UNIVERSITY OF CALIFORNIA

Los Angeles

Resilience of Stormwater Treatment Systems under Changing Climates

A dissertation submitted in partial satisfaction of the

requirements for the degree of Doctor of Philosophy

in Civil Engineering

by

Renan Lucas Valenca

2022

ABSTRACT OF THE DISSERTATION

Resilience of Stormwater Treatment Systems under Changing Climates

by

Renan Lucas Valenca

Doctor of Philosophy in Civil Engineering

University of California, Los Angeles, 2022

Professor Sanjay K. Mohanty, Chair

Access to water is critical for societal development. Urban areas, where more than half of the world's population currently lives, are projected to increase to 70% by 2050. This growth indicates that the water scarcity issue in urban areas will get worse unless alternative water resources are utilized. Stormwater may serve as an alternative water source, but stormwater often contains many contaminants including pathogens, heavy metals, motor oils, nutrients, pesticides, herbicides, polyaromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), per- and polyfluorinated substances (PFAS), microplastics, and other emerging organic contaminants. To manage and treat stormwater in urban areas, stormwater control measures (SCM) or green infrastructures have been used. However, traditional stormwater control measures are highly unreliable. The performance variability of SCM is due to varying stormwater, weather conditions, and design factors. To reduce the performance variability and make SCM more reliable, the development of climate resilient is required, which will result in increased water security in urban

areas. In stormwater treatment systems, the resilience concept involves four central elements: stressors, indicators of resilience, metrics and the intervention. In this dissertation, I researched about each of the four central elements of resilience for stormwater treatment systems in order to develop climate resilient SCM.

I researched about the stressor elements in Chapter 2 and 3. In Chapter 2, I showed that both design and local climate can explain nitrate removal variability by critically analyzing data reported on the international BMP database for nitrate removal by four common types of SCM: bioretention cells, grass swales, media filters, and retention ponds. In Chapter 3, I analyzed 7,421 data collected from 19 retention ponds across North America showed that FIB removal in retention ponds is sensitive to weather conditions or seasons, but temperature and precipitation data failed to describe the variable FIB removal.

In Chapter 4 and 5, I quantified the performance variability of SCM through metrics. In Chapter 4, I examined how post-wildfire runoff containing burned residues affect the transport and survival of indicator bacteria, resulting in changes in the microbial quality of surface water and subsurface soil. In Chapter 5, I demonstrated how the deposition of wildfire residues could increase methane emissions in wetland sediments by up to 56%, but the emission depended on the amount of wildfire residues deposited.

Finally, in Chapter 6 I researched about the last resilience concept: intervention or mitigation strategies. I showed in Chapter 6 that biochar's capacity to remove pathogens from stormwater can vary by orders of magnitude, but the usage of machine learning techniques can predict biochar's performance based in their commonly reported properties: surface area, carbon content, ash content, and volatile organic carbon content. This dissertation advances the science applied to climate resilient stormwater treatment systems.

The dissertation of Renan Lucas Valenca is approved.

Shaily Mahendra

Richard Ambrose

David Jassby

Sanjay K. Mohanty, Committee Chair

University of California, Los Angeles

2022

Table of Contents

List of Figures

List of Tables

Acknowledgments

First and most importantly, I would like to thank the endless support from my wife and soul mate, Janie – you have always gone above and beyond to help me achieve my goals, encouraged me in every second of my life, and I will forever love you. I would also like to thank my parents, Ray and Max, who have taught me from day one the importance of integrity, education, and passion. I also want to thank my brother and best friend, Caio, who introduced me to the engineering world and serves as an inspiration to me every day. I would also like to thank my corgi, Charlie, who helped me stay calm and go for walks to clear my head. I am also very thankful to my extended family, Martha, Daniel, Will, Ali, and Juliana, who have always been by my side guiding me through this journey. Thank you also to my family and friends in Brazil, Italy, and across the United States that always send me words of encouragement and cheered my achivements from afar.

Before and during my graduate studies, I had the pleasure to learn from and work aside some of the most incredible people that I have ever met in my life. I would like to thank my Ph.D. advisor, Prof. Sanjay K. Mohanty, for all the meetings, guidance, and advice that helped me inside and outside the laboratory – I sincerely could not have asked for a better mentor. Thank you also to all my laboratory colleagues from SEALab: Alex, Annesh, Onja, Vera, Jamie, Cami, Maryam, Samiha, Tonoy, and all the undergrads who assisted me with experiments. I am also very grateful to Prof. Shaily Mahendra, Prof. David Jassby, and Prof. Richard Ambrose, as well as their mentees, for their support during my doctorate studies. I would like to thank my undergraduate advisor, Dr. Fabio Ferraço, for introducing me to the research and development world, which led me to graduate school. Thank you also to my high school chemistry teacher, Mario Lopes Macedo, who was the first person to instigate my interest in chemistry and shaped my curiosity for science.

Last but not least, I would like to thank three dogs that are not among us anymore, but that immensely marked my life: Lilica, Tudão, and Steve. Thank you all from the bottom of my heart. This dissertation is dedicated to all of you.

VITA

EDUCATION

2013	B.S. in Science and Technology, Federal University of Alfenas, Brazil
2016	B.S. in Chemical Engineering, Federal University of Alfenas, Brazil
2018	M.S. in Civil and Environmental, University of California, Los Angeles

AWARDS

2013	Science Without Borders Undergraduate Scholarship, Italy
2016	Fundação de Amparo à Pesquisa do Estado de Minas, Brazil
2020	Martin Rubin Scholarship
2021	Los Angeles Urban Center Fellowship
2021	UCLA NRT-INFEWS Fellowship
2021	UCLA Dissertation Year Fellowship

PUBLICATIONS

Valença, R.L. and Ferraço, F. (2018) Reutilization of the Solid Waste Materials Produced by a Ceramic Tiles Industry as a Raw Material for the Production of New Ceramic Tiles. *Materials Science Forum*, 912, 180-184.

Mohanty, S.K., **Valenca, R.**, Berger, A.W., Yu, I.K.M., Xiong, X.N., Saunders, T.M. and Tsang, D.C.W. (2018) Plenty of room for carbon on the ground: Potential applications of biochar for stormwater treatment. *Science of the Total Environment* 625, 1644-1658.

Berger, A.W., **Valenca, R.**, Miao, Y., Ravi, S., Mahendra, S. and Mohanty, S.K. (2019) Biochar increases nitrate removal capacity of woodchip biofilters during high-intensity rainfall. *Water Research* 165, 115008.

Baalousha, M., Wang, J., Nabi, M.M., Loosli, F., **Valenca, R.**, Mohanty, S.K., Afrooz, N., Cantando, E. and Aich, N. (2020) Stormwater green infrastructures retain high concentrations of TiO_2 engineered (nano)-particles. *Journal of Hazardous Materials* 392, 122335

Valenca, R., Ramnath, K., Dittrich, T.M., Taylor, R.E. and Mohanty, S.K. (2020) Microbial quality of surface water and subsurface soil after wildfire. *Water Research* 175, 115672.

Ghavanloughajar, M., **Valenca, R.,** Le, H., Rahman, M., Borthakur, A., Ravi, S., Stenstrom, M.K. and Mohanty, S.K. (2020) Compaction conditions affect the capacity of biochar-amended sand filters to treat road runoff. *Science of the Total Environment* 735, 139180.

Le, H., **Valenca, R.**, Ravi, S., Stenstrom, M.K. and Mohanty, S.K. (2020) Size-dependent biochar breaking under compaction: Implications on clogging and pathogen removal in biofilters. *Environmental Pollution* 266, 115195.

Valenca, R., Borthakur, A., Zu, Y., Matthiesen, E.A., Stenstrom, M.K. and Mohanty, S.K. (2021) Biochar Selection for *Escherichia coli* Removal in Stormwater Biofilters. *Journal of Environmental Engineering* 147(2), 06020005.

Tirpak, R.A., Afrooz, A.R.M.N., Winston, R.J., **Valenca, R.**, Schiff, K. and Mohanty, S.K. (2021) Conventional and amended bioretention soil media for targeted pollutant treatment: A critical review to guide the state of the practice. *Water Research* 189, 116648.

Valenca, R., Le, H., Zu, Y., Dittrich, T.M., Tsang, D.C.W., Datta, R., Sarkar, D. and Mohanty, S.K. (2021) Nitrate removal uncertainty in stormwater control measures: Is the design or climate a culprit? *Water Research* 190, 116781.

Ghavanloughajar, M., Borthakur, A., **Valenca, R.**, McAdam, M., Khor, C.M., Dittrich, T.M., Stenstrom, M.K. and Mohanty, S.K. (2021) Iron amendments minimize the first-flush release of pathogens from stormwater biofilters. *Environmental Pollution*, 281, 116989.

Valenca, R., Borthakur, A., Le, H., & Mohanty, S. K. (2021). Biochar role in improving pathogens removal capacity of stormwater biofilters. In *Advances in Chemical Pollution, Environmental Management and Protection* (Vol. 7, pp. 175-201). Elsevier.

1. CHAPTER 1 – RESILIENCE OF STORMWATER TREATMENT SYSTEMS

1.1. Introduction

1.1.1. Urbanization and water scarcity

Access to water is critical for societal development. In history, civilizations have been developed around water resources such as rivers and lakes so that water is readily available to support agricultural, residential and industrial activities. With the development of advanced technologies to extract, distribute, and treat water, water can now be supplied to remote areas and areas of greatest demand: urban areas. Nearly one-fifth of the world's population lives in water-stressed areas, and one-fourth of the world's population faces water shortages at least one month out of a year (United Nations 2014a). Although a sufficient amount of fresh water is available on Earth for human consumption, water is not frequently available where it is needed the most: urban areas (McDonald et al. 2014). Urban areas, where more than half of the world's population currently lives, are projected to increase to 70% by 2050 (United Nations 2014b). This growth indicates that the water scarcity issue in urban areas will almost certainly get worse unless alternative water resources are utilized (Oppenheimer et al. 2017). Urbanization also increases the pollution of water resources (Yazdanfar and Sharma 2015). Urbanization increases impervious surfaces, which not only limit the natural infiltration of water to groundwater, thereby depleting groundwater level, but also covey pollutants deposited on impervious surfaces via stormwater runoff to rivers and lakes (Sharma and Malaviya 2021). Stormwater often contains many contaminants including pathogens, heavy metals, motor oils, nutrients, pesticides, herbicides, polyaromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), per- and polyfluorinated substances (PFAS), microplastics, and other emerging organic contaminants (Borthakur et al. 2021, Grebel et al. 2013, Koutnik et al. 2021). As these contaminants are typically originated from non-point sources in urban areas, treatment using centralized treatment systems including traditional wastewater treatment systems is impractical.

Climate change is expected to exacerbate the water scarcity issues in urban areas. Based on climate models, the frequency of extreme events such as high-intensity rainfall, drought, and wildfires is projected to increase (IPCC 2007). With an increase in rainfall intensity, urban runoff volume and subsurface infiltration rate would increase, if the hydraulic conductivity of the soil is not limited. This could decrease the removal of contaminants in the subsurface (Garcia et al. 2010). Coupled effects of urbanization and climate change could worsen the water scarcity issue in urban areas unless alternative water resources such as recycled water and stormwater are utilized.

1.1.2. Stormwater treatment systems

To manage and treat stormwater in urban areas, stormwater control measures (SCM) or green infrastructures have been used. Among different types of SCM, infiltration-based treatment systems such as biofilters are popular because of their limited space requirements and better pollutant removal performance than other SCMs (US EPA 2000). A biofilter is a depressed area designed by replacing a section of soil with a mixture of sand and compost and growing plants on the top (Figure 1.1). Stormwater infiltrates through the filter media to either an underdrain to supplement surface waters or to underneath soil to recharge groundwater. There are three design factors that can be manipulated to increase contaminant removal: (1) filter media type and depth, (2) relative saturation of filter media by using submerged layer, and (3) biological components such as filter media microbiome and plants. Traditional or conventional biofilter media include sand, to increase infiltration, and compost, to increase contaminant removal and provide nutrients for plants. However, traditional biofilter media have a limited contaminant removal capacity (Roy-Poirier et al. 2010). The addition of amendments, such as biochar, activated carbon, zeolite, and iron filings to biofilter, has been shown to improve contaminant removal in the short term. However, the long-term removal capacity of amendments could decrease due to the exhaustion of adsorption sites with pollutants and other stormwater constituents (US NRC 2009). Frequent replacement of exhausted filter media or restorative maintenance to sustain contaminant removal can become expensive (Brown and Hunt 2012). Thus, an alternative in situ approach to regenerating the adsorption capacity of filter media must be developed.

The performance of biofilter depends on stormwater composition (e.g., pH, ionic strengths, and turbidity) (Okaikue-Woodi et al. 2020) and weather conditions (e.g., rainfall intensity, antecedent drying) (Kratky et al. 2017, Zinger et al. 2020). During high-intensity rainfall, runoff volume increases rapidly. An increase in elevation of ponding water level and relative saturation of filter media cause stormwater to infiltrate rapidly through the filter media, thereby limiting the capacity of filter media to remove contaminants (Berger et al. 2019). Thus, it is critical to examine how different weather conditions may affect contaminant removal in stormwater treatment systems.

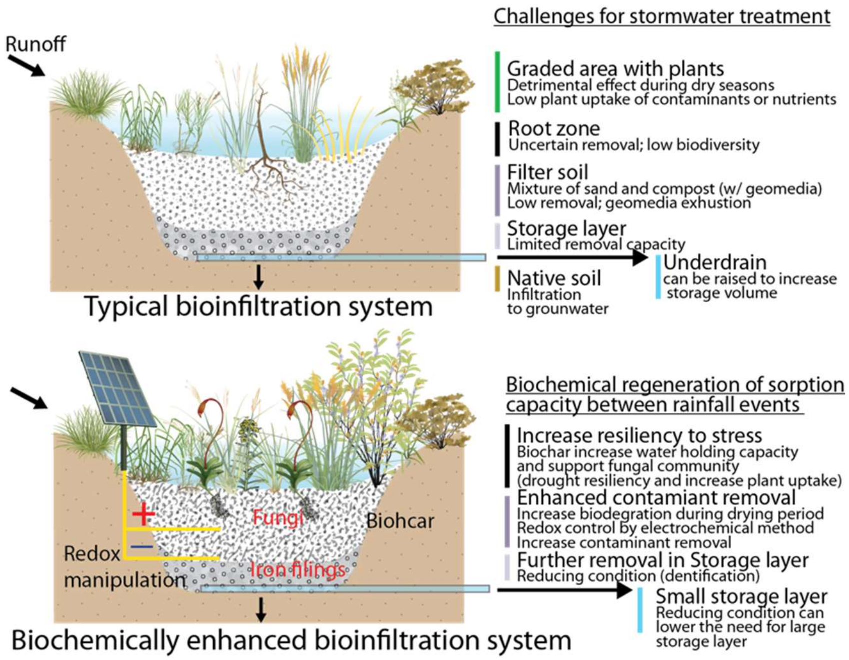


Figure 1.1 – Challenges to remove stormwater contaminants in traditional biofilters, and potential methods to improve contaminants removal.

1.1.3. Resilience of stormwater treatment systems

Resilience is the capacity of an engineered system to withstand disturbance and reorganize while undergoing changes so as to still retain the same function, structure, identity, and feedback (Walker et al. 2004). In stormwater treatment systems, the resilience concept involves four central elements: stressors, indicators of resilience, metrics and the intervention (Juan-García et al. 2017). A stressor can be defined as a pressure on the system caused by either human activities, such as changes in land use and accidental contaminant spill, or by natural events, such as drought, high-intensity rainfall, and wildfires (Figure 1.2). The resilience indicator in a stormwater treatment system can be infiltration capacity, filter media sorption capacity and lifetime, and robust microbiome, all of which will provide an indication of whether the treatment system can withstand, respond to, and adapt more readily to stressors. Metrics are related to the quantification of the system's recovery time and failure magnitude, which can be estimated by measuring the time required for stormwater treatment systems to regain their original contaminant removal capacity after exhaustion or the number of times effluent water quality exceeds a fixed water quality standards. Finally, interventions are the manipulation of the design in order to alter the properties or increase system capacities to stressors such as recharge of filter media capacity, microbiome manipulation, and electrochemical treatment.

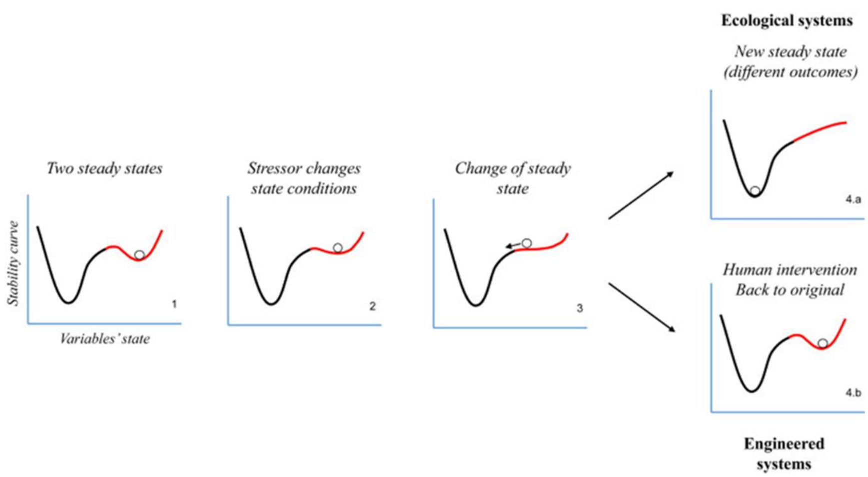


Figure 1.2 – Effect of stressors on the stability of a system and how engineering methods can be used to prevent a permanent shift in the condition of an ecological system such as stormwater biofilters. Source: (Juan-García et al. 2017).

1.2. Research gaps

Stormwater treatment systems such as biofilters are designed to reduce pollution of water supply, but whether stormwater treatment systems would be resilient under climate change is not clear. For example, extreme weather events such as heavy rainfall and prolonged drying, which are projected to be more frequent under climate change (Prein et al. 2017), could adversely affect the treatment efficiency of stormwater treatment systems and consequently increase the risk of groundwater and surface water contamination. Antecedent drying conditions have been shown to have detrimental effects on the removal of pollutants, possibly because of remobilization of the particle-associated contaminants during intermittent rainfall events (Mohanty et al. 2014) and reduction in biological activities during prolonged drying (Badin et al. 2011). Prolonged drying could kill plants and inactivate microbes. These biological components of stormwater treatment systems are essential to not only maintain the system's function but also for the removal of many pollutants (Chandrasena et al. 2017, Glaister et al. 2014, Ulrich et al. 2017, Zhang et al. 2011). Moreover, post-wildfire runoff could transport wildfire residues into the stormwater treatment system, which may reduce their microbial biomass and enzyme activity (Fultz et al. 2016). Thus, it is critical to developing engineering methods to maintain the biological communities in biofilters and utilize them for contaminant removal.

Little is known about the dynamic of biological communities in stormwater treatment systems subjected to environmental stressors (Hills et al. 2017). In fact, there is no study to date that has examined the shift in biofilter microbiomes subjected to extreme weather conditions. Improving the knowledge gap will help develop engineering control to manipulate the diversity and abundance of biological communities capable of degrading stormwater pollutants. For instance, biological stimulation during rainless or drying periods could provide an opportunity to regenerate the adsorption capacity of filter media via biological degradation of sequestered contaminants (Ulrich et al. 2015). But, for in situ regeneration to be practical, the biological communities must survive on contaminated filter media subjected to harsh weather conditions. Yet, how and to what extent climate stressors (e.g., rainfall intensity, antecedent drying duration, and wildfire) may affect the microbial community of stormwater treatment systems has not been assessed to date.

1.3. Objectives.

The overarching goals of the dissertation are to examine how the performance of stormwater treatment systems may change due to natural stressors that are projected to increase or intensify during climate change and develop engineering interventions to increase the system's resilience. The dissertation consists of five research chapters that examine three pillars of resilience: stressors, metrics, and intervention. Chapter 2 and 3 examine the stressors that are causing performance variability in stormwater treatment systems. Chapter 4 and 5 quantifies the performance variability through metrics. At the end, Chapter 6 proposes interventions or mitigation strategies to increase the resiliency of stormwater treatment systems. Specific goals are described below.

Chapter 2 examines how local weather based on *Köppen–Geiger* classification can affect nitrate removal in stormwater treatment systems including media filters, bioretention/biofilter, grass swales, and retention ponds. The results of this chapter identify local weather as a climate stressor for the performance of stormwater treatment systems. The outcome of Chapter 2:

> **Valenca, R.**, Le, H., Zu, Y., Dittrich, T. M., Tsang, D. C. W., Datta, R., & Mohanty, S. K. (2021). Nitrate removal uncertainty in stormwater control measures: Is the design or climate a culprit? *Water Research, 190*, 116781. https://doi.org/10.1016/j.watres.2020.116781

Chapter 3 explores how local weather conditions such as precipitation and temperature can impact the performance of stormwater retention ponds in removing indicator bacteria. This chapter also investigates how machine learning techniques could predict the removal of bacteria in retention ponds in different local weather conditions. The results of this chapter inform how the synergy between precipitation and temperature can influence the fate and transport of bacteria through stormwater treatment systems. The outcome of Chapter 3:

Valenca, R., Garcia, L., Espinosa, C., Flor, D., & Mohanty, S. K. (2022) Can water composition and weather factors predict fecal indicator bacteria removal in retention ponds in variable weather conditions? *Science of The Total Environment*. (*under revision*)

Chapter 4 examines how post-wildfire runoff containing burned residues affect the transport and survival of indicator bacteria, resulting in changes in the microbial quality of surface water and subsurface soil. The results of this chapter help to quantify the implications of wildfire residues on the microbial community of stormwater treatment systems impacted by post-wildfire runoff. The outcome of Chapter 4:

Valenca, R., Ramnath, K., Dittrich, T. M., Taylor, R. E., & Mohanty, S. K. (2020). Microbial quality of surface water and subsurface soil after wildfire. *Water Research, 175*, 115672. https://doi.org/10.1016/j.watres.2020.115672

Chapter 5 examines how the deposition of wildfire residues in wetlands, a type of stormwater treatment system, can affect the emission of methane and the microbial community. The results of this chapter will quantify the resilience of wetlands in treating post-wildfire runoff and inform public and private institutions of the threat of enhance methane emission from this type of stormwater treatment system. The outcome of Chapter 5:

Valenca, R., McKnight, Q., Indiresan, S., Kwok, I. K., Mahendra, S., & Mohanty, S. K. (2022) Enhanced methane emissions from deposited wildfire residues in wetlands: implications to climate change. *Water Research*. (*in preparation*).

<u>Chapter 6</u> explores how biochar – a sustainable soil amendment – can help remove contaminants and increase the resilience of stormwater treatment systems. The chapter also uses machine learning principles to create a model that predicts the removal performance of any given biochar. The results of this chapter present intervention or mitigation strategies to reduce the performance variability of stormwater treatment systems, making them more reliable. There are two outcomes of Chapter 6:

> **Valenca, R.**, Borthakur, A., Zu, Y., Matthiesen, E. A., Stenstrom, M. K., & Mohanty, S. K. (2021). Biochar Selection for Escherichia coli Removal in Stormwater Biofilters. *Journal of Environmental Engineering, 147*(2), 06020005. https://doi.org/10.1061/(ASCE)EE.1943-7870.0001843
>
> **Valenca, R.**, Borthakur, A., Le, H., & Mohanty, S. K. (2021). Chapter Seven - Biochar role in improving pathogens removal capacity of stormwater biofilters. In A. K. Sarmah (Ed.), *Advances in Chemical Pollution, Environmental Management and Protection* (Vol. 7, pp. 175-201): Elsevier. https://doi.org/10.1016/bs.apmp.2021.08.007

1.4. References

Badin, A.L., Monier, A., Volatier, L., Geremia, R.A., Delolme, C. and Bedell, J.P. (2011) Structural Stability, Microbial Biomass and Community Composition of Sediments Affected by the Hydric Dynamics of an Urban Stormwater Infiltration Basin. Microbial Ecology 61(4), 885-897.

Berger, A.W., Valenca, R., Miao, Y., Ravi, S., Mahendra, S. and Mohanty, S.K. (2019) Biochar increases nitrate removal capacity of woodchip biofilters during high-intensity rainfall. Water Research 165, 8.

Borthakur, A., Wang, M., He, M., Ascencio, K., Blotevogel, J., Adamson, D.T., Mahendra, S. and Mohanty, S.K. (2021) Perfluoroalkyl acids on suspended particles: Significant transport pathways in surface runoff, surface waters, and subsurface soils. Journal of Hazardous Materials 417, 126159.

Brown, R.A. and Hunt, W.F. (2012) Improving bioretention/biofiltration performance with restorative maintenance. Water Science and Technology 65(2), 361-367.

Chandrasena, G.I., Shirdashtzadeh, M., Li, Y.L., Deletic, A., Hathaway, J.M. and McCarthy, D.T. (2017) Retention and survival of E. coli in stormwater biofilters: Role of vegetation, rhizosphere microorganisms and antimicrobial filter media. Ecological Engineering 102, 166-177.

Fultz, L.M., Moore-Kucera, J., Dathe, J., Davinic, M., Perry, G., Wester, D., Schwilk, D.W. and Rideout-Hanzak, S. (2016) Forest wildfire and grassland prescribed fire effects on soil biogeochemical processes and microbial communities: Two case studies in the semi-arid Southwest. Applied Soil Ecology 99, 118-128.

Garcia, J., Rousseau, D.P.L., Morato, J., Lesage, E., Matamoros, V. and Bayona, J.M. (2010) Contaminant Removal Processes in Subsurface-Flow Constructed Wetlands: A Review. Critical Reviews in Environmental Science and Technology 40(7), 561-661.

Glaister, B.J., Fletcher, T.D., Cook, P.L. and Hatt, B.E. (2014) Co-optimisation of phosphorus and nitrogen removal in stormwater biofilters: the role of filter media, vegetation and saturated zone. Water Science and Technology 69(9), 1961-1969.

Grebel, J.E., Mohanty, S.K., Torkelson, A.A., Boehm, A.B., Higgins, C.P., Maxwell, R.M., Nelson, K.L. and Sedlak, D.L. (2013) Engineered Infiltration Systems for Urban Stormwater Reclamation. Environmental Engineering Science 30(8), 437-454.

Hills, M., Allen, V. and Lenhart, J. (2017) Investigating the Microbiology of Bioretention, pp. 416-430.

IPCC (2007) Climate Change 2007: Impacts, Adaptation and Vulnerability. Contribution of Working Group II to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change.

Juan-García, P., Butler, D., Comas, J., Darch, G., Sweetapple, C., Thornton, A. and Corominas, L. (2017) Resilience theory incorporated into urban wastewater systems management. State of the art. Water Research 115, 149-161.

Koutnik, V.S., Leonard, J., Alkidim, S., DePrima, F.J., Ravi, S., Hoek, E.M.V. and Mohanty, S.K. (2021) Distribution of microplastics in soil and freshwater environments: Global analysis and framework for transport modeling. Environmental Pollution 274, 116552.

Kratky, H., Li, Z., Chen, Y., Wang, C., Li, X. and Yu, T. (2017) A critical literature review of bioretention research for stormwater management in cold climate and future research recommendations. Frontiers of Environmental Science & Engineering 11(4), 16.

McDonald, R.I., Weber, K., Padowski, J., Flörke, M., Schneider, C., Green, P.A., Gleeson, T., Eckman, S., Lehner, B. and Balk, D. (2014) Water on an urban planet: Urbanization and the reach of urban water infrastructure. Global Environmental Change 27, 96-105.

Mohanty, S.K., Saiers, J.E. and Ryan, J.N. (2014) Colloid-facilitated mobilization of metals by freeze-thaw cycles. Environmental Science & Technology 48(2), 977-984.

Okaikue-Woodi, F.E.K., Cherukumilli, K. and Ray, J.R. (2020) A critical review of contaminant removal by conventional and emerging media for urban stormwater treatment in the United States. Water Research 187, 116434.

Oppenheimer, J., Da Silva, A., Yu, Z.L., Hanna, M. and Susilo, K. (2017) Total Water Solutions--Capturing Alternative Water Sources to Supplement Drinking Water Supply. Journal-American Water Works Association 109(1), 18-25.

Prein, A.F., Rasmussen, R.M., Ikeda, K., Liu, C.H., Clark, M.P. and Holland, G.J. (2017) The future intensification of hourly precipitation extremes. Nature Climate Change 7(1), 48-+.

Roy-Poirier, A., Champagne, P. and Filion, Y. (2010) Review of Bioretention System Research and Design: Past, Present, and Future. Journal of Environmental Engineering-Asce 136(9), 878-889.

Sharma, R. and Malaviya, P. (2021) Management of stormwater pollution using green infrastructure: The role of rain gardens. Wiley Interdisciplinary Reviews: Water 8(2), e1507.

Ulrich, B.A., Im, E.A., Werner, D. and Higgins, C.P. (2015) Biochar and Activated Carbon for Enhanced Trace Organic Contaminant Retention in Stormwater Infiltration Systems. Environmental Science & Technology 49(10), 6222-6230.

Ulrich, B.A., Loehnert, M. and Higgins, C.P. (2017) Improved contaminant removal in vegetated stormwater biofilters amended with biochar. Environmental Science-Water Research & Technology 3(4), 726-734.

United Nations (2014a) The United Nations world water development report 2014: water and energy. United Nations, Paris.

United Nations (2014b) World Urbanization Prospects: The 2014 Revision, Highlights. Department of Economic and Social Affairs. Population Division, United Nations.

US EPA (2000) Low Impact Development (LID): A Literature Review., United States Environmental Protection Agency.

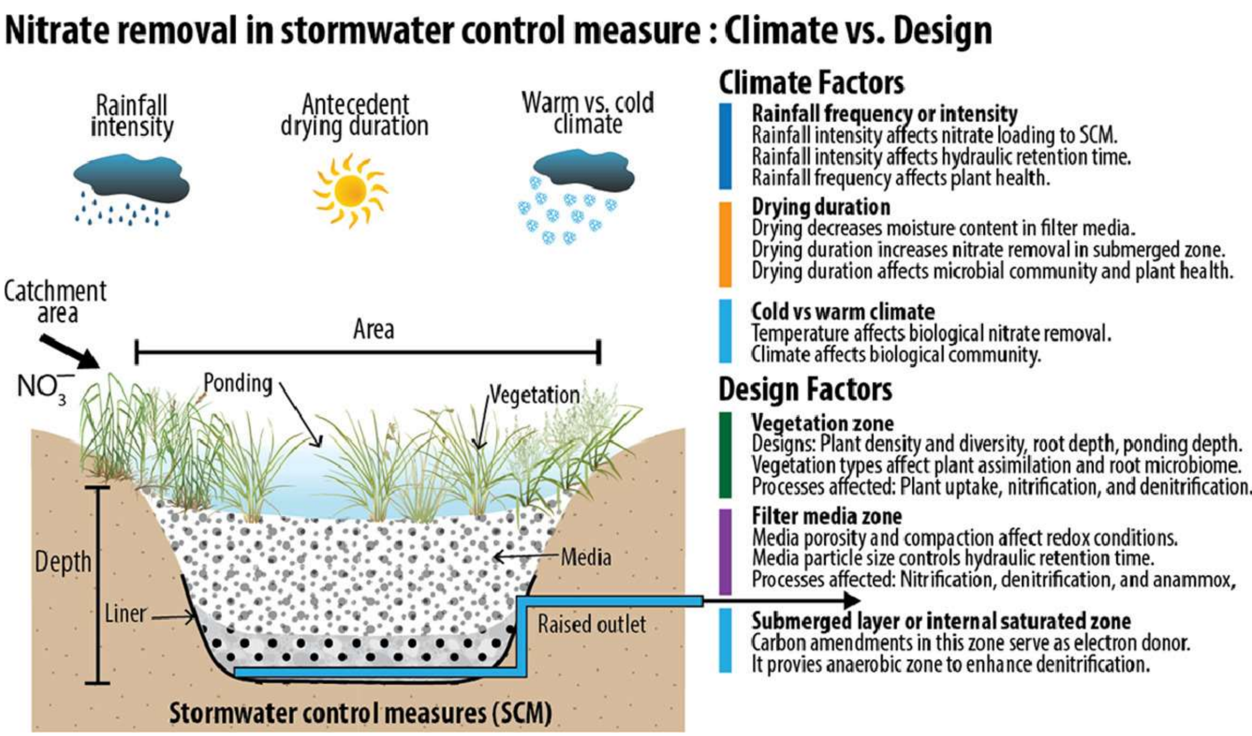
US NRC (2009) Urban Stormwater Management in the United States, p. 598, National Academies Press, Washington, D.C.

Yazdanfar, Z. and Sharma, A. (2015) Urban drainage system planning and design – challenges with climate change and urbanization: a review. Water Science and Technology 72(2), 165-179.

Zhang, Z., Rengel, Z., Liaghati, T., Antoniette, T. and Meney, K. (2011) Influence of plant species and submerged zone with carbon addition on nutrient removal in stormwater biofilter. Ecological Engineering 37(11), 1833-1841.

Zinger, Y., Prodanovic, V., Zhang, K., Fletcher, T.D. and Deletic, A. (2020) The effect of intermittent drying and wetting stormwater cycles on the nutrient removal performances of two vegetated biofiltration designs. Chemosphere, 129294.

2. CHAPTER 2 – NITRATE REMOVAL UNCERTAINTY IN STORMWATER CONTROL MEASURES: IS THE DESIGN OR CLIMATE A CULPRIT?



Valenca, R., Le, H., Zu, Y., Dittrich, T. M., Tsang, D. C. W., Datta, R., & Mohanty, S. K. (2021). Nitrate removal uncertainty in stormwater control measures: Is the design or climate a culprit? *Water Research, 190*, 116781. https://doi.org/10.1016/j.watres.2020.116781

Abstract

Eutrophication is largely caused by excess nitrate and other nutrient export via stormwater runoff to surface waters and is projected to increase as a result of climate change. Despite recent increases in implementation of stormwater control measures (SCM), nutrient export has not abated. This indicates poor or inconsistent removal capacities of SCM for nitrate; however, the cause of the variability is unclear. We show that both design and local climate can explain nitrate removal variability by critically analyzing data reported on the international BMP database for nitrate removal by four common types of SCM: bioretention cells, grass swales, media filters, and retention ponds. The relative importance of climate or design on nitrate removal depends on the SCM type. Nitrate removal in grass swales and bioretention systems are more sensitive to local climate than design specifications, while nitrate removal in the retention ponds is less sensitive to climate and more sensitive to design features such as vegetation and pond volume. Media filters without amendment have the least capacity compared to other SCM surveyed and their removal capacity was independent of the local climate. Adding amendments made up of carbon biomass, iron-based media, or a mixture of these amendments can significantly improve nitrate removal. The type of carbon biomass is also a factor since biochar, a popular amendment, does not affect nitrate removal. This analysis can help inform the selection of SCM and adequate modification of their design based on local and projected climate factors to maximize nitrate removal and minimize eutrophication of surface waters.

2.1. Introduction

Excess nutrients in stormwater from nonpoint sources cause eutrophication (Boeykens et al. 2017), resulting in significant financial losses (Deegan et al. 2012, Dodds et al. 2009). Eutrophication is projected to get worse because of climate change (Michalak et al. 2013, Sinha et al. 2017). To manage stormwater, different stormwater control measures (SCM) have been widely implemented (Bowles et al. 2018). However, the implementation of SCM on a watershed scale has often not resulted in improved water quality (Lintern et al. 2020). Among many reasons (Lintern et al. 2020), a wide variation in nitrate removal in all SCM is a primary factor (Manka et al. 2016). The cause of wide variation is often attributed to inadequate design (Zhang et al. 2020) or local climate (Blecken et al. 2010, Kratky et al. 2017). For instance, an increase in temperature and the frequency of high-intensity rainfall events is projected to accelerate eutrophication (Ballard et al. 2019, Sinha et al. 2017). Currently, SCM is rarely designed based on local climate information or projected climate changes (Brudler et al. 2016, Kerkez et al. 2016, Zhang et al. 2019a), partly because it is not clear how nitrate removal is affected by the coupled effect of climate and design or whether any design modification could minimize the detrimental effect of changing climates on nitrate removal.

Local climate conditions and the SCM's design have been shown to affect removal of some contaminants (Rippy 2015, Roseen et al. 2009, Valtanen et al. 2017b) , so it is expected they could also affect nitrate removal (McPhillips and Walter 2015, Payne et al. 2018, Shrestha et al. 2018). However, relative importance of these factors on nitrate removal is unknow. In SCM, nitrate can be removed via abiotic processes such as ion exchange (Hu et al. 2020) but such process is sensitive to chloride concentration (Samatya et al. 2006). Nitrate can adsorb on filter media with net positive surface charge (Hassapak et al. 2015, Mahdy et al. 2008, Ordonez et al. 2020, Yan et al. 2016), but most media in biofilters have net negative surface charges or become negatively charge after adsorption of organic carbon (Kaiser and Guggenberger 2003). Thus, contribution of these abiotic processes for nitrate removal is typically low. Nitrate are typically removed in SCM via biotic processes such as denitrification (Mangum et al. 2020), dissimilatory nitrate reduction to ammonium (DNRA) (Bu et al. 2017, Burgin and Hamilton 2007), or by direct plant uptake (Morse et al. 2018) (Figure 2.1). Among these processes, nitrate removal by DNRA can be temporary as oxidation of ammonium by nitrifiers can produce nitrate (Payne et al. 2014a, Payne et al. 2014b, Rahman et al. 2019). Abiotic processes are governed by amendment types and quantity (Yang et al. 2017) and the design configuration (Noubactep et al. 2012). Biotic processes are governed by fluctuations in pH, dissolved oxygen, and moisture content – all factors influenced by local climate. These factors result in soil microbial community shifts (Glassman et al. 2018) and changes in contaminant removal rates (Garfí et al. 2012). The coupled effect of climate and design modifications may act as the main cause of the high nitrate removal variability in SCM; however, to what extent climate or design factors contribute to nitrate removal uncertainty remains unclear (Gold et al. 2019, Schifman et al. 2016).

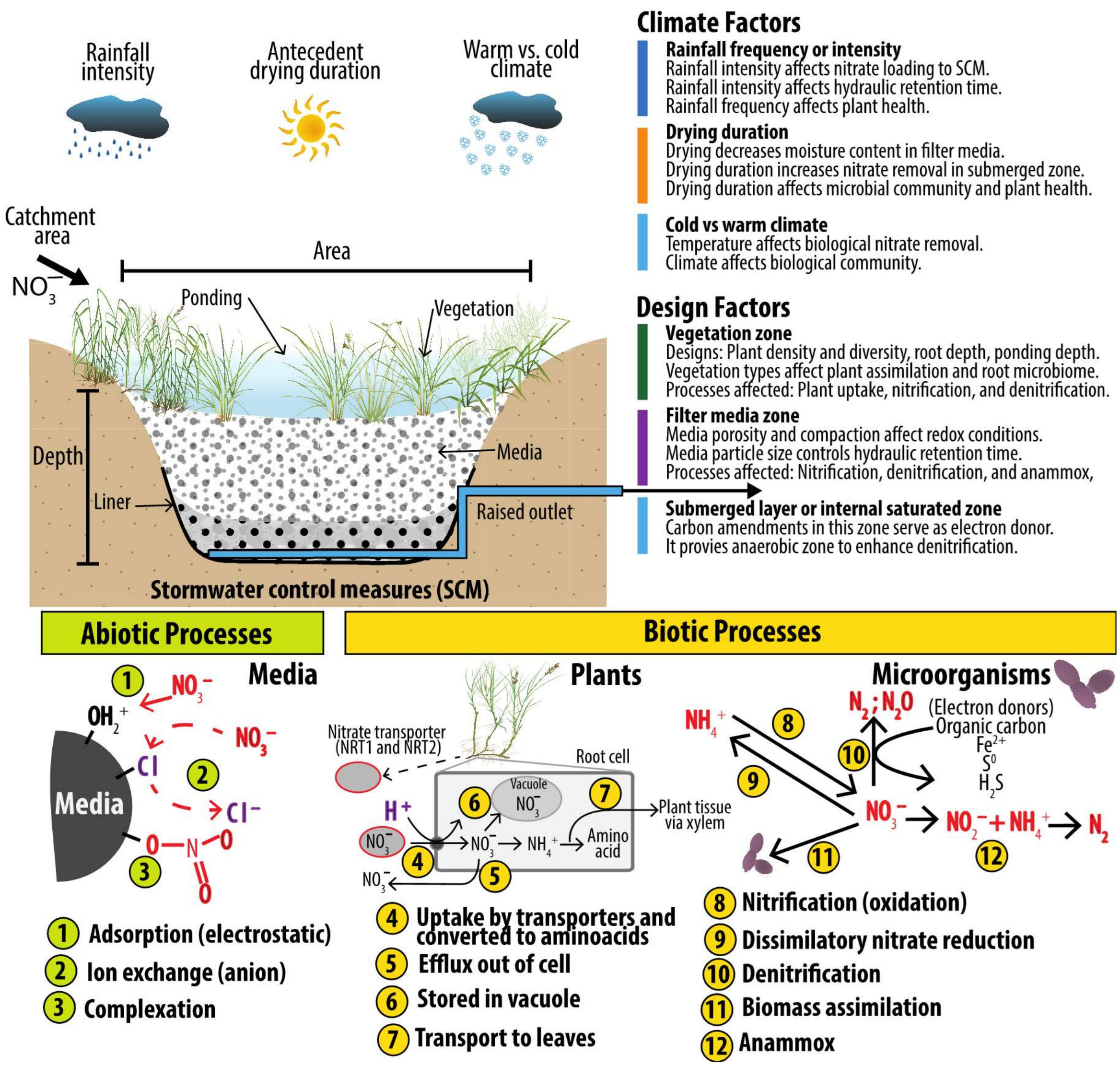


Figure 2.1 – Abiotic and biotic processes in stormwater treatment systems that could affect nitrate concentration in the effluent and the role of climate and design in modulating those processes. A major fraction of nitrate removal is contributed by biotic processes involving plants and soil microorganisms. Design factors such as area, depth, saturation, amendments, and plants can affect the effectiveness of chemical and biological processes. Climate such as temperature, rainfall intensity, and drying duration can affect the nitrate removal kinetics and health of microorganisms and plants that help remove nitrate from stormwater.

