTR	68	72	76	83
Imm+25g/kg WTR	60	63	70	78
Imm+50g/kg WTR	59	62	69	77
Imm+Man-25g/kgTR	64	65	68	75
Imm+ Man-50g/kg WTR	63	64	68	74
Belle+no WTR	65	69	72	80
Belle+25g/kg WTR	64	68	70	78
Belle+50g/kg WTR	63	65	69	77
Belle+ Man-25g/kg WTR	62	64	68	75
Belle+ Man-50g/kg WTR	62	63	68	75
Time 0.5 Y	TTC leaching (%)			
Treatment (22.5 mg/kg TTC)	DI	KCl	MeOH	EDTA
Imm+ noWTR	60	65	68	78
Imm+25g/kg WTR	10	12	14	20
Imm+50g/kg WTR	8	10	11	18
Imm+Man-25g/kgTR	9	10	12	17
Imm+ Man-50g/kg WTR	8	9	11	14
Belle+no WTR	55	58	64	72
Belle+25g/kg WTR	9	11	13	18
Belle+5%WTR	8	10	12	16
Belle+ Man-25g/kg WTR	10	11	14	17
Belle+ Man-50g/kg WTR	9	10	12	13
Time 1Y	TTC leaching (%)			
Treatment (22.5 mg/kg TTC)	DI	KCl	MeOH	EDTA
Imm+ noWTR	50	54	59	63
Imm+25g/kg WTR	8	9	11	13
Imm+50g/kg WTR	6	8	9	11
Imm+Man-25g/kgTR	8	10	11	12
Imm+ Man-50g/kg WTR	7	8	10	12
Belle+no WTR	49	51	55	60
Belle+25g/kg WTR	8	9	11	12
Belle+5%WTR	7	8	10	11
Belle+ Man-25g/kg WTR	8	9	11	12
Belle+ Man-50g/kg WTR	6	8	9	11

D.8: Different extractions conducted on soils and soils amended with OTC rich manure

Time Zero	Initial Conc 22.5 mg/kg			
	TTC leaching (%)			
Treatment (22.5 mg/kg OTC)	DI	KCl	MeOH	EDTA
Imm+ noWTR	66	71	75	82
Imm+25g/kg WTR	59	62	69	77
Imm+50g/kg WTR	57	60	67	75
Imm+Man-25g/kgTR	63	64	67	74
Imm+ Man-50g/kg WTR	62	63	67	73
Belle+no WTR	64	67	71	78
Belle+25g/kg WTR	62	67	68	77
Belle+50g/kg WTR	62	64	68	76
Belle+ Man-25g/kg WTR	60	62	67	73
Belle+ Man-50g/kg WTR	61	62	67	74
Time 0.5 Y	TTC leaching (%)			
Treatment (22.5 mg/kg OTC)	DI	KCl	MeOH	EDTA
Imm+ noWTR	58	64	67	77
Imm+25g/kg WTR	9	11	13	19
Imm+50g/kg WTR	6	8	9	16
Imm+Man-25g/kgTR	8	9	11	16
Imm+ Man-50g/kg WTR	7	8	10	13
Belle+no WTR	54	56	63	70
Belle+25g/kg WTR	7	10	11	17
Belle+5%WTR	7	9	11	15
Belle+ Man-25g/kg WTR	8	9	13	15
Belle+ Man-50g/kg WTR	8	9	11	12
Time 1Y	TTC leaching (%)			
Treatment (22.5 mg/kg OTC)	DI	KCl	MeOH	EDTA
Imm+ noWTR	48	53	58	62
Imm+25g/kg WTR	7	8	10	12
Imm+50g/kg WTR	4	6	7	9
Imm+Man-25g/kgTR	7	9	10	11
Imm+ Man-50g/kg WTR	6	7	9	11
Belle+no WTR	48	49	54	58
Belle+25g/kg WTR	6	8	9	11
Belle+5%WTR	6	7	9	10
Belle+ Man-25g/kg WTR	6	7	10	10
Belle+ Man-50g/kg WTR	5	7	8	10