Nitrate removal in SCM fluctuates widely (Collins et al. 2010a, Lopez-Ponnada et al. 2020, Manka et al. 2016, Tian et al. 2019), and even a large scale implementation of SCM has not lowered the nutrient loading to water bodies significantly (Lintern et al. 2020). The cause of this wide variability has been difficult to attribute at a specific site but can be attributed to several factors (Table 2.1). First, field-scale SCM are rarely monitored long enough to accurately measure their removal potential. Even in controlled laboratory studies (Bock et al. 2015, Davis et al. 2001), nitrate removal varies widely. Second, differences in design specifications such as hydraulic retention time (McPhillips and Walter 2015), usage of amendments (Morgan et al. 2020), and configuration (Palmer et al. 2013, Wissler et al. 2020) could lead to variable nitrate removal. Third, the local climate may influence the rainfall intensity, dry periods, and temperature (Liao et al. 2018, Xie et al. 2003). Thus, a variation in local climate could cause fluctuation in nitrate removal (Berger et al. 2019, Blecken et al. 2007, Mangangka et al. 2015), but it is unclear if the combined effects of design and climate can improve or worsen nitrate removal (Tanner and Kadlec 2013). Previous reviews have investigated specific details of SCM including the sources, cycling process, and fate and transport of nitrogen-based nutrients (Nestler et al. 2011, Reisinger et al. 2016, Yang and Lusk 2018), overall design (Collins et al. 2010b), the effect of media type and vegetation (Osman et al. 2019, Skorobogatov et al. 2020), biotic and abiotic removal mechanisms (Burgin and Hamilton 2007, Lee et al. 2009, Tang et al. 2020), and more recently the coupled effect of infiltration rate and design characteristics (Zhang et al. 2020). While many articles have recognized the lack of studies examining the effect of climate (Gold et al. 2019, Yang and Lusk 2018) and design factors (Osman et al. 2019) on the performance of SCM, previous reviews have rarely compared the importance of either factor on nitrate removal in the most common SCM.

Table 2.1 – Summary of the mechanisms and process involved on nitrate natural pathway and how mechanisms are impacted by the changes in design and local climate. (+) indicates the factor favors nitrate removal; (-) indicates the factor inhibits nitrate removal.

Processes	Design Factors	Climate Factors
Adsorption: Attachment of NO_3^- on amendments' surface positively charged by electrostatic attraction	(+) Positive surface charge of amendment (Eick et al. 1999, Long et al. 2019) (-) Presence of DOC or anions that compete for NO_3^- (+) Amendments that decrease pH of porewater (-) Contact time (Öztürk and Bektaş 2004) (+) Amendments ratio (more active sites and surface area) (Harmayani and Anwar 2010, Hu et al. 2015)	(-) Warmer climate that increase DOC production (+) temperature are important factors in maximizing nitrate removal
Ion exchange: Exchange of NO_3^- with anion on amendment surface	(+) Anion exchange cations (Jackson and Bolto 1990, Samatya et al. 2006) (-) Stormwater TDS (Hsieh et al. 2007)	Temperature is not important (Bhatnagar and Sillanpää 2011)
Nitrification: Production of NO_3^- from NH_4^+	(+) High Oxygen (Stenstrom and Poduska 1980) (-) Low compacted media (+) High pH (6.5 – 9.5) (Sharma and Ahlert 1977) (+) Aerobic microorganisms: genus *Brocadia* (van Kessel et al. 2015), *Nitrosomonas* and *Nitrobacter* species (Sharma and Ahlert 1977)	(+) Warmer temperature (28 – 42°C) (Sharma and Ahlert 1977) (-) Light exposure as inhibitor (Alleman et al. 1987, Kaplan et al. 2000)

Continuation of Table 2.1 - Summary of the mechanisms and process involved on nitrate natural pathway and how mechanisms are impacted by the changes in design and local climate. (+) indicates the factor favors nitrate removal; (-) indicates the factor inhibits nitrate removal.

Processes	Design Factors	Climate Factors
Denitrification: Biological conversion of NO_3^- to $N_{2(g)}$	(+) Low oxygen (Brady et al. 2008, Seitzinger et al. 2006) (+) Electron donors: DOC or Sulfur/Iron compounds (+) pH (7 – 8 is optimum) (Knowles 1982) (+) Anerobic microorganisms: genus *Pseudomonas* (Knowles 1982), *Alcaligenes faecalis* (Gamble et al. 1977), *Paracoccus denitrifican* and *Thibacillus denitrifican* (Tiedje 1983)	(+) Warmer Temperature (65 – 70°C) (Knowles 1982, Tan et al. 2020) (-) Environment inhibitors such as pesticide and toxic compounds (Knowles 1982) (+) Soil moisture, relating to rainfall intensity or irrigation (Ekpete and Cornfield 1964, Smith and Tiedje 1979)
Anammox: Anaerobic oxidation of NH_4^+ to $N_{2(g)}$ without the production of NO_3^-.	(+) Low Oxygen (Kuenen 2008, Tang et al. 2011) (+) Slightly high pH (6.7 – 8.3) (Strous et al. 1999) (+) Availability of CO_2(Kraft et al. 2014) (+) C/NO_2^- concentration ratio (Strous et al. 1999) (+) Anerobic microorganisms: *Panctomycetales* phylum (Kuenen 2008) as Brocadia, Kuenenia (Strous et al. 2006), Anamoxoglobus, and Jettenia species in fresh water	(+)Temperature (20 – 52°C) (Jaeschke et al. 2009, Strous et al. 1999)
Plant uptake: Uptake of NO_3^- for health and growth	(+) Availability of N-source in soil bulk flow (Mantelin and Touraine 2004) (-) Soil compaction (root health) (Lipiec and Stępniewski 1995) (+) Water holding capacity (Cui and Caldwell 1997, Månsson et al. 2014) (+) Terrestrial plants dominate aquatic plants (Mantelin and Touraine 2004) (+) Depending on vegetation species	(-) Extreme dry weather as rainfall promotes leaching of N-source (Mantelin and Touraine 2004) (+) Temperature (4 – 22°C) (Tischner 2000) (+) Biotic and abiotic activities affect the concentration of N-source before it reaches the root system: pH, temperature, and oxygen

The overall objective of this review is to evaluate the relative importance of climate and design on nitrate removal in SCM. This article compares the nitrate removal of four commonly used SCM – bioretention, grass swales, media filters, and retention ponds – based on field data reported on the BMP database from 1982 through 2018 combined with peer-reviewed articles published before June 30, 2020. By identifying the local climate of those SCM based on *Köppen-Geiger* climate classification, we link nitrate removal capacity to local climate and design configurations. We have analyzed the data against numerous design configurations that may affect nitrate removal in SCM, but we reported only selected data where sufficient data is available for statistical analysis (Table 2.2). The selected design configurations include the presence and depth of ponding/saturated zone, watershed area, infiltration rate, vegetation density, amendment type, length, area-to-depth ratio, and the presence of plastic linen.

Table 2.2 – Summary of design factors that can influence nitrate removal in SCM. Bold factors represent the design that were included in the current analysis.

SCM Type	Possible Design Factors Influencing Nitrate Removal	
Bioretention	• **Presence or absence of plastic linen** • **Presence or absence of saturated zone** • **Presence and absence of vegetation** • Hydraulic retention time (HRT)	• **Depth of saturated zone** • **Area-to-depth ratio** • **Amendment type** • **Infiltration rate**
Grass swale	• **Geometry (length)** • Presence or absence of check dams • Composition of soil materials	• Type of vegetation • Centerline slope • HRT
Filter media	• **Runoff peak flow** • Amendment type	• HRT
Retention pond	• **Pond depth** • **Vegetation density**	• **Watershed area** • HRT

2.2. Data collection and analysis method

To analyze the effect of different climate and design variables on nitrate removal, we used data from the BMP Database updated by June 30, 2020. The BMP Database is an open-access website initiated in cooperation between the USEPA and ASCE (Clary et al. 2011). Based on the availability of sufficient data for statistical analysis, we chose four stormwater control measures (SCM) — bioretention, grass swale, media filter, and retention pond. These SCM have a unique design or configuration for the removal or release of nitrate (Table 2.3). Although other types of SCM can also remove nitrate, the lack of sufficient nitrate data limited our analysis. Among types of SCM surveyed, bioretention systems are the most common, including bioretention and infiltration basins. Media filters permit rapid infiltration of stormwater through packed sand, where pollutants can be removed by physiochemical filtration and adsorption (Sabiri et al. 2017). Media filters are mostly sand filters or sand mixed with soil to increase the hydraulic conductivity of native soil for the rapid infiltration of stormwater. Thus, they are not optimized to remove dissolved nutrients due to the low adsorption capacity of sand and low hydraulic retention time. Bioswales include both grass swales and grass strips and they are common in the roadside environment. They do not have much design consideration other than the length and depth of the depressed area. Bioretention systems can remove nitrate by filtration, adsorption, and biotransformation mechanisms (Davis et al. 2006, Kim et al. 2003, Palmer et al. 2013), whereas grass swales can treat stormwater via sedimentation and filtration (Barrett et al. 1998, Deletic and Fletcher 2006, Stagge et al. 2012). Retention ponds include wetland basins and detention basins. They are particularly useful to handle a large volume of stormwater and lower the peak flow. Thus, the design factors for these SCM include the depth or volume of the pond and the presence or absence of plants. Retention ponds can lower nitrate concentration by dilution, photolysis, and other reactions in an aqueous medium (Chrétien et al. 2016, Krometis et al. 2009), but they can simultaneously increase nitrate concentration through nitrification and decomposition of organic matter (Bettez and Groffman 2012). In this study, we catalog all SCM data based on their design specifications such as watershed area, SCM length, volume, internal water storage (IWS) zone or submerged zone and its depth, area, depth of SCM, and hydraulic conductivity of filter media.

Table 2.3 – Summary of four common SCM, nitrate removal mechanisms, specifications of design, and project.

Properties	**Stormwater Control Measures (SCM)**			
	Bioretention	**Grass Swale**	**Media Filter**	**Retention Pond**
Design configuration	Vertical flow-through solid media with or without amendment, submerged layer, and plants	Shallow, horizontal channel with planted grass	Vertical flow-through of mixed or layered sand, peat, and/or soil	Deep and long water pool without geomedia and with surrounding vegetation
Fraction of contaminants removed (d/s)	Dissolved, suspended	Suspended	Suspended	Dissolved and suspended
Main contaminants removed	Nutrients, heavy metals, suspended solids, bacteria	Heavy metals, suspended solids	Suspended solids, bacteria	Nutrients, suspended solids
Main removal mechanisms	Adsorption, plant uptake, biodegradation, filtration	Adsorption, filtration, plant uptake, sedimentation	Adsorption, filtration, direct interception, inertial impaction, and diffusion by Brownian motion	Biodegradation, sedimentation
Project area (m^2)	100 – 1,000	5 – 320	0.8 – 150	100 – 10,000
Cost ($)	100K – 300K	57K – 63K	161K – 485K	297K – 1.78M
Retention time	1 – 12 hours	1 – 3 hours	0.5 – 1.5 hours	1 – 7 days
Vegetation	Yes	Yes	No	Yes
# of sites analyzed from BMP database	10	17	15	18
# of nitrate removal data	62	217	186	247
Anoxic environment	Yes/No	Mostly No	No	Yes/No
Common locations	Rural areas, adjacent to rives	Next to roadways	Maintenance stations	Urban areas

We used *Köppen-Geiger* climate classification because it reflects the biome distribution of each region (Beck et al. 2018). *Köppen-Geiger* classifies the climate into five main groups including tropical, dry, temperate, continental, and polar, which are further divided into 30 sub-types depending on local seasonal precipitation and local temperature (Peel et al. 2007). Using the global positioning system (GPS) coordinates of each SCM from the BMP database, we designated each SCM surveyed to one of the *Köppen-Geiger* climate categories.

To analyze the performance of each SCM in removing nitrate, we calculated the log removal of nitrate (LRN) as follows: $LRN_t = -\log_{10}\left(\frac{Ce_t}{Ci_t}\right)$; where C_e and C_i represent the concentration of nitrate in the effluent and influent, respectively, in a given day (t). The removal calculation assumes a steady state; that is, the influent concentration remains consistent or does not vary within the time scale of hydraulic residence time. This assumption could introduce significant error particularly if the residence time is much longer than the sampling frequency. Thus, a composite sample should be used to account for such fluctuation in influent concentration. Data without both influent and effluent for the given day was excluded from the analysis. To verify the change in performance due to design, we compared SCM located near each other and within the same climate classification. Nitrate removal was compared using Wilcoxon Test where *p-values* lower than 0.05 represent a statistical difference. To provide mechanistic insight into the link between nitrate removal and bioretention system design, results were analyzed from 29 peer-reviewed studies. These studies were collected from Web of Science based on keyword combinations of the terms “nitrate and biofilters”, “mesocosm”, or “biofiltration”. The complete dataset used in the analysis is provided in an online open-access repository, Figshare (https://doi.org/10.6084/m9.figshare.13167608.v1).

2.3. The extent of nitrate removal uncertainty in SCM

SCM are expected to have different nitrate removal capacity because of a difference in their design configurations. Our analysis reveals that, irrespective of SCM type, nitrate removal (95 percentile) varied by two orders of magnitude ranging from net positive removal to net negative removal (Figure 2.2). A fluctuation in DO concentration diurnally could affect denitrification. However, a lack of data on variation of DO in urban stormwater and corresponding changes in nitrate concentration prevented us linking diurnal fluctuation in DO with denitrification. Among the four SCM, the retention pond has a net positive median log removal of nitrate (~ 0.2). For all other SCM, the median log removal is negative, indicating they act as a source of nitrate in most cases. The media filters perform the worst but they are also the most consistent among all SCM. The removal performance of nitrate by media filters is similar ($p > 0.05$) to the removal performance of bioretention systems, but statistically ($p < 0.05$) different compared to the removal performance by grass swales and retention ponds.

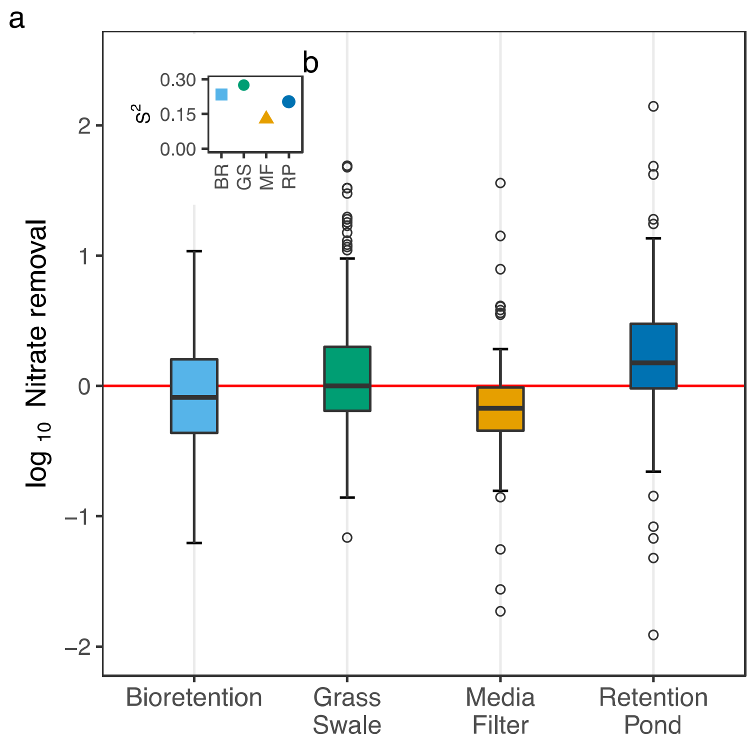


Figure 2.2 – (a) Log nitrate removal of four types of SCM. Log removal was calculated as the ratio between effluent and influent concentration of nitrate. The horizontal red line represents conditions where influent concentration is the same as the effluent (log removal = 0). (b) The variance (S^2) of log-removal data calculated for bioretention (BR), grass swale (GS), media filter (MF), and retention pond (RP).

We attribute the wide variability of the nitrate removal to a difference in design and climate. The rainfall intensity can vary widely between sampling events for the same SCM, which likely affects the nitrate loading and hydraulic retention time (Spieles and Mitsch 1999), thereby varying the nitrate removal (Berger et al. 2019). The variability of nitrate removal in grass swales could also be related to varying temperature and water salinity between seasons (Roseen et al. 2009) as salinity could lower the abundance of denitrifiers (von Ahnen et al. 2019). On the other hand, these SCM may have different types of amendments (Kameyama et al. 2016), vegetation (Flite III et al. 2001, Shrestha et al. 2018), and size that dictate residence time (Kjellin et al. 2007). All these factors could add uncertainty to nitrate removal by these systems. We evaluate the contribution of each factor separately in the following sections.

2.4. Extent to which climate affects nitrate removal in SCM

Comparing the nitrate removal of SCM to local climate (Figure 2.3) suggests that climate does not affect nitrate removal in most SCM. The result is in contrast to the results from previously published studies (Collings et al. 2020, Shrestha et al. 2018). For instance, there is no significant ($p > 0.05$) difference between nitrate removal by bioretention systems in hot-summer Mediterranean and cold-semi arid climate. In contrast, nitrate removal of grass swales in a humid tropical climate (median log removal, 0.23) is significantly ($p < 0.05$) better than the removal (–0.35) in a warm-summer humid continental climate. In contrast to plants in other climates, native plants in tropical climates are adapted to switch nitrogen sources based on precipitation patterns and thus are more efficient at removing nitrate (Houlton et al. 2007). This probably explains why grass swales remove more nitrate in tropical climates than in any other climate condition. Irrespective of climate, media filters exhibit a negative removal of nitrate in most climates, indicating that adding media filters could potentially lead to an increase in nitrate pollution. Since media filters are typically made up of sand with aerobic features, nitrate removal via most of the biological mechanisms is not feasible. In aerobic conditions, ammonium in stormwater can be rapidly oxidized to nitrate by nitrifiers and could explain the net negative removal of nitrate. One study shows that nitrification can occur within 0.7 h (Jin et al. 2012), which is in the range of overall stormwater retention time in media filters. Thus, ammonia oxidation can make filter media a net source of nitrate.

Comparing the performance of different SCM under the same climate classification, we found that some SCM are more efficient than others in removing nitrate (Figure **2.3**). For instance, in a cold-semi arid climate, a retention pond provides a median net positive (0.16) nitrate removal while the other three SCM exhibit net negative removal (source of nitrate). In this climate, a combination of low annual precipitation (less than 508 mm) and low mean annual temperature

(Collings et al. 2020) could lead to low biological activity and explain low nitrate removal. In contrast, in humid subtropical climates, grass swales and retention ponds show a net positive median removal of 0.23 and 0.19, respectively. While warm and moist climates increase nitrate concentration due to organic matter decomposition (Bulseco et al. 2019, Joslin and Wolfe 1993, Luo et al. 1999), cold climates retard denitrification (Collings et al. 2020). Denitrification rates can vary with the season due to a difference in mean water temperature. Denitrification is typically greatest in the spring and lowest in the summer and early autumn (Zhong et al. 2010). A decrease in denitrification at low temperature can be compensated by an increase in hydraulic retention time (Wicke et al. 2015). Thus, the climate can affect the extent to which a design modification is effective for nitrate removal in SCM. The ability of SCM in removing nitrate is limited under high-intensity rainfall when most of the runoff overflows the system or infiltrates at a faster rate, thereby limiting reaction time in the SCM. In contrast, rainfall promotes the leaching of N-source from soil bulk and favors nitrate uptake by plants (Mantelin and Touraine 2004). In addition to rainfall intensity, increasing the dry duration between rainfall events can improve the nitrate removal from trapped pore water (Berger et al. 2019, Norton et al. 2017). Consequently, a longer antecedent dry period between rainfall events partially explains why nitrate removal in hot-summer Mediterranean climates is higher than the removal in a humid subtropical climate. Collectively, these results indicate that climate can influence moisture content in SCM and affect nitrate removal by biological processes.

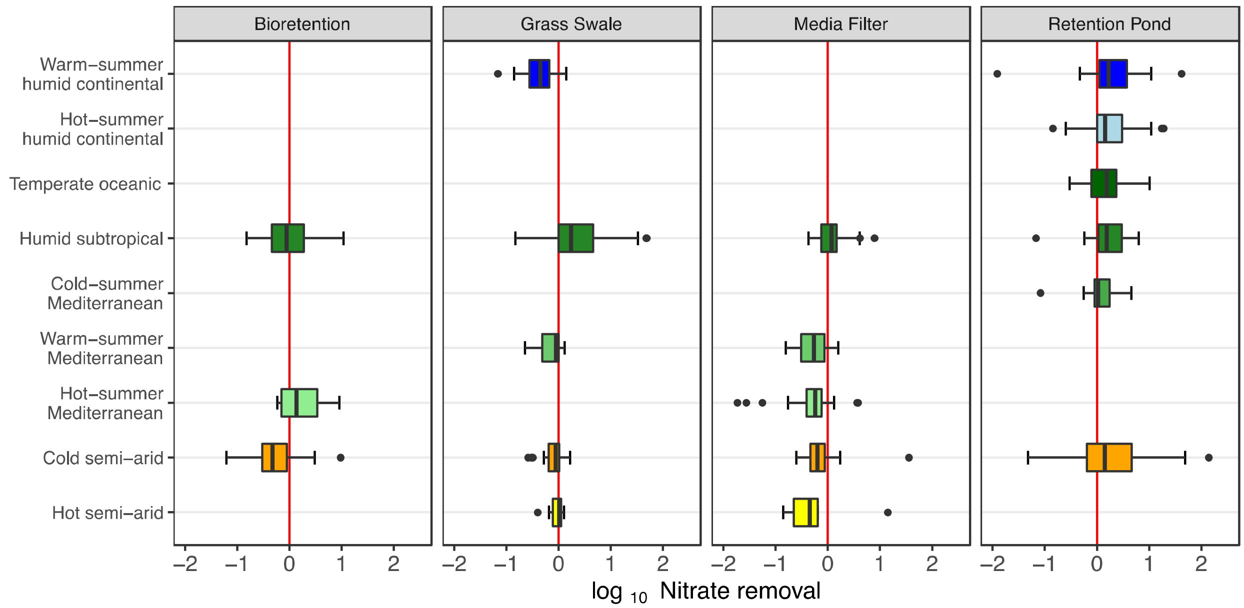


Figure 2.3 **– Effect of climate on the performance of different green stormwater infrastructure in removing nitrate from stormwater runoff. Climate is classified based on Köppen-Geiger climate classification. The vertical red line indicates no log removal of nitrate. Numbers between parenthesis represent n-values of boxplot analysis.**

2.5. Effect of SCM design on nitrate removal

To isolate the effect of specific design factors, we selected SCM under the same climate conditions from the BMP database and compared the nitrate removal between SCM as a function of different design variables. The analysis reveals specific design parameters that could change the SCM from a net sink of nitrate to a net source.

2.5.1. Which design factors affect nitrate removal in retention ponds?

To isolate the effect of designs, we compared nitrate removal between ponds within the same climate regions that differed by a single design factor. Due to the lack of available data for all climate regions, our data analysis was possible for retention ponds located in 2 climate regions: hot-summer humid continental climate and humid subtropical climate. Our analysis reveals that nitrate removal in retention ponds varies based on the pond’s depth, vegetation density, and watershed area (Figure **2.4**). The removal decreases significantly ($p < 0.01$) when the depth of the pond increases from 0.3 m to 0.7 m. An increase in depth limits mass transfer of nitrate to the reactive zone, interface between biofilm or sediment and water columns, thereby decreasing nitrate removal (Cubas et al. 2019). A low volume of water column per anoxic zone near sediments (Chen et al. 2019b) is critical for enhancing denitrification (Mayo 2020). This could make a shallow pond or wetland more effective in removing nitrate than deeper ponds (Chen et al. 2019a). An increase in vegetation density significantly ($p < 0.05$) increases the removal of nitrate in retention ponds, showing that plants can play a critical role in nitrate biotransformation (Vymazal 2020). Plants boost denitrification by providing endogenous carbon through root exudates for root microbiome to get energy via denitrification (Wu et al. 2017) and increasing the hydraulic retention time (HRT) by blocking the flow in the pond (Khan et al. 2019), all of which could explain the positive effect of vegetation on nitrate removal in retention ponds. The carbon released from decomposition of plant materials provides carbon source critical for nitrogen mineralization (Hooker and Stark 2008). However, decomposition of plants detritus, unless removed, can also release nitrogen into water (Knops et al. 2002). Thus, the pond should be maintained to prevent excessive accumulation of plant debris. Retention ponds located in smaller watershed areas also remove more nitrate than those located in larger watershed areas, possibly because of the increase of nutrient loading in larger watersheds (Zhang et al. 2019b). Our analysis reveals that a retention pond connected to a small watershed (5.2×10^5 m^2) removes nitrate, whereas a pond connected to a larger watershed (1.75×10^6 m^2) exports nitrate. We attribute the pond size-dependent removal capacity to the increased loading of nitrate from larger watershed due to deposition and biodegradation of plant debris (Jani et al. 2020, Krometis et al. 2009) and faster exhaustion of pond capacity to remove nitrate. In this case, pre-treatment of influent water by an algal pond could fully nitrify the influent and increase the overall nitrate removal (Mayo 2020). Based on our prior analysis, the nitrate removal capacity of retention ponds is nearly similar in all climates. The design factor, rather than climate, explains the variability in the nitrate removal capacity of retention ponds.

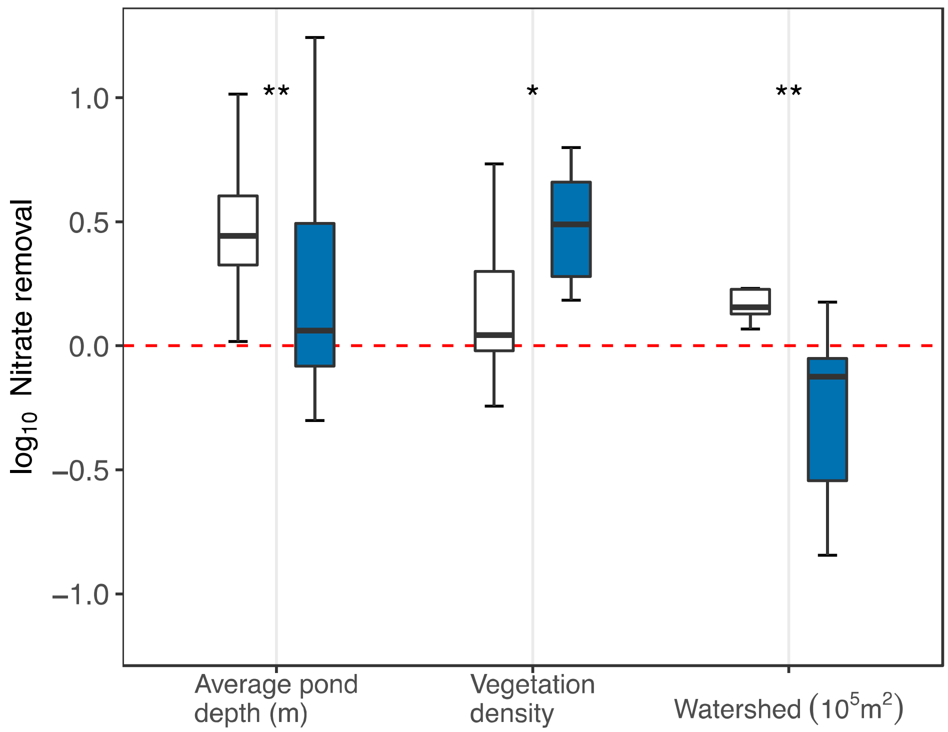


Figure 2.4 – Nitrate removal of retention ponds within the same climate classification is sensitive to design. Watershed area and pond depth were imported from the BMP Database summary report. The average pond depth was calculated as the ratio between permanent pool volume (m^3) and the permanent pool surface area (m^2). Vegetation density was evaluated using Google Earth Software. The SCM involved in the pond depth and watershed area analysis were located in a hot-summer humid continental climate and the SCM involved in the analysis of the vegetation density were located in a humid subtropical climate. Statistical analysis representation: *p-value < 0.05, **p-value < 0.01.

2.5.2. Does grass swale length affect nitrate removal?

Our analysis shows that nitrate removal by grass swales is highly sensitive to local climate conditions (Figure **2.5**). Grass swales remove nitrate only in humid subtropical climates and an increase in the length of a grass swale increases nitrate removal. In other climates, grass swales can become a source of nitrate. Although moisture is critical for plant health and may explain the nitrate removal variability, many plants are still healthy even in absence of water as a result of the plant's phenotype and physiological characteristics. In humid subtropic climates, a high decomposition rate of organic debris provides carbon source essential for dentification. Moisture, carbon and nitrogen availably has been shown to increase abundance of microorganism responsible for denitrification (ATTARD et al. 2011, Shrewsbury et al. 2016). High moisture content or a submerged layer in the soil is needed to create local anoxic conditions (Hsieh et al. 2007), which can facilitate denitrification. Thus, we speculated that soil conditions in subtropic climate is more favorable for denitrification than other dry climate because of soil moisture, carbon and nitrogen abundance that shape the denitrifier communities in soil. High variability in nitrate removal can be attributed to the difference in a contact time as the data shows that increasing the length of a grass swale in a humid subtropical climate increased nitrate removal. However, other unexplored design parameters such as centerline slope and vegetation cover can affect in what manner and for how long the stormwater is detained on the grass swale, and hence their ability to remove nitrate (Wicke et al. 2015). Thus, the length of the grass swale may not be the only key factor affecting nitrate removal.

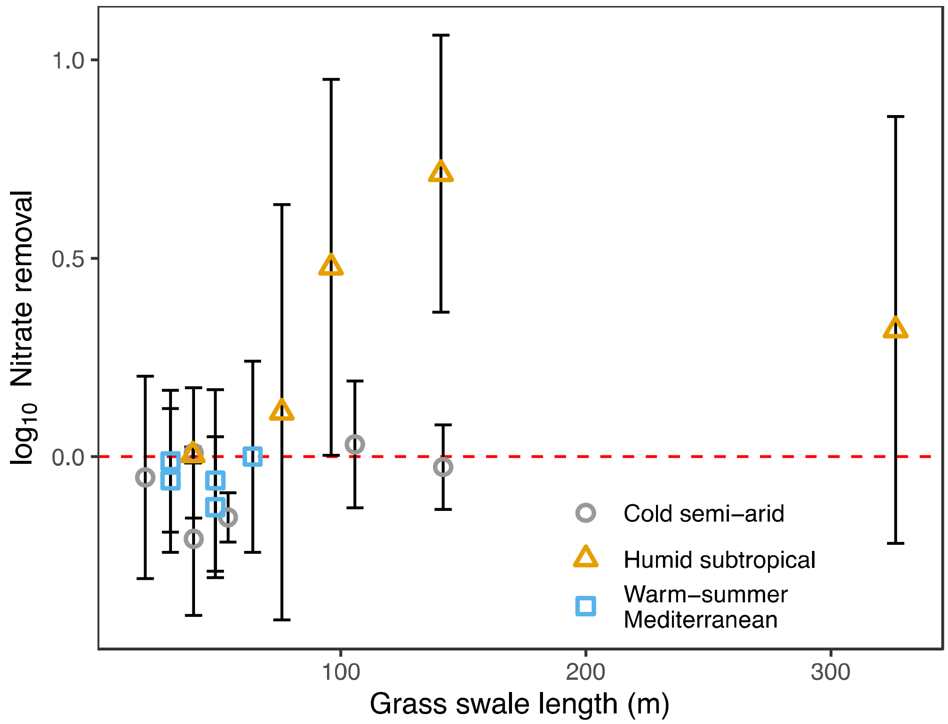


Figure 2.5 – **Removal of nitrate by grass swale varies with the length of grass swale in a humid subtropical climate, and grass swale acts as a source (a net negative removal) in cold semi-arid or warm-summer Mediterranean climates. Sixteen (16) grass swales from the BMP database were analyzed based on average nitrate removal. The horizontal red dashed line represents no nitrate removal, whereas positive and negative values represent net-removal and net-export of nitrate, respectively.**

2.5.3. Does flow rate affect nitrate removal in the media filter?

The flow rate through media filters can depend on rainfall intensity and watershed area. An increase in these variables can lead to increases in the discharge rate or nitrogen loading to media filters. We compared the nitrate removal of paired sand filters located in the same local climate based on the Rational Method ($Q = CiA$), which permits the calculation of the peak runoff Q (Chin 2019). The precipitation (i) for the paired sand filters was assumed to be the same, and the BMP database provided the impervious area (e.g., equivalent to the runoff coefficient, C) and the watershed area (A) for each sand filter. Our analysis shows that media filters act as a net source of nitrate in all climates and flow conditions, except in a humid subtropical climate with an average flow rate of 0.33 m^3 min^{-1} (Figure 2.6). In humid subtropical climates, low runoff volumes may cause a positive removal of nitrate, but heavy precipitation could reduce the removal rates (Feng et al. 2012) possibly because of a decrease in hydraulic retention time (Gottinger et al. 2011, Nakhla and Farooq 2003). As media filters mainly consist of sand, they have limited capacity to remove nitrate by adsorption or biotransformation. Furthermore, oxidation of ammonium to nitrate (Landsman and Davis 2018a) can make the filter itself a source of nitrate leaching. Therefore, filter media should not be used for nitrate removal from stormwater unless amendments are added to sand filters which can substantially increase nitrate removal (Palmer et al. 2013, Ulrich et al. 2017).

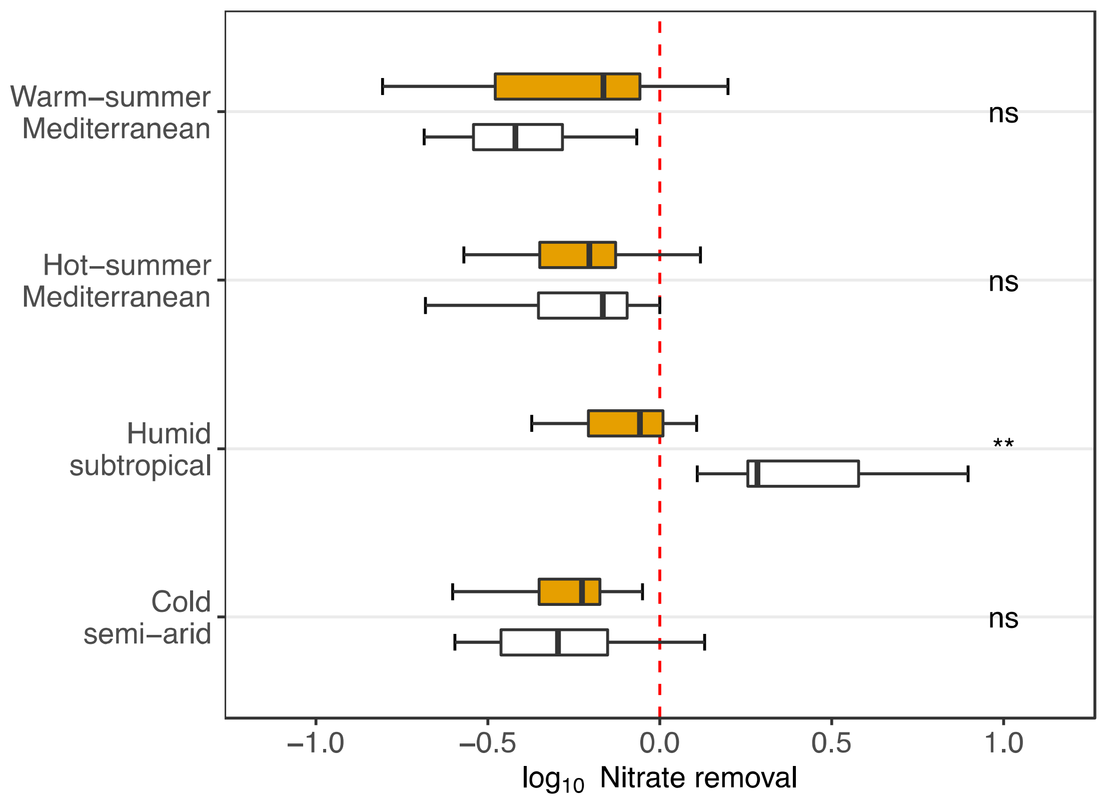


Figure 2.6 – Effect of peak runoff (Q, m^3 min^{-1}) on nitrate removal of media filter within different climates. Vertical dashed line represents no nitrate removal, while positive and negative values represent net-removal and net-source of nitrate, respectively. Statistical analysis representation: ns = no-significance, **p-value < 0.01.

2.5.4. Which design factors of bioretention systems are critical for total nitrogen removal?

For bioretention systems, we analyzed total nitrogen (TN) instead of nitrate due to a lack of sufficient paired data in the BMP database linking design parameters and nitrate removal. We compared the TN removal of paired bioretention systems that were located within the same local climate but differed in only one design factor. We extracted the following data for the design parameters from published studies that analyzed the same bioretention systems reported on the BMP database: the presence of IWS or submerged zone (Hunt et al. 2006) or plastic linen (Li et al. 2009), surface infiltration (< 0.13 or > 0.15 mm h^{-1}) and depth (0.3 or 0.6 m) of the saturated/submerged zone and surface infiltration (Brown and Hunt 2008), and biofilter area-to-depth ratio of 230 or 480 (Brown and Hunt 2012). Our analysis shows that TN removal in bioretention systems can vary based on design factors, but the presence of plastic linen and the depth of the saturated zone do not vary TN removal significantly (Figure 2.7). The presence of a saturated zone slightly improved log TN removal from -0.63 to -0.24. Previous laboratory studies have demonstrated an improvement in nitrate removal by adding a saturated zone (Alikhani et al. 2020, Lopez-Ponnada et al. 2020, Nabiul Afrooz and Boehm 2017, Zinger et al. 2013, Zinger et al. 2020). The saturated zone typically helps maintain anoxic conditions (Ding et al. 2019) and improves nitrate removal (Palmer et al. 2013). It should be noted that in the presence of a submerged zone, denitrification only accounts for 23% of nitrogen removal in bioretention systems (Norton et al. 2017), and DNRA may dominate nitrate removal in areas with rewetting occurrence (Friedl et al. 2018). Our analysis reveals that an increase in depth of the saturated zone from 0.3 m to 0.6 m did not significantly improve TN removal, indicating an increase in nitrate removal is probably offset by a decrease in ammonium removal in the saturated layer. A comparison of bioretention systems with area-to-depth ratios of 230 and 480 shows that an increase in bioretention area significantly improves TN removal, possibly because the removal of particulate N occurs near the surface instead of the deep layer (Landsman and Davis 2018b).

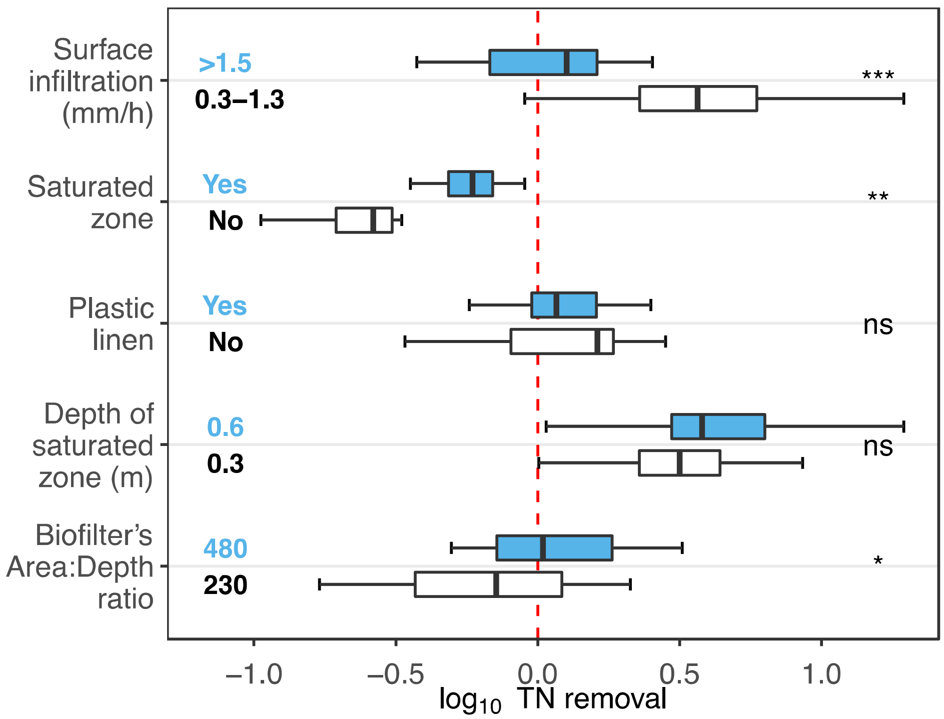


Figure 2.7 – Effect of various design parameters on the removal of total nitrogen (TN) in bioretention systems. Vertical red dashed line represents no removal of TN. Statistical analysis representation: ns = no-significance, *p-value < 0.05, **p-value < 0.01, ***p-value < 0.001.

TN removal is highly sensitive to stormwater infiltration, which is controlled by the hydraulic conductivity of filter media. An increase in retention time typically improves nitrate removal in bioretention systems (Alikhani et al. 2020, Berger et al. 2019, Ding et al. 2019, Lopez-Ponnada et al. 2020, Shrestha et al. 2018). Our analysis shows that infiltration rates exceeding 0.15 cm h^{-1} rapidly decrease TN removal from net positive to negative. Hydraulic retention time typically increases with an increase in rainfall intensity or catchment area as both factors produce larger runoff volume and reduce TN removal (Alikhani et al. 2020). Additionally, rainfall patterns could affect the levels of dissolved organic carbon (DOC) in stormwater (Lipczynska-Kochany 2018) and the infiltration rate through SCM, which would have further implications on nitrate removal (Fidel et al. 2018). In summary, bioretention systems should be designed with a greater area, shallow submerged layer, and a relatively small catchment area, if possible.

2.5.5. How does vegetation influence the removal of different N species?

Nearly all SCM contains vegetation which can directly uptake dissolved nitrogen species such as nitrate and ammonium via roots (Parker and Newstead 2014, Wang et al. 2012). The presence of vegetation typically increases the removal of nitrogen by more than 75% (Barron et al. 2019, Davis et al. 2006), although some studies observed no benefits of plants (Palmer et al. 2013, Valtanen et al. 2017a). Our analysis shows that the presence of vegetation – mostly emergent macrophytes – can significantly increase TN removal (Figure 2.8), but it does not affect the removal of NH_4^+, NO_3^-, and total dissolved nitrogen (TDN). The discrepancy could be attributed to the soil pH as DNRA can contribute to 18% of nitrate removal if soil pH is neutral or alkaline (Zhang et al. 2015). In addition, the lack of removal of nitrogen-based compounds could also be attributed to the leaching of different N species from the fertilizer that may have been applied to maintain the plant health and to the complexities of nitrate uptake mechanisms in plants. Compost is often added to support plant growth, which can leach nitrate (Shrestha et al. 2018). In those cases, organic biomass such as woodchips, bark, mulch, wood dust, compost, or biochar can be used to improve denitrification (Greenan et al. 2009, He et al. 2019).

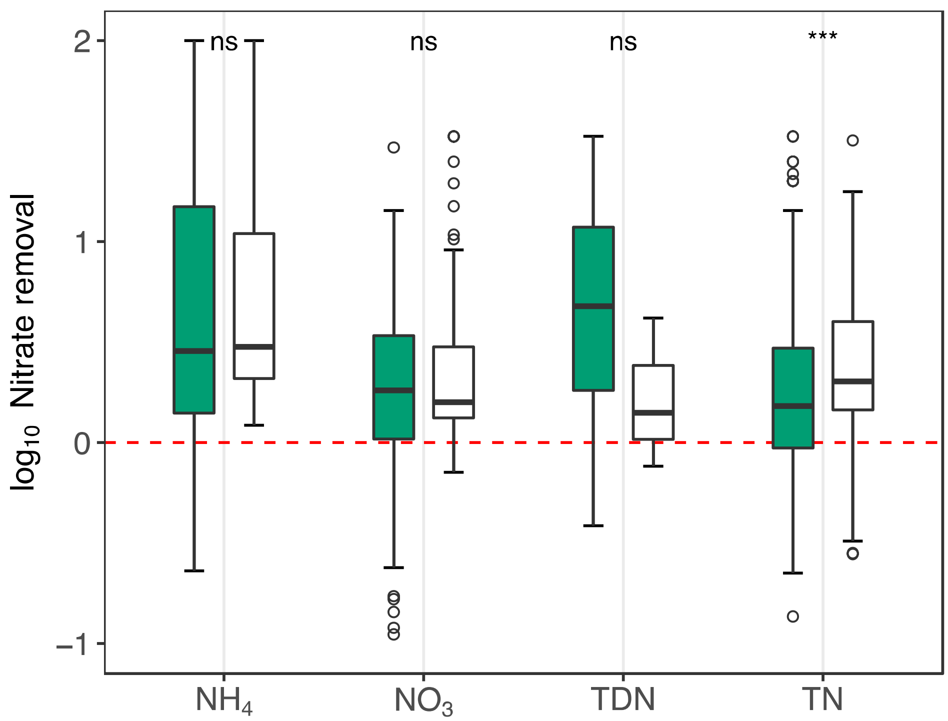


Figure 2.8 – Effect of presence or absence of vegetation (emergent macrophytes) on the removal of different nitrogen-based contaminants in bioretention systems. Green-filled boxplots represent bioretention systems with vegetation, while empty boxplot represents bioretention systems without vegetation. Data analysis was based on 29 peer-reviewed articles. A horizontal red dashed line represents no nitrate removal. Negative values for nitrate removal represent export or leaching of nitrate, while positive values represent net-positive removal of nitrate. Statistical analysis representation: ns = no-significance, ***p-value < 0.001.

Nitrate uptake capacity of a plant is sensitive to the functional properties of the transporters in roots, density in the plasma membrane of root cells, the surface and architecture of the root, plants types, root depth, and leave density (Cardinale 2011, Hallin et al. 2015, Morse et al. 2018, Noguero and Lacombe 2016), because they all directly or indirectly influence nitrate assimilation pathways. The nitrate assimilation pathway in plants occurs in three steps: (1) nitrate uptake, (2) nitrate reduction, and (3) nitrate storage (Crawford and Glass 1998, Tischner 2000). First, anionic nitrate in the soil is carried toward the root systems by bulk flow and actively transported across the plasma membrane. Roots use transporters (Crawford 1995) encoded by *NRT1* (low affinity) or *NRT2* (high affinity) genes that bind to nitrate and transport them through the plasma membrane of the root cells to the root symplast. Nitrate can either be utilized into amino acids or effluxed out of the cell by loading it into the xylem and the transporter cells translocate nitrate to the leave system where it is stored in the vacuole as nitrite. Furthermore, previous studies have shown that higher biomass content in plant and microbial community diversity enhances nitrate removal rates (Deng et al. 2020, Wen et al. 2010, Zhang et al. 2016), but plant's root development depends on the presence of inorganic nitrogen (e.g. nitrate and ammonium), pH and redox potential conditions (Bloom et al. 2002) which are likely to experience seasonal variability (Fatubarin and Olojugba 2014, Fernandes et al. 2002) and affect microbial communities (Mellado-Vázquez et al. 2019). Thus, root health, plant species and their growth rate in different SCM could affect nitrate removal.

To maintain charge balance during nitrate uptake, a proton is transported into root cells. As amendments can alter pH to affect plant uptake of nitrate (Revell et al. 2012), they can indirectly affect the ability of the plant to uptake nitrate. Plants can also indirectly affect nitrate removal by altering the moisture content in the filter media via evapotranspiration and the hydraulic conductivity of the soil (Valtanen et al. 2017a) by root architecture (Wang et al. 2020). The selection of plant types should be used as a design factor to increase nitrate removal.

2.5.6. Which amendments have maximum removal capacity?

Nitrate removal by bioretention systems or filter media can be increased by the addition of amendments. Through literature review, we divided amendments into five categories: (1) no amendment: only sand and/or soil; (2) organic amendments: compost, mulch, bark, and woodchips; (3) media mixture: a mix of three or more amendments including zeolite, tire crumb, printed paper, fly ash, bark, and water treatment residuals; (4) biochar; (5) iron-based amendments: zero-valent iron, iron fillings, iron-oxide from water treatment residues. Our analysis shows that the median removal capacity of amendments decreases in the following order: iron-based media > media mixture > organic amendment > biochar (Figure 2.9). Compared to control (no amendment), iron-based media, media mixture, and organic amendments removed significantly more nitrate ($p < 0.05$), whereas biochar offered no significant improvement in nitrate removal. These results indicate that biochar may not necessarily improve nitrate removal, although it may remove other N species such as NH_4^+ due to electrostatic adsorption based on the opposite surface charge between NH_4^+ and biochar (Hina et al. 2015, Vu et al. 2017).

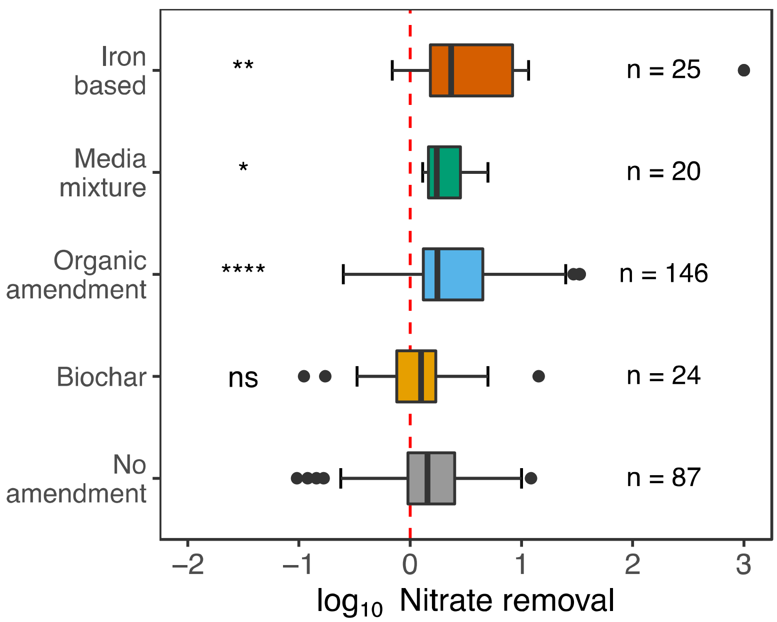


Figure 2.9 – Removal of nitrate in flow-through bioretention systems amended with diverse types of geomedia by analyzing the results of 24 articles. Negative values represent net export of nitrate, while positive values represent net removal. No amendment includes sand or soil. The organic amendments include compost, mulch, and other organics. Media mixture represents the mix of three or more amendments including zeolite, tire crumb, printed paper, fly ash, bark, and water treatment residuals. Statistical analysis representation: ns = no-significance, *p-value < 0.05, **p-value < 0.01, ***p-value < 0.001.

Bioretention systems with organic amendments show significantly more ($p < 0.05$) nitrate removal than bioretention systems without amendment. One possibility for high variability is the difference in the types of organic amendments used. Organic amendments typically provide dissolved organic carbon (an electron donor) to facilitate the reduction of nitrate (Chang et al. 2018, Pfenning and McMahon 1997); however, some organic amendments such as compost can also be a source of nitrate (Chahal et al. 2016). The amendment should be carefully selected to ensure they do not contribute nitrate to effluent. The addition of biochar to compost may decrease the leaching of nitrate from compost (Iqbal et al. 2015b), but such an alternative may not be enough to reduce the effluent nitrate concentration due to continuous net export of nitrate (Shrestha et al. 2018).

Our analysis revealed that the benefits of biochar on nitrate removal observed in laboratory studies may not be translated to field studies. Biochar rarely removes nitrate by adsorption due to a net negative surface charge (Iqbal et al. 2015a). However, biochar can modify microbial activity and affect denitrification. Based on laboratory studies, biochar can increase total nitrogen removal by increasing the enzyme activity and reduction of ammonium nitrogen, but the extent of the enzyme activity depends on biochar feedstock and vegetation growth stage (Jing et al. 2020). Biochar addition can increase total nitrogen removal due to the higher mineralization of organic N to NH_4^+ and NO_X which is subsequently denitrified (de Rozari et al. 2018). Similarly, the addition of biochar can slow down nitrate leaching from the biofilter and increase nitrate utilization by the denitrifying community (Berger et al. 2019). Our analysis indicates that the extent to which biochar can affect nitrate removal is limited in field conditions and biochar alone may not be an appropriate amendment for the removal of nitrate (Poor and Mohamed 2020).

Iron-based amendment shows significant improvement compared to any other amendments for nitrate removal. Several laboratory studies confirmed the advantage of iron-based media for nitrate removal (Chen et al. 2020, Shrestha et al. 2018). The improvement can be attributed to several mechanisms including electrochemical reduction, ligand complexation, coupled microbial reduction of nitrate and iron-oxidation, and nitrate sorption onto precipitated metal oxides (Reddy et al. 2014, Scholz et al. 2016, Valencia et al. 2020, Westerhoff and James 2003). For instance, oxidation of Fe^0 to Fe^{2+} releases two electrons that assist the electrochemical reduction of NO_3^-

to NH_4^+ (Westerhoff 2003). Ligand complexation can occur when Fe^{III} binds to nitrate to form a complex (Song et al. 2017), although nitrate adsorption can be greatly reduced in the presence of other anions such as sulfate (Kalaruban et al. 2016). Some studies have shown that media mixtures (e.g., mixtures of two or more amendments like spongy iron with pine bark, or zero-valent iron powder with activated carbon) can achieve more than 95% nitrate removal from stormwater runoff (Huang et al. 2015, Huno et al. 2018, Liu et al. 2013). Mixing iron amendments with biochar could help slow down the flow as well as increase the interactions between nitrate with iron amendment material, thereby improving overall capacity (Tian et al. 2019) even under extreme weather conditions.

2.6. Opportunities

Despite the challenges of variable performance, SCM are a cost-effective method to protect natural water bodies and improve water quality and quantity. Future studies should explore the effect of climate by conducting field experiments with similar variables in different climate regions. Thus, a collaboration between researchers from different institutions at multiple climates could help design the experiments to evaluate the effect of climate. Long-term monitoring of these systems in field conditions could help determine how future climate change extremes such as prolonged drought or high-intensity storms can affect the performance of SCM.

Plants in stormwater biofilters can have a significant association with fungi that could affect nitrate utilization. The fundamental process of nitrate uptake by fungi (Garrett and Amy 1979) and plants (Crawford and Glass 1998) have been explored separately in earlier studies; however, there is a lack of fundamental studies on the coexistence of fungi and plants in SCM and their role in denitrification (Fochi et al. 2017). Beneficial interactions between biochar and fungi have been observed (Gujre et al. 2020). This is particularly important because fungi have been shown to increase a plant tolerance in high salinity and drought conditions (Martínez-García et al. 2017). Fungi could facilitate nitrate uptake by actively transporting nitrate and by helping plants survive in harsh climates (Bücking and Kafle 2015). Future studies should explore whether and how the presence of fungi could increase nitrate removal in SCM and how to increase the abundance of fungi in the system.

Our analysis reveals that bioretention systems are not efficient at removing nitrate. Studies that optimize the design of bioretention systems related to media amendment, selected vegetation, and submerged layer controls would be helpful to gain further insight into these systems. Subsurface wetlands – a combination of retention ponds and bioretention systems – may maximize nitrate removal due to synergy between the removal mechanisms and added design flexibility (Saeed and Sun 2012). By actively supplying electrons or inducing reducing conditions via an external power source charged by solar panels, nitrate removal capacity may be enhanced particularly during the rainfall period (Yang et al. 2019). However, further cost-benefit analysis should be performed to evaluate the feasibility of such approach.

Although grass swales have poor performance in nitrate removal, this control measure is widely used as roadside infrastructure due to its simplicity and low maintenance requirements (Stagge et al. 2012). However, no study to date has analyzed the effect of the local climate on the performance of grass swales. Future studies should examine the specific mechanisms by which local climate can affect nitrate removal in SCM as the climate would affect the conditions and activities of plants and root microbes, which in turn could affect biological nitrate removal. In particular, future studies should examine how SCM that can operate under different hydraulic conditions that may occur in different climate scenarios (Okaikue-Woodi et al. 2020). Furthermore, local carbon dioxide and humidity are the driving force for plant evapotranspiration and water conductance, respectively (de Boer et al. 2011, Patanè 2011). While evapotranspiration remains poorly understood in SCM (Ebrahimian et al. 2019), no study in the context of SCM has shown how plants and local CO_2 levels may affect the performance of SCM that contain plants.

Climate change is expected to alter *Köppen-Geiger* climate classification (Beck et al. 2018), which was used in this study to evaluate the effect of climate on nitrate removal capacities of SCM. In some regions, precipitation frequency or intensity is expected to increase (Tabari 2020), while other regions are expected to experience more drying duration between rainfall events (Hari et al. 2020). The resulting changes in moisture content in SCM and the loading of nitrate to SCM are expected to affect nitrate removal (Berger et al. 2019, He et al. 2020). However, SCM are rarely designed to account for changes in these variables due to climatic changes (Yazdanfar and Sharma 2015). Our analysis shows that retention ponds can be more effective in treating high nitrate loading in regions with greater rainfall events through modification of the water depth and vegetation in the ponds. The analysis also reveals that nitrate removal capacity of bioretention systems, the most commonly used SCM, is sensitive to changing climate. The addition of specific amendments can increase their capacity in all climate conditions. The analysis also shows specific design conditions that could improve nitrate removal. Because consistent moisture is needed to improve nitrate removal, alternative innovative designs such as dual-mode stormwater-greywater biofilters could be used in a dry climate (Barron et al. 2019, Barron et al. 2020). There is also a lack of mechanistic study on how climate conditions affect the denitrifying community in SCM. Future studies should also evaluate whether climate conditions or amendments explain changes in the microbial community in SCM.

2.7. Conclusions

Analysis of the performance of the four most common SCM from 60 locations listed in the BMP database reveals the following conclusions:

- Climate and design both affect the nitrate removal capacity of SCM, but the extent to which they are critical varies between SCM type.
- Low efficiency of SCM in removing nitrate could be mostly related to nitrification in oxic conditions and low efficiency of removal of nitrate in high flow conditions.
- Retention ponds provide the best nitrate removal rates partially because of the long residence time. Their removal is more sensitive to design than climate. The shallow depth and smaller catchment area improve the nitrate removal capacity of retention ponds.
- Media filter (sand filter) mostly exports nitrate irrespective of the local climate or design specifications.
- Bioretention systems are highly unreliable for the removal of nitrate. Optimizing their design by adding submerged layers and amendments, increasing area to depth, and lowering infiltration rates could significantly improve their nitrate removal capacity and make them resilient in different climates or seasons.
- Nitrate removal capacities of filter media or bioretention systems can be improved by adding amendments including organic biomass, iron-based media, and media mixtures; however, biochar addition provides no benefits for nitrate removal.
- To alleviate the detrimental effect of changing climate on nitrate removal, retention ponds and bioretention systems with amendments should be implemented.

2.8. References

Alikhani, J., Nietch, C., Jacobs, S., Shuster, B. and Massoudieh, A. (2020) Modeling and Design Scenario Analysis of Long-Term Monitored Bioretention System for Rainfall-Runoff Reduction to Combined Sewer in Cincinnati, OH. Journal of Sustainable Water in the Built Environment 6(2), 17.

Alleman, J.E., Keramida, V. and Pantea-Kiser, L. (1987) Light induced Nitrosomonas inhibition. Water Research 21(4), 499-501.

ATTARD, E., RECOUS, S., CHABBI, A., DE BERRANGER, C., GUILLAUMAUD, N., LABREUCHE, J., PHILIPPOT, L., SCHMID, B. and LE ROUX, X. (2011) Soil environmental conditions rather than denitrifier abundance and diversity drive potential denitrification after changes in land uses. Global Change Biology 17(5), 1975-1989.

Ballard, T.C., Sinha, E. and Michalak, A.M. (2019) Long-Term Changes in Precipitation and Temperature Have Already Impacted Nitrogen Loading. Environmental Science & Technology 53(9), 5080-5090.

Barrett, M.E., Walsh, P.M., Malina, J.F. and Charbeneau, R.J. (1998) Performance of Vegetative Controls for Treating Highway Runoff. Journal of Environmental Engineering 124(11), 1121-1128.

Barron, N.J., Deletic, A., Jung, J., Fowdar, H., Chen, Y. and Hatt, B.E. (2019) Dual-mode stormwater-greywater biofilters: The impact of alternating water sources on treatment performance. Water Research 159, 521-537.

Barron, N.J., Hatt, B., Jung, J., Chen, Y. and Deletic, A. (2020) Seasonal operation of dual-mode biofilters: The influence of plant species on stormwater and greywater treatment. Science of the Total Environment 715, 136680.

Beck, H.E., Zimmermann, N.E., McVicar, T.R., Vergopolan, N., Berg, A. and Wood, E.F. (2018) Present and future Köppen-Geiger climate classification maps at 1-km resolution. Scientific Data 5(1), 180214.

Berger, A.W., Valenca, R., Miao, Y., Ravi, S., Mahendra, S. and Mohanty, S.K. (2019) Biochar increases nitrate removal capacity of woodchip biofilters during high-intensity rainfall. Water Research 165, 8.

Bettez, N.D. and Groffman, P.M. (2012) Denitrification Potential in Stormwater Control Structures and Natural Riparian Zones in an Urban Landscape. Environmental Science & Technology 46(20), 10909-10917.

Bhatnagar, A. and Sillanpää, M. (2011) A review of emerging adsorbents for nitrate removal from water. Chemical Engineering Journal 168(2), 493-504.

Blecken, G.-T., Zinger, Y., Deletić, A., Fletcher, T.D., Hedström, A. and Viklander, M. (2010) Laboratory study on stormwater biofiltration: Nutrient and sediment removal in cold temperatures. Journal of Hydrology 394(3), 507-514.

Blecken, G.-T., Zinger, Y., Muthanna, T.M., Deletic, A., Fletcher, T.D. and Viklander, M. (2007) The influence of temperature on nutrient treatment efficiency in stormwater biofilter systems. Water Science and Technology 56(10), 83-91.

Bloom, A.J., Meyerhoff, P.A., Taylor, A.R. and Rost, T.L. (2002) Root Development and Absorption of Ammonium and Nitrate from the Rhizosphere. Journal of Plant Growth Regulation 21(4), 416-431.

Bock, E., Smith, N., Rogers, M., Coleman, B., Reiter, M., Benham, B. and Easton, Z.M. (2015) Enhanced nitrate and phosphate removal in a denitrifying bioreactor with biochar. J Environ Qual 44(2), 605-613.

Boeykens, S.P., Piol, M.N., Samudio Legal, L., Saralegui, A.B. and Vázquez, C. (2017) Eutrophication decrease: Phosphate adsorption processes in presence of nitrates. Journal of Environmental Management 203, 888-895.

Bowles, T.M., Atallah, S.S., Campbell, E.E., Gaudin, A.C.M., Wieder, W.R. and Grandy, A.S. (2018) Addressing agricultural nitrogen losses in a changing climate. Nature Sustainability 1(8), 399-408.

Brady, N.C., Weil, R.R. and Weil, R.R. (2008) The nature and properties of soils, Prentice Hall Upper Saddle River, NJ.

Brown, R.A. and Hunt, W.F. (2008) Low Impact Development for Urban Ecosystem and Habitat Protection, pp. 1-10.

Brown, R.A. and Hunt, W.F. (2012) Improving bioretention/biofiltration performance with restorative maintenance. Water Science and Technology 65(2), 361-367.

Brudler, S., Arnbjerg-Nielsen, K., Hauschild, M.Z. and Rygaard, M. (2016) Life cycle assessment of stormwater management in the context of climate change adaptation. Water Research 106, 394-404.

Bu, C., Wang, Y., Ge, C., Ahmad, H.A., Gao, B. and Ni, S.-Q. (2017) Dissimilatory Nitrate Reduction to Ammonium in the Yellow River Estuary: Rates, Abundance, and Community Diversity. Scientific Reports 7(1), 6830.

Bücking, H. and Kafle, A. (2015) Role of arbuscular mycorrhizal fungi in the nitrogen uptake of plants: current knowledge and research gaps. Agronomy 5(4), 587-612.

Bulseco, A.N., Giblin, A.E., Tucker, J., Murphy, A.E., Sanderman, J., Hiller-Bittrolff, K. and Bowen, J.L. (2019) Nitrate addition stimulates microbial decomposition of organic matter in salt marsh sediments. Global Change Biology 25(10), 3224-3241.

Burgin, A.J. and Hamilton, S.K. (2007) Have we overemphasized the role of denitrification in aquatic ecosystems? A review of nitrate removal pathways. Frontiers in Ecology and the Environment 5(2), 89-96.

Cardinale, B.J. (2011) Biodiversity improves water quality through niche partitioning. Nature 472(7341), 86-89.

Chahal, M.K., Shi, Z.Q. and Flury, M. (2016) Nutrient leaching and copper speciation in compost-amended bioretention systems. Science of the Total Environment 556, 302-309.

Chang, N.-B., Wen, D., McKenna, A.M. and Wanielista, M.P. (2018) The Impact of Carbon Source as Electron Donor on Composition and Concentration of Dissolved Organic Nitrogen in Biosorption-Activated Media for Stormwater and Groundwater Co-Treatment. Environmental Science & Technology 52(16), 9380-9390.

Chen, L., D'Aoust, P.M., Rennie, C.D., Poulain, A., Pick, F., Wang, R. and Delatolla, R. (2019a) Numerical investigation on the impact of wind-induced hydraulics on dissolved oxygen characteristics in a shallow stormwater pond. Water Quality Research Journal 54(4), 309-325.

Chen, L.Y., Delatolla, R., D'Aoust, P.M., Wang, R., Pick, F., Poulain, A. and Rennie, C.D. (2019b) Hypoxic conditions in stormwater retention ponds: potential for hydrogen sulfide emission. Environmental Technology 40(5), 642-653.

Chen, Y., Shao, Z., Kong, Z., Gu, L., Fang, J. and Chai, H. (2020) Study of pyrite based autotrophic denitrification system for low-carbon source stormwater treatment. Journal of Water Process Engineering 37, 101414.

Chin, D.A. (2019) Estimating Peak Runoff Rates Using the Rational Method. Journal of Irrigation and Drainage Engineering 145(6), 04019006.

Chrétien, F., Gagnon, P., Thériault, G. and Guillou, M. (2016) Performance Analysis of a Wet-Retention Pond in a Small Agricultural Catchment. Journal of Environmental Engineering 142(4), 04016005.

Clary, J., Leisenring, M., Poresky, A., Earles, A. and Jones, J. (2011) World Environmental and Water Resources Congress 2011, pp. 441-449.

Collings, E.J., Bunce, J.T., Jong, M.-C. and Graham, D.W. (2020) Impact of Cold Temperatures on Nitrogen Removal in Denitrifying Down-Flow Hanging Sponge (DDHS) Reactors. Water 12(7), 2029.

Collins, K.A., Lawrence, T.J., Stander, E.K., Jontos, R.J., Kaushal, S.S., Newcomer, T.A., Grimm, N.B. and Cole Ekberg, M.L. (2010a) Opportunities and challenges for managing nitrogen in urban stormwater: A review and synthesis. Ecological Engineering 36(11), 1507-1519.

Collins, K.A., Lawrence, T.J., Stander, E.K., Jontos, R.J., Kaushal, S.S., Newcomer, T.A., Grimm, N.B. and Ekberg, M.L.C. (2010b) Opportunities and challenges for managing nitrogen in urban stormwater: A review and synthesis. Ecological Engineering 36(11), 1507-1519.

Crawford, N.M. (1995) Nitrate: nutrient and signal for plant growth. The Plant Cell 7(7), 859-868.

Crawford, N.M. and Glass, A.D.M. (1998) Molecular and physiological aspects of nitrate uptake in plants. Trends in Plant Science 3(10), 389-395.

Cubas, F.J., Holbrook, R.D., Novak, J.T., Godrej, A.N. and Grizzard, T.J. (2019) Effective depth controls the nitrate removal rates in a water supply reservoir with a high nitrate load. Science of the Total Environment 673, 44-53.

Cui, M. and Caldwell, M.M. (1997) A large ephemeral release of nitrogen upon wetting of dry soil and corresponding root responses in the field. Plant and Soil 191(2), 291-299.

Davis, A.P., Shokouhian, M., Sharma, H. and Minami, C. (2001) Laboratory study of biological retention for urban stormwater management. Water Environment Research 73(1), 5-14.

Davis, A.P., Shokouhian, M., Sharma, H. and Minami, C. (2006) Water Quality Improvement through Bioretention Media: Nitrogen and Phosphorus Removal. Water Environment Research 78(3), 284-293.

de Boer, H.J., Lammertsma, E.I., Wagner-Cremer, F., Dilcher, D.L., Wassen, M.J. and Dekker, S.C. (2011) Climate forcing due to optimization of maximal leaf conductance in subtropical vegetation under rising CO₂. Proceedings of the National Academy of Sciences 108(10), 4041-4046.

de Rozari, P., Greenway, M. and El Hanandeh, A. (2018) Nitrogen removal from sewage and septage in constructed wetland mesocosms using sand media amended with biochar. Ecological Engineering 111, 1-10.

Deegan, L.A., Johnson, D.S., Warren, R.S., Peterson, B.J., Fleeger, J.W., Fagherazzi, S. and Wollheim, W.M. (2012) Coastal eutrophication as a driver of salt marsh loss. Nature 490(7420), 388-392.

Deletic, A. and Fletcher, T.D. (2006) Performance of grass filters used for stormwater treatment—a field and modelling study. Journal of Hydrology 317(3), 261-275.

Deng, Q., Wan, L., Li, X., Cao, X., Zhou, Y. and Song, C. (2020) Metagenomic evidence reveals denitrifying community diversity rather than abundance drives nitrate removal in stormwater biofilters amended with different organic and inorganic electron donors. Chemosphere 257, 127269.

Ding, B., Rezanezhad, F., Gharedaghloo, B., Van Cappellen, P. and Passeport, E. (2019) Bioretention cells under cold climate conditions: Effects of freezing and thawing on water infiltration, soil structure, and nutrient removal. Science of the Total Environment 649, 749-759.

Dodds, W.K., Bouska, W.W., Eitzmann, J.L., Pilger, T.J., Pitts, K.L., Riley, A.J., Schloesser, J.T. and Thornbrugh, D.J. (2009) Eutrophication of U.S. Freshwaters: Analysis of Potential Economic Damages. Environmental Science & Technology 43(1), 12-19.

Ebrahimian, A., Wadzuk, B. and Traver, R. (2019) Evapotranspiration in green stormwater infrastructure systems. Science of the Total Environment 688, 797-810.

Eick, M.J., Brady, W.D. and Lynch, C.K. (1999) Charge Properties and Nitrate Adsorption of Some Acid Southeastern Soils. Journal of Environmental Quality 28(1), 138-144.

Ekpete, D.M. and Cornfield, A.H. (1964) Losses, through Denitrification, from Soil of Applied Inorganic Nitrogen even at Low Moisture Contents. Nature 201(4916), 322-323.

Fatubarin, A. and Olojugba, M. (2014) Effect of rainfall season on the chemical properties of the soil of a Southern Guinea Savanna ecosystem in Nigeria. Journal of Ecology and the Natural Environment 6(4), 182-189.

Feng, S., Xie, S., Zhang, X., Yang, Z., Ding, W., Liao, X., Liu, Y. and Chen, C. (2012) Ammonium removal pathways and microbial community in GAC-sand dual media filter in drinking water treatment. Journal of Environmental Sciences 24(9), 1587-1593.

Fernandes, S.A.P., Bernoux, M., Cerri, C.C., Feigl, B.J. and Piccolo, M.C. (2002) Seasonal variation of soil chemical properties and CO2 and CH4 fluxes in unfertilized and P-fertilized pastures in an Ultisol of the Brazilian Amazon. Geoderma 107(3), 227-241.

Fidel, R.B., Laird, D.A. and Spokas, K.A. (2018) Sorption of ammonium and nitrate to biochars is electrostatic and pH-dependent. Scientific Reports 8(1), 17627.

Flite III, O.P., Shannon, R.D., Schnabel, R.R. and Parizek, R.R. (2001) Nitrate Removal in a Riparian Wetland of the Appalachian Valley and Ridge Physiographic Province. Journal of Environmental Quality 30(1), 254-261.

Fochi, V., Chitarra, W., Kohler, A., Voyron, S., Singan, V.R., Lindquist, E.A., Barry, K.W., Girlanda, M., Grigoriev, I.V., Martin, F., Balestrini, R. and Perotto, S. (2017) Fungal and plant gene expression in the Tulasnella calospora–Serapias vomeracea symbiosis provides clues about nitrogen pathways in orchid mycorrhizas. New Phytologist 213(1), 365-379.

Friedl, J., De Rosa, D., Rowlings, D.W., Grace, P.R., Müller, C. and Scheer, C. (2018) Dissimilatory nitrate reduction to ammonium (DNRA), not denitrification dominates nitrate reduction in subtropical pasture soils upon rewetting. Soil biology and biochemistry 125, 340-349.

Gamble, T.N., Betlach, M.R. and Tiedje, J.M. (1977) Numerically Dominant Denitrifying Bacteria from World Soils. Applied and Environmental Microbiology 33(4), 926-939.

Garfí, M., Pedescoll, A., Bécares, E., Hijosa-Valsero, M., Sidrach-Cardona, R. and García, J. (2012) Effect of climatic conditions, season and wastewater quality on contaminant removal efficiency of two experimental constructed wetlands in different regions of Spain. Science of the Total Environment 437, 61-67.

Garrett, R.H. and Amy, N.K. (1979) Advances in Microbial Physiology. Rose, A.H. and Morris, J.G. (eds), pp. 1-65, Academic Press.

Glassman, S.I., Weihe, C., Li, J., Albright, M.B.N., Looby, C.I., Martiny, A.C., Treseder, K.K., Allison, S.D. and Martiny, J.B.H. (2018) Decomposition responses to climate depend on microbial community composition. Proceedings of the National Academy of Sciences 115(47), 11994-11999.

Gold, A.C., Thompson, S.P. and Piehler, M.F. (2019) Nitrogen cycling processes within stormwater control measures: A review and call for research. Water Research 149, 578-587.

Gottinger, A.M., McMartin, D.W., Price, D. and Hanson, B. (2011) The effectiveness of slow sand filters to treat Canadian rural prairie water. Canadian Journal of Civil Engineering 38(4), 455-463.

Greenan, C.M., Moorman, T.B., Parkin, T.B., Kaspar, T.C. and Jaynes, D.B. (2009) Denitrification in Wood Chip Bioreactors at Different Water Flows. Journal of Environmental Quality 38(4), 1664-1671.

Gujre, N., Soni, A., Rangan, L., Tsang, D.C.W. and Mitra, S. (2020) Sustainable improvement of soil health utilizing biochar and arbuscular mycorrhizal fungi: A review. Environmental Pollution, 115549.

Hallin, S., Hellman, M., Choudhury, M.I. and Ecke, F. (2015) Relative importance of plant uptake and plant associated denitrification for removal of nitrogen from mine drainage in sub-arctic wetlands. Water Research 85, 377-383.

Hari, V., Rakovec, O., Markonis, Y., Hanel, M. and Kumar, R. (2020) Increased future occurrences of the exceptional 2018–2019 Central European drought under global warming. Scientific Reports 10(1), 12207.

Harmayani, K.D. and Anwar, A.H.M.F. (2010) Adsorption of nutrients from stormwater using sawdust. International Journal of Environmental Science and Development (IJESD) 1(1).

Hassapak, C., Chetpattananondh, P., Chongkhong, S. and Chalermyanont, T. (2015) Performance of iron filings and activated sludge as media for permeable reactive barriers to treat zinc contaminated groundwater. Songklanakarin Journal of Science & Technology 37(1).

He, K., Qin, H., Wang, F., Ding, W. and Yin, Y. (2020) Importance of the Submerged Zone during Dry Periods to Nitrogen Removal in a Bioretention System. Water 12(3), 876.

He, L., Shan, J., Zhao, X., Wang, S. and Yan, X. (2019) Variable responses of nitrification and denitrification in a paddy soil to long-term biochar amendment and short-term biochar addition. Chemosphere 234, 558-567.

Hina, K., Hedley, M., Camps-Arbestain, M. and Hanly, J. (2015) Comparison of Pine Bark, Biochar and Zeolite as Sorbents for NH4+-N Removal from Water. CLEAN – Soil, Air, Water 43(1), 86-91.

Hooker, T.D. and Stark, J.M. (2008) Soil C and N cycling in three semiarid vegetation types: Response to an in situ pulse of plant detritus. Soil biology and biochemistry 40(10), 2678-2685.

Houlton, B.Z., Sigman, D.M., Schuur, E.A.G. and Hedin, L.O. (2007) A climate-driven switch in plant nitrogen acquisition within tropical forest communities. Proceedings of the National Academy of Sciences 104(21), 8902-8906.

Hsieh, C.-h., Davis, A.P. and Needelman, B.A. (2007) Nitrogen Removal from Urban Stormwater Runoff Through Layered Bioretention Columns. Water Environment Research 79(12), 2404-2411.

Hu, Q., Chen, N., Feng, C. and Hu, W. (2015) Nitrate adsorption from aqueous solution using granular chitosan-Fe3+ complex. Applied Surface Science 347, 1-9.

Hu, Q.L., Liu, H.Y., Zhang, Z.Y. and Xie, Y.H. (2020) Nitrate removal from aqueous solution using polyaniline modified activated carbon: Optimization and characterization. Journal of Molecular Liquids 309, 11.

Huang, G., Huang, Y., Hu, H., Liu, F., Zhang, Y. and Deng, R. (2015) Remediation of nitrate–nitrogen contaminated groundwater using a pilot-scale two-layer heterotrophic–autotrophic denitrification permeable reactive barrier with spongy iron/pine bark. Chemosphere 130, 8-16.

Huno, S.K.M., Rene, E.R., van Hullebusch, E.D. and Annachhatre, A.P. (2018) Nitrate removal from groundwater: a review of natural and engineered processes. Journal of Water Supply: Research and Technology-Aqua 67(8), 885-902.

Hunt, W.F., Jarrett, A.R., Smith, J.T. and Sharkey, L.J. (2006) Evaluating Bioretention Hydrology and Nutrient Removal at Three Field Sites in North Carolina. Journal of Irrigation and Drainage Engineering 132(6), 600-608.

Iqbal, H., Garcia-Perez, M. and Flury, M. (2015a) Effect of biochar on leaching of organic carbon, nitrogen, and phosphorus from compost in bioretention systems. Science of the Total Environment 521-522, 37-45.

Iqbal, H., Garcia-Perez, M. and Flury, M. (2015b) Effect of biochar on leaching of organic carbon, nitrogen, and phosphorus from compost in bioretention systems. Science of the Total Environment 521, 37-45.

Jackson, M.B. and Bolto, B.A. (1990) Effect of ion-exchange resin structure on nitrate selectivity. Reactive Polymers 12(3), 277-290.

Jaeschke, A., Op den Camp, H.J.M., Harhangi, H., Klimiuk, A., Hopmans, E.C., Jetten, M.S.M., Schouten, S. and Sinninghe Damsté, J.S. (2009) 16S rRNA gene and lipid biomarker evidence for anaerobic ammonium-oxidizing bacteria (anammox) in California and Nevada hot springs. Fems Microbiology Ecology 67(3), 343-350.

Jani, J., Lusk, M.G., Yang, Y.Y. and Toor, G.S. (2020) Wet season nitrogen export from a residential stormwater pond. Plos One 15(4), 12.

Jin, R.-C., Yang, G.-F., Yu, J.-J. and Zheng, P. (2012) The inhibition of the Anammox process: A review. Chemical Engineering Journal 197, 67-79.

Jing, Y.L., Zhang, Y.H., Han, I., Wang, P., Mei, Q.W. and Huang, Y.J. (2020) Effects of different straw biochars on soil organic carbon, nitrogen, available phosphorus, and enzyme activity in paddy soil. Scientific Reports 10(1), 12.

Joslin, J.D. and Wolfe, M.H. (1993) TEMPERATURE INCREASE ACCELERATES NITRATE RELEASE FROM HIGH-ELEVATION RED SPRUCE SOILS. Canadian Journal of Forest Research-Revue Canadienne De Recherche Forestiere 23(4), 756-759.

Kaiser, K. and Guggenberger, G. (2003) Mineral surfaces and soil organic matter. European Journal of Soil Science 54(2), 219-236.

Kalaruban, M., Loganathan, P., Shim, W.G., Kandasamy, J., Naidu, G., Nguyen, T.V. and Vigneswaran, S. (2016) Removing nitrate from water using iron-modified Dowex 21K XLT ion exchange resin: Batch and fluidised-bed adsorption studies. Separation and Purification Technology 158, 62-70.

Kameyama, K., Miyamoto, T., Iwata, Y. and Shiono, T. (2016) Influences of feedstock and pyrolysis temperature on the nitrate adsorption of biochar. Soil Science and Plant Nutrition 62(2), 180-184.

Kaplan, D., Wilhelm, R. and Abeliovich, A. (2000) Interdependent environmental factors controlling nitrification in waters. Water Science and Technology 42(1-2), 167-172.

Kerkez, B., Gruden, C., Lewis, M., Montestruque, L., Quigley, M., Wong, B., Bedig, A., Kertesz, R., Braun, T., Cadwalader, O., Poresky, A. and Pak, C. (2016) Smarter Stormwater Systems. Environmental Science & Technology 50(14), 7267-7273.

Khan, S., Shoaib, M., Khan, M.M., Melville, B.W. and Shamseldin, A.Y. (2019) Hydraulic investigation of the impact of retrofitting floating treatment wetlands in retention ponds. Water Science and Technology 80(8), 1476-1484.

Kim, H., Seagren, E.A. and Davis, A.P. (2003) Engineered Bioretention for Removal of Nitrate from Stormwater Runoff. Water Environment Research 75(4), 355-367.

Kjellin, J., Hallin, S. and Wörman, A. (2007) Spatial variations in denitrification activity in wetland sediments explained by hydrology and denitrifying community structure. Water Research 41(20), 4710-4720.