Appendix E

E.1: Pictures from the incubation study

E.2: Pictures from the greenhouse study.
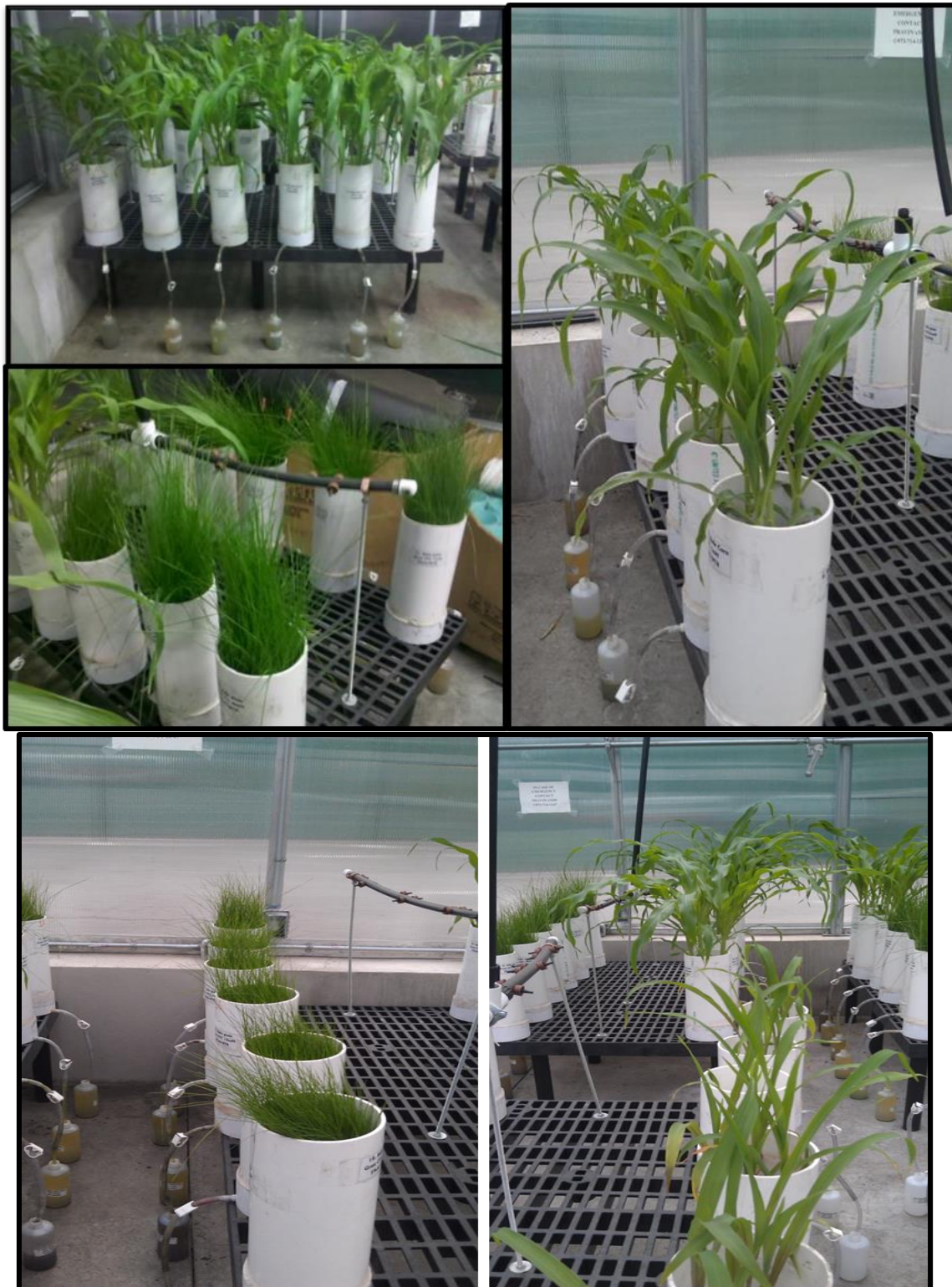
APPENDIX F