Knops, J.M.H., Bradley, K.L. and Wedin, D.A. (2002) Mechanisms of plant species impacts on ecosystem nitrogen cycling. Ecology Letters 5(3), 454-466.

Knowles, R. (1982) Denitrification. Microbiological reviews 46(1), 43-70.

Kraft, B., Tegetmeyer, H.E., Sharma, R., Klotz, M.G., Ferdelman, T.G., Hettich, R.L., Geelhoed, J.S. and Strous, M. (2014) The environmental controls that govern the end product of bacterial nitrate respiration. Science 345(6197), 676-679.

Kratky, H., Li, Z., Chen, Y., Wang, C., Li, X. and Yu, T. (2017) A critical literature review of bioretention research for stormwater management in cold climate and future research recommendations. Frontiers of Environmental Science & Engineering 11(4), 16.

Krometis, L.-A.H., Drummey, P.N., Characklis, G.W. and Sobsey, M.D. (2009) Impact of Microbial Partitioning on Wet Retention Pond Effectiveness. Journal of Environmental Engineering 135(9), 758-767.

Kuenen, J.G. (2008) Anammox bacteria: from discovery to application. Nature Reviews Microbiology 6(4), 320-326.

Landsman, M.R. and Davis, A.P. (2018a) Evaluation of Nutrients and Suspended Solids Removal by Stormwater Control Measures Using High-Flow Media. Journal of Environmental Engineering 144(10), 04018106.

Landsman, M.R. and Davis, A.P. (2018b) Evaluation of Nutrients and Suspended Solids Removal by Stormwater Control Measures Using High-Flow Media. Journal of Environmental Engineering 144(10), 12.

Lee, C.G., Fletcher, T.D. and Sun, G.Z. (2009) Nitrogen removal in constructed wetland systems. Engineering in Life Sciences 9(1), 11-22.

Li, H., Sharkey, L.J., Hunt, W.F. and Davis, A.P. (2009) Mitigation of Impervious Surface Hydrology Using Bioretention in North Carolina and Maryland. Journal of Hydrologic Engineering 14(4), 407-415.

Liao, R.H., Miao, Y., Li, J., Li, Y., Wang, Z., Du, J., Li, Y.M., Li, A.M. and Shen, H.J. (2018) Temperature dependence of denitrification microbial communities and functional genes in an expanded granular sludge bed reactor treating nitrate-rich wastewater. Rsc Advances 8(73), 42087-42094.

Lintern, A., McPhillips, L., Winfrey, B., Duncan, J. and Grady, C. (2020) Best Management Practices for Diffuse Nutrient Pollution: Wicked Problems Across Urban and Agricultural Watersheds. Environmental Science & Technology 54(15), 9159-9174.

Lipczynska-Kochany, E. (2018) Effect of climate change on humic substances and associated impacts on the quality of surface water and groundwater: A review. Science of the Total Environment 640, 1548-1565.

Lipiec, J. and Stępniewski, W. (1995) Effects of soil compaction and tillage systems on uptake and losses of nutrients. Soil and Tillage Research 35(1), 37-52.

Liu, S.-J., Zhao, Z.-Y., Li, J., Wang, J. and Qi, Y. (2013) An anaerobic two-layer permeable reactive biobarrier for the remediation of nitrate-contaminated groundwater. Water Research 47(16), 5977-5985.

Long, L., Xue, Y., Hu, X. and Zhu, Y. (2019) Study on the influence of surface potential on the nitrate adsorption capacity of metal modified biochar. Environmental Science and Pollution Research 26(3), 3065-3074.

Lopez-Ponnada, E.V., Lynn, T.J., Ergas, S.J. and Mihelcic, J.R. (2020) Long-term field performance of a conventional and modified bioretention system for removing dissolved nitrogen species in stormwater runoff. Water Research 170, 11.

Luo, J., Tillman, R.W. and Ball, P.R. (1999) Factors regulating denitrification in a soil under pasture. Soil Biology & Biochemistry 31(6), 913-927.

Mahdy, A., Elkhatib, E. and Fathi, N. (2008) Drinking water treatment residuals as an amendment to alkaline soils: Effects on bioaccumulation of heavy metals and aluminum in corn plants. Plant Soil and Environment 54(6), 234.

Mangangka, I.R., Liu, A., Egodawatta, P. and Goonetilleke, A. (2015) Performance characterisation of a stormwater treatment bioretention basin. Journal of Environmental Management 150, 173-178.

Mangum, K.R., Yan, Q., Ostrom, T.K. and Davis, A.P. (2020) Nutrient Leaching from Green Waste Compost Addition to Stormwater Submerged Gravel Wetland Mesocosms. Journal of Environmental Engineering 146(3), 11.

Manka, B.N., Hathaway, J.M., Tirpak, R.A., He, Q. and Hunt, W.F. (2016) Driving forces of effluent nutrient variability in field scale bioretention. Ecological Engineering 94, 622-628.

Månsson, K.F., Olsson, M.O., Falkengren-Grerup, U. and Bengtsson, G. (2014) Soil moisture variations affect short-term plant-microbial competition for ammonium, glycine, and glutamate. Ecology and Evolution 4(7), 1061-1072.

Mantelin, S. and Touraine, B. (2004) Plant growth-promoting bacteria and nitrate availability: impacts on root development and nitrate uptake. J Exp Bot 55(394), 27-34.

Martínez-García, L.B., De Deyn, G.B., Pugnaire, F.I., Kothamasi, D. and van der Heijden, M.G.A. (2017) Symbiotic soil fungi enhance ecosystem resilience to climate change. Global Change Biology 23(12), 5228-5236.

Mayo, A.W. (2020) Effect of pre-treatment of wastewater in HRAP on nitrogen removal in subsurface flow gravel bed constructed wetland. Physics and Chemistry of the Earth 117, 8.

McPhillips, L. and Walter, M.T. (2015) Hydrologic conditions drive denitrification and greenhouse gas emissions in stormwater detention basins. Ecological Engineering 85, 67-75.

Mellado-Vázquez, P.G., Lange, M. and Gleixner, G. (2019) Soil microbial communities and their carbon assimilation are affected by soil properties and season but not by plants differing in their photosynthetic pathways (C3 vs. C4). Biogeochemistry 142(2), 175-187.

Michalak, A.M., Anderson, E.J., Beletsky, D., Boland, S., Bosch, N.S., Bridgeman, T.B., Chaffin, J.D., Cho, K., Confesor, R., Daloğlu, I., DePinto, J.V., Evans, M.A., Fahnenstiel, G.L., He, L., Ho, J.C., Jenkins, L., Johengen, T.H., Kuo, K.C., LaPorte, E., Liu, X., McWilliams, M.R., Moore, M.R., Posselt, D.J., Richards, R.P., Scavia, D., Steiner, A.L., Verhamme, E., Wright, D.M. and Zagorski, M.A. (2013) Record-setting algal bloom in Lake Erie caused by agricultural and meteorological trends consistent with expected future conditions. Proceedings of the National Academy of Sciences 110(16), 6448-6452.

Morgan, C., Poor, C., Giudice, B. and Bibb, J. (2020) Agricultural Byproducts as Amendments in Bioretention Soils for Metal and Nutrient Removal. Journal of Environmental Engineering 146(6), 10.

Morse, N., Payne, E., Henry, R., Hatt, B., Chandrasena, G., Shapleigh, J., Cook, P., Coutts, S., Hathaway, J., Walter, M.T. and McCarthy, D. (2018) Plant-Microbe Interactions Drive Denitrification Rates, Dissolved Nitrogen Removal, and the Abundance of Denitrification Genes in Stormwater Control Measures. Environmental Science & Technology 52(16), 9320-9329.

Nabiul Afrooz, A.R.M. and Boehm, A.B. (2017) Effects of submerged zone, media aging, and antecedent dry period on the performance of biochar-amended biofilters in removing fecal indicators and nutrients from natural stormwater. Ecological Engineering 102, 320-330.

Nakhla, G. and Farooq, S. (2003) Simultaneous nitrification–denitrification in slow sand filters. Journal of Hazardous Materials 96(2), 291-303.

Nestler, A., Berglund, M., Accoe, F., Duta, S., Xue, D., Boeckx, P. and Taylor, P. (2011) Isotopes for improved management of nitrate pollution in aqueous resources: review of surface water field studies. Environmental Science and Pollution Research 18(4), 519-533.

Noguero, M. and Lacombe, B. (2016) Transporters Involved in Root Nitrate Uptake and Sensing by Arabidopsis. Frontiers in Plant Science 7(1391).

Norton, R.A., Harrison, J.A., Keller, C.K. and Moffett, K.B. (2017) Effects of storm size and frequency on nitrogen retention, denitrification, and N2O production in bioretention swale mesocosms. Biogeochemistry 134(3), 353-370.

Okaikue-Woodi, F.E.K., Cherukumilli, K. and Ray, J.R. (2020) A critical review of contaminant removal by conventional and emerging media for urban stormwater treatment in the United States. Water Research 187, 116434.

Ordonez, D., Valencia, A., Chang, N.-B. and Wanielista, M.P. (2020) Synergistic effects of aluminum/iron oxides and clay minerals on nutrient removal and recovery in water filtration media. Journal of Cleaner Production 275, 122728.

Osman, M., Yusof, K.W., Takaijudin, H., Goh, H.W., Malek, M.A., Azizan, N.A., Ab Ghani, A. and Abdurrasheed, A.S. (2019) A Review of Nitrogen Removal for Urban Stormwater Runoff in Bioretention System. Sustainability 11(19).

Öztürk, N. and Bektaş, T.E.l. (2004) Nitrate removal from aqueous solution by adsorption onto various materials. Journal of Hazardous Materials 112(1), 155-162.

Palmer, E.T., Poor, C.J., Hinman, C. and Stark, J.D. (2013) Nitrate and Phosphate Removal through Enhanced Bioretention Media: Mesocosm Study. Water Environment Research 85(9), 823-832.

Parker, J.L. and Newstead, S. (2014) Molecular basis of nitrate uptake by the plant nitrate transporter NRT1.1. Nature 507(7490), 68-72.

Patanè, C. (2011) Leaf Area Index, Leaf Transpiration and Stomatal Conductance as Affected by Soil Water Deficit and VPD in Processing Tomato in Semi Arid Mediterranean Climate. Journal of Agronomy and Crop Science 197(3), 165-176.

Payne, E.G.I., Fletcher, T.D., Cook, P.L.M., Deletic, A. and Hatt, B.E. (2014a) Processes and Drivers of Nitrogen Removal in Stormwater Biofiltration. Critical Reviews in Environmental Science and Technology 44(7), 796-846.

Payne, E.G.I., Fletcher, T.D., Russell, D.G., Grace, M.R., Cavagnaro, T.R., Evrard, V., Deletic, A., Hatt, B.E. and Cook, P.L.M. (2014b) Temporary Storage or Permanent Removal? The Division of Nitrogen between Biotic Assimilation and Denitrification in Stormwater Biofiltration Systems. Plos One 9(3), e90890.

Payne, E.G.I., Pham, T., Deletic, A., Hatt, B.E., Cook, P.L.M. and Fletcher, T.D. (2018) Which species? A decision-support tool to guide plant selection in stormwater biofilters. Advances in Water Resources 113, 86-99.

Peel, M.C., Finlayson, B.L. and McMahon, T.A. (2007) Updated world map of the Koppen-Geiger climate classification. Hydrology and Earth System Sciences 11(5), 1633-1644.

Pfenning, K.S. and McMahon, P.B. (1997) Effect of nitrate, organic carbon, and temperature on potential denitrification rates in nitrate-rich riverbed sediments. Journal of Hydrology 187(3), 283-295.

Poor, C. and Mohamed, M. (2020) World Environmental and Water Resources Congress 2020, pp. 73-83.

Rahman, M.M., Roberts, K.L., Warry, F., Grace, M.R. and Cook, P.L.M. (2019) Factors controlling dissimilatory nitrate reduction processes in constructed stormwater urban wetlands. Biogeochemistry 142(3), 375-393.

Reddy, K.R., Xie, T. and Dastgheibi, S. (2014) Nutrients Removal from Urban Stormwater by Different Filter Materials. Water Air and Soil Pollution 225(1), 14.

Reisinger, A.J., Groffman, P.M. and Rosi-Marshall, E.J. (2016) Nitrogen-cycling process rates across urban ecosystems. Fems Microbiology Ecology 92(12).

Revell, K.T., Maguire, R.O. and Agblevor, F.A. (2012) Influence of Poultry Litter Biochar on Soil Properties and Plant Growth. Soil Science 177(6), 402-408.

Rippy, M.A. (2015) Meeting the criteria: linking biofilter design to fecal indicator bacteria removal. WIREs Water 2(5), 577-592.

Roseen, R.M., Ballestero, T.P., Houle, J.J., Avellaneda, P., Briggs, J., Fowler, G. and Wildey, R. (2009) Seasonal Performance Variations for Storm-Water Management Systems in Cold Climate Conditions. Journal of Environmental Engineering 135(3), 128-137.

Sabiri, N.-E., Monnier, E., Raimbault, V., Massé, A., Séchet, V. and Jaouen, P. (2017) Effect of filtration rate on coal-sand dual-media filter performances for microalgae removal. Environmental Technology 38(3), 345-352.

Saeed, T. and Sun, G. (2012) A review on nitrogen and organics removal mechanisms in subsurface flow constructed wetlands: Dependency on environmental parameters, operating conditions and supporting media. Journal of Environmental Management 112, 429-448.

Samatya, S., Kabay, N., Yüksel, Ü., Arda, M. and Yüksel, M. (2006) Removal of nitrate from aqueous solution by nitrate selective ion exchange resins. Reactive and Functional Polymers 66(11), 1206-1214.

Schifman, L.A., Kasaraneni, V.K., Sullivan, R.K., Oyanedel-Craver, V. and Boving, T.B. (2016) Bacteria Removal from Stormwater Runoff Using Tree Filters: A Comparison of a Conventional and an Innovative System. Water 8(3), 16.

Scholz, F., Löscher, C.R., Fiskal, A., Sommer, S., Hensen, C., Lomnitz, U., Wuttig, K., Göttlicher, J., Kossel, E., Steininger, R. and Canfield, D.E. (2016) Nitrate-dependent iron oxidation limits iron transport in anoxic ocean regions. Earth and Planetary Science Letters 454, 272-281.

Seitzinger, S., Harrison, J.A., Böhlke, J.K., Bouwman, A.F., Lowrance, R., Peterson, B., Tobias, C. and Drecht, G.V. (2006) DENITRIFICATION ACROSS LANDSCAPES AND WATERSCAPES: A SYNTHESIS. Ecological Applications 16(6), 2064-2090.

Sharma, B. and Ahlert, R.C. (1977) Nitrification and nitrogen removal. Water Research 11(10), 897-925.

Shrestha, P., Hurley, S.E. and Wemple, B.C. (2018) Effects of different soil media, vegetation, and hydrologic treatments on nutrient and sediment removal in roadside bioretention systems. Ecological Engineering 112, 116-131.

Shrewsbury, L.H., Smith, J.L., Huggins, D.R., Carpenter-Boggs, L. and Reardon, C.L. (2016) Denitrifier abundance has a greater influence on denitrification rates at larger landscape scales but is a lesser driver than environmental variables. Soil biology and biochemistry 103, 221-231.

Sinha, E., Michalak, A.M. and Balaji, V. (2017) Eutrophication will increase during the 21st century as a result of precipitation changes. Science 357(6349), 405-408.

Skorobogatov, A., He, J.X., Chu, A., Valeo, C. and van Duin, B. (2020) The impact of media, plants and their interactions on bioretention performance: A review. Science of the Total Environment 715.

Smith, M.S. and Tiedje, J.M. (1979) Phases of denitrification following oxygen depletion in soil. Soil biology and biochemistry 11(3), 261-267.

Song, X., Chen, Z., Wang, X. and Zhang, S. (2017) Ligand effects on nitrate reduction by zero-valent iron: Role of surface complexation. Water Research 114, 218-227.

Spieles, D.J. and Mitsch, W.J. (1999) The effects of season and hydrologic and chemical loading on nitrate retention in constructed wetlands: a comparison of low- and high-nutrient riverine systems. Ecological Engineering 14(1), 77-91.

Stagge, J.H., Davis, A.P., Jamil, E. and Kim, H. (2012) Performance of grass swales for improving water quality from highway runoff. Water Research 46(20), 6731-6742.

Stenstrom, M.K. and Poduska, R.A. (1980) The effect of dissolved oxygen concentration on nitrification. Water Research 14(6), 643-649.

Strous, M., Kuenen, J.G. and Jetten, M.S. (1999) Key physiology of anaerobic ammonium oxidation. Appl Environ Microbiol 65(7), 3248-3250.

Strous, M., Pelletier, E., Mangenot, S., Rattei, T., Lehner, A., Taylor, M.W., Horn, M., Daims, H., Bartol-Mavel, D., Wincker, P., Barbe, V., Fonknechten, N., Vallenet, D., Segurens, B., Schenowitz-Truong, C., Médigue, C., Collingro, A., Snel, B., Dutilh, B.E., Op den Camp, H.J.M., van der Drift, C., Cirpus, I., van de Pas-Schoonen, K.T., Harhangi, H.R., van Niftrik, L., Schmid, M., Keltjens, J., van de Vossenberg, J., Kartal, B., Meier, H., Frishman, D., Huynen, M.A., Mewes, H.-W., Weissenbach, J., Jetten, M.S.M., Wagner, M. and Le Paslier, D. (2006) Deciphering the evolution and metabolism of an anammox bacterium from a community genome. Nature 440(7085), 790-794.

Tabari, H. (2020) Climate change impact on flood and extreme precipitation increases with water availability. Scientific Reports 10(1), 13768.

Tan, E., Zou, W., Zheng, Z., Yan, X., Du, M., Hsu, T.-C., Tian, L., Middelburg, J.J., Trull, T.W. and Kao, S.-j. (2020) Warming stimulates sediment denitrification at the expense of anaerobic ammonium oxidation. Nature Climate Change 10(4), 349-355.

Tang, C.-J., Zheng, P., Chen, T.-T., Zhang, J.-Q., Mahmood, Q., Ding, S., Chen, X.-G., Chen, J.-W. and Wu, D.-T. (2011) Enhanced nitrogen removal from pharmaceutical wastewater using SBA-ANAMMOX process. Water Research 45(1), 201-210.

Tang, S., Liao, Y., Xu, Y., Dang, Z., Zhu, X. and Ji, G. (2020) Microbial coupling mechanisms of nitrogen removal in constructed wetlands: A review. Bioresource Technology, 123759.

Tanner, C.C. and Kadlec, R.H. (2013) Influence of hydrological regime on wetland attenuation of diffuse agricultural nitrate losses. Ecological Engineering 56, 79-88.

Tian, J., Jin, J., Chiu, P.C., Cha, D.K., Guo, M.X. and Imhoff, P.T. (2019) A pilot-scale, bi-layer bioretention system with biochar and zero-valent iron for enhanced nitrate removal from stormwater. Water Research 148, 378-387.

Tiedje, J.M. (1983) Methods of Soil Analysis, pp. 1011-1026.

Tischner, R. (2000) Nitrate uptake and reduction in higher and lower plants. Plant, Cell & Environment 23(10), 1005-1024.

Ulrich, B.A., Loehnert, M. and Higgins, C.P. (2017) Improved contaminant removal in vegetated stormwater biofilters amended with biochar. Environmental Science: Water Research & Technology 3(4), 726-734.

Valencia, A., Ordonez, D., Wen, D., McKenna, A.M., Chang, N.-B. and Wanielista, M.P. (2020) The interaction of dissolved organic nitrogen removal and microbial abundance in iron-filings based green environmental media for stormwater treatment. Environmental Research 188, 109815.

Valtanen, M., Sillanpaa, N. and Setala, H. (2017a) A large-scale lysimeter study of stormwater biofiltration under cold climatic conditions. Ecological Engineering 100, 89-98.

Valtanen, M., Sillanpää, N. and Setälä, H. (2017b) A large-scale lysimeter study of stormwater biofiltration under cold climatic conditions. Ecological Engineering 100, 89-98.

van Kessel, M.A.H.J., Speth, D.R., Albertsen, M., Nielsen, P.H., Op den Camp, H.J.M., Kartal, B., Jetten, M.S.M. and Lücker, S. (2015) Complete nitrification by a single microorganism. Nature 528(7583), 555-559.

von Ahnen, M., Aalto, S.L., Suurnäkki, S., Tiirola, M. and Pedersen, P.B. (2019) Salinity affects nitrate removal and microbial composition of denitrifying woodchip bioreactors treating recirculating aquaculture system effluents. Aquaculture 504, 182-189.

Vu, T.M., Trinh, V.T., Doan, D.P., Van, H.T., Nguyen, T.V., Vigneswaran, S. and Ngo, H.H. (2017) Removing ammonium from water using modified corncob-biochar. Science of the Total Environment 579, 612-619.

Vymazal, J. (2020) Removal of nutrients in constructed wetlands for wastewater treatment through plant harvesting – Biomass and load matter the most. Ecological Engineering 155, 105962.

Wang, X.H., Ma, C., Wang, Y.Q., Wang, Y.J., Li, T., Dai, Z.S. and Li, M.Y. (2020) Effect of root architecture on rainfall threshold for slope stability: variabilities in saturated hydraulic conductivity and strength of root-soil composite. Landslides 17(8), 1965-1977.

Wang, Y.-Y., Hsu, P.-K. and Tsay, Y.-F. (2012) Uptake, allocation and signaling of nitrate. Trends in Plant Science 17(8), 458-467.

Wen, Y., Chen, Y., Zheng, N., Yang, D. and Zhou, Q. (2010) Effects of plant biomass on nitrate removal and transformation of carbon sources in subsurface-flow constructed wetlands. Bioresource Technology 101(19), 7286-7292.

Westerhoff, P. (2003) Reduction of Nitrate, Bromate, and Chlorate by Zero Valent Iron (Fe0). Journal of Environmental Engineering 129(1), 10-16.

Westerhoff, P. and James, J. (2003) Nitrate removal in zero-valent iron packed columns. Water Research 37(8), 1818-1830.

Wicke, D., Rouault, P., Camilo, B.K., Pagotto, C., Dechesne, M. and Soyeux, E. (2015) Nitrate reduction in reactive swales at low temperatures: full-size field system vs. technical scale. Water Science and Technology-Water Supply 15(3), 642-648.

Wissler, A.D., Hunt, W.F. and McLaughlin, R.A. (2020) Water Quality and Hydrologic Performance of Two Dry Detention Basins Receiving Highway Stormwater Runoff in the Piedmont Region of North Carolina. Journal of Sustainable Water in the Built Environment 6(2), 11.

Wu, H., Wang, X., He, X., Zhang, S., Liang, R. and Shen, J. (2017) Effects of root exudates on denitrifier gene abundance, community structure and activity in a micro-polluted constructed wetland. Science of the Total Environment 598, 697-703.

Xie, S.G., Zhang, X.J. and Wang, Z.S. (2003) Temperature effect on aerobic denitrification and nitrification. Journal of Environmental Sciences 15(5), 669-673.

Yan, Q., Davis Allen, P. and James Bruce, R. (2016) Enhanced Organic Phosphorus Sorption from Urban Stormwater Using Modified Bioretention Media: Batch Studies. Journal of Environmental Engineering 142(4), 04016001.

Yang, Y.-Y. and Lusk, M.G. (2018) Nutrients in Urban Stormwater Runoff: Current State of the Science and Potential Mitigation Options. Current Pollution Reports 4(2), 112-127.

Yang, Y., Zhao, Y., Tang, C., Mao, Y. and Shen, C. (2019) Significance of water level in affecting cathode potential in electro-wetland. Bioresource Technology 285, 121345.

Yazdanfar, Z. and Sharma, A. (2015) Urban drainage system planning and design – challenges with climate change and urbanization: a review. Water Science and Technology 72(2), 165-179.

Zhang, C., Yin, Q., Wen, Y., Guo, W., Liu, C. and Zhou, Q. (2016) Enhanced nitrate removal in self-supplying carbon source constructed wetlands treating secondary effluent: The roles of plants and plant fermentation broth. Ecological Engineering 91, 310-316.

Zhang, J., Lan, T., Müller, C. and Cai, Z. (2015) Dissimilatory nitrate reduction to ammonium (DNRA) plays an important role in soil nitrogen conservation in neutral and alkaline but not acidic rice soil. Journal of Soils and Sediments 15(3), 523-531.

Zhang, K., Liu, Y., Deletic, A., McCarthy, D.T., Hatt, B.E., Payne, E.G.I., Chandrasena, G., Li, Y., Pham, T., Jamali, B., Daly, E., Fletcher, T.D. and Lintern, A. (2020) The impact of stormwater biofilter design and operational variables on nutrient removal - a statistical modelling approach. Water Research, 116486.

Zhang, K., Manuelpillai, D., Raut, B., Deletic, A. and Bach, P.M. (2019a) Evaluating the reliability of stormwater treatment systems under various future climate conditions. Journal of Hydrology 568, 57-66.

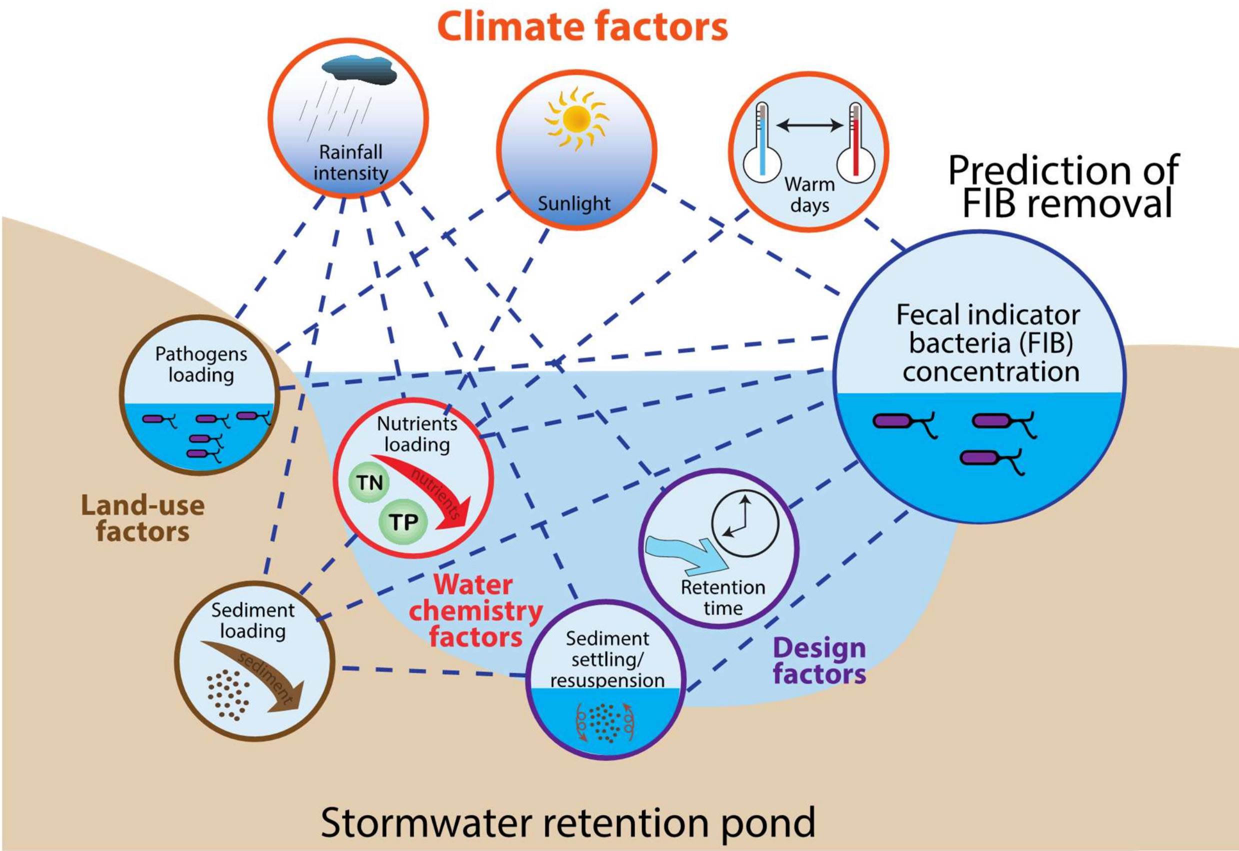
Zhang, W.S., Pueppke, S.G., Li, H.P., Geng, J.W., Diao, Y.Q. and Hyndman, D.W. (2019b) Modeling phosphorus sources and transport in a headwater catchment with rapid agricultural expansion. Environmental Pollution 255, 12.

Zhong, J.C., Fan, C.X., Liu, G.F., Zhang, L., Shang, J.G. and Gu, X.Z. (2010) Seasonal variation of potential denitrification rates of surface sediment from Meiliang Bay, Taihu Lake, China. Journal of Environmental Sciences 22(7), 961-967.

Zinger, Y., Blecken, G.-T., Fletcher, T.D., Viklander, M. and Deletić, A. (2013) Optimising nitrogen removal in existing stormwater biofilters: Benefits and tradeoffs of a retrofitted saturated zone. Ecological Engineering 51, 75-82.

Zinger, Y., Prodanovic, V., Zhang, K., Fletcher, T.D. and Deletic, A. (2020) The effect of intermittent drying and wetting stormwater cycles on the nutrient removal performances of two vegetated biofiltration designs. Chemosphere, 129294.

3. CHAPTER 3 – CAN WATER COMPOSITION AND WEATHER FACTORS PREDICT FECAL INDICATOR BACTERIA REMOVAL IN RETENTION PONDS IN VARIABLE WEATHER CONDITIONS?



Citation: Valenca, R., Garcia, L., Espinosa, C., Flor, D., & Mohanty, S. K. (2022) Can water composition and weather factors predict fecal indicator bacteria removal in retention ponds in variable weather conditions? *Science of The Total Environment*. (*under revision*)

Abstract

Retention ponds provide benefits including flood control, groundwater recharge, and water quality improvement, but changes in weather conditions could limit the effectiveness in improving microbial water quality metrics. The concentration of fecal indicator bacteria (FIB), which is used as regulatory standards to assess microbial water quality in retention ponds, could vary widely based on many factors including local weather and influent water chemistry and composition due to their proven effects on FIB removal processes. In this critical review, we analyzed 7,421 data collected from 19 retention ponds across North America listed in the International Stormwater BMP Database to examine if variable FIB removal in the field conditions can be predicted based on changes in these weather and water composition factors. Our analysis confirms that FIB removal in retention ponds is sensitive to weather conditions or seasons, but temperature and precipitation data failed to describe the variable FIB removal. These weather conditions affect suspended solid and nutrient concentrations, which in turn could affect FIB concentration in the ponds. However, removal of total suspended solids and total P only explained 5% and 12% of FIB removal data, respectively, and TN removal had no correlation with FIB removal. These results indicate that regression-based modeling with a single parameter as input has limited use to predict FIB removal due to the interactive nature of their effects on FIB removal. In contrast, machine learning algorithms such as the random forest method were able to predict 65% of the data. The overall analysis indicates that the machine learning model could play a critical role in predicting microbial water quality of surface waters under complex conditions where the variation of both water composition and weather conditions could deem regression-based modeling less effective.

3.1. Introduction

Retention or detention ponds have been used to reduce flooding and support biodiversity (Sun et al. 2019), with multiple benefits including improving water quality (Hathaway and Hunt 2012, Lusk and Toor 2016, Valenca et al. 2021) and recharging groundwater (Herrmann 2012, Sun et al. 2019) to augment drinking water supply (Hartmann et al. 2017). However, a change in the microbial water quality of ponds could pose human health risks (EPA 2002) such as gastrointestinal illnesses (Ishii et al. 2006, Oster et al. 2014) and eventually lead to groundwater pollution (Stephens et al. 2012). Because most pathogens are not culturable in the laboratory, regulatory agencies use fecal indicator bacteria (FIB) such as coliform, total coliform, *E. coli*, and *enterococcus* as pathogen indicators to inform the public about microbial contamination risks (Rochelle-Newall et al. 2015, Tiefenthaler et al. 2010). As the measurement of the FIB could take 24 h, predictive models that link weather variables (e.g., temperature and rainfall intensity) and water composition variables (e.g., turbidity and nutrients) could help develop a management tool to provide advanced notice based on weather data (Searcy and Boehm 2021). Thus, it is critical to determine how FIB removal in retention ponds is related to local weather conditions and water composition of runoffs (Merriman et al. 2017, Saxton et al. 2016, Sharma et al. 2016, Vander Meer et al. 2021).

Based on data collected in the International BMP database, FIB removal in retention ponds could vary by 5 orders of magnitude (Clary et al. 2011). The International BMP Database is an open-access platform created by the USEPA and ASCE where private and public institutions report the concentration of contaminants in the influent water and the effluent water of BMP systems, as well as the date and the location of the sample collection. Depending on site conditions, retention ponds may either act as a source (Serrano and DeLorenzo 2008) or a sink for FIB (Hathaway and

Hunt 2010) due to the variation in removal processes in those conditions (Figure 3.1). FIB can be removed in ponds by physical, chemical, and biological processes. The physical process includes sedimentation of bacteria attached to suspended solids (Krometis et al. 2009), where both the concentration (Jiang et al. 2015) and the size (Walters et al. 2013) of the solids affect FIB removal. Suspended solids concentration and size depend on land use, season, and local weather (Ciupa et al. 2020, Pizarro et al. 2014). The chemical process involves the inactivation of bacteria under exposure to radicals or ultraviolet (UV) radiation from sunlight (Nguyen et al. 2015). The rate of photoinactivation could change diurnally (Maraccini et al. 2012) and seasonally (Rincón and Pulgarin 2004). Thus, local weather conditions can affect photoinactivation by affecting the amount of sunlight exposure at a place, the temperature of the water, and the presence of suspended solids that could block sunlight (Huovinen et al. 2006, Tala et al. 2017, Vione and Scozzaro 2019). Lastly, FIB may be removed through biological processes via starvation as a result of nutrient limitation (Cornforth and Foster 2013), predation by protozoa and zooplankton (Brookes et al. 2004, Šimek et al. 2001), and natural bacterial selection (Hibbing et al. 2010). Alternatively, FIB may also grow naturally by utilizing nutrients and organic carbon present in retention ponds (Song et al. 2019). The links between these controlling variables and FIB removal are evaluated using batch studies that do not simulate complex changes in site conditions (Jang et al. 2017). The FIB removal can be affected by water composition or local weather conditions, which vary dynamically and interactively in the field. Thus, further analysis is needed to estimate what fraction of FIB removal in the field can be predicted based on the changes in the controlling variables.

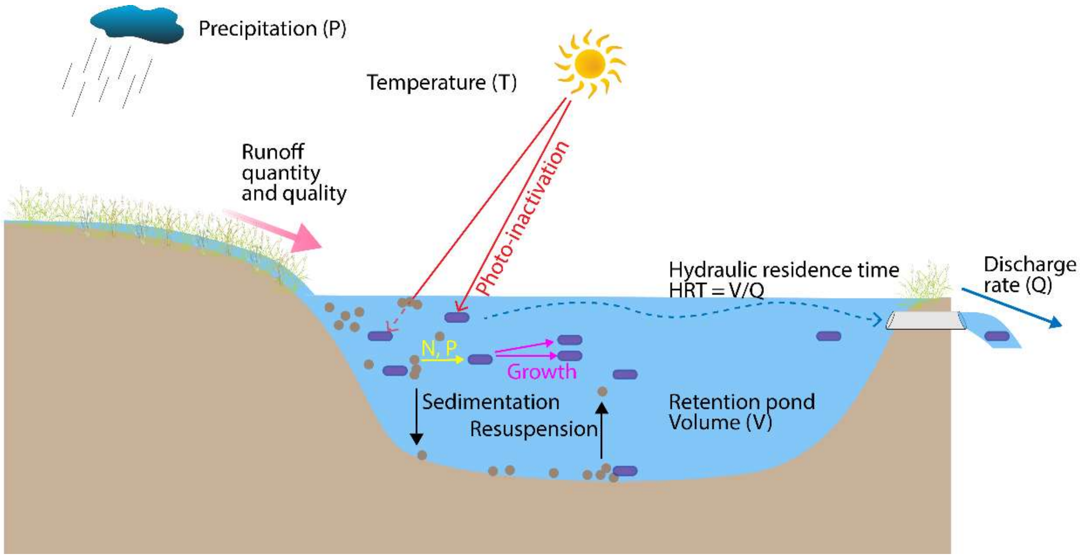


Figure 3.1 – Fecal indicator bacteria removal in retention ponds can be affected by different factors: climate (precipitation, temperature), land use types that determine the runoff quality and suspended sediments, water chemistry such as nutrients and pH, and retention pond design such as volume and depth.

The extent to which local weather conditions can affect FIB concentration in ponds is interlinked with their effect on water chemistry or the composition of runoff and FIB removal processes in ponds. Establishing these links required analysis of data in the field conditions where all these factors co-occur. The interactive effects of the confounding factors make it difficult to predict the concentration of indicator bacteria in retention ponds in changing climates. In this case, machine learning can be useful to predict FIB concentration in ponds. Machine learning techniques have been used to identify the drivers for groundwater well contamination (White et al. 2021), hydraulic performance of green infrastructure (Li et al. 2019), optimum stormwater treatment geomedia for biofilters (Valencia et al. 2021), environmental performances of pond ash treatment (Suthar 2019) and wastewater treatment (Sundui et al. 2021), recovery of plant communities (Peaple et al. 2021) and thermal profile of ponds (Stajkowski et al. 2021). However, the same method has not been tested to predict FIB removal or concentration in retention ponds.

The objectives of this critical analysis are to estimate the fraction of FIB removal data that can be linked to changes in controlling factors such as rainfall intensity, temperature, and associated changes in water compositions, and to test the utility of common machine learning models to predict FIB removal in ponds. To achieve the objectives, we correlate FIB removal with weather variables and associated changes in water composition in 19 retention ponds in the North American continent archived in International BMP Database. Furthermore, we utilized five different machine learning algorithms to identify the best method to predict FIB removal based on the environmental variables, which could inform how changes in weather patterns in projected climate change may affect microbial risk in retention ponds (Whitman et al. 2008).

3.2. Data Collection and Analysis

We collected data from 19 retention ponds in North America before May 5, 2021 (Clary et al. 2011). The dataset from the database has been previously used to assess the performance of best management practices (BMP) in removing nutrients (Valenca et al. 2021), heavy metals (Tirpak et al. 2021), and suspended particles (Ramesh et al. 2021) in field conditions. The name and location of each retention pond were provided in Table 3.1. The data consisted of influent and effluent concentrations of the following parameters on a given sampling day in each pond: 5 types of indicator bacteria including *Enterococcus, Escherichia coli, Fecal coliform, Total coliform,* and *Fecal Streptococcus*, total suspended solids (TSS), total nitrogen (TN), total phosphorus (TP), dissolved phosphorus, nitrate, nitrite, ammonia, dissolved organic nitrogen (DON), and total organic nitrogen (TON). The data reported ranged over 29 years from 1981 through 2010. Although the database did not report the time of sample collection which could affect contaminant measurement, we calculated the removal based on influent and effluent measurements, and thus the variability due to the time of sample collection is minimized. Monthly average data for temperature and precipitation for the location or city where the ponds are located were collected from the National Weather Service Forecast hosted by the National Oceanic and Atmospheric Administration (NOAA). Although daily data would be more accurate to verify the effect of temperature and precipitation on FIB removal, weekly or daily data was not reported by NOAA. Thus, average monthly temperature and precipitation were used in the analysis as a proxy. If the temperature or precipitation data was not available for a given city, the same data from the closest city was collected. We used air temperature as a proxy to water temperature (Rosencranz et al. 2021), as water temperature data was not reported in the database.

Table 3.1 – Location of the retention ponds included in this study.

Retention Pond Name	City	State	Country
Heritage Estates Pond	Richmond Hill	Ontario	Canada
I-5 / La Costa (east)	Encinitas	California	United States
Dem. Urban SW Treatment (DUST) Marsh	Fremont	California	United States
North Natomas Water Quality Basin 4	Sacramento	California	United States
Duval County Pond 1	Jacksonville	Florida	United States
Central Park BMP	Largo	Florida	United States
Largo Regional STF	Largo	Florida	United States
FL Blvd Detention Pond	Merrit Island	Florida	United States
Jungle Lake	St. Petersburg	Florida	United States
Shawnee Ridge	Suwanee	Georgia	United States
Northeast Creek Pond 1	Durham	North Carolina	United States
NCSU Wilmington	Wilmington	North Carolina	United States
Convention Center	Austin	Texas	United States
WH	Austin	Texas	United States
Sand Beach Wet Pond (SB)	Austin	Texas	United States
BO BMP	Austin	Texas	United States
Greens Bayou Wetland Mitigation Bank	Houston	Texas	United States

We used log FIB removal (LBR_t) to determine the retention pond's ability to reduce the fecal indicator bacteria contamination: $LBR_t = -log\left(\frac{CB_{et}}{CB_{it}}\right)$, where CB_{it} and CB_{et} are, respectively, the concentration of bacteria in the influent and effluent water on a given day (t). The removal of all other contaminants such as TSS, TN, TP, dissolved phosphorus, nitrate, nitrite, ammonia, DON, and TON was calculated as percentage removal (R_t,%): $R_t = \left(\frac{C_{et} - C_{it}}{C_{it}}\right) \times 100$, where C_{et} and C_{it} respectively represent the concentration of specific parameters in the effluent and influent water of retention ponds on a given day (t).

Statistical analysis of the available data included the creation of regression models to analyze trends and the calculation of the coefficient of determination (R^2) to identify the correlation between variables. All analyses were done using the Caret and ggplot2 packages in RStudio (version 1.4.1106). The summary of all data collected and used in this study can be found in the online repository (Figshare: https://doi.org/10.6084/m9.figshare.14787411.v1).

3.3. Machine Learning Applications in Environmental Science

Machine Learning (ML) models are data-driven algorithms that connect input and output variables to extract hidden relationships and patterns from large data sets (Nouraki et al. 2021), which makes them a promising tool to revolutionize the environmental science field (Liu et al. 2022). Although the application of ML in complex systems like environmental science and engineering is feasible and highly encouraged (Table 3.2), no study to date has applied ML techniques to predict how retention ponds perform under varying weather conditions. To predict the FIB removal performance of retention ponds under different weather conditions, we utilized five supervised ML algorithms: Random Forest, Support Vector Machines, Classification and Regression Trees, Linear Discriminant Analysis, and k-Nearest Neighbors. Each ML model uses a different learning technique, which affects the decision time and accuracy depending on the complexity of the dataset. The dataset used for the ML models consisted of 1,448 data points from 19 retention ponds with the following parameters: monthly precipitation, monthly temperature, bacteria concentration in influent stormwater, and the daily log removal of bacteria associated with each retention pond. Pre-data treatment was performed manually to ensure that the ML model had accurate measurements for the input variables and to remove possible outliers that could distort the analysis. Any data point that missed any of the input parameters were removed from the dataset. Algorithms were trained using 80% of the data (randomly selected), and the assessment of the trained model was performed with the remaining 20% of the data. Machine Learning models were run using the package Caret in RStudio (version 1.4.1106).

Random forests (RF) models use bootstrapping and bagging methods to create several hundred to thousand decision trees that are trained with randomly chosen sub-datasets to reduce the variance of the data (Breiman 2001). Further details about the RF model can be found elsewhere (Ao et al. 2019, Breiman 2001, Fox et al. 2017).

Support vector machines (SVM) can solve complex regression problems by using the statistical learning theory (Raghavendra. N and Deka 2014). It uses the kernel function to determine a hyperplane function with a high marginal distance from the targets to increase the dimensions of the dataset (Noble 2006). An in-depth explanation of the model is given elsewhere (Mohammadpour et al. 2015).

Classification and Regression Trees (CART) uses a non-linear and non-parametric statistical approach to partition the dependent variables into homogeneous subclasses that are used to create multiple simple regression models (Ji et al. 2013). The model utilizes multiple nodes (classification questions) to categorize the data into branches, and the terminal node (leaves) is the ultimate classification where the regression is fitted (Breiman et al. 2017). Further description of CART can be found elsewhere (Choubin et al. 2018, Timofeev 2004).

Linear Discriminant Analysis (LDA) aims to create a discriminant function that linearly transforms two variables and create a new set of transformed values that are more accurate than each variable alone (Bhattacharyya and Rahul 2013) – similar to methods used in the principal component analysis (Yang and Yang 2003). By grouping samples that share common properties, LDA can transform an original dataset into a single discriminant score (Boyacioglu and Boyacioglu 2010). LDA has been discussed in detail in previous studies (Singh et al. 2004, Tabachnick and Fidell 2013).

k-Nearest Neighbors (kNN) creates regression models by calculating the distance between a data point and the closest neighbors, followed by the calculation of the mean for that neighbors' dataset which simulates the final value (Towler et al. 2009). The selection of the numbers of neighbors, as well as the probability metric used to weight each neighbor's importance, depends on each model application. Further explanation on kNN can be found elsewhere (Sharif and Burn 2007).

Table 3.2 – Application of machine learning models in 30 environmental science studies.

ML Model	Parameters Predicted	Prediction accuracy*	Reference
Random Forest	Infiltration rates of permeable stormwater channels	0.89[a]	(Yaseen et al. 2021)
	Heavy metals removal in stormwater biofilters	0.64 – 0.99[b]	(Fang et al. 2021).
	Drivers of nitrate contamination in surface and groundwater	0.21 – 0.52[a]	(Pennino et al. 2020)
	Prediction of total dissolved solids in surface water	0.98 [a]	(Nouraki et al. 2021)
	Fecal indicator bacteria prediction in beach water	0.52 – 0.78[c]	(Searcy and Boehm 2021)
	Suspended solids concentration in stormwater	0.64[a]	(Moeini et al. 2021)
	Source of fecal contamination in the environmental samples	NR	(Roguet et al. 2018)
	Conventional water quality indices	0.70 – 0.86[d]	(Wang et al. 2021)
Support Vector Machines	Generation of municipal solid waste	0.75 – 0.78[a]	(Noori et al. 2009)
	Water quality in constructed wetlands	0.79 – 0.99[a]	(Mohammadpour et al. 2015)
	Sediment loads in three rivers	0.96[a]	(Azamathulla et al. 2010)
	Water quality index in rivers	0.80 – 0.92[a]	(Leong et al. 2021)
	Level of algal bloom in reservoirs	67.5 – 75.3[e]	(Kim et al. 2021)
	Suspended solids concentration in stormwater	0.58[a]	(Moeini et al. 2021)
	Sodium adsorption ratio in surface water	0.99[a]	(Nouraki et al. 2021)
	Flooding susceptibility in watersheds	88[e]	(Choubin et al. 2019)
Classification and Regression Trees	Flooding risk assessment	NR	(Ji et al. 2013)
	Flooding susceptibility in watersheds	83[e]	(Choubin et al. 2019)
	Seasonal variability of E. coli in irrigation ponds	0.38 – 0.86[a]	(Stocker et al. 2019)
	Bioaccumulation of organic contaminants in plant roots	0.77[a]	(Gao et al. 2021)
	Suspended solids concentration in stormwater	0.36[a]	(Moeini et al. 2021)
	Distribution of plant species	0.66 – 0.87[f]	(Vayssières et al. 2000)

Linear Discriminant Analysis	Effect of seasons on water quality	NR	(Boyacioglu and Boyacioglu 2010)
	River water quality	0.85 – 0.88[a]	(Djarum et al. 2021)
	Marine water quality index	0.41[a]	(Samsudin et al. 2019)
	Redox conditions in groundwater that could favor denitrification processes	42 – 69[e]	(Wilson et al. 2018).
k-Nearest Neighbors	Future rainfall intensity	NR	(Chen et al. 2021)
	Precipitation forecast	49.5[e]	(Huang et al. 2017)
	Water quality classification in aquifers	NR	(Modaresi and Araghinejad 2014)
	Prediction of total organic carbon and alkalinity in watersheds	NR	(Towler et al. 2009)
	Water quality classification	80 – 85[e]	(Aldhyani et al. 2020)
	Total suspended solids concentration in stormwater runoff	0.36 – 0.56[a]	(Moeini et al. 2021)

***Accuracy is given in terms of [a]Coefficient of determination (R^2), [b]Nash–Sutcliffe model efficiency coefficient (NSE), [c]Root-mean-square deviation (RMSE), [d]Kappa, [e]Percentage, [f]Specificity. NR: not reported.**

3.4. Factors affecting FIB removal processes in retention ponds based on laboratory studies

FIB removal in ponds could be influenced by three factors: pond design, local weather conditions, and water composition (Figure 3.1).

Retention pond design: In urban areas, stormwater retention ponds are built by excavating soil to create a depression so that runoff from surrounding areas can naturally flow into it (Figure 1). Their size can vary between 100 to 10,000 m^2 with depth between 1 – 10 m, and they are often surrounded by vegetation (Valenca et al. 2021). Stormwater typically enters the ponds in one end and exits at the other end via spillways or pipes. The average time stormwater stays in a pond, also called retention time (θ, days), could vary between 1 to 7 days. Thus, the retention time increases with an increase in the size of the pond (V) and decreases with the discharge rate (Q), which in turn depends on the intensity of the current rainfall event and the catchment area that contributes to the runoff to the pond. As retention time is an indication of the time pollutants could undergo reactions in the ponds, they play a critical role in the removal of FIB. In ponds, indicator bacteria or pathogens can be removed by adsorption, sedimentation, and inactivation (Ahmed et al. 2019). As sedimentation rates in a retention pond depend on the size of the particulate contaminant and the retention time (Ahn 2012, Cheng 2008), making the retention pond bigger and deeper can increase the removal of sediments. The inactivation rates of FIB can depend on abiotic and biotic factors. For instance, abiotic factors like water turbidity, water temperature, sunlight intensity, pH, dissolved oxygen levels, and retention time may interfere with the inactivation rates of FIB in retention ponds (Reed 1997, Ross et al. 2008, Stocker et al. 2019, Zhang et al. 2012). On the other hand, biotic factors like bacteria types, competition, predation, and vegetation can also influence the fate of FIB (Abia et al. 2016, Avelar et al. 2014, Tunçsiper et al. 2012) . Thus, the presence of vegetation, which is part of pond design, can block sunlight and lower inactivation. In summary, increasing the size of the pond and removing vegetation that could block sunlight by periodic maintenance could improve the overall FIB removal capacity of ponds.

Local weather conditions. Precipitation and temperature may critically affect FIB removal mechanisms in a retention pond (Dean and Mitchell 2022). Local weather conditions can affect the influent water composition such as suspended solids and particulate nutrients to ponds (Gong et al. 2016, Zanon et al. 2020), which could affect FIB removal in ponds. While increased particulate concentration may increase bacteria removal by sedimentation (Krometis et al. 2009), it could decrease photoinactivation by blocking sunlight and increasing loading of carbon, nitrogen, and phosphorus to ponds that could stimulate bacterial growth (Heisler et al. 2008). Similarly, intense rainfall events could dilute the bacteria concentration in influent stormwater under source limited conditions. It could also decrease the hydraulic retention time (Sønderup et al. 2016, Su et al. 2009) and deliver more growth stimulants (Liu et al. 2014)—both factors can increase FIB concentration in the pond. High water flow could resuspend previously settled bacteria due to turbulent flow (Banas et al. 2010). The area receiving frequent rainfall also supports vegetation, delivering a high amount of DOC and nutrients to ponds and supporting the growth of FIB (Glick 2012). If the FIB sources are depleted, precipitation could dilute the concentration of FIB. The temperature of water affects the biodegradation of organic matter in retention ponds, which provides nutrients to FIB and affects the growth rate of the FIB (Nydahl et al. 2013). As the temperature of water increases on sunny days or summer seasons, the same conditions can also increase inactivation by sunlight (Maraccini et al. 2016a). Thus, similarly to precipitation, temperature could have an ambiguous effect on FIB removal in retention ponds.

Water composition: Local weather may change the influent stormwater composition (Morison et al. 2017), which could affect the removal of FIB in retention ponds. The presence of suspended solids can have opposite effects on removal. They could facilitate sedimentation of particle-associated FIB and increase removal (Walters et al. 2013). However, suspended solids could protect FIB from inactivation (Henao et al. 2018) and facilitate the transport of bacteria (Ahn 2012). Suspended solids leach nutrients that may induce bacterial growth (Li and Zuo 2020) including natural FIB predators. If FIB is outcompeted for nutrients and become susceptible to predation, their concentration could decrease in ponds (Bauer and Forchhammer 2021). Thus, water chemistry – which can vary based on local weather – may also ambiguously affect FIB removal in retention ponds.

3.5. Correlation between FIB removal and factors affecting the removal processes in field conditions

3.5.1. Types of FIB did not explain their removal in field conditions

The survival and inactivation of gram-positive and gram-negative bacteria could differ in laboratory experiments based on the types of reactive oxygen species present (Huang et al. 2012), sunlight exposure (Maraccini et al. 2016b), and water chemistry (Chen et al. 2013). Analyzing 3 types of gram-negative bacteria (*Escherichia coli, Fecal coliform, Total coliform*) and 2 types of gram-positive bacteria (*Enterococcus, Fecal Streptococcus*), we showed that retention ponds could remove all types, but the removal capacity varied widely irrespective of species (Figure 3.2 – A and B). Removal of all 5 species was similar ($p > 0.05$), indicating that FIB type did not affect their removal. However, laboratory studies showed that removal could vary with changes in bacterial cell wall properties (Dörr et al. 2019), which affect their adsorption on suspended particles (Ploux et al. 2010). A lack of agreement between field and laboratory data is attributed to confounding factors in field conditions related to water compositions, which are typically not varied in laboratory experiments. For instance, a laboratory study concluded that the photoinactivation rate of *E. coli* was faster than *Enterococcus faecalis* when dissolved organic matter was present (Maraccini et al. 2016a). However, dissolved organic matter may be photodegraded under field conditions due to exposure to sunlight (Porcal et al. 2015) which would likely make the photoinactivation rate between *E. coli* and *Enterococcus faecalis* similar. Therefore, laboratory experiments tend to overestimate the photoinactivation rate (Fisher et al. 2012) because confounding factors could inhibit inactivation in the field. Thus, variation of water chemistry (Bertilsson and Widenfalk 2002) and local weather (Dias et al. 2017) should be reported along with FIB removal data.

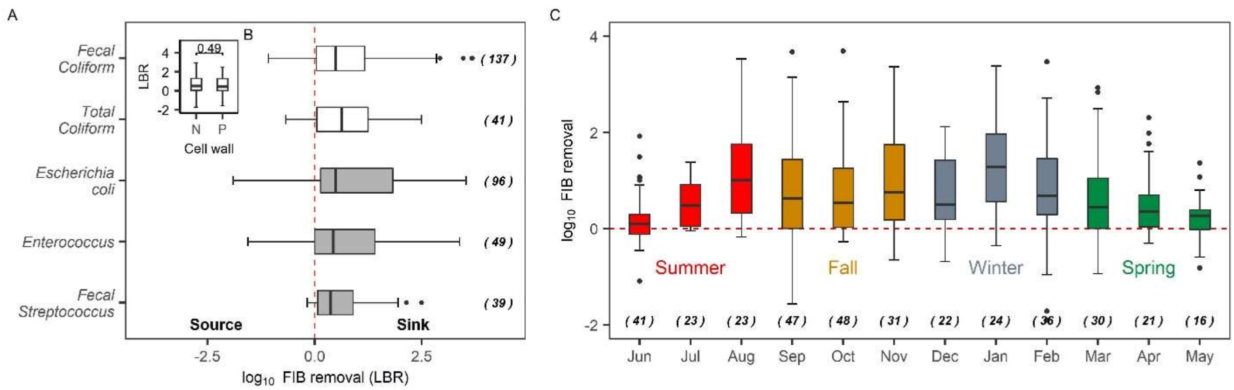


Figure 3.2 – (A) Log removal for 5 types of fecal indicator bacteria (FIB) in 19 retention ponds. Values between parentheses represent the n-values used to create each boxplot. (B) Comparison between the $\log_{10}$ FIB removal (LBR) of gram-positive (P) bacteria (*fecal coliform* and *total coliform*) and gram-negative (N) bacteria (*Escherichia coli, enterococcus*, and *fecal streptococcus*) in retention ponds. The value between the boxplots represents the p-value based on the Wilcoxon t-test (p-value < 0.05 represents statistically different removal rates). (C) Monthly removal of FIB in retention ponds located in North America is divided between summer (June, July, and August), fall (September, October, and November), winter (December, January, and February), and spring (March, April, and May). Horizontal dashed lines represent no removal of bacteria. Positive values represent that the retention pond act as a sink for the FIB, while negative values represent that the retention pond acts as a source of FIB.

3.5.2. FIB removal rates depended on seasons but monthly precipitation and temperature data did not explain FIB removal

Our analysis showed that FIB removal in retention ponds varied with seasons (Figure 3.2 – C). The removal was highest in winter and the lowest in spring (median removal, $\mu = 0.55$). Differences in precipitation and temperature across the year could explain the seasonal variability (DeLorenzo et al. 2012). While the pond hydraulic retention time is susceptible to seasonal precipitation levels, microbial activity is greatly influenced by temperature as higher temperatures can increase metabolism rates and enhance enzyme activity (Smith et al. 2019). These factors affect FIB removal or persistence in ponds. Because late spring and early summer may provide warm days coupled with spring snowmelt and summer storms, the period in between both seasons may be unfavorable for FIB removal in ponds due to accelerated bacterial growth and reduced retention time. In contrast, high removal rates during the winter can be explained in terms of slow bacterial growth attributed to low temperatures (Membré et al. 2005), long exposure to UV-light can occur during the winter (Malinović-Milićević et al. 2022), and increased bacteria dilution during winter rainfall events (Leandro et al. 2022). Overall, these results indicate that seasonal variability is expected in all ponds with the highest FIB removal observed in winter. Thus, the effect of temperature and water compositions in different seasons could partially explain the variability in removal between seasons.

Monthly precipitation or temperature data for each retention pond explained less than 2.5% (or $R^2 < 0.025$) of FIB removal in the field (Figure 3.3). R^2 values below 0.25 are considered to have a weak correlation between variables (Purwanto and Sudargini 2021), showing that monthly precipitation or temperature could not predict FIB removal alone. We attributed a lack of correlation to both positive and negative effects of temperature and precipitation on FIB removal based on the removal mechanisms. FIB concentration in ponds could be higher in wet-weather than dry-weather precipitation events (Huang et al. 2016), possibly because an increase in the antecedent drying period could facilitate FIB die-off rate (Hou et al. 2018). However, antecedent drying periods were not reported in the database. While elevated precipitation could increase the discharge of dissolved organic matter and nutrients that could favor bacterial growth and increase FIB concentration in ponds (Nydahl et al. 2013), it could also dilute the source and lower FIB concentration. Intense rainfall could increase flow rates and decrease hydraulic retention time (Grzywna 2019), which could decrease FIB removal (Ferguson et al. 2003, Powers et al. 2020). Higher temperatures may increase the growth of bacteria including FIB, thereby raising their concentration (Hou et al. 2018, Rousk et al. 2012), but FIB concentration could also decrease by inactivation at higher temperature as it is typically associated with sunny days (Madoshi et al. 2021). It should be noted that we used monthly average temperature and precipitation data for the location or city where the ponds are located. This may not match with water temperature and precipitation data related to the sampling event. This could explain a lack of correlation. Thus, future studies should report water temperature, precipitation, and antecedent drying days corresponding to each sampling event. Nevertheless, the results confirmed that precipitation or temperature data poorly predicted seasonal variation of FIB removal in field conditions, indicating a limited utility of regression-based modeling for microbial water quality of surface waters in variable weather conditions.

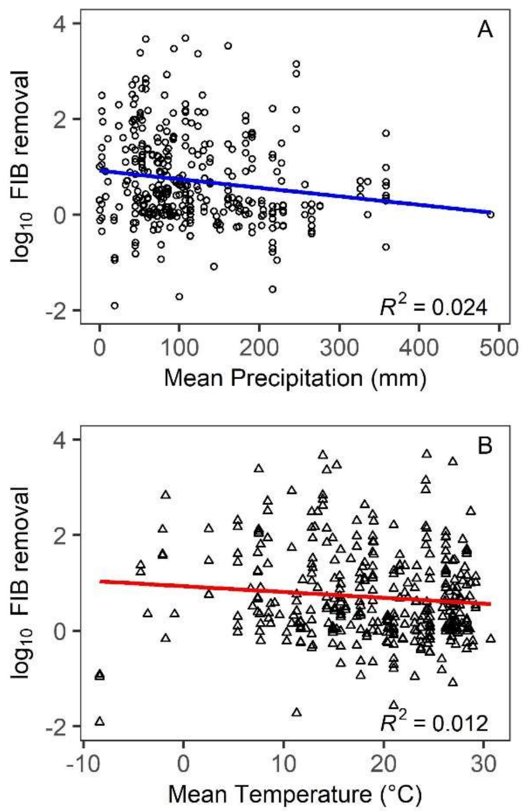


Figure 3.3 - Effect of mean precipitation (A) and temperature (B) on fecal indicator bacteria (FIB) removal in 19 retention ponds located in North America. R^2 values represent the variance of the linear regression model.

3.5.3. Suspended solid loading explained only 5% of FIB removal data

Laboratory experiments showed that the presence of suspended solids may induce bacterial growth by providing nutrients (Valenca et al. 2020) and reduce the photoinactivation of bacteria as the presence of solids protect bacteria from UV light (Walters et al. 2013). Suspended solids should strongly correlate with reduction in bacteria removal in retention ponds in field conditions. Analyzing the performance of retention ponds on the simultaneous removal of FIB and total suspended solids (TSS), we showed that an increase in solid removal increased the FIB removal, but the correlation between FIB and TSS removal was less than 5% (Figure 3.4). Coincidentally, an increase in precipitation also exhibited a weak correlation to an increase in the concentration of FIB and total suspended solids (TSS) in influent water (Figure 3.5), indicating weather conditions could dictate the loading of TSS, some of which may carry FIB into the ponds. Thus, the removal of suspended solid could also remove a fraction of total FIB in water. Our analysis suggests that removal of TSS only explained 5% of the FIB removal. The low correlation suggests that TSS can either help or inhibit the FIB removal based on the conditions. Previous studies showed that suspended solid removal occurred in retention ponds in all seasons (Nayeb Yazdi et al. 2021) but the extent of removal changed with the local precipitation (Carpenter et al. 2013). High flow, which occurred during heavy precipitation events, could also affect suspended sediment removal by settling (He et al. 2010). Consequently, high flow could lower the likelihood of FIB removal by settling in associated with sediments (Ausland et al. 2002). However, a high concentration of suspended solids could also decrease the photoinactivation of FIB in ponds (Walters et al. 2013) as particles may serve as a shield for the bacteria from the sunlight (Gutiérrez-Cacciabue et al. 2016). Moreover, suspended solids were found to carry more than half of the bacteria available in surface water depending on the concentration of the suspended solids (Jiang et al. 2015).

Suspended solids can also affect the die-off or decay of bacteria, but the decay rate depends on the particle size as FIB associated with smaller particles have faster decay than pathogen associated with large particles (Walters et al. 2013). The resuspension of sediment by turbulent flow during high-intensity rainfall could also increase FIB concentration. For instance, a previous study has shown that older ponds retain 10 to 50% fewer particulates than young ponds (Sønderup et al. 2016) possibly due to the resuspension of settled sediments in older ponds. Nevertheless, the overall analysis indicates that TSS removal explained only a small fraction of FIB removal, indicating settling of particle-associated FIB is not the dominant removal process, and that blocking of sunlight and resuspension and transport of sediment-associated FIB could complicate the prediction of FIB removal based on TSS concentration. Because all these processes are related to weather conditions such as sunny days or rainfall intensity, FIB removal may not be predicted well based on TSS removal data in the field.

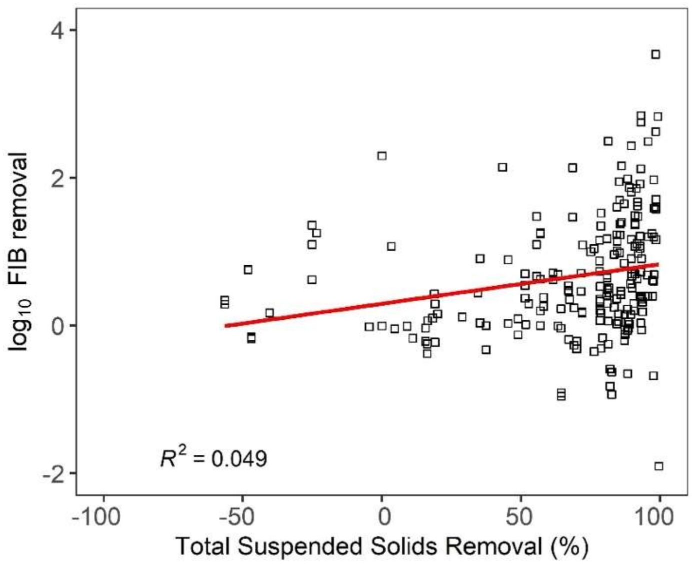


Figure 3.4 – Correlation between the removal of total suspended solids and the log removal of fecal indicator bacteria (FIB) in 19 different retention ponds located in North America. The removal of FIB and suspended solids were calculated based on the influent and effluent concentration of the contaminants measured in the same sample at the same location. R^2 values represent the variance of the linear regression model.

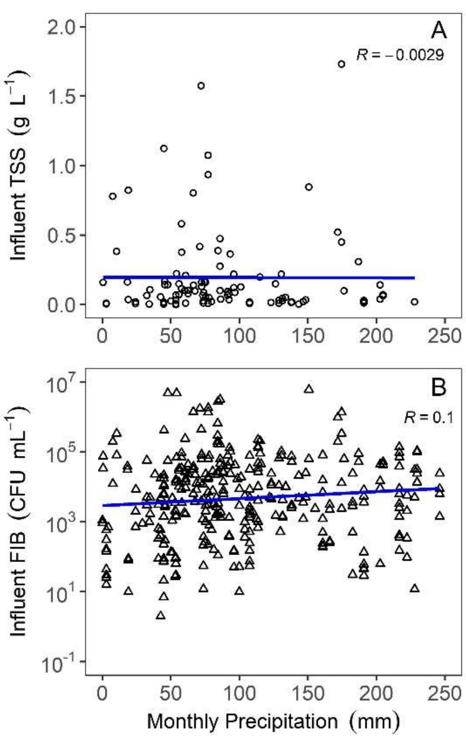


Figure 3.5 – Effect of local precipitation on the influent concentration of (A) total suspended solids (TSS) and (B) fecal indicator bacteria (FIB) in 19 retention ponds located in North America. Local precipitation was calculated as yearly average precipitation of each city. R-values represent the Pearson correlation of each linear regression model.

3.5.4. Removal of P and N did not explain FIB removal

Bioavailable nutrients in surface waters can stimulate the growth of bacteria (Westrich Jason et al. 2016), and local weather can affect nutrient loading into retention ponds (Fong et al. 2020). Nutrients including both nitrogen and phosphorus may present in stormwater runoff in dissolved and particulate form. Similar to FIB, nutrients could be removed by adsorption and sedimentation, and their removal is sensitive to local weather and the design of retention ponds (Valenca et al. 2021). Herein, we compared the removal of FIB to the removal of nutrient species to indicate if nutrient removal can be used as a proxy to the FIB removal capacity of the ponds or whether removal of nutrient species could explain FIB removal. Our analysis confirmed that FIB removal was not correlated ($R^2 < 0.005$) to TN and ammonia removal, and the correlation between FIB and NO_3^- removals was weak ($R^2 < 0.06$) (Figure 3.6). A weak correlation between FIB and nitrogen species removal could be partially attributed to a difference in mechanisms of N removal and FIB removal. In ponds, most nitrogen species could be removed via biological assimilation or transformation (Kadlec and Wallace 2008) and uptake by plants (Collins et al. 2010, Saunders and Kalff 2001). TN can be removed as sedimentation upon formation of particulates (Kadlec and Wallace 2008, Troitsky et al. 2019). However, the sedimentation of organic residues or plant detritus, which constitute most particulate TN, is expected to be slower than that of soil minerals due to the low density of plant detritus. Furthermore, particulate nitrogen could also release dissolved nutrients that could stimulate FIB growth. Thus, a difference in magnitude and type of removal processes affecting N removal and FIB removal results in a poor correlation between both parameters, showing that regression-based modeling using nutrient concentration is unlikely to predict FIB removal in retention ponds.

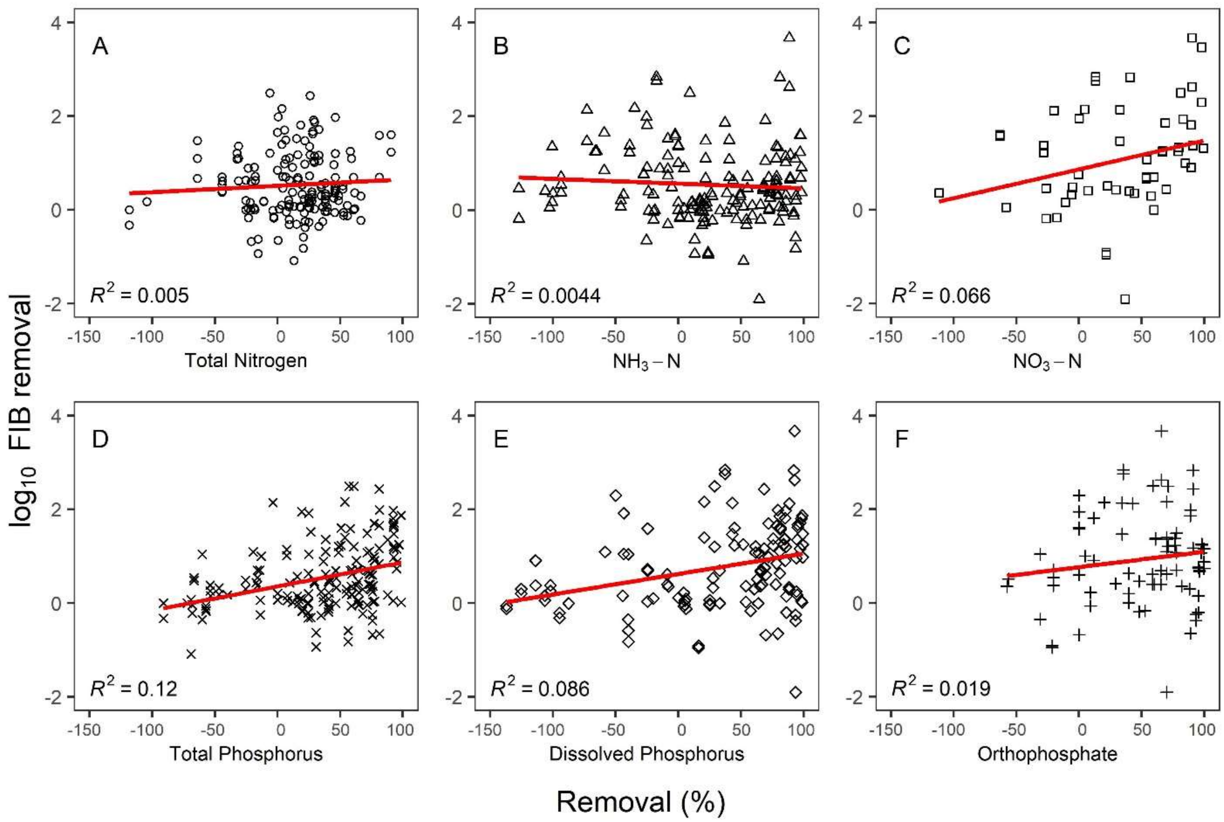


Figure 3.6 – Fecal indicator bacteria (FIB) removal variability in retention ponds based on the removal of (A) Total Nitrogen, (B) Ammonia, (C) Nitrate, (D) Total Phosphorus, (E) Dissolved Phosphorus, and (F) Orthophosphate as P. Positive removal values represent a positive removal of the pollutant by the retention pond, while negative removal values represent the retention pond acts as a source of the pollutant. R^2 values represent the variance of the linear regression model.

In contrast to TN removal, a better correlation (R^2 = 0.12) was observed between TP removal and FIB removal, indicating an increase in TP removal predicted 12% of the FIB removal data (Figure 3.6). However, an increase in dissolved P and phosphate removal predicted only 8% and 2% of the FIB removal data, respectively. We attributed the improved correlation between TP and FIB removal owing to similar removal mechanisms such as sedimentation and adsorption. Bacterial outer cell walls are composed of phospholipids, where the hydrophilic end with phosphate group interacts with particles or sediments (Cagnasso et al. 2010). Thus, their interaction with settled and suspended sediments can be similar to that of P species. As a fraction of TP is particulate phosphorus in urban retention ponds (Song et al. 2017), sedimentation can contribute to part of TP removal. This explained why 12% of FIB removal data can be explained by TP removal. Phosphate and other dissolved P species could bind with iron oxide and aluminum oxides present in suspended sediments (Weng et al. 2012) and be removed by settling, similar to particle-facilitated sedimentation of FIB. However, the presence of dissolved P such as orthophosphate could also limit the binding of FIB to particulates (Appenzeller et al. 2002) and stimulate the growth of FIB in pond water (Croft et al. 2005, Ramanan et al. 2016). Both processes could lower FIB removal. This explained a low correlation of phosphate removal with FIB removal, compared with that of TP removal. In summary, the analysis indicates that the presence of nutrients can affect FIB removal by altering FIB interaction with particles and stimulating their growth, but the changes in these processes due to the presence of nutrients might not predict FIB removal. This signifies again the limited utility of regression-based modeling to predict FIB removal in ponds under complex environmental conditions.

3.6. Best machine learning method to predict FIB removal

Using 80% of our data to train the machine learning models, we showed that the Random Forest (RF) model achieved the highest training accuracy (0.58 ± 0.04), while k-Nearest Neighbors achieved the lowest training accuracy (0.29 ± 0.05) (Figure 3.7). The advantage of RF compared to the other models is due to its learning ability and robustness against strong data errors (Ao et al. 2019). Because environmental datasets are unique to locations, the learning ability of RF in creating unique decision trees within the dataset may play an advantage in predicting environmental data. Among all ML models, RF could reduce the bias and the variance of datasets that may arise from cofounding environmental factors (Gama 2004), thereby increasing the training accuracy. In the dataset, input data vary widely: monthly precipitation (0 to 489.2 mm), monthly temperature (-8.4 to 30.7 ºC), and influent FIB concentration (0.02 to 1.7 x 105 CFU mL-

1). Despite the high variability of the data fed into the RF model that could reduce the model's accuracy (Fang et al. 2021), the RF model predicted FIB removal with an accuracy of 65%. Unlike random forest that uses multiple decision trees to find hidden interaction between data points, both supporting vector machines and k-nearest neighbors rely on the distance between data points which could explain their inferior performance when using data with high variability. For instance, SVM is usually suited for two-class problems, while random forest is intrinsically suited for multiclass problems which could explain why RF overperformed SVM when calculating FIB removal based on 4 input variables. On the other hand, kNN typically requires a large number of training examples which was not possible to achieve with the current data set. Lastly, random forest commonly outperforms classification and regression trees because RF uses multiple decision trees (e.g., a combination of multiple CART) which can significantly improve the accuracy of the results. The RF performance will likely continue to improve with the usage of additional inputs like total phosphorus removal data. However, future work needs to be done to evaluate the extent to which the prediction performance will increase with additional input data. To improve the predicting power of ML algorithm on how weather conditions affect FIB removal, ML can be performed on a single pond, which requires extensive data. In that case, the effect of other confounding factors such as land use and pond design can be eliminated. Future studies should also optimize the hyperparameter for each machine learning model to further enhance the prediction results. Nonetheless, the RF model significantly outperformed all regression-based models previously presented in this study. RF could be used to predict pathogen concentration in retention ponds during changing climates and inform the regulatory agencies and institutions for improved management methods and data collection strategies.

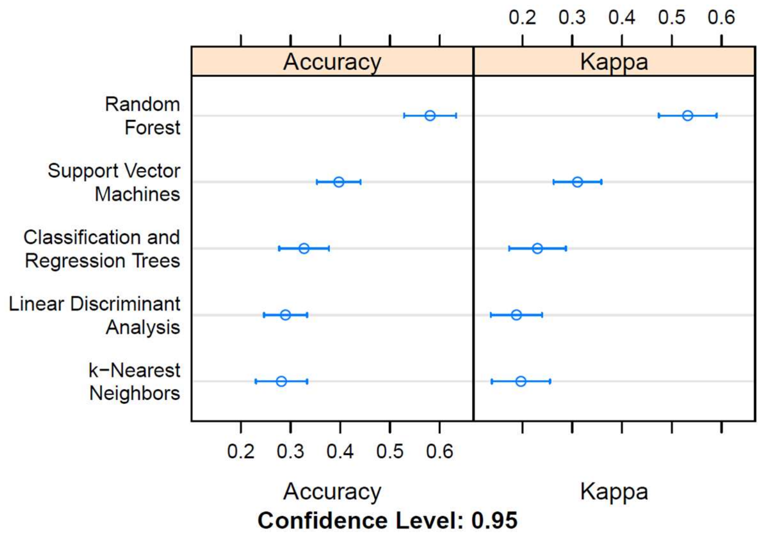


Figure 3.7 – Comparison between the accuracy of machine learning models in predicting FIB removal in retention ponds based on the average monthly temperature, precipitation, and daily bacterial concentration in influent stormwater. The training dataset consisted of 298 rows which were equivalent to 80% of the entire dataset. Models were created in RStudio with the Caret package (version 1.4.1106).

3.7. Summary

This critical analysis of water quality field data from 19 retention ponds reveals that FIB removal varies widely. The FIB removal varied with seasons with the highest removal occurring during winter. Changes in local weather conditions such as an increase in rainfall intensity and temperature affect FIB removal, but the correlation is weak possibly due to the effects of confounding variables. The correlation between FIB removal and the removal of water quality parameters such as nutrients and suspended solids is weak (R2 < 0.12). Thus, regression-based modeling with these factors as input variables is ineffective in predicting FIB removal in retention ponds in field conditions where interactions between the factors effect FIB removal. Because weather conditions affect the chemical and biological processes of FIB removal in retention ponds, as well as the influent water composition, weather conditions should be included in the reported data to improve prediction of FIB removal in retention ponds. Our results showed that the application of machine learning models utilizing weather conditions as input variables could predict FIB removal in retention ponds. All five machine learning models implemented outperformed regression-based modeling, but the Random Forest was the best prediction model, with an accuracy of 65%. The accuracy percentage would likely increase with an increase in data availability including data related to water composition, accurate water temperature, and UV-light intensity. Thus, a better field data collection would increase the accuracy of machine learning models and help protect water resources. Overall, the results could help stormwater managers and government agencies to track the performance of retention ponds during climate change and inform the management methods best suited to reduce the risk of microbial pollution of water resources.

3.8. References

Abia, A.L.K., Ubomba-Jaswa, E. and Momba, M.N.B. (2016) Competitive Survival of Escherichia coli, Vibrio cholerae, Salmonella typhimurium and Shigella dysenteriae in Riverbed Sediments. Microbial Ecology 72(4), 881-889.

Ahmed, W., Hamilton, K., Toze, S., Cook, S. and Page, D. (2019) A review on microbial contaminants in stormwater runoff and outfalls: Potential health risks and mitigation strategies. Science of the Total Environment 692, 1304-1321.

Ahn, J.H. (2012) Size distribution and settling velocities of suspended particles in a tidal embayment. Water Research 46(10), 3219-3228.

Aldhyani, T.H.H., Al-Yaari, M., Alkahtani, H. and Maashi, M. (2020) Water Quality Prediction Using Artificial Intelligence Algorithms. Applied Bionics and Biomechanics 2020, 6659314.

Ao, Y., Li, H., Zhu, L., Ali, S. and Yang, Z. (2019) The linear random forest algorithm and its advantages in machine learning assisted logging regression modeling. Journal of Petroleum Science and Engineering 174, 776-789.

Appenzeller, B.M.R., Duval, Y.B., Thomas, F. and Block, J.-C. (2002) Influence of Phosphate on Bacterial Adhesion onto Iron Oxyhydroxide in Drinking Water. Environmental Science & Technology 36(4), 646-652.

Ausland, G., Stevik, T.K., Hanssen, J.F., Køhler, J.C. and Jenssen, P.D. (2002) Intermittent filtration of wastewater—removal of fecal coliforms and fecal streptococci. Water Research 36(14), 3507-3516.

Avelar, F.F., de Matos, A.T., de Matos, M.P. and Borges, A.C. (2014) Coliform bacteria removal from sewage in constructed wetlands planted with Mentha aquatica. Environmental Technology 35(16), 2095-2103.

Azamathulla, H.M., Ghani, A.A., Chang, C.K., Hasan, Z.A. and Zakaria, N.A. (2010) Machine Learning Approach to Predict Sediment Load – A Case Study. CLEAN – Soil, Air, Water 38(10), 969-976.

Banas, D., Marin, B., Skraber, S., Chopin, E.I.B. and Zanella, A. (2010) Copper mobilization affected by weather conditions in a stormwater detention system receiving runoff waters from vineyard soils (Champagne, France). Environmental Pollution 158(2), 476-482.

Bauer, A. and Forchhammer, K. (2021) Bacterial Predation on Cyanobacteria. Microbial Physiology 31(2), 99-108.

Bertilsson, S. and Widenfalk, A. (2002) Photochemical degradation of PAHs in freshwaters and their impact on bacterial growth – influence of water chemistry. Hydrobiologia 469(1), 23-32.

Bhattacharyya, S.K. and Rahul, K. (2013) Face recognition by linear discriminant analysis. International Journal of Communication Network Security 2(2), 31-35.

Boyacioglu, H. and Boyacioglu, H. (2010) Detection of seasonal variations in surface water quality using discriminant analysis. Environmental Monitoring and Assessment 162(1), 15-20.

Breiman, L. (2001) Random Forests. Machine Learning 45(1), 5-32.

Breiman, L., Friedman, J.H., Olshen, R.A. and Stone, C.J. (2017) Classification and regression trees, Routledge.

Brookes, J.D., Antenucci, J., Hipsey, M., Burch, M.D., Ashbolt, N.J. and Ferguson, C. (2004) Fate and transport of pathogens in lakes and reservoirs. Environment International 30(5), 741-759.

Cagnasso, M., Boero, V., Franchini, M.A. and Chorover, J. (2010) ATR-FTIR studies of phospholipid vesicle interactions with α-FeOOH and α-Fe2O3 surfaces. Colloids and Surfaces B: Biointerfaces 76(2), 456-467.

Carpenter, J.F., Vallet, B., Pelletier, G., Lessard, P. and Vanrolleghem, P.A. (2013) Pollutant removal efficiency of a retrofitted stormwater detention pond. Water Quality Research Journal 49(2), 124-134.