PREFACE

"This Doctoral Dissertation was produced in accordance with guidelines which permit the inclusion as part of the Doctoral Dissertation the text of an original paper, or papers, submitted for publication. Doctoral Dissertation must still conform to all other requirements explained in the "Guide for the Preparation of the Doctoral Dissertation at The Montclair State University." It must include a comprehensive abstract, a full introduction and literature review, and a final overall conclusion. Additional material (procedural and design data as well as descriptions of equipment) must be provided in sufficient detail to allow a clear and precise judgment to be made of the importance and originality of the research reported.

It is acceptable for this Doctoral Dissertation to include as chapters authentic copies of papers already published, provided these meet type size, margin, and legibility requirements. In such cases, connecting texts, which provide logical bridges between different manuscripts, are mandatory. Where the student is not the sole author of a manuscript, the student is required to make an explicit statement in the introductory material to that manuscript describing the student's contribution to the work and acknowledging the contribution of the other author(s). The signatures of the Supervising Committee which precede all other material in the Doctoral Dissertation attest to the accuracy of this statement."

<u>Punamiya, P.,</u> Sarkar, D., Rakshit, S., Datta, R., 2013. Effectiveness of Al- based drinking water treatment residuals as a novel sorbent to remove tetracyclines from aqueous medium, J. Environ. Qual Vol. 42 No. 5, p. 1449-1459 (DOI: 10.2134/jeq201).

<u>Punamiya, P.,</u> Sarkar, D., Rakshit, S., and Datta, R. 2015. Effect of Solution Properties, Competing Ligands, and Complexing Metal on Sorption of Tetracyclines on Al-Based Drinking Water Treatment Residuals. Environ. Sci. Pollut. Res. (DOI 10.1007/s11356-015-4145-z)

<u>Punamiya, P.,</u> Sarkar, D., Rakshit, S., Elzinga, E. J., and Datta, R. 2015. Immobilization of Tetracyclines in Manure and Manure-Amended Soils using Aluminum-based Drinking Water Treatment Residuals. Environ. Sci. Pollut. Res. (In Review)

VITA

Pravin Anraj Punamiya was born in Pune, India. He graduated from Nowrosjee Wadia College of Arts and Science, Pune, India with a high school degree in 2000. He received his Bachelor of Science degree with major in Microbiology and minor in Chemistry from Modern College of Arts and Science, Pune University, India in 2003. He received his Master of Science degree in Environmental Science from Pune University, India in 2005. After completion of his Masters, he worked as a Junior Environmental Scientist for a waste water treatment plant and then a drinking water treatment plant for 6 months, respectively. After working for a year, he came to the United States in August 2006 to pursue Ph.D. in Environmental Science and Engineering at the University of Texas at San Antonio (UTSA). However, he completed Master of Science in Environmental Science at UTSA in summer 2008 and moved to Montclair State University (MSU) for his Ph.D. In fall 2008, he joined the Ph.D. program in Environmental Management, MSU, NJ. While doing his research, he also worked as a laboratory manager in Environmental Geochemistry Laboratory, MSU for five years. In 2011, he received the "Doctoral Student Research Award" from the College of Mathematics and Sciences, MSU for his academic excellence and dissertation research. He also received student research grants from the Geological Society of America (GSA) in 2009 and New Jersey Water Resources Research Institute (NJWRRI) in 2010, respectively. He has so far authored /co-authored 19 (4 authored and 15 co-authored) peer-reviewed journal articles and 43 technical abstracts. In addition, he has made numerous presentations at international, national and regional conferences and won graduate student oral (two) and poster (four) awards. At present, he has accepted an offer for the position of a Post-Doctoral Research Associate in the Department of Earth and Environmental Studies, MSU, NJ.

This Page Intentionally Left Blank