Chen, P.-Y., Tung, C.-P., Tsao, J.-H. and Chen, C.-J. (2021) Assessing Future Rainfall Intensity–Duration–Frequency Characteristics across Taiwan Using the k-Nearest Neighbor Method. Water 13(11), 1521.

Chen, Q., Li, J., Wu, Y., Shen, F.X. and Yao, M.S. (2013) Biological responses of Gram-positive and Gram-negative bacteria to nZVI (Fe-0), Fe2+ and Fe3+. Rsc Advances 3(33), 13835-13842.

Cheng, Y. (2008) Sediment Discharge from a Storm-Water Retention Pond1. Journal of Irrigation and Drainage Engineering 134(5), 606-612.

Choubin, B., Moradi, E., Golshan, M., Adamowski, J., Sajedi-Hosseini, F. and Mosavi, A. (2019) An ensemble prediction of flood susceptibility using multivariate discriminant analysis, classification and regression trees, and support vector machines. Science of the Total Environment 651, 2087-2096.

Choubin, B., Zehtabian, G., Azareh, A., Rafiei-Sardooi, E., Sajedi-Hosseini, F. and Kişi, Ö. (2018) Precipitation forecasting using classification and regression trees (CART) model: a comparative study of different approaches. Environmental Earth Sciences 77(8), 314.

Ciupa, T., Suligowski, R. and Wałek, G. (2020) Impact of an urban area on the dynamics and features of suspended solids transport in a small catchment during floods. Ecohydrology & Hydrobiology.

Clary, J., Leisenring, M., Poresky, A., Earles, A. and Jones, J. (2011) World Environmental and Water Resources Congress 2011, pp. 441-449.

Collins, K.A., Lawrence, T.J., Stander, E.K., Jontos, R.J., Kaushal, S.S., Newcomer, T.A., Grimm, N.B. and Ekberg, M.L.C. (2010) Opportunities and challenges for managing nitrogen in urban stormwater: A review and synthesis. Ecological Engineering 36(11), 1507-1519.

Cornforth, D.M. and Foster, K.R. (2013) Competition sensing: the social side of bacterial stress responses. Nature Reviews Microbiology 11(4), 285-293.

Croft, M.T., Lawrence, A.D., Raux-Deery, E., Warren, M.J. and Smith, A.G. (2005) Algae acquire vitamin B12 through a symbiotic relationship with bacteria. Nature 438(7064), 90-93.

Dean, K. and Mitchell, J. (2022) Identifying water quality and environmental factors that influence indicator and pathogen decay in natural surface waters. Water Research 211, 118051.

DeLorenzo, M.E., Thompson, B., Cooper, E., Moore, J. and Fulton, M.H. (2012) A long-term monitoring study of chlorophyll, microbial contaminants, and pesticides in a coastal residential stormwater pond and its adjacent tidal creek. Environmental Monitoring and Assessment 184(1), 343-359.

Dias, D.F.C., Passos, R.G. and von Sperling, M. (2017) A review of bacterial indicator disinfection mechanisms in waste stabilisation ponds. Reviews in Environmental Science and Bio/Technology 16(3), 517-539.

Djarum, D.H., Ahmad, Z. and Zhang, J. (2021) Computer Aided Chemical Engineering. Türkay, M. and Gani, R. (eds), pp. 1491-1496, Elsevier.

Dörr, T., Moynihan, P.J. and Mayer, C. (2019) Editorial: Bacterial Cell Wall Structure and Dynamics. Frontiers in Microbiology 10(2051).

EPA, U. (2002) National water quality inventory 2000 report.

Fang, H., Jamali, B., Deletic, A. and Zhang, K. (2021) Machine learning approaches for predicting the performance of stormwater biofilters in heavy metal removal and risk mitigation. Water Research 200, 117273.

Ferguson, C., Husman, A.M.d.R., Altavilla, N., Deere, D. and Ashbolt, N. (2003) Fate and Transport of Surface Water Pathogens in Watersheds. Critical Reviews in Environmental Science and Technology 33(3), 299-361.

Fisher, M.B., Iriarte, M. and Nelson, K.L. (2012) Solar water disinfection (SODIS) of Escherichia coli, Enterococcus spp., and MS2 coliphage: effects of additives and alternative container materials. Water Res 46(6), 1745-1754.

Fong, C.R., Gaynus, C.J. and Carpenter, R.C. (2020) Extreme rainfall events pulse substantial nutrients and sediments from terrestrial to nearshore coastal communities: a case study from French Polynesia. Scientific Reports 10(1), 2955.

Fox, E.W., Hill, R.A., Leibowitz, S.G., Olsen, A.R., Thornbrugh, D.J. and Weber, M.H. (2017) Assessing the accuracy and stability of variable selection methods for random forest modeling in ecology. Environmental Monitoring and Assessment 189(7), 316.

Gama, J. (2004) Functional Trees. Machine Learning 55(3), 219-250.

Gao, F., Shen, Y., Sallach, J.B., Li, H., Liu, C. and Li, Y. (2021) Direct Prediction of Bioaccumulation of Organic Contaminants in Plant Roots from Soils with Machine Learning Models Based on Molecular Structures. Environmental Science & Technology.

Glick, B.R. (2012) Plant Growth-Promoting Bacteria: Mechanisms and Applications. Scientifica 2012, 963401.

Gong, Y., Liang, X., Li, X., Li, J., Fang, X. and Song, R. (2016) Influence of Rainfall Characteristics on Total Suspended Solids in Urban Runoff: A Case Study in Beijing, China. Water 8(7), 278.

Grzywna, A. (2019) Indicators of Variability of Water Flow Rate in the Piwonia River. Journal of Ecological Engineering 20(7), 205-210.

Gutiérrez-Cacciabue, D., Cid, A.G. and Rajal, V.B. (2016) How long can culturable bacteria and total DNA persist in environmental waters? The role of sunlight and solid particles. Science of the Total Environment 539, 494-502.

Hartmann, A., Gleeson, T., Wada, Y. and Wagener, T. (2017) Enhanced groundwater recharge rates and altered recharge sensitivity to climate variability through subsurface heterogeneity. Proceedings of the National Academy of Sciences 114(11), 2842-2847.

Hathaway, J.M. and Hunt, W.F. (2010) World Environmental and Water Resources Congress 2010, pp. 3218-3225.

Hathaway, J.M. and Hunt, W.F. (2012) Indicator Bacteria Performance of Storm Water Control Measures in Wilmington, North Carolina. Journal of Irrigation and Drainage Engineering 138(2), 185-197.

He, J., Valeo, C., Chu, A. and Neumann, N.F. (2010) Characteristics of Suspended Solids, Microorganisms, and Chemical Water Quality in Event-Based Stormwater Runoff from an Urban Residential Area. Water Environment Research 82(12), 2333-2345.

Heisler, J., Glibert, P.M., Burkholder, J.M., Anderson, D.M., Cochlan, W., Dennison, W.C., Dortch, Q., Gobler, C.J., Heil, C.A., Humphries, E., Lewitus, A., Magnien, R., Marshall, H.G., Sellner, K., Stockwell, D.A., Stoecker, D.K. and Suddleson, M. (2008) Eutrophication and harmful algal blooms: A scientific consensus. Harmful Algae 8(1), 3-13.

Henao, L.D., Cascio, M., Turolla, A. and Antonelli, M. (2018) Effect of suspended solids on peracetic acid decay and bacterial inactivation kinetics: Experimental assessment and definition of predictive models. Science of the Total Environment 643, 936-945.

Herrmann, J. (2012) Chemical and biological benefits in a stormwater wetland in Kalmar, SE Sweden. Limnologica 42(4), 299-309.

Hibbing, M.E., Fuqua, C., Parsek, M.R. and Peterson, S.B. (2010) Bacterial competition: surviving and thriving in the microbial jungle. Nature Reviews Microbiology 8(1), 15-25.

Hou, X., Chen, L., Wei, G., Gong, Y. and Shen, Z. (2018) Factors affecting microbial and physico-chemical pollutants in stormwater in a typical Chinese urban catchment. Environmental Science: Processes & Impacts 20(12), 1697-1707.

Huang, H., Chow, C.W.K. and Jin, B. (2016) Characterisation of dissolved organic matter in stormwater using high-performance size exclusion chromatography. Journal of Environmental Sciences 42, 236-245.

Huang, L., Xuan, Y., Koide, Y., Zhiyentayev, T., Tanaka, M. and Hamblin, M.R. (2012) Type I and Type II mechanisms of antimicrobial photodynamic therapy: An in vitro study on gram-negative and gram-positive bacteria. Lasers in Surgery and Medicine 44(6), 490-499.

Huang, M., Lin, R., Huang, S. and Xing, T. (2017) A novel approach for precipitation forecast via improved K-nearest neighbor algorithm. Advanced Engineering Informatics 33, 89-95.

Huovinen, P., Gómez, I. and Lovengreen, C. (2006) A Five-year Study of Solar Ultraviolet Radiation in Southern Chile (39° S): Potential Impact on Physiology of Coastal Marine Algae? Photochemistry and Photobiology 82(2), 515-522.

Ishii, S., Yan, T., Shively, D.A., Byappanahalli, M.N., Whitman, R.L. and Sadowsky, M.J. (2006) Cladophora (Chlorophyta) spp. harbor human bacterial pathogens in nearshore water of Lake Michigan. Applied and Environmental Microbiology 72(7), 4545-4553.

Jang, J., Hur, H.-G., Sadowsky, M.J., Byappanahalli, M.N., Yan, T. and Ishii, S. (2017) Environmental Escherichia coli: ecology and public health implications—a review. Journal of Applied Microbiology 123(3), 570-581.

Ji, Z., Li, N., Xie, W., Wu, J. and Zhou, Y. (2013) Comprehensive assessment of flood risk using the classification and regression tree method. Stochastic Environmental Research and Risk Assessment 27(8), 1815-1828.

Jiang, L., Chen, Y.C., Zhu, D.J. and Liu, Z.W. (2015) Faecal coliform attachment to settleable suspended sediments in fresh surface waters: linear partition model validation and sediment concentration effects. Water Supply 15(4), 864-870.

Kadlec, R.H. and Wallace, S. (2008) Treatment wetlands, CRC press.

Kim, J.H., Shin, J.-K., Lee, H., Lee, D.H., Kang, J.-h., Cho, K.H., Lee, Y.-G., Chon, K., Baek, S.-S. and Park, Y. (2021) Improving the performance of machine learning models for early warning of harmful algal blooms using an adaptive synthetic sampling method. Water Research, 117821.

Krometis, L.-A.H., Drummey, P.N., Characklis, G.W. and Sobsey, M.D. (2009) Impact of microbial partitioning on wet retention pond effectiveness. Journal of Environmental Engineering 135(9), 758-767.

Leandro, J., Hotta, C.I., Pinto, T.A. and Ahadzie, D.K. (2022) Expected Annual Probability of Infection: a flood-risk approach to waterborne infectious diseases. Water Research, 118561.

Leong, W.C., Bahadori, A., Zhang, J. and Ahmad, Z. (2021) Prediction of water quality index (WQI) using support vector machine (SVM) and least square-support vector machine (LS-SVM). International Journal of River Basin Management 19(2), 149-156.

Li, J. and Zuo, Q. (2020) Forms of Nitrogen and Phosphorus in Suspended Solids: A Case Study of Lihu Lake, China. Sustainability 12(12), 5026.

Li, S., Kazemi, H. and Rockaway, T.D. (2019) Performance assessment of stormwater GI practices using artificial neural networks. Science of the Total Environment 651, 2811-2819.

Liu, R., Wang, J., Shi, J., Chen, Y., Sun, C., Zhang, P. and Shen, Z. (2014) Runoff characteristics and nutrient loss mechanism from plain farmland under simulated rainfall conditions. Science of the Total Environment 468-469, 1069-1077.

Liu, X., Lu, D., Zhang, A., Liu, Q. and Jiang, G. (2022) Data-Driven Machine Learning in Environmental Pollution: Gains and Problems. Environmental Science & Technology 56(4), 2124-2133.

Lusk, M.G. and Toor, G.S. (2016) Biodegradability and Molecular Composition of Dissolved Organic Nitrogen in Urban Stormwater Runoff and Outflow Water from a Stormwater Retention Pond. Environmental Science & Technology 50(7), 3391-3398.

Madoshi, P.B., Muhairwa, A.P., Mtambo, M.M. and Forslund, A. (2021) Inactivation of Escherichia Coli, Coliforms and Coliphages During Storage of Animal Wastes Under Tropical Climatic Conditions. Frontiers in Environmental Microbiology 7(1), 35.

Malinović-Milićević, S., Mijatović, Z., Stanojević, G., Radovanović, M.M. and Popović, V. (2022) Health risks of extended exposure to low-level UV radiation – An analysis of ground-based and satellite-derived data. Science of the Total Environment 831, 154899.

Maraccini, P.A., Ferguson, D.M. and Boehm, A.B. (2012) Diurnal Variation in Enterococcus Species Composition in Polluted Ocean Water and a Potential Role for the Enterococcal Carotenoid in Protection against Photoinactivation. Applied and Environmental Microbiology 78(2), 305-310.

Maraccini, P.A., Mattioli, M.C.M., Sassoubre, L.M., Cao, Y., Griffith, J.F., Ervin, J.S., Van De Werfhorst, L.C. and Boehm, A.B. (2016a) Solar Inactivation of Enterococci and Escherichia coli in Natural Waters: Effects of Water Absorbance and Depth. Environmental Science & Technology 50(10), 5068-5076.

Maraccini, P.A., Wenk, J. and Boehm, A.B. (2016b) Exogenous indirect photoinactivation of bacterial pathogens and indicators in water with natural and synthetic photosensitizers in simulated sunlight with reduced UVB. Journal of Applied Microbiology 121(2), 587-597.

Membré, J.-M., Leporq, B., Vialette, M., Mettler, E., Perrier, L., Thuault, D. and Zwietering, M. (2005) Temperature effect on bacterial growth rate: quantitative microbiology approach including cardinal values and variability estimates to perform growth simulations on/in food. International Journal of Food Microbiology 100(1), 179-186.

Merriman, L.S., Moore, T.L.C., Wang, J.W., Osmond, D.L., Al-Rubaei, A.M., Smolek, A.P., Blecken, G.T., Viklander, M. and Hunt, W.F. (2017) Evaluation of factors affecting soil carbon sequestration services of stormwater wet retention ponds in varying climate zones. Science of the Total Environment 583, 133-141.

Modaresi, F. and Araghinejad, S. (2014) A Comparative Assessment of Support Vector Machines, Probabilistic Neural Networks, and K-Nearest Neighbor Algorithms for Water Quality Classification. Water Resources Management 28(12), 4095-4111.

Moeini, M., Shojaeizadeh, A. and Geza, M. (2021) Supervised Machine Learning for Estimation of Total Suspended Solids in Urban Watersheds. Water 13(2), 147.

Mohammadpour, R., Shaharuddin, S., Chang, C.K., Zakaria, N.A., Ghani, A.A. and Chan, N.W. (2015) Prediction of water quality index in constructed wetlands using support vector machine. Environmental Science and Pollution Research 22(8), 6208-6219.

Morison, M.Q., Macrae, M.L., Petrone, R.M. and Fishback, L. (2017) Capturing temporal and spatial variability in the chemistry of shallow permafrost ponds. Biogeosciences 14(23), 5471-5485.

Nayeb Yazdi, M., Scott, D., Sample, D.J. and Wang, X. (2021) Efficacy of a retention pond in treating stormwater nutrients and sediment. Journal of Cleaner Production 290, 125787.

Nguyen, M.T., Jasper, J.T., Boehm, A.B. and Nelson, K.L. (2015) Sunlight inactivation of fecal indicator bacteria in open-water unit process treatment wetlands: Modeling endogenous and exogenous inactivation rates. Water Research 83, 282-292.

Noble, W.S. (2006) What is a support vector machine? Nature Biotechnology 24(12), 1565-1567.

Noori, R., Abdoli, M.A., Ghasrodashti, A.A. and Jalili Ghazizade, M. (2009) Prediction of municipal solid waste generation with combination of support vector machine and principal component analysis: A case study of Mashhad. Environmental Progress & Sustainable Energy 28(2), 249-258.

Nouraki, A., Alavi, M., Golabi, M. and Albaji, M. (2021) Prediction of water quality parameters using machine learning models: a case study of the Karun River, Iran. Environmental Science and Pollution Research.

Nydahl, A., Panigrahi, S. and Wikner, J. (2013) Increased microbial activity in a warmer and wetter climate enhances the risk of coastal hypoxia. Fems Microbiology Ecology 85(2), 338-347.

Oster, R.J., Wijesinghe, R.U., Haack, S.K., Fogarty, L.R., Tucker, T.R. and Riley, S.C. (2014) Bacterial Pathogen Gene Abundance and Relation to Recreational Water Quality at Seven Great Lakes Beaches. Environmental Science & Technology 48(24), 14148-14157.

Peaple, M.D., Tierney, J.E., McGee, D., Lowenstein, T.K., Bhattacharya, T. and Feakins, S.J. (2021) Identifying plant wax inputs in lake sediments using machine learning. Organic Geochemistry 156, 104222.

Pennino, M.J., Leibowitz, S.G., Compton, J.E., Hill, R.A. and Sabo, R.D. (2020) Patterns and predictions of drinking water nitrate violations across the conterminous United States. Science of the Total Environment 722, 137661.

Pizarro, J., Vergara, P.M., Morales, J.L., Rodríguez, J.A. and Vila, I. (2014) Influence of land use and climate on the load of suspended solids in catchments of Andean rivers. Environmental Monitoring and Assessment 186(2), 835-843.

Ploux, L., Ponche, A. and Anselme, K. (2010) Bacteria/Material Interfaces: Role of the Material and Cell Wall Properties. Journal of Adhesion Science and Technology 24(13-14), 2165-2201.

Porcal, P., Dillon, P.J. and Molot, L.A. (2015) Temperature Dependence of Photodegradation of Dissolved Organic Matter to Dissolved Inorganic Carbon and Particulate Organic Carbon. Plos One 10(6), e0128884.

Powers, N.C., Wallgren, H.R., Marbach, S. and Turner, J.W. (2020) Relationship between Rainfall, Fecal Pollution, Antimicrobial Resistance, and Microbial Diversity in an Urbanized Subtropical Bay. Applied and Environmental Microbiology 86(19), e01229-01220.

Purwanto, A. and Sudargini, Y. (2021) Partial Least Squares Structural Squation Modeling (PLS-SEM) Analysis for Social and Management Research : A Literature Review. Journal of Industrial Engineering & Management Research 2(4), 114 - 123.

Raghavendra. N, S. and Deka, P.C. (2014) Support vector machine applications in the field of hydrology: A review. Applied Soft Computing 19, 372-386.

Ramanan, R., Kim, B.-H., Cho, D.-H., Oh, H.-M. and Kim, H.-S. (2016) Algae–bacteria interactions: Evolution, ecology and emerging applications. Biotechnology Advances 34(1), 14-29.

Ramesh, R., Kalin, L., Hantush, M. and Chaudhary, A. (2021) A secondary assessment of sediment trapping effectiveness by vegetated buffers. Ecological Engineering 159, 106094.

Reed, R.H. (1997) Solar inactivation of faecal bacteria in water: the critical role of oxygen. Letters in Applied Microbiology 24(4), 276-280.

Rincón, A.-G. and Pulgarin, C. (2004) Field solar E. coli inactivation in the absence and presence of TiO2: is UV solar dose an appropriate parameter for standardization of water solar disinfection? Solar Energy 77(5), 635-648.

Rochelle-Newall, E., Nguyen, T.M.H., Le, T.P.Q., Sengteheuanghoung, O. and Ribolzi, O. (2015) A short review of fecal indicator bacteria in tropical aquatic ecosystems: knowledge gaps and future directions. Frontiers in Microbiology 6.

Roguet, A., Eren, A.M., Newton, R.J. and McLellan, S.L. (2018) Fecal source identification using random forest. Microbiome 6(1), 185.

Rosencranz, J., Cuddington, K., Brook, M., Koops, M.A. and Drake, D.A. (2021) Data-limited models to predict river temperatures for aquatic species at risk. Canadian Journal of Fisheries and Aquatic Sciences 78(9), 1268-1277.

Ross, T., Zhang, D. and McQuestin, O.J. (2008) Temperature governs the inactivation rate of vegetative bacteria under growth-preventing conditions. International Journal of Food Microbiology 128(1), 129-135.

Rousk, J., Frey, S.D. and Bååth, E. (2012) Temperature adaptation of bacterial communities in experimentally warmed forest soils. Global Change Biology 18(10), 3252-3258.

Samsudin, M.S., Azid, A., Khalit, S.I., Sani, M.S.A. and Lananan, F. (2019) Comparison of prediction model using spatial discriminant analysis for marine water quality index in mangrove estuarine zones. Marine Pollution Bulletin 141, 472-481.

Saunders, D. and Kalff, J. (2001) Nitrogen retention in wetlands, lakes and rivers. Hydrobiologia 443(1), 205-212.

Saxton, M.A., Naqvi, N.S., Rahman, F., Thompson, C.P., Chambers, R.M., Kaste, J.M. and Williamson, K.E. (2016) Site-specific environmental factors control bacterial and viral diversity in stormwater retention ponds. Aquatic Microbial Ecology 77(1), 23-36.

Searcy, R.T. and Boehm, A.B. (2021) A Day at the Beach: Enabling Coastal Water Quality Prediction with High-Frequency Sampling and Data-Driven Models. Environmental Science & Technology 55(3), 1908-1918.

Serrano, L. and DeLorenzo, M.E. (2008) Water quality and restoration in a coastal subdivision stormwater pond. Journal of Environmental Management 88(1), 43-52.

Sharif, M. and Burn, D.H. (2007) Improved K -Nearest Neighbor Weather Generating Model. Journal of Hydrologic Engineering 12(1), 42-51.

Sharma, A.K., Vezzaro, L., Birch, H., Arnbjerg-Nielsen, K. and Mikkelsen, P.S. (2016) Effect of climate change on stormwater runoff characteristics and treatment efficiencies of stormwater retention ponds: a case study from Denmark using TSS and Cu as indicator pollutants. SpringerPlus 5(1), 1984.

Šimek, K., Pernthaler, J., Weinbauer, M.G., Hornák, K., Dolan, J.R., Nedoma, J., Mašín, M. and Amann, R. (2001) Changes in Bacterial Community Composition and Dynamics and Viral Mortality Rates Associated with Enhanced Flagellate Grazing in a Mesoeutrophic Reservoir. Applied and Environmental Microbiology 67(6), 2723-2733.

Singh, K.P., Malik, A., Mohan, D. and Sinha, S. (2004) Multivariate statistical techniques for the evaluation of spatial and temporal variations in water quality of Gomti River (India)—a case study. Water Research 38(18), 3980-3992.

Smith, T.P., Thomas, T.J.H., García-Carreras, B., Sal, S., Yvon-Durocher, G., Bell, T. and Pawar, S. (2019) Community-level respiration of prokaryotic microbes may rise with global warming. Nature Communications 10(1), 5124.

Sønderup, M.J., Egemose, S., Hansen, A.S., Grudinina, A., Madsen, M.H. and Flindt, M.R. (2016) Factors affecting retention of nutrients and organic matter in stormwater ponds. Ecohydrology 9(5), 796-806.

Song, K., Winters, C., Xenopoulos, M.A., Marsalek, J. and Frost, P.C. (2017) Phosphorus cycling in urban aquatic ecosystems: connecting biological processes and water chemistry to sediment P fractions in urban stormwater management ponds. Biogeochemistry 132(1), 203-212.

Song, Y., Du, X. and Ye, X. (2019) Analysis of Potential Risks Associated with Urban Stormwater Quality for Managed Aquifer Recharge. International Journal of Environmental Research and Public Health 16(17), 3121.

Stajkowski, S., Laleva, A., Farghaly, H., Bonakdari, H. and Gharabaghi, B. (2021) Modelling dry-weather temperature profiles in urban stormwater management ponds. Journal of Hydrology 598, 126206.

Stephens, D.B., Miller, M., Moore, S.J., Umstot, T. and Salvato, D.J. (2012) Decentralized Groundwater Recharge Systems Using Roofwater and Stormwater Runoff1. JAWRA Journal of the American Water Resources Association 48(1), 134-144.

Stocker, M.D., Pachepsky, Y.A., Hill, R.L., Sellner, K.G., Macarisin, D. and Staver, K.W. (2019) Intraseasonal variation of E. coli and environmental covariates in two irrigation ponds in Maryland, USA. Science of the Total Environment 670, 732-740.

Su, T.-M., Yang, S.-C., Shih, S.-S. and Lee, H.-Y. (2009) Optimal design for hydraulic efficiency performance of free-water-surface constructed wetlands. Ecological Engineering 35(8), 1200-1207.

Sun, Z., Sokolova, E., Brittain, J.E., Saltveit, S.J., Rauch, S. and Meland, S. (2019) Impact of environmental factors on aquatic biodiversity in roadside stormwater ponds. Scientific Reports 9(1), 5994.

Sundui, B., Ramirez Calderon, O.A., Abdeldayem, O.M., Lázaro-Gil, J., Rene, E.R. and Sambuu, U. (2021) Applications of machine learning algorithms for biological wastewater treatment: Updates and perspectives. Clean Technologies and Environmental Policy 23(1), 127-143.

Suthar, M. (2019) Applying several machine learning approaches for prediction of unconfined compressive strength of stabilized pond ashes. Neural Computing and Applications, 1-10.

Tabachnick, B.G. and Fidell, L.S. (2013) Using multivariate statistics: International edition. Pearson2012.

Tala, F., Penna-Díaz, M.A., Luna-Jorquera, G., Rothäusler, E. and Thiel, M. (2017) Daily and seasonal changes of photobiological responses in floating bull kelp Durvillaea antarctica (Chamisso) Hariot (Fucales: Phaeophyceae). Phycologia 56(3), 271-283.

Tiefenthaler, L., Stein, E.D. and Schiff, K.C. (2010) Levels and patterns of fecal indicator bacteria in stormwater runoff from homogenous land use sites and urban watersheds. Journal of Water and Health 9(2), 279-290.

Timofeev, R. (2004) Classification and regression trees (CART) theory and applications. Humboldt University, Berlin, 1-40.

Tirpak, R.A., Afrooz, A.R.M.N., Winston, R.J., Valenca, R., Schiff, K. and Mohanty, S.K. (2021) Conventional and amended bioretention soil media for targeted pollutant treatment: A critical review to guide the state of the practice. Water Research 189, 116648.

Towler, E., Rajagopalan, B., Seidel, C. and Summers, R.S. (2009) Simulating Ensembles of Source Water Quality Using a K-Nearest Neighbor Resampling Approach. Environmental Science & Technology 43(5), 1407-1411.

Troitsky, B., Zhu, D.Z., Loewen, M., van Duin, B. and Mahmood, K. (2019) Nutrient processes and modeling in urban stormwater ponds and constructed wetlands. Canadian Water Resources Journal 44(3), 230-247.

Tunçsiper, B., Ayaz, S. and Akca, L. (2012) Coliform bacteria removal from septic wastewater in a pilot-scale combined constructed wetland system. Environmental Engineering and Management Journal 11, 1873-1879.

Valenca, R., Le, H., Zu, Y., Dittrich, T.M., Tsang, D.C.W., Datta, R., Sarkar, D. and Mohanty, S.K. (2021) Nitrate removal uncertainty in stormwater control measures: Is the design or climate a culprit? Water Research 190, 116781.

Valenca, R., Ramnath, K., Dittrich, T.M., Taylor, R.E. and Mohanty, S.K. (2020) Microbial quality of surface water and subsurface soil after wildfire. Water Research 175, 115672.

Valencia, A., Zhang, W. and Chang, N.-B. (2021) Integration of machine learning classifiers and higher order tensors for screening the optimal recipe of filter media in stormwater treatment. Science of the Total Environment 771, 145423.

Vander Meer, L., DeHeer, K., Mellinger, J., Gibes, S., Paasch, B., Wildschut, J., Miller, W.L., He, S.-Y. and DuBois, K.N. (2021) Indicator species characterization and removal in a detention pond in the Plaster Creek watershed. Journal of Environmental Management 298, 113503.

Vayssières, M.P., Plant, R.E. and Allen-Diaz, B.H. (2000) Classification trees: An alternative non-parametric approach for predicting species distributions. Journal of Vegetation Science 11(5), 679-694.

Vione, D. and Scozzaro, A. (2019) Photochemistry of Surface Fresh Waters in the Framework of Climate Change. Environmental Science & Technology 53(14), 7945-7963.

Walters, E., Graml, M., Behle, C., Müller, E. and Horn, H. (2013) Influence of Particle Association and Suspended Solids on UV Inactivation of Fecal Indicator Bacteria in an Urban River. Water, Air, & Soil Pollution 225(1), 1822.

Wang, C., Mao, G., Liao, K., Ben, W., Qiao, M., Bai, Y. and Qu, J. (2021) Machine learning approach identifies water sample source based on microbial abundance. Water Research, 117185.

Weng, L., Van Riemsdijk, W.H. and Hiemstra, T. (2012) Factors Controlling Phosphate Interaction with Iron Oxides. Journal of Environmental Quality 41(3), 628-635.

Westrich Jason, R., Ebling Alina, M., Landing William, M., Joyner Jessica, L., Kemp Keri, M., Griffin Dale, W. and Lipp Erin, K. (2016) Saharan dust nutrients promote Vibrio bloom formation in marine surface waters. Proceedings of the National Academy of Sciences 113(21), 5964-5969.

White, K., Dickson-Anderson, S., Majury, A., McDermott, K., Hynds, P., Brown, R.S. and Schuster-Wallace, C. (2021) Exploration of E. coli contamination drivers in private drinking water wells: An application of machine learning to a large, multivariable, geo-spatio-temporal dataset. Water Research 197, 117089.

Whitman, R.L., Przybyla-Kelly, K., Shively, D.A., Nevers, M.B. and Byappanahalli, M.N. (2008) Sunlight, season, snowmelt, storm, and source affect E. coli populations in an artificially ponded stream. Science of the Total Environment 390(2), 448-455.

Wilson, S.R., Close, M.E. and Abraham, P. (2018) Applying linear discriminant analysis to predict groundwater redox conditions conducive to denitrification. Journal of Hydrology 556, 611-624.

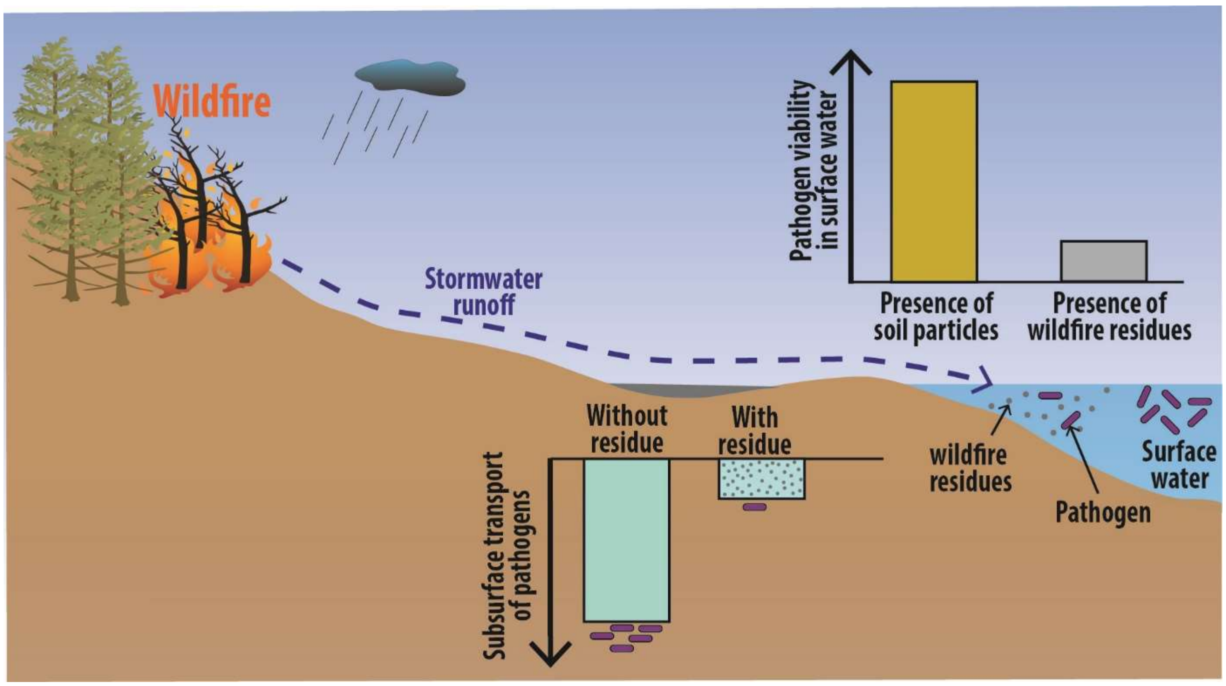
Yang, J. and Yang, J.-y. (2003) Why can LDA be performed in PCA transformed space? Pattern Recognition 36(2), 563-566.

Yaseen, Z.M., Sihag, P., Yusuf, B. and Al-Janabi, A.M.S. (2021) Modelling infiltration rates in permeable stormwater channels using soft computing techniques*. Irrigation and Drainage 70(1), 117-130.

Zanon, J.A., Favaretto, N., Democh Goularte, G., Dieckow, J. and Barth, G. (2020) Manure application at long-term in no-till: Effects on runoff, sediment and nutrients losses in high rainfall events. Agricultural Water Management 228, 105908.

Zhang, L., Seagren, E.A., Davis, A.P. and Karns, J.S. (2012) Effects of Temperature on Bacterial Transport and Destruction in Bioretention Media: Field and Laboratory Evaluations. Water Environment Research 84(6), 485-496.

4. CHAPTER 4 – MICROBIAL QUALITY OF SURFACE WATER AND SUBSURFACE SOIL AFTER WILDFIRE



Valenca, R., Ramnath, K., Dittrich, T. M., Taylor, R. E., & Mohanty, S. K. (2020). Microbial quality of surface water and subsurface soil after wildfire. Water Research, 175, 115672. https://doi.org/10.1016/j.watres.2020.115672

Abstract

Runoff from the wildfire affected areas typically carries high concentrations of fine burned residues or eroded sediment and deposits them in surface water bodies or on subsurface soils. Although the role of wildfire residues in increasing the concentration of chemical contaminants in both environments is known, whether and to what degree wildfire residues may affect microbial contaminants is poorly understood. To examine the effect of wildfire residues on growth and die-off of *Escherichia coli* (*E. coli)* —a pathogen indicator, we mixed stormwater *E. coli* contaminated stormwater and suspended particles from the pre- and post-wildfire area in batch reactors and monitored *E. coli* concentration. *E. coli* grew initially in the presence of particles, but the relative *E. coli* concentration was 10 times lower in the presence of wildfire residues than in natural soil from unaffected areas. Wildfire residues also decreased the persistence of *E. coli* during a 15-day incubation period. These results indicate that the growth or persistence of *E. coli* in surface water in the presence of wildfire residues was lesser than that in the presence of unburned soil particles, potentially due to depletion of nutrient concentration and loss of viability of bacteria in the presence of wildfire residues. To examine the transport potential of wildfire residues and their ability to facilitate the transport of *E. coli* in the subsurface system, suspension containing wildfire residues and/or *E. coli* were injected through unsaturated sand columns—a model subsurface system. Transport of wildfire residues in sand columns increased with decreases in the depth and increases in the concentration of particles, but increased transport of wildfire residues did not result in the transport of *E. coli*, suggesting wildfire residues did not facilitated transport of *E. coli*. Overall, the results indicate that wildfire residues may not increase the risk of the microbial contamination of surface water or groundwater via subsurface infiltration.

4.1. Introduction

Wildfire frequency is likely to increase by more than 15% based on a 2000-2050 meteorology prediction (Huang et al. 2015). Wildfire removes vegetation, increases soil hydrophobicity, and reduces infiltration, thereby increasing the volume of stormwater runoff (Rodrigues et al. 2019). Furthermore, compared with pre-fire runoff, post-fire runoff could contain 1000 times more suspended particles and contaminants including traces of metals, nutrients, total suspended solids, and polycyclic aromatic hydrocarbons (Burke et al. 2013), and it could affect the water quality of receiving water bodies and affect aquatic species (Silva et al. 2016). Although numerous studies have examined the effect of wildfire on transport of chemical contaminants to surface waters (Burke et al. 2013, Earl and Blinn 2003, Hernandez et al. 1997, Ilstedt et al. 2003, Stein et al. 2012, Tsai et al. 2019), no study to date has examined the effect of wildfire on microbial contamination of surface water and groundwater via subsurface infiltration of stormwater containing wildfire residues and pathogen.

Post wildfire, runoff from wildfire affected area can carry wildfire residues and deposit them in surface waters or on subsurface, from where they can infiltrate into groundwater (Figure 4.1). Thus, wildfire residues could mix with pathogens present in surface waters or subsurface environment and affect their fate in these systems. The fate of pathogens in surface water depends on water chemistry (Wang et al. 2019), sunlight exposure (Nelson et al. 2018), and the presence of particles, which may protect pathogens from inactivation by sunlight exposure (Bohrerova and Linden 2006) or provide nutrients (Chua et al. 2009) for microbial growth. On the other hand, particles may release chemicals that are toxic to bacteria and kill them. Although wildfire events shift the microbial community in soil (Fultz et al. 2016), it is not clear whether wildfire residues could increase or decrease the survival of pathogens in surface waters. Wildfire residues primarily consist of burnt biomass such as ash, black carbon or charcoal, and soil minerals. All these particles have different chemical properties (Bodi et al. 2014, Preston et al. 2017), which can affect the growth or decay of pathogen in water. Bacteria can colonize on black carbon and form biofilm, which could protect them from disinfectants (Lechevallier et al. 1984). Small suspended solids (< 20 μm) might induce the agglomeration of bacteria by acting as a condensation nucleus offering protection from antibacterial effects (Henao et al. 2018). Wildfire residues can also change the nutrient concentration in water. Thus, wildfire residues could either help increase or decrease the viability of pathogen in surface water based on the chemical composition of water containing wildfire residues.

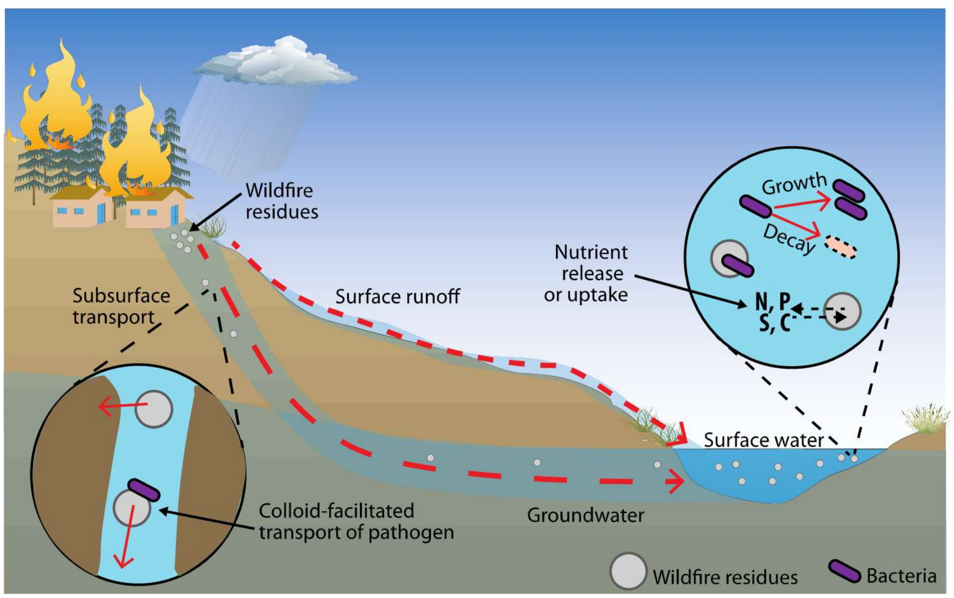


Figure 4.1 – Illustration of potential routes for the transport of wildfire residues in environments and their impact on microbial water quality.

Wildfire residues, like natural soil colloids, could be transported through subsurface and facilitate the transport of pathogens to groundwater. Microbial contamination of groundwater has been associated with heavy rainfall events, particularly in the beginning of the wet season (Wu et al. 2016). Under this condition, the concentration of *E. coli* and coliform bacteria in groundwater could increase, partly due to particle-facilitated transport of bacteria (Zeleznik et al. 2011). The same process could be relevant after wildfire, where the concentration of particles in the runoff increases by orders of magnitude due to the presence of wildfire residues. For colloid-facilitated transport to be important in the subsurface environment, wildfire residues or colloids must be transported through the subsurface, and pathogens must remain attached and viable on wildfire residues. However, little is known about the effect of wildfire residues on growth or decay of pathogens in water.

Conditions that may affect the infiltration of pathogens through subsurface soils include soil chemical properties (Clark and Pitt 2007), infiltration rate (Mohanty and Boehm 2014), and pH or chemical composition of infiltrating water (Pitt et al. 1999). Suspended particle type and concentration (AbuSharar and Salameh 1995) are critical in determining the relevance of the facilitated transport of pathogens in subsurface soil. Post wildfire, the concentration of particles in runoff increases due to an increase in erosion (Lee et al. 2016). An increase in particle concentration could decrease pathogen removal in the subsurface (Muirhead et al. 2006), thereby facilitating infiltration through the subsurface (Fries et al. 2006, Jeng et al. 2005). Particles may also be filtered through subsurface soils. Thus, it is important to compare the effect of wildfire residues with that of natural soil particles to help determine the fate and transport of pathogens in surface water and subsurface environment, so that the effect of wildfire on the microbial quality of surface waters and groundwater can be assessed.

This study examines the effect of wildfire particles on bacterial viability and their transport through the subsurface. We hypothesized that the microbial transport through the subsurface will increase in the presence of particles, but the transport will depend on particle type due to the bacteria-particle association. Residues from wildfire regions will decrease the viability of pathogens in surface waters due to the depletion of nutrients in wildfire-eroded soils. To test these hypotheses, bacteria-laden stormwater was injected at varying particle concentrations and subsurface depths, and the effluent was monitored for *E. coli* – a pathogen indicator – and total particle concentration. The viability of *E. coli* in stormwater was monitored over time in the presence of unburned soil, wildfire residues and biochar, a surrogate for black carbon generated during a wildfire.

4.2. Material and Methods

4.2.1. Stormwater collection

Stormwater was collected using 20-L carboys from Ballona Creek located in Los Angeles, CA (34 0’36’’ N 118 23’29’’ W) The stormwater from urban area could have very different composition from stormwater from forest watershed or other type of catchment. Nevertheless, the stormwater provides a natural water matrix for use in the study to examine the fate of *E. coli* when the stormwater is mixed runoff from fire affected area. The stormwater was kept at least 24 hours to settle large particles, and the supernatant was transferred into 1-L glass containers and autoclaved at 121 °C for 45 minutes. The sterile stormwater was kept at 4°C before being used in the experiment. We autoclaved the water to kill native microorganisms so that they don't compete for the available nutrients with the added *E. coli*. Although autoclaving water may change the nutrient composition of stormwater, the same water was used for all the study to ensure comparison between results.

4.2.2. Post-wildfire residues collection

For the control study, the soil was collected from the Ballona Wetlands (33.9709, -118.4367), where there has been no fire occurrence in the last several decades. After the Woolsey

Fire in November 2018, recently burned soil with wildfire residues were collected from Corral Canyon Park (34.0389, 118.7300) and from Malibu Lagoon (34.0347, -118.6828). The sampling locations were chosen based on the stormwater runoff route. The samples were collected from the top 10 cm of soil using a sterilized spatula and stored at 4°C. Biochar particles (Biochar Supreme, Everson, WA) were used as a control for black carbon without soil.

4.2.3. Characterization of wildfire residues

The postfire residues were characterized using Nuclear Magnetic Resonance (NMR) spectroscopy. Briefly, the ^{13}C Cross Polarization with Magic-Angle Spinning NMR spectrum was acquired on a Bruker AV-III HD 600 NMR spectrometer at a frequency of 150.9 MHz. The fine wildfire residue was mix thoroughly, and a homogenous wildfire sample weighing 33.7 mg was packed in a 3.2mm (outside diameter) zirconia rotor with a Vespel cap. A total of 59,657 scans were acquired with a sample spinning rate of 10 kHz using a variable amplitude cross-polarization sequence with a contact time of 2 ms and a recycle delay of 1 second. The data was processed with 50 Hz of line broadening.

To analyze the nutrient concentration leached from soil samples, 4.0 g of different particle types were suspended into 40 mL of Milli-Q water using a 50 mL centrifuge tube, and the solution was shaken (Wrist Action Shaker, Burrel Scientific) for 24 hours. The particles were removed from the supernatant by centrifugation (5,000 g for 15 min), and the water chemistry of the supernatant was analyzed for nutrients (nitrate, nitrite, and phosphate), dissolved organic carbon, and total nitrogen (Table 4.1).

Table 4.1 – Nutrient concentration leached from different particle type and real stormwater.

	Concentration ($mg\ L^{-1}$)					
	Nitrite	**Nitrate**	**Sulfate**	**Phosphate**	**Dissolved Organic Carbon**	**Total Nitrogen**
Unburned soil	0.00	27.09	83.99	6.45	50.57	9.07
Biochar particles	4.05	5.43	11.70	15.54	6.84	1.40
Wildfire #1	2.76	17.13	6.47	4.93	27.75	5.09
Wildfire #2	0.00	4.40	0.00	16.18	107.80	9.63
Wildfire #3	0.00	0.00	108.70	0.00	20.28	2.52
Autoclaved stormwater	0.00	8.23	0.00	0.00	3.66	1.41

4.2.4. Suspended particles solution preparation

To prepare suspended wildfire residues, samples were first sieved to remove particulates larger than 45 µm, and 2 g of the sieved sample was suspended in 1 L deionized water. The suspension was placed in an ice-bath and sonicated using a probe (Branson Digital Sonifier) to enhance the dispersion of particles for 15 minutes (on for 1.0 s, off for 3.0 s). The suspension was transferred into a 500-mL graduated cylinder, and particles with a size greater than 10 µm were settled based on Stokes Law (**Equation 4.1**). Particles with size lower than 10 µm were isolated for transport study because larger particles have limited potential for subsurface transport due to filtration and gravitational settling and are expected to be deposited on the surface. 200 mL of the suspension was transferred into 50 mL centrifuge tubes, centrifuged at 4,000 g for 15 minutes, and 45 mL of the supernatant was discarded, leaving behind 5 mL of concentrated particle suspension. The stock suspension was shaken by hand and then sonicated for 1 min prior to its use in the experiments. The particle size distribution of influent and effluent samples containing suspended solids was determined by analyzing 1.0 mL of the solution using a Particle Sizing Analyzer System (AccuSizer Model 770, Particle Sizing Systems), which determines the concentration of particles per mL and the diameter of each particle ranging from 0.55 µm to 500 µm.

$$v = \frac{2}{9}\frac{(\rho_p - \rho_f)}{\mu} g R^2$$ **Equation 4.1**

4.2.5. *E. coli* K-12 suspension

Suspension of *E. coli* K-12 with resistance to kanamycin (CAS: 25389-94-0, Fisher BioReagents) was prepared following the method described in a previous study (Mohanty and Boehm 2014). Although growth of bacteria could vary with strains (Foppen et al. 2010), and stormwater contains a wide range of bacterial strain, we used this particular stain to eliminate growth of environmental *E. coli* or potential contamination from natural dust during experiment. Briefly, a single colony of *E. coli* was grown in Luria-Bertani growth media (LB Agar, Miller, Fisher BioReagents), and the *E. coli* was separated from the media by centrifugation to remove the supernatant and washed with a phosphate-buffered saline (PBS) solution. The *E. coli* stock solution was added to stormwater containing particles to achieve the desired final concentration (10^3 to 10^5 CFU mL^{-1}). The range used in this study is within expected concentration of *E. coli* in stormwater or surface waters (Grebel et al. 2013). For the column experiments, the suspension was mixed for 120 minutes using an automated shaker to ensure attachment of bacteria on particles (Vasiliadou and Chrysikopoulos 2011).

4.2.6. Growth and decay of *E. coli* in the presence of post-wildfire residues

To examine if the presence of wildfire residues affects the growth and die-off of *E. coli* in the stormwater, 50 mL of autoclaved stormwater spiked with 10^3 CFU mL^{-1} of *E. coli* and 100 mg L^{-1} of suspended particles from different origins (a control soil, 3 soils with wildfire residues, or biochar) were mixed at 150 rpm in 200 mL glass flasks at 37°C for 15 days. To identify the growth and die-off of *E. coli* in stormwater without particles, the experiment was repeated without particles. To monitor any change in concentration of *E. coli*, 500 μL samples were pipetted and analyzed for *E. coli* at the following time intervals: 0.3, 0.8, 1, 2, 4, 7, 9, 11, and 15 days. Bacteria concentration was analyzed by inoculating 50 μL of the sample into LB agar plates with kanamycin, following spread plate and counting techniques (2 plates per sample). When the concentration was expected to be high to count, the PBS solution was used to dilute the sample to achieve bacteria counts between 30-300 CFU per plate. However, sample with low concentration was not concentrated due to low sample volume, and the resulting low colony count below 30 was included to estimate the concentration.

4.2.7. Sand columns as a model for subsurface

Sand filters were used as a model to examine if the wildfire residues could migrate through the subsurface into groundwater. A coarse sand (20-30 Standard Sand, Certified MTP) with size between 0.6 to 0.85 mm was used in this study to examine the worst-case condition for subsurface infiltration. Sand was washed using deionized water for 10 minutes, soaked in 1M HCl solution for 6 hours and then washed multiple times with Milli-Q water until the pH was near neutral. PVC pipes (2.0 cm diameter and 35 cm height) were used as columns. A screen (100 μm pore size) was placed at the bottom of the column before packing to prevent the sand particles from being washed off with the effluent. Columns were packed with sand in 15 g intervals to ensure they were packed uniformly. Deionized water was applied on the top of the sand surfaces at 9.0 mL min^{-1} for 4 hours using a peristaltic pump (Masterflex L/S Digital Drive, Cole Parmer) in order to equilibrate the flow and wash out any small sand colloids generated during packing. Details about the pore volume (PV) estimation by the bromide tracer are provided in the Supplementary Material (Figure 4.2 and Table 4.2).

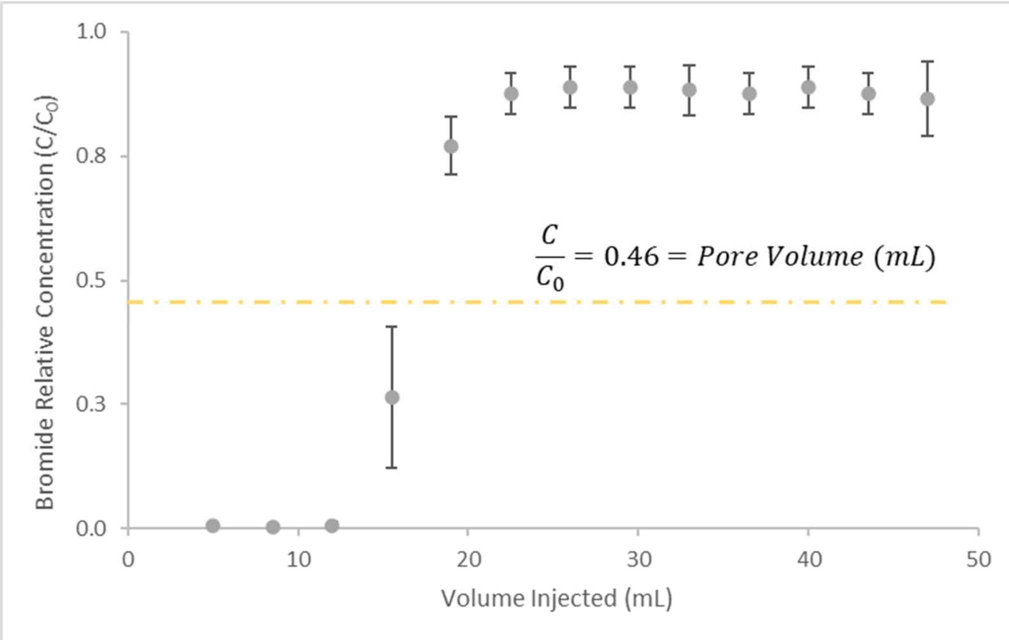


Figure 4.2 – Breakthrough curve based off bromide injection to determine pore volume of 20-cm sand column. PV = 16.7 ± 0.6 mL.

Table 4.2 – Pore volume of columns with different depths.

Sand Mass (g)	Column Depth (cm)	Pore Volume (mL)
44.0	10	8.4
66.0	15	12.5
88.0	20	16.7
110.0	25	20.9
132.0	30	25.1

4.2.8. Effect of sand filters depth and suspended particle concentration on particle removal

To examine the effect of subsurface depth on particle removal, duplicated sand columns with different heights (10, 15, 20, 25, and 30 cm) were assembled, and autoclaved stormwater water was constantly injected at 9.0 mL min^{-1}. To simulate pulse input, 1.0 mL of suspension containing control (unburned) soil (4.0 g L^{-1}) was injected on the top of the column using a pipette controller. 1.0 mL is sufficiently high to detect effluent concentration and low to prevent temporary ponding layer on filter layer, which could increase the flow rate and affect transport of particles or bacteria. Ten effluent samples were collected at the bottom of the column every 0.3 PV using 15-mL centrifuge tubes. The injection of suspended solids was repeated 5 times per column.

To investigate the change on particle removal due to particle type and concentration, duplicated sand columns with 20-cm depth were used, and suspensions of control soil and biochar particles were created at different concentrations: 0.01, 0.05, 1.0, 2.0, 3.0 and 4.0 g L^{-1}. The concentration range represents the concentration of particles measured in stormwater (Huey and Meyer 2010). 1.0 mL of each particle concentration solution was injected per column, and samples were collected every 0.3 PV at the bottom of the column. Each injection was repeated 5 times per column. The volume and particle concentration of samples were measured in order to calculate the total mass of solids removed during the injection.

4.2.9. Transport of *E. coli* and suspended particles through sand columns

To examine the transport of bacteria with and without wildfire residues, 1 mL of suspension containing 10^5 CFU mL^{-1} of *E. coli* with 2.0 g L^{-1} of particles of either type was injected on top of the column using a pipette controller, while deionized water was continuously injected at 9.0 mL min^{-1} using a peristaltic pump. Effluent samples were collected at the bottom of the column. Influent and effluent samples were analyzed for volume and bacteria and particle concentration in order to calculate the mass balance for each contaminant during the infiltration process.

4.2.10. Water sample analysis

The pH of the solutions used for column and batch experiments was measured using an Ion-Selective Electrode (Fisher Scientific #9107BN), and the concentration of particles was measured using a spectrophotometer (PerkinElmer Lambda 365 UV-Visible Spectrophotometer) based on absorbance at 890nm. The high wavelength is typically used for turbidity measurement (Mohanty et al. 2015) because the absorbance by color (dissolved organic carbon) is insignificant at high wavelength. Calibration curves were used for unburned and burned particles to accurately estimate the particle concentration based on the absorbance (Figure 4.3 and Figure 4.4). The concentration of nutrients (nitrate, nitrite, and phosphate) was analyzed using an Ion Chromatography (Dionex™ Integrion™ HPIC™ System, ThermoFisher). The concentration of dissolved organic carbon, total nitrogen, and total organic carbon was analyzed using a Total Organic Carbon Analyzer (TOC-L, Shimadzu).

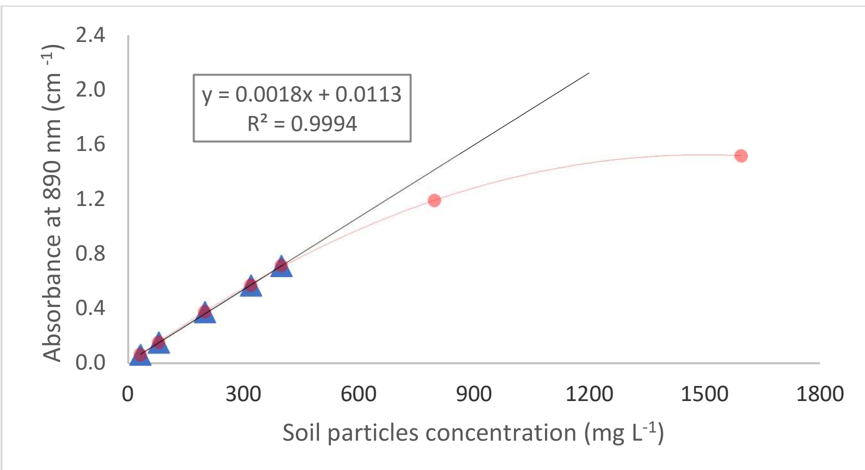


Figure 4.3 – Calibration graph used to measure soil particles concentration in water based on absorbance at 890nm.

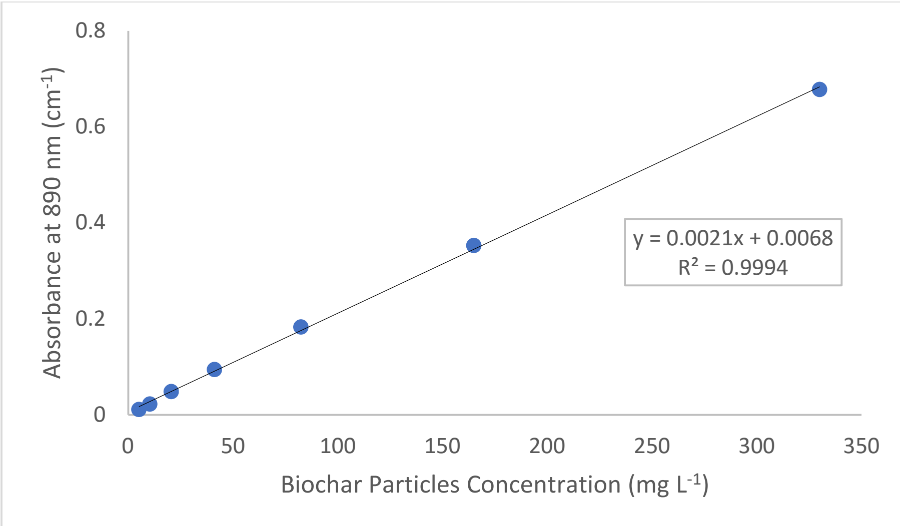


Figure 4.4 – Calibration graph used to measure biochar particles concentration in water based on absorbance at 890nm.

4.2.11. Data and statistical analysis

The bacteria concentration in samples was calculated by multiplying the average of colonies counted in two plates and presented as colony forming units (CFU) per mL. The relative concentration of bacteria during batch experiments was determined by calculating the ratio of the *E. coli* concentration (C) in the sample and the initial *E. coli* concentration (C_0). Total removal (R) of suspended solids through column experiments was calculated as $R = 1 - \frac{\sum C_e V_e}{C_i V_i}$ (%), where C = particle concentration (mg L^{-1}), V = volume (mL), i = influent and e = effluent. Statistical analysis was conducted using R (version 3.5.3).

4.3. Results

4.3.1. Growth and die-off of fecal bacteria is affected by particle types

Nutrient leaching results (Table 4.1) showed that more nutrients were leached from unburned soil than wildfire residues or fire depletes the nutrient availability in soil. NMR analysis of burned residues (Figure 4.5 and Figure 4.6) of wildfire residues confirmed these changes:

presence of aliphatic (0-50 ppm), substituted aliphatic (50-110 ppm), aromatic/substituted aromatic (110-165 ppm), and carboxylic and carbonyl (165-215 ppm).

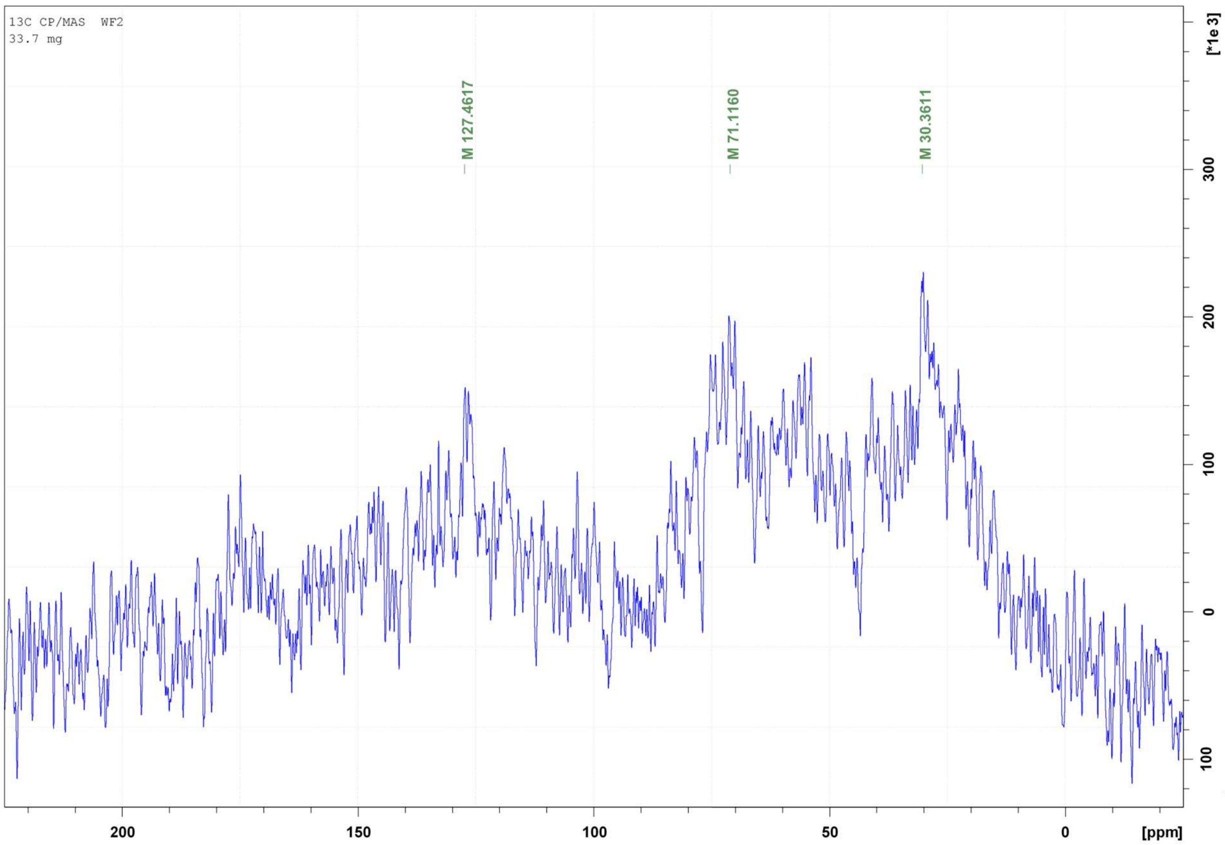


Figure 4.5 – NMR spectrum from wildfire residue sample (Wildfire #2). Mass analyzed: 33.7 mg.

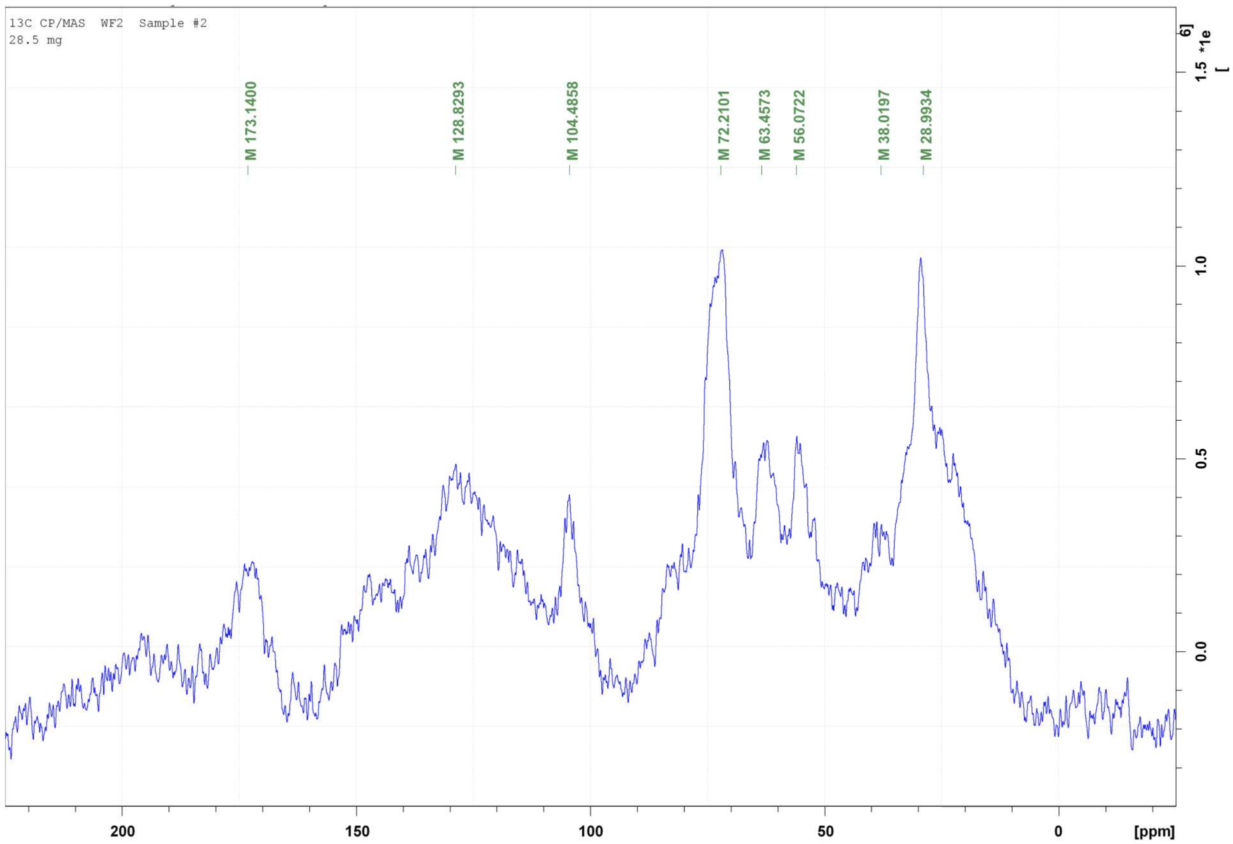


Figure 4.6 – NMR spectrum from wildfire residue sample (Wildfire #2). Mass analyzed: 28.5 mg.

E. coli in stormwater grew in the presence of particles, but the extent of growth varied with particle origin (Figure 4.7). Irrespective of particle types, *E. coli* concentration increased for 2 – 3 days (growth phase) and remained constant (stationary phase) for an additional 1 – 7 days based on particle types before a decrease in concentration indicating the die-off phase. The lag, growth and stationary phases (Table 4.3) of *E. coli* were determined following a method described elsewhere (Buchanan et al. 1997). The die-off phase was stipulated as the total concentration of bacteria started decreasing.

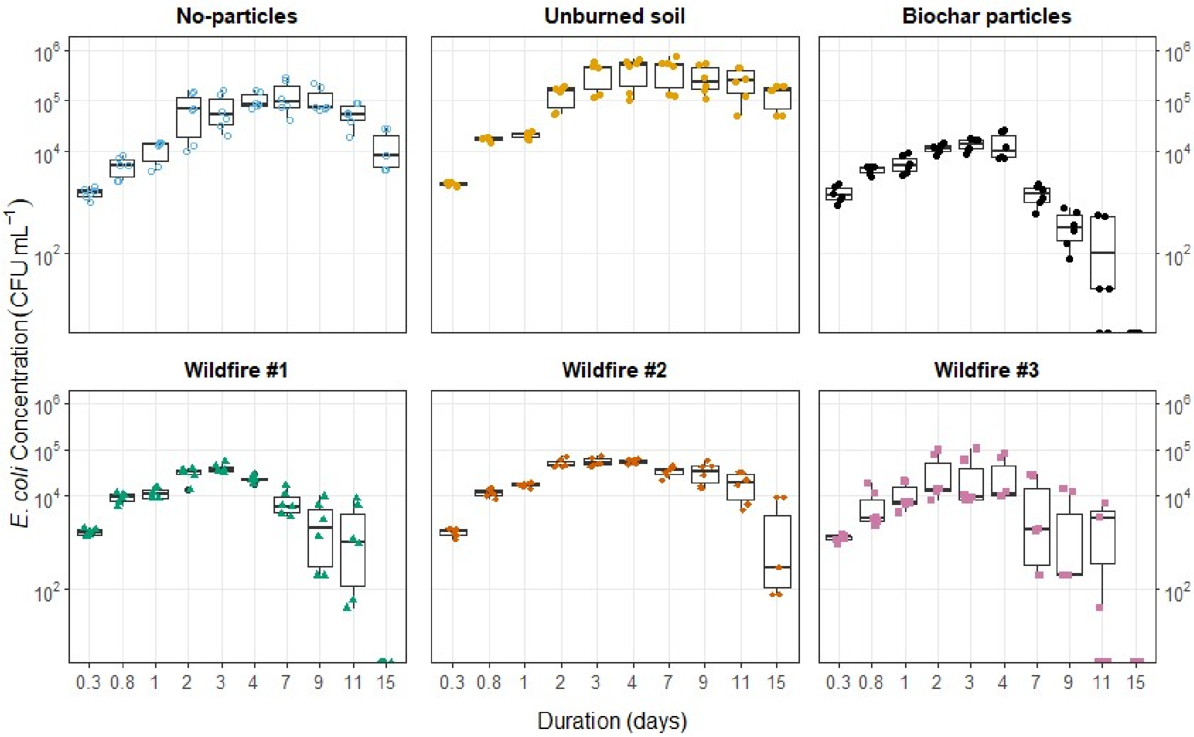


Figure 4.7 – Growth and die-off of *E. coli* in the stormwater without and with soil particles from wildfire affected areas, unburned soil, and biochar particles. Initial *E. coli* concentration was ~ 10^3 CFU mL^{-1}. Box-plot represents the concentration of *E. coli* in triplicated batches, with duplicate measurements per time point (n = 6). The detection limit is 20 CFU mL^{-1}, and the limit of statistically significant quantification on agar plate was 30 CFU on plate, which corresponds to 600 CFU mL^{-1}.

The extent to which the concentration increased initially or decreased after the stationary phase depended on the particle origin or type. In the absence of added particles in the stormwater, *E. coli* grew to 79 times its initial concentration by the end of 7 days; whereas in the presence of unburned soil particles, *E. coli* grew faster: the concentration increased by a factor of 250 by the end of 7 days. However, in the presence of wildfire residues or biochar particles, *E. coli* grew only by 20 – 30 times, which is nearly 10 times less than that observed in the presence of unburned soil particles. Additionally, the growth phase of *E. coli* was shorter in the presence of wildfire residues compared with unburned soil: in the presence of wildfire residues, the *E. coli* concentration started to decrease after 4 – 7 days, compared to 11 days in the presence of unburned soil particles. While there is no significance difference (P = .578) between black carbon particles and wildfire residues regarding *E. coli* concentration between day 4 and 11, *E. coli* concentration was significantly different (P <<< .05) in presence of unburned soil particles compared to wildfire residues in the same time period. After 11 days, the concentration of *E. coli* was drastically lowered in the presence of wildfire residues (1 – 10 times its initial concentration), but the *E. coli* concentration remained high in the presence of unburned soil particles: the concentration remained 147 times the initial concentration. Furthermore, the survival rate of *E. coli* was lower in the presence of wildfire residues than in unburned soil particles after 15 days of incubation. After 15 days, the *E. coli* concentration was below the detection limit when wildfire residues and biochar particles were present, but the concentration of *E. coli* remained high (77 times the initial concentration) in the presence of unburned soil particles after 15 days.

Table 4.3 – Phases of *E. coli* growth curve determined based on *E. coli* concentration in the presence of different particle types using a model (Buchanan et al. 1997). Statistical analysis shows significance difference on data compared to "No-particles" results. * P values between 0.05 and 0.01, ** P values between 0.01 and 0.005, *** P values below 0.005.

	Approximated Phase Duration (days)			
Particle Type	**Lag**	**Growth**	**Stationary**	**Die-off**
No-particles	0.3	3.7	5	6+
Unburned soil	0.3**	2.7*	8***	4+***
Biochar particles	0.3	2.7**	1***	11***
Wildfire #1	0.3	2.7	1***	11***
Wildfire #2	0.3	1.7	5***	8**
Wildfire #3	0.3	1.7	5***	8***

4.3.2. Removal of suspended solids depends on the subsurface depth

Suspended particles from unburned soil were removed during infiltration through sand filters, but the removal decreased with a decrease in the filter media depth (Figure 4.8). The depth of filter media was negatively correlated (Pearson correlation coefficient, $r = -0.940$) to effluent peak concentration, but positively correlated (Spearman correlation $\rho = 1$) to the removal of suspended particles. The trend is similar for *E. coli* without the presence of particles (Figure 3b), although removal of *E. coli* (also a type of particle) was consistently higher than soil particles, indicating greater adsorption or filtration of soil particles compared with *E. coli*. A three times increase in filter media depth decreased the peak height by half. An addition of a 1 cm sand layer increased the removal of suspended solids by 1.9%. Increases in concentration of influent particle did not change the concentration of effluent particles but shifted the distribution (Figure 4.9): When the influent concentration was high, a greater number of larger particles were passed through the sand filters.

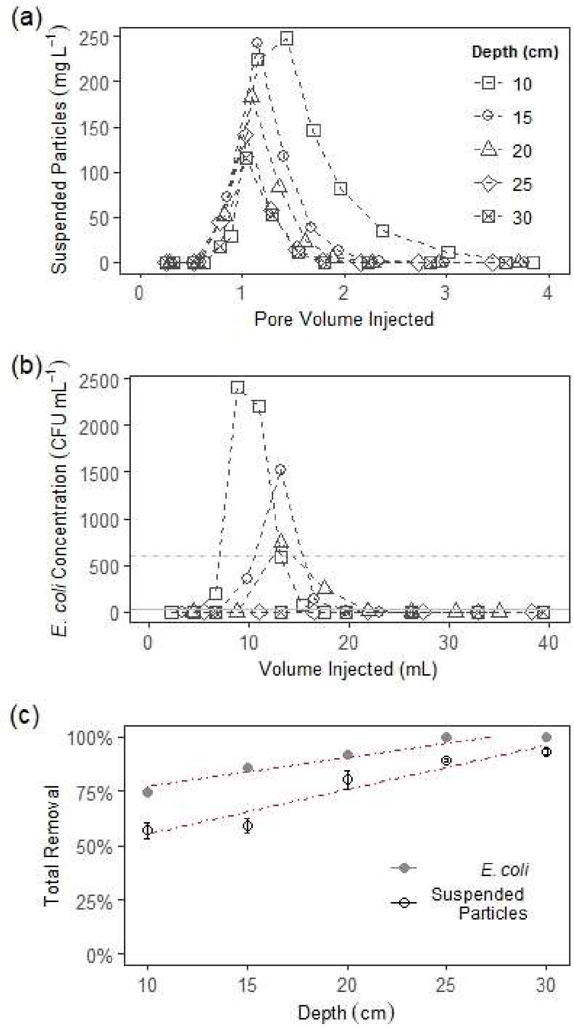


Figure 4.8 – Effect of sand filter depth on the removal of suspended soil particles when 1.0 mL of particle suspension (4.0 g L^{-1}) was spiked on sand column receiving particle-free water at 9.0 mL min-1. (a) Suspended solid concentration peak decreased, and the centroid of peak appeared earlier with an increase in sand filter depth. (b) Decrease in bacterial transport with increase in sand filter depth. The solid line indicates detection limit (1 CFU on plate or 20 CFU mL^{-1}), whereas gray dashed line indicates quantification limit with statistical certainty (30 CFU on plate or 600 CFU mL^{-1}). (c) The removal (n = 10) of suspended particles and bacteria increased with increases in sand filter depth (Spearman correlation $\rho = 1$).

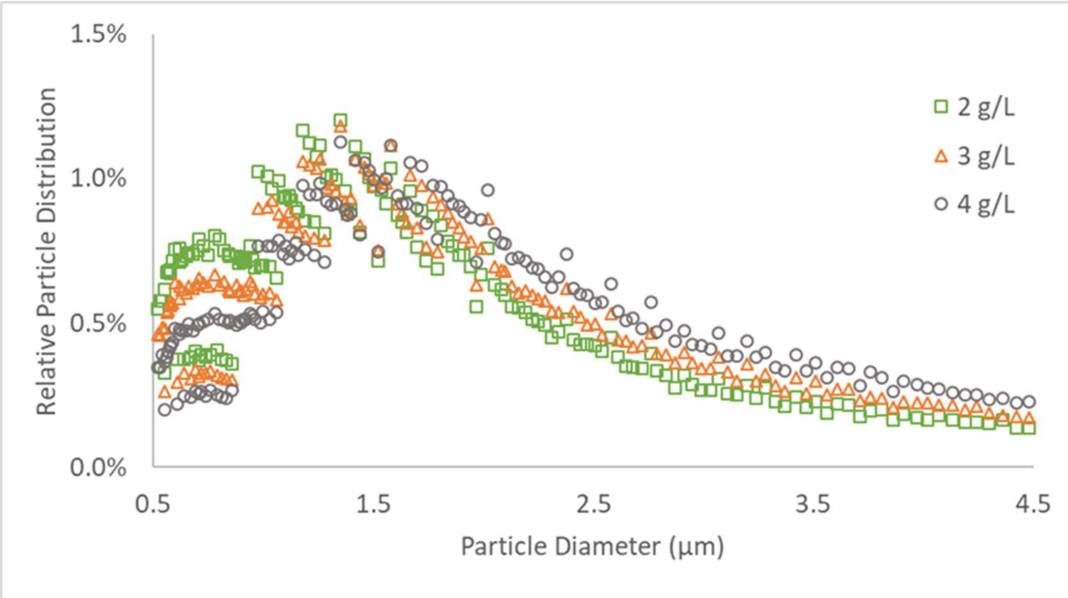


Figure 4.9 – Particle size distribution of effluents with varying particle concentration in the influent.

4.3.3. Particle type affects the removal of suspended particles

Removal of suspended particles depended on particle type and concentration, but colloid-facilitated transport of *E. coli* was not observed in this study despite the transport of particles through sand filters (Figure 4.10). Removal of suspended particles was 100% when particles concentration was below 0.7 g L^{-1} irrespective of the particle origin. However, particle removal decreased with increases in influent particle concentration above 0.7 g L^{-1}, and the removal rate depended on particle origin. For instance, while unburned soil was completely removed when solids concentration was 0.5 g L^{-1}, removal decreased to 62% when the suspended solids concentration increased to 3.0 g L^{-1}. Similarly, 100% of biochar particles were removed with suspended particles concentration of 0.5 g L^{-1}, but the removal slightly decreased to 96% when particles concentration was 2.9 g L^{-1}. When suspended particles concentration was above 2.0 g L^{-1}, sand filter removed biochar particles 10 times more efficiently than unburned soil. Removal of wildfire residues was closer to the removal of unburned soil rather than biochar particles. Although particle removal decreased with increases in particle concentration in the influent, *E. coli* were completely removed, suggesting colloid-facilitated transport of *E. coli* is unlikely in the presence of wildfire residues.

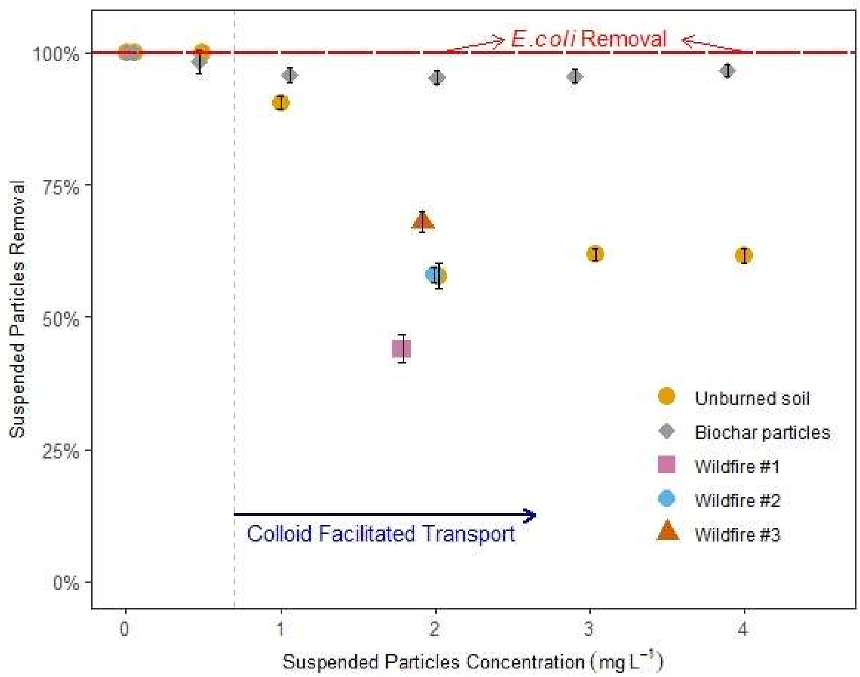


Figure 4.10 – Removal of particles of different origin and E. coli in 20-cm sand columns. Arrow mark represents region (right side of the vertical dashed line) where colloid-facilitated transport of bacteria is possible. The mean removal (n = 10) of particles varied with particle types and concentrations. Error bars represent standard deviation over mean. The horizontal red line indicates 100% removal of E. coli, owing to concentration of E. coli in the effluent below detection limit (20 CFU mL^{-1}). Thus, maximum removal that can be detected was 98% assuming input concentration is 1000 CFU mL^{-1}.

Sand columns removed most of the suspended particles irrespective of the particle origin, leaving only fine particles (diameter < 3 µm) to pass through the sand filter (Figure **4.11**). For all particle type analyzed, the mode of particle size in the influent was higher than that of effluent samples, indicating removal of particles by sand filter. For influent solutions, the mode of the particle size distribution varied from 6.53 µm for wildfire particle #1 to 15.08 µm for biochar particles, whereas the mode of the particle size distribution of effluent was smaller, ranging from 0.55 µm for biochar particles to 1.35 µm for natural soil and wildfire residues. An increase in particle concentration in the influent solution did not significantly (p = 0.14) affect the particle distribution in the effluent (Figure 4.9).

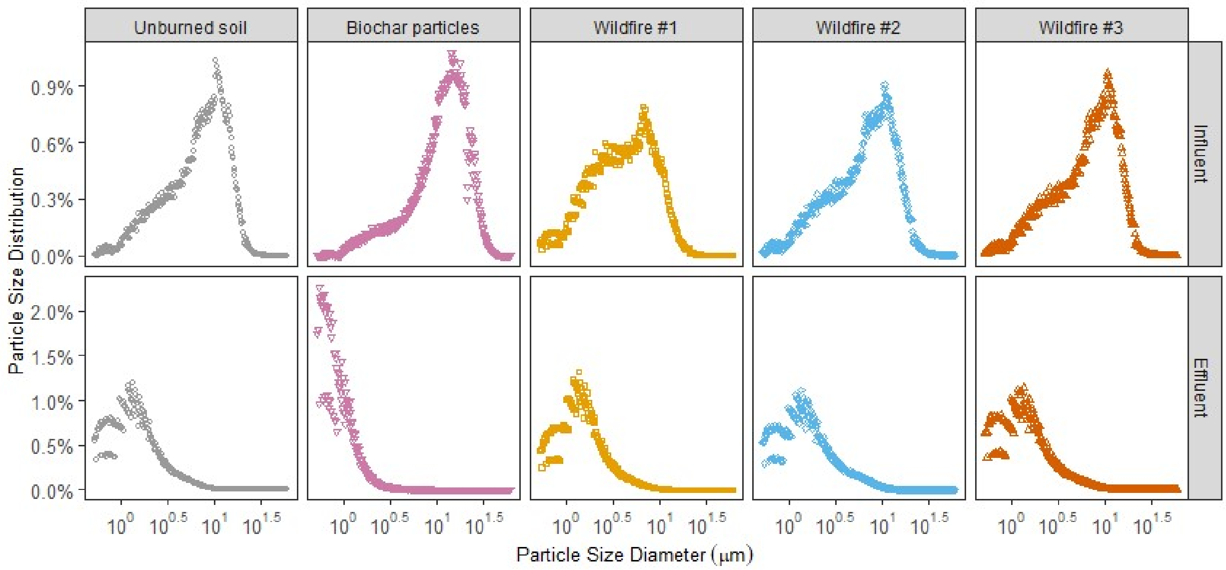


Figure 4.11 **– Size distribution of suspended particles in influent (top) and effluent (bottom) samples varied with particle origin. Particle concentration in influent samples was 2.0 g L^{-1}.**

4.4. Discussion

4.4.1. Presence of wildfire residues in surface water suppresses bacterial growth

Our results showed that the fate of fecal indicator bacteria in surface waters depended on not only the presence of suspended particles but also the source of particles or more particularly whether the soil contained wildfire residues. Wildfire residues suppressed bacteria growth and accelerated their die-off compared with unburned or unaffected soil. Natural soil particles typically contain organic matter and soil minerals, which can serve as a source of dissolved nutrients for bacteria (Friedrich et al. 1999). An alteration in nutrient concentration in water due to the presence of wildfire residue could be attributed to the observed change in microbial growth and persistence in this study. Similar results were observed in other studies with natural soil particles. For instance, increases in suspended particles content had been shown to increase nitrifying bacteria population in rivers (Xia et al. 2004) and phytoplankton growth in marine waters (Garzon-Garcia et al. 2018). However, wildfire residues are mostly composed by burned organic matter such as ash and char that has low nutrients such as carbon and nitrogen than soil (Homann et al. 2011, Ilstedt et al. 2003). Thus, decrease in nutrient leaching could suppress *E. coli* growth in our experiment. Wildfire residues including ash could also leach chemicals such as heavy metals that could be toxic to bacteria (Mitic et al. 2015). Mixing nitrate and phosphate to suspension containing similar concentration of wildfire, we observed negligible difference nutrient concentration after mixing, indicating adsorption of nutrients present in stormwater on wildfire residues has negligible effect on the result. One other possibility is that a wildfire residue or soil particle might attach multiple *E. coli* and make one colony on agar plate, thereby underpredicting the actual concentration of *E. coli*. This is particularly possible for biochar particles, which has higher adsorption capacity for *E. coli* than soil particles (Abit et al. 2012). In contrast to biochar, wildfire residues contain soil, ash, and a small quantity of black carbon. Thus, the resulting mixture would have lower affinity to *E. coli* than biochar. Thus, the result could vary with nature of burnt materials. Nevertheless, used agar-plate method to examine the effect of wildfire residues on viability of *E. coli*. Overall, the result indicates that the export of wildfire residues to surface water would not increase pathogen concentration more than it does due to the deposition of eroded soil.

4.4.2. Particle removal increases with increases in subsurface depth and lowers suspended particle concentration

Subsurface soil depth could vary from less than a meter to hundreds of meters. Thus, it is important to understand whether subsurface depth can influence potential groundwater contamination from wildfire residues. Injecting biochar or unburned soil residues (two extreme cases), we showed that increases in subsurface depth increased the removal of wildfire residues, but the removal decreased with increases in particle concentration. Increases in removal with increases in subsurface depth can be attributed to longer hydraulic retention time and increase in adsorption sites (Li and Davis 2008, Mitchell et al. 2011). However, the removal decreased with increases in particle concentration, potentially due to the exhaustion of attachment sites. When the influent particle concentration was 2.0 g L^{-1}, only 62% of unburned soil particles were removed, which is significantly lower than the removal of biochar particles (96%) under the same condition. The result indicates that burned residues have a stronger interaction with sand particles and are easier to remove in subsurface soils.

4.4.3. Subsurface removal of wildfire residues are similar to unburned soil rather than biochar

The removal of wildfire residues by sand filters was similar to that of unburned soil particles than biochar particles. At particle concentration higher than 0.7 g L^{-1}, the removal of biochar particles was around 96%, which is significantly higher than the removal of unburned soil and wildfire particles (44% to 68%). Biochar particles can serve as the nucleus of aggregation (Lehmann et al. 2011), forming larger colloids that are more likely to be removed than fine wildfire residues or soil particles. Furthermore, a change in surface properties of soil during wildfire could affect their removal. NMR analysis of wildfire residues confirmed the changes consistent with a previous study (Otto et al. 2006). The result suggests that a significant portion of wildfire residues contain black carbon and ash in addition to soil. Wildfire residues have been shown to have a high content of aromatic carbon, which decreases their polarity and increases their water repellency properties (Knicker et al. 2006). An increase in water repellency was previously attributed to increased removal of wildfire resides in sands (Goebel et al. 2013).

Removal of biochar particles was higher than that of wildfire residues suggesting biochar may not be a good surrogate to predict the transport of wildfire residues in subsurface soil. A difference is attributed to how both are formed under intense heat. Although wildfire residues and biochar are formed under similar temperature conditions (~ 800ºC), biochar is produced in the absence of oxygen while wildfire residues are formed in the presence of oxygen, resulting in higher ash content. This key difference in production conditions appears to affect their removal during infiltration through the subsurface.

4.4.4. Colloidal particles (< 3 μm) are poorly removed through subsurface infiltration

During subsurface infiltration, most particles regardless of their origin or types with size greater than 3 μm were removed. The particle size distribution of effluents indicates that finer particles were present in larger quantity when biochar was injected compared with unburned soil and wildfire residues. The effluent particle size range is similar to that of bacteria, which indicates that bacteria could move through the sand filter under the same conditions unless the interaction of bacteria with sand is stronger than the interaction of wildfire residues with sand. However, *E. coli* concentration in the effluent was below the detection limit in the effluent, indicating bacterial interaction with the sand surface was high. The presence of fine colloids in the effluent did not increase bacteria transport, suggesting colloid-facilitated transport of *E. coli* in the presence of wildfire residues is unlikely. In fact, colloid retarded transport of bacteria was observed in our study. Without soil colloids, bacteria removal in 10-cm columns was around 75%, which increased to near 100% with an increase in depth by 10 cm (Figure 4.12). In the presence of suspended particles and bacteria, the removal of bacteria in 20-cm sand columns remained at 100% irrespective of nature of particles. The results contradicted the result in the previous studies (Muirhead et al. 2006, Walters et al. 2013), which showed that *E. coli* predominantly attached to suspended solids with particle diameter lower than 12 or 20 μm. In our study, particles larger than 3 μm were filtered out on top of the filter layer, and the deposited particles could block pores or flow paths in sand layer, thereby increasing removal of suspended bacteria and colloid-associated bacteria. The particle size distribution analysis showed that biochar particles were more mobile than wildfire residues and unburned soil, and the relative size in the effluent for biochar particles was smaller than wildfire residues and unburned soil. Soils from affected and unaffected regions have similar particle size distribution, which suggests that the effluent might be dominated by soil minerals rather than burned black carbon that might be a small fraction of total mass. But the presence of these particles did not affect *E. coli* concentration in the effluent, suggesting their deposition in subsurface soil would not increase microbial risk.

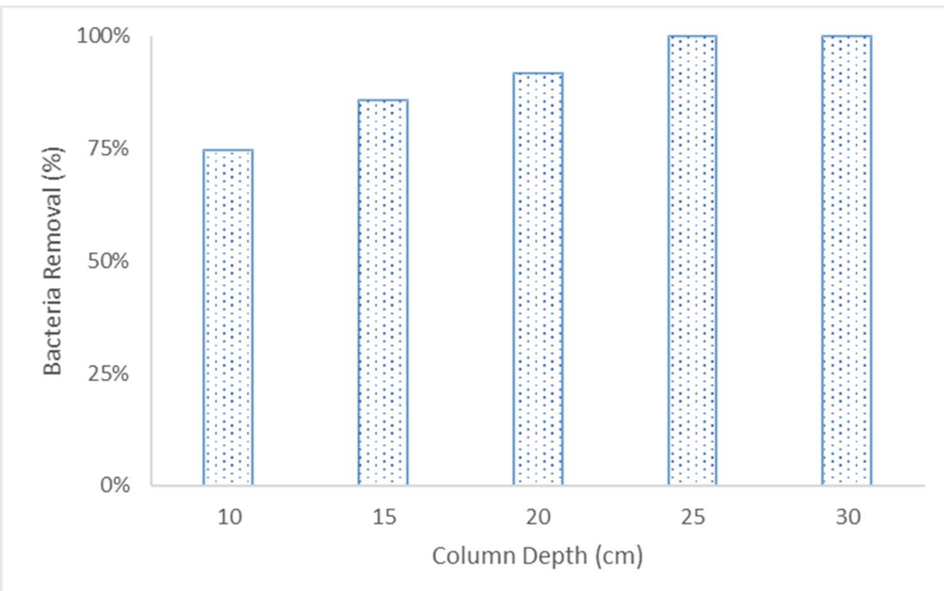


Figure 4.12 – Removal of bacteria among different column depths.

We used one strain of *E. coli.* The fate and transport behavior of *E. coli* could vary based on type of strains within species (Bolster et al. 2009) or by different types of pathogen species (Haznedaroglu et al. 2009). Thus, the result presented in this study could vary based on strain types. It should be noted that transport of virus in the presence of wildfire could be much higher, as virus is much smaller than bacteria and their removal by straining is minimal (Sasidharan et al.

2016). Thus, future studies should include virus and actual pathogens, instead of indicator bacteria used in this study.

4.5. Conclusion and Environmental Implications

The study answered the question of whether rainfall event following a wildfire would increase the risk of microbial contamination of surface waters, subsurface soil, and consequently groundwater. Specific conclusions are:

- The presence of wildfire residues in surface water reduces the growth of indicator bacteria and accelerates their die-off when compared to unburned soil, suggesting microbial risk post-wildfire is minimal.
- Wildfire residues have a limited effect on the transport of pathogen through subsurface soil, although transport of these particles increased when their concentration exceeded 0.7 g L^{-1}.
- Transport of biochar particles in subsurface soil was less than wildfire residues, indicating biochar may not be a good surrogate to study the transport of wildfire residues in subsurface soils.

This study is the first study to examine potential implication of wildfire residues on microbial water quality of receiving water bodies. The result shows that wildfire residue may not have any negative impact on microbial water quality because of decrease in subsurface transport and viability of indicator bacteria in surface water relative to natural soil particles. However, the result may have wide implications on other natural processes in nature. The result proves that wildfire residues can impair the growth of bacteria. Naturally, soil and water contain billions of non-pathogenic bacteria, which serve many ecosystem functions such as biodegradation of chemical pollutants and nutrient cycling. Thus, presence of wildfire residues could also have detrimental effect on these processes. Furthermore, wildfire residues and receiving water chemistry can vary based on the condition and sources. Thus, future studies should examine the effect of wildfire on the basis of different components such as ash type, black carbon, soil mineralogy.

4.6. References

Abit, S.M., Bolster, C.H., Cai, P. and Walker, S.L. (2012) Influence of feedstock and pyrolysis temperature of biochar amendments on transport of Escherichia coli in saturated and unsaturated soil. Environmental Science and Technology 46(15), 8097-8105.

AbuSharar, T.M. and Salameh, A.S. (1995) Reductions in hydraulic conductivity and infiltration rate in relation to aggregate stability and irrigation water turbidity. Agricultural Water Management 29(1), 53-62.

Bodi, M.B., Martin, D.A., Balfour, V.N., Santin, C., Doerr, S.H., Pereira, P., Cerda, A. and Mataix-Solera, J. (2014) Wild land fire ash: Production, composition and eco-hydro-geomorphic effects. Earth-Science Reviews 130, 103-127.

Bohrerova, Z. and Linden, K.G. (2006) Ultraviolet and chlorine disinfection of Mycobacterium in wastewater: Effect of aggregation. Water Environment Research 78(6), 565-571.

Bolster, C.H., Haznedaroglu, B.Z. and Walker, S.L. (2009) Diversity in Cell Properties and Transport Behavior among 12 Different Environmental Escherichia coli Isolates. Journal of Environmental Quality 38(2), 465-472.

Buchanan, R.L., Whiting, R.C. and Damert, W.C. (1997) When is simple good enough: a comparison of the Gompertz, Baranyi, and three-phase linear models for fitting bacterial growth curves. Food Microbiology 14(4), 313-326.

Burke, M.P., Hogue, T.S., Kinoshita, A.M., Barco, J., Wessel, C. and Stein, E.D. (2013) Pre- and post-fire pollutant loads in an urban fringe watershed in Southern California. Environmental Monitoring and Assessment 185(12), 10131-10145.

Chua, L.H.C., Lo, E.Y.M., Shuy, E.B. and Tan, S.B.K. (2009) Nutrients and suspended solids in dry weather and storm flows from a tropical catchment with various proportions of rural and urban land use. Journal of Environmental Management 90(11), 3635-3642.

Clark, S.E. and Pitt, R. (2007) Influencing factors and a proposed evaluation methodology for predicting groundwater contamination potential from stormwater infiltration activities. Water Environment Research 79(1), 29-36.

Earl, S.R. and Blinn, D.W. (2003) Effects of wildfire ash on water chemistry and biota in South-Western USA streams. Freshwater Biology 48(6), 1015-1030.

Foppen, J.W., Lutterodt, G., Roling, W.F.M. and Uhlenbrook, S. (2010) Towards understanding inter-strain attachment variations of Escherichia coli during transport in saturated quartz sand. Water Research 44(4), 1202-1212.

Friedrich, U., Schallenberg, M. and Holliger, C. (1999) Pelagic bacteria-particle interactions and community-specific growth rates in four lakes along a trophic gradient. Microbial Ecology 37(1), 49-61.

Fries, J.S., Characklis, G.W. and Noble, R.T. (2006) Attachment of fecal indicator bacteria to particles in the Neuse River Estuary, NC. Journal of Environmental Engineering-Asce 132(10), 1338-1345.

Fultz, L.M., Moore-Kucera, J., Dathe, J., Davinic, M., Perry, G., Wester, D., Schwilk, D.W. and Rideout-Hanzak, S. (2016) Forest wildfire and grassland prescribed fire effects on soil biogeochemical processes and microbial communities: Two case studies in the semi-arid Southwest. Applied Soil Ecology 99, 118-128.

Garzon-Garcia, A., Burton, J., Franklin, H.M., Moody, P.W., De Hayr, R.W. and Burford, M.A. (2018) Indicators of phytoplankton response to particulate nutrient bioavailability in fresh and marine waters of the Great Barrier Reef. Science of the Total Environment 636, 1416-1427.

Goebel, M.-O., Woche, S.K., Abraham, P.M., Schaumann, G.E. and Bachmann, J. (2013) Water repellency enhances the deposition of negatively charged hydrophilic colloids in a water-saturated sand matrix. Colloids and Surfaces A: Physicochemical and Engineering Aspects 431, 150-160.

Grebel, J.E., Mohanty, S.K., Torkelson, A.A., Boehm, A.B., Higgins, C.P., Maxwell, R.M., Nelson, K.L. and Sedlak, D.L. (2013) Engineered Infiltration Systems for Urban Stormwater Reclamation. Environmental Engineering Science 30(8), 437-454.

Haznedaroglu, B.Z., Kim, H.N., Bradford, S.A. and Walker, S.L. (2009) Relative Transport Behavior of Escherichia coli O157:H7 and Salmonella enterica Serovar Pullorum in Packed Bed Column Systems: Influence of Solution Chemistry and Cell Concentration. Environmental Science and Technology 43(6), 1838-1844.

Henao, L.D., Cascio, M., Turolla, A. and Antonelli, M. (2018) Effect of suspended solids on peracetic acid decay and bacterial inactivation kinetics: Experimental assessment and definition of predictive models. Science of the Total Environment 643, 936-945.

Hernandez, T., Garcia, C. and Reinhardt, I. (1997) Short-term effect of wildfire on the chemical, biochemical and microbiological properties of Mediterranean pine forest soils. Biology and Fertility of Soils 25(2), 109-116.

Homann, P.S., Bormann, B.T., Darbyshire, R.L. and Morrissette, B.A. (2011) Forest Soil Carbon and Nitrogen Losses Associated with Wildfire and Prescribed Fire. Soil Science Society of America Journal 75(5), 1926-1934.

Huang, Y.X., Wu, S.L. and Kaplan, J.O. (2015) Sensitivity of global wildfire occurrences to various factors in the context of global change. Atmospheric Environment 121, 86-92.

Huey, G.M. and Meyer, M.L. (2010) Turbidity as an Indicator of Water Quality in Diverse Watersheds of the Upper Pecos River Basin. Water 2(2), 273-284.

Ilstedt, U., Giesler, R., Nordgren, A. and Malmer, A. (2003) Changes in soil chemical and microbial properties after a wildfire in a tropical rainforest in Sabah, Malaysia. Soil Biology & Biochemistry 35(8), 1071-1078.

Jeng, H.A.C., Englande, A.J., Bakeer, R.M. and Bradford, H.B. (2005) Impact of urban stormwater runoff on estuarine environmental quality. Estuarine Coastal and Shelf Science 63(4), 513-526.

Knicker, H., Almendros, G., González-Vila, F.J., González-Pérez, J.A. and Polvillo, O. (2006) Characteristic alterations of quantity and quality of soil organic matter caused by forest fires in continental Mediterranean ecosystems: a solid-state 13C NMR study. European Journal of Soil Science 57(4), 558-569.

Lechevallier, M.W., Hassenauer, T.S., Camper, A.K. and McFeters, G.A. (1984) DISINFECTION OF BACTERIA ATTACHED TO GRANULAR ACTIVATED CARBON. Applied and Environmental Microbiology 48(5), 918-923.

Lee, C.S., Lee, Y.C. and Chiang, H.M. (2016) Abrupt state change of river water quality (turbidity): Effect of extreme rainfalls and typhoons. Science of the Total Environment 557, 91-101.

Lehmann, J., Rillig, M.C., Thies, J., Masiello, C.A., Hockaday, W.C. and Crowley, D. (2011) Biochar effects on soil biota – A review. Soil Biology and Biochemistry 43(9), 1812-1836.

Li, H. and Davis, A.P. (2008) Urban particle capture in bioretention media. I: Laboratory and field studies. Journal of Environmental Engineering-Asce 134(6), 409-418.

Mitchell, G.F., Riefler, R.G. and Russ, A. (2011) Removal, by Vegetated Biofilter, of Medium and Low Concentrations of Pollutants from Simulated Highway Runoff. Transportation Research Record (2262), 214-224.

Mitic, V.D., Stankov Jovanovic, V.P., Ilic, M.D. and Nikolic Mandic, S.D. (2015) Impact of wildfire on soil characteristics and some metal content in selected plants species of Geraniaceae family. Environmental Earth Sciences 73(8), 4581-4594.

Mohanty, S.K. and Boehm, A.B. (2014) Escherichia coli Removal in Biochar-Augmented Biofilter: Effect of Infiltration Rate, Initial Bacterial Concentration, Biochar Particle Size, and Presence of Compost. Environmental Science & Technology 48(19), 11535-11542.

Mohanty, S.K., Bulicek, M.C.D., Metge, D.W., Harvey, R.W., Ryan, J.N. and Boehm, A.B. (2015) Mobilization of microspheres from a fractured soil during intermittent infiltration events. Vadose Zone Journal 14(1), doi: 10.2136/vzj2014.2105.0058.

Muirhead, R.W., Collins, R.P. and Bremer, P.J. (2006) The association of E-coli and soil particles in overland flow. Water Science and Technology 54(3), 153-159.

Nelson, K.L., Boehm, A.B., Davies-Colley, R.J., Dodd, M.C., Kohn, T., Linden, K.G., Liu, Y., Maraccini, P.A., McNeill, K., Mitch, W.A., Nguyen, T.H., Parker, K.M., Rodriguez, R.A., Sassoubre, L.M., Silverman, A.I., Wigginton, K.R. and Zepp, R.G. (2018) Sunlight-mediated inactivation of health-relevant microorganisms in water: a review of mechanisms and modeling approaches. Environmental Science: Processes & Impacts 20(8), 1089-1122.

Otto, A., Gondokusumo, R. and Simpson, M.J. (2006) Characterization and quantification of biomarkers from biomass burning at a recent wildfire site in Northern Alberta, Canada. Applied Geochemistry 21(1), 166-183.

Pitt, R., Clark, S. and Field, R. (1999) Groundwater contamination potential from stormwater infiltration practices. Urban Water 1(3), 217-236.

Preston, C.M., Simard, M., Bergeron, Y., Bernard, G.M. and Wasylishen, R.E. (2017) Charcoal in Organic Horizon and Surface Mineral Soil in a Boreal Forest Fire Chronosequence of Western Quebec: Stocks, Depth Distribution, Chemical Properties and a Synthesis of Related Studies. Frontiers in Earth Science 5, 20.

Rodrigues, E.L., Jacobi, C.M. and Figueira, J.E.C. (2019) Wildfires and their impact on the water supply of a large neotropical metropolis: A simulation approach. Science of the Total Environment 651, 1261-1271.

Sasidharan, S., Torkzaban, S., Bradford, S.A., Kookana, R., Page, D. and Cook, P.G. (2016) Transport and retention of bacteria and viruses in biochar-amended sand. Science of the Total Environment 548, 100-109.

Silva, V., Abrantes, N., Costa, R., Keizer, J.J., Goncalves, F. and Pereira, J.L. (2016) Effects of ash-loaded post-fire runoff on the freshwater clam Corbicula fluminea. Ecological Engineering 90, 180-189.

Stein, E.D., Brown, J.S., Hogue, T.S., Burke, M.P. and Kinoshita, A. (2012) Stormwater contaminant loading following southern California wildfires. Environmental Toxicology and Chemistry 31(11), 2625-2638.

Tsai, K.P., Uzun, H., Chen, H., Karanfil, T. and Chow, A.T. (2019) Control wildfire-induced Microcystis aeruginosa blooms by copper sulfate: Trade-offs between reducing algal organic matter and promoting disinfection byproduct formation. Water Research 158, 227-236.

Vasiliadou, I.A. and Chrysikopoulos, C.V. (2011) Cotransport of Pseudomonas putida and kaolinite particles through water-saturated columns packed with glass beads. Water Resources Research 47, 14.

Walters, E., Graml, M., Behle, C., Müller, E. and Horn, H. (2013) Influence of Particle Association and Suspended Solids on UV Inactivation of Fecal Indicator Bacteria in an Urban River. Water, Air, & Soil Pollution 225(1), 1822.

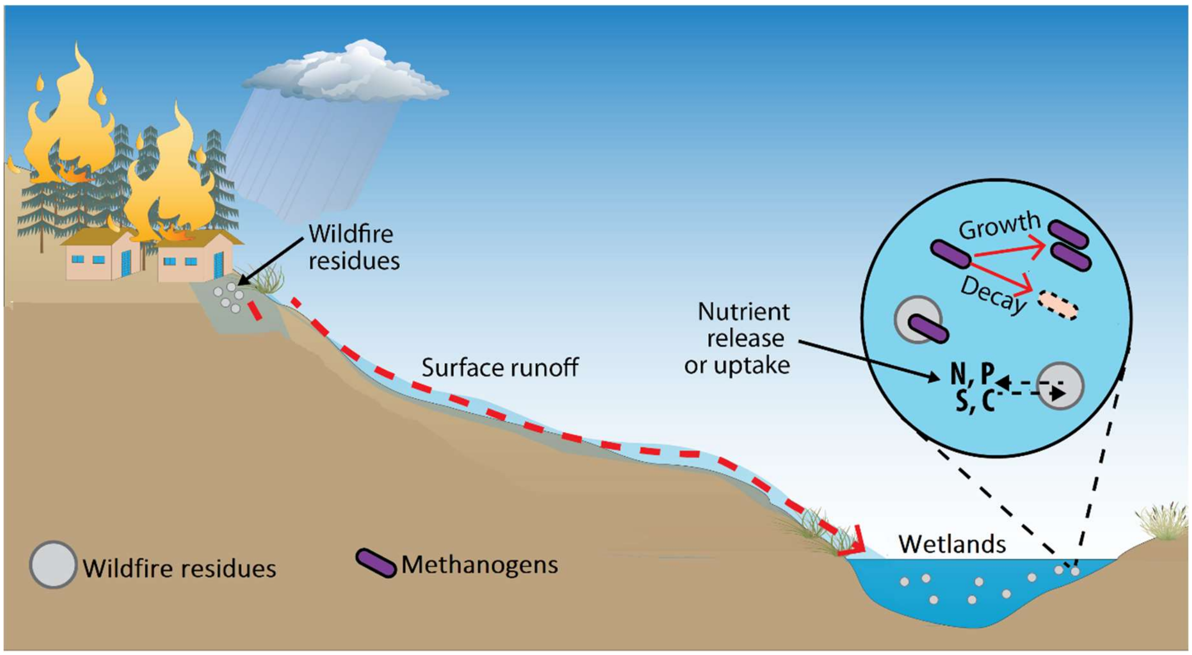
Wang, P., Zhao, J., Xiao, H.Y., Yang, W.J. and Yu, X.F. (2019) Bacterial community composition shaped by water chemistry and geographic distance in an anthropogenically disturbed river. Science of the Total Environment 655, 61-69.

Wu, J.Y., Yunus, M., Islam, M.S. and Emch, M. (2016) Influence of Climate Extremes and Land Use on Fecal Contamination of Shallow Tubewells in Bangladesh. Environmental Science & Technology 50(5), 2669-2676.

Xia, X.H., Yang, Z.F., Huang, G.H., Zhang, X.Q., Yu, H. and Rong, X. (2004) Nitrification in natural waters with high suspended-solid content - A study for the Yellow River. Chemosphere 57(8), 1017-1029.

Zeleznik, B.B., Curk, B.C. and Souvent, P. (2011) Water Quality: Current Trends and Expected Climate Change Impacts. Peters, N.E., Krysanova, V., Lepisto, A., Prasad, R., Thoms, M., Wilby, R. and Zandaryaa, S. (eds), pp. 101-+.

5. CHAPTER 5 – ENHANCED METHANE EMISSIONS FROM WETLANDS AFTER DEPOSITION OF WILDFIRE RESIDUES – IMPLICATIONS TO CLIMATE CHANGE



Citation: Valenca, R., McKnight, Q., Indiresan, S., Kwok, I. K., Mahendra, S., & Mohanty, S. K. (2022) Enhanced methane emissions from deposited wildfire residues in wetlands: implications to climate change. *In preparation*.

Abstract

Wetlands store over a third of the world's terrestrial carbon, but they also contribute to one-third of methane emissions. A critical balance between carbon storage and methane emission could affect global warming and its impact on wildfire. Many wetlands are located downstream of wildfire-affected areas that could deposit tons of wildfire residues annually and shift the critical carbon balance. Yet, no study to date has examined whether and how wildfire residue deposition could affect methane emissions in wetlands. We conducted a series of batch incubation experiments in the laboratory and in-situ in the field by adding wildfire residues to wetland sediments and monitored changes in methane emission, pore water chemistry, and microbial community. The results showed that the deposition of wildfire residues could increase methane emission in wetland sediments by up to 56%, but the emission depended on the amount of wildfire residues deposited. Mechanistic studies confirmed that increased methane emission is linked to the wildfire-induced production of hydrogen peroxide and increased concentration of silica, both of which could accelerate the breakdown of organic carbon, a precursor for methane production. Deposition of wildfire residue increased the pH of sediment initially, but the effect did not last long due to the buffer capacity of sediment, indicating an initial increase in pH did not inhibit long-term methane emission. Analysis of the microbiome showed that the presence of wildfire residues may affect the abundance of *mcrA* genes, possibly due to the effect of gene competition under a high nutrient environment. Overall, the results suggest that global methane production in wetland can increase following wildfires, thereby exacerbating global warming that causes an increase in wildfire frequency and intensity. This feedback process that could accelerate global warming and make wildfire season go from bad to worse.

5.1. Introduction

Wetlands are a critical part of the ecosystem and provide many functions including water treatment, nutrient recycling, carbon balance, and supporting biodiversity (Perron and Pick 2020). They store one-third of total terrestrial carbon and contribute to nearly 30% of methane emissions globally. Thus, they provide a critical balance to either mitigate or exacerbate greenhouse gas emissions, thereby affecting global warming (Nahlik and Fennessy 2016). Thus, disruption of the wetland ecosystem could alter the carbon balance. As many wetlands are located downstream of wildfire-prone areas, they could receive polluted runoff from wildfire-affected areas carrying the ash or burned residues created during wildfire (Garcia et al. 2021). These residues could increase the runoff pH (Mendez 2010), nutrients concentration (Basso et al. 2020), dissolved organic carbon (Uzun et al. 2020), and other contaminants including polycyclic aromatic hydrocarbons (PAHs) and heavy metals (Burton et al. 2016, Mansilha et al. 2019). Although some studies have examined the direct impact of fire on wetland plants and wetland functions (Li et al. 2020, Medvedeff et al. 2015), limited studies to date have examined the effect of wildfire residues on the functions of downstream wetlands.

Wetland sediments contain a rich microbiome including methanogenic archaea or methanogens (Nazaries et al. 2013) and provide anoxic conditions, which is ideal to break down the stored carbon and release them as methane– a powerful greenhouse gas (Narrowe et al. 2019). In fact, wetlands are projected to become the primary methane emitter by 2100 (Dean et al. 2018). Production of methane in wetlands can be impacted by the presence and type of substrate (Narrowe et al. 2019), pH (Ye et al. 2012), redox conditions (Angle et al. 2017), and nutrients (Ramsay et al. 2021) – all of which can change following the deposition of wildfire residues via the runoff from wildfire affected areas.

How and to what extent the presence of wildfire residues can affect methane production in wetlands remains unknown. Post-wildfire runoff typically contains a high concentration of nitrate (Basso et al. 2020), which could reduce methane production (Wenner et al. 2020). On the other hand, ash in post-wildfire runoff could induce the generation of hydroxyl radicals and hydrogen peroxide (Leonard et al. 2007) and accelerate the breaking down of complex carbon to acetate, a methane precursor, thereby favoring the methane production (Wormald and Humphreys 2019). Direct wetland fires could also affect the microbiome of this stormwater treatment system (Li et al. 2020). A previous study showed that methane emission from peatlands remained the same after a wildfire despite an increase in sulfate reducers (Belova et al. 2014). Unlike direct fire that burns the vegetation in wetland and affects the net biomass in wetland, deposition of wildfire residues would not affect the biomass loading. Yet, no study to date has examined the extent to which methane emission can be affected by the deposition of wildfire residues.

This study aims to quantify the methane production from wetland sediments when exposed to wildfire residues. We hypothesized that the deposition of wildfire residues in wetland sediments would increase methane emission due to the wildfire-induced hydrogen peroxide production and increased silica content. Laboratory and manipulative field experiments were conducted using natural wetlands sediments and wildfire residues to quantify the production of methane and identify the threshold amount of wildfire residues necessary to affect the production. In the field experiments, we tracked the abundance of methanogens when exposed to wildfire residues. Our results improve the understanding of how the deposition of wildfire residues may affect methane emission from wetlands.

5.2. Materials and Methods

5.2.1. Wildfire site and residue collection

We collected wildfire residues following a natural wildfire: the Pacific Palisades fire started on May 14, 2021, in the Topanga Canyon area (34.097764, -118.584847) in Los Angeles, USA, and was fully contained by May 26, 2021. The fire burned over 1,200 acres of land and damaged more than 700 structures. The local climate is classified as a warm-summer Mediterranean climate according to the *Köppen–Geiger* classification and the local vegetation is classified as chaparral and grassland. The wildfire residues samples were collected from the top 10 cm of soil using a sterilized spatula, sieved using a 2 mm opening sieve (#10 ASTM E11 standards) to remove large debris, and stored at 4 ºC. The wildfire residues samples were characterized for their pH, electrical conductivity, nutrient leaching, and heavy metals leaching following methods described elsewhere (Valenca et al. 2020).

5.2.2. Collection of wetland sediments and wetland water

Wetland sediment used in this study was collected from the Ballona Freshwater Marsh (33.970851, -118.431141) in Los Angeles, USA, which receives urban stormwater runoff and discharges the effluent into the Pacific Ocean. The marsh compromises nearly an area of 133,000 m^2 and was designed to treat stormwater from a 1,000-acre watershed based on a 1-year rainfall return period. There has been no fire occurrence in the last several decades in the area surrounding the marsh. The stormwater was collected using 5-L plastic bottles by submerging the bottle into the marsh. The wetland sediment from the Ballona Freshwater Marsh was collected from a location approximately 1.5 meters from the shoreline and 0.5 m below the water level. The sediment was placed in ziplock bags and transported in a cooler to the laboratory immediately to avoid microbial decay. The sediment was sieved using a sieve #10 (2 mm opening) to separate small particles (<

2.0 mm) from large particles (> 2.0 mm). In the experiment, we used only small particles (< 2.0 mm) because large particles contain large organic biomass (Supporting Information). The sediment was centrifuged in 50-mL sterile plastic tubes for 10 minutes at 5,300 rpm to separate from the supernatant and stored at 4 ºC. The sediment was used in laboratory experiments within 24 hours of the collection.

5.2.3. Batch studies to track methane emission

To analyze methane emission from wetland sediments, 30 g of wetland sediment was added to the 200-mL glass flasks with rubber caps using a sterilized spatula. 60 mL of stormwater from the wetland was added to the flask along with 3 mL of concentrated acetate solution (1.0 g L^{-1}), which served as the substrate for the methanogens. To examine the threshold amount of wildfire residues needed to cause an effect, different amounts of wildfire residues —0 (control), 0.1%, 1%, 5%, or 10% (by weight)— were added to batches. To create an anoxic condition, nitrogen gas was purged for 10 minutes using a syringe. The flasks were kept closed with rubber caps and a metal seal. Before placing the batch in the incubator, 15-mL of air was removed from the headspace of the flask using a syringe to ensure that there was enough space for methane gas to form inside the flask. The batches were mixed in an incubator at 120 RPM and 30 ºC. Methane concentration was assessed daily followed by a 5-minute nitrogen gas purge to ensure that all methane was removed, and anoxic conditions were kept constant.

5.2.4. Effect of silica and hydrogen peroxide on methane emission

To analyze if silica could interfere with methane emissions, we conducted batch experiments with wildfire residues or silica and compared the results to the control experiment. Unlike previous experiments, acetate was spiked only one time at day zero, and methane concentration was tracked for 21 days. This approach ensured that methanogens were active but created a substrate-limited environment with time so that the effect of silica can be deciphered. Briefly, we created triplicated batch experiments containing either 0.5 mg L^{-1} of silicic acid (H_4SiO_4) or 1.5 grams of wildfire residues, spiked acetated, tracked daily methane emission, and compared the results with the control experiments with no added silicic acid or wildfire residues. To analyze the effect of hydrogen peroxide on methane emission, we first quantified how much hydrogen peroxide was leached from wildfire residues by doing 24 hours leaching experiment. Briefly, 8 grams of wildfire residues were suspended in 40 mL of water and shacked by an automated machine (Wrist Action Shaker, Burrel Scientific) for 24 hours. 1.0 mL samples were collected at different times, diluted in 5.0 mL of DI water, centrifuged at 5,300 rpm for 5 minutes, and the supernatant was analyzed for hydrogen peroxide. Leaching experiments showed that each gram of wildfire residues may release nearly 75.7 ± 16.2 µmol of hydrogen peroxide (H_2O_2) when suspended in water (Figure S4). Thus, 5 grams of wildfire residues would release nearly 370 µmol of H_2O_2. To quantify the effect of H_2O_2 on methane emission, 370 µmol H_2O_2 was spiked into sediment without wildfire residues, and methane emission was assessed daily in the triplicated batch experiments.

5.2.5. Microbial community shift following wildfire residues deposition

In-situ field experiments were conducted at the Ballona Marsh to track the response of methanogens following the deposition of wildfire residues in field conditions. Using a 1-L bottle, nearly 100 g of wetland residues and 5 g of wildfire residues were suspended in 1 L of wetland stormwater. The bottles were closed with glass wool and submerged in the wetland inside mesh boxes (**Figure 5.1**). The glass wool permitted water exchange between the natural wetland and the bottle, but it trapped all sediments inside the bottle. The mesh boxes were used to keep the bottles in place and for an easy access to the experiment. The bottles were submerged on August 20th, 2021 and samples were collected after 2, 6, 10, and 30 weeks. Samples from wetland sediments were also collected on August 20th and served as the time zero of the experiment. The samples were transferred into a 50-mL centrifuge tube and kept at -80 ºC freezer for microbial community analysis.

Figure 5.1 – Picture of the experimental bottles that were submerged in the Ballona Wetland for 130 days. 6 identical bottles were used for the control experiment, and other 6 identical bottles were used for the experiment with wetland residues. The containers (control and wildfire) were submerged next to each other in the wetland.

5.2.6. Microbial community analysis in field and laboratory samples

To quantify the effect of added wildfire residues on methanogens, we extracted total nucleic acid from the sediments from batch and field studies. The previously frozen samples were retrieved from storage at –80 ºC and brought to room temperature. 0.5 g of each sample was used to perform nucleic acid extraction using a modified phenol-chloroform extraction as described in full elsewhere (Gedalanga et al. 2014). Briefly, cells (solid sample) were lysed chemically and mechanically by incubating at 65 ºC with lysis buffer, SDS, phenol, and zirconia-silica beads for 2 min followed by bead-beating for an additional 2 min. This was repeated with an 8-min incubation and 2-min bead-beating. The cell suspension was centrifuged for 5 minutes, and the supernatant was transferred to new DNase- and RNase-free tubes. Nucleic acids were extracted twice using a phenol-chloroform-alcohol extraction and with chloroform and alcohol. The samples were stored in isopropanol and sodium acetate at –20 ºC overnight to precipitate. The precipitate was then collected, purified, and measured for purity and concentration on a NanoDrop 2000C spectrophotometer. Following total nucleic acid quantification, DNA was removed using DNase, and the RNA alone was synthesized into cDNA, then quantified on the spectrophotometer. cDNA samples were then stored at –80 ºC.

The genes of interest selected were *mcrA*, or methyl coenzyme M reductase subunit A, as well as 16s rRNA to estimate abundance and *rpoD* as a housekeeping gene to estimate activity. The *mcrA* primers selected were ME1F and ME2R, while the 16S rRNA primers selected were 783F and 984R (Hales et al. 1996). qPCR was run using DNA to quantify the abundance of methanogens, and cDNA to quantify the expression of methanogenic genes. qPCR was run using a reaction mixture of primers, SYBR green, BSA, and the DNA and cDNA samples. Each sample for control and wildfire was amplified using real-time quantitative polymerase chain reactions (qPCR) conducted on a StepOnePlus thermocycler (Life Technologies, Carlsbad, CA) in triplicates along with controls. A standard curve was made by serially diluting nucleic acids extracted from a pure culture of methanogenic archaea, *Methanosarcina acetivorans*. No-template negative controls were also included to verify there was no extraneous contamination or residual nucleic acid. qPCR was run with 10x master mix (SYBR Green), 0.3 µM primer, 10 ng RNA template, and 0.2 µg mL^{-1} bovine serum albumin (BSA) to mitigate inhibition in environmental samples. For *mcrA*, qPCR was run for 30 cycles of 94 ºC for 40 s, 50 ºC for 1.5 min, 72 ºC for 3 min, and a final extension step at 72 ºC for 10 min.

5.2.7. Water sample and methane analysis

The pH of the solutions was measured using an Ion-Selective Electrode (Fisher Scientific #9107BN). Acetate concentration was assessed with Ion Chromatography (Dionex™ Integrion™ HPIC™ System, ThermoFisher). To determine methane in the air inside the laboratory batch experiments, a gas chromatography equipped with an array detector was used with the peak of detection around 1.3 min. Briefly, 0.5 mL of air sample was injected into the GC-FID where it was burned in the flame and emitted a spectrum that was detected at the array detector. To determine hydrogen peroxide concentration in samples, we followed methods described elsewhere (Tanner and Wong 1998). Briefly, a reagent was created by suspending 0.3 g of NH_4VO_3 and 1.3 g of 2,6-Pyridine dicarboxylic acid diluted in 150 mL of water and 15 mL of concentrated H_2SO_4. The concentration of H_2O_2 was analyzed by spiking 1.0 mL of reagent into 1.0 mL of the water sample, and the mixture was shacked manually for 1 min. Absorbance was scanned from 200nm to 600nm using a UV-spectrophotometer (PerkinElmer Lambda 365 UV-Visible Spectrophotometer), and the peak at 432nm was correlated with hydrogen peroxide concentration.

5.3. Results

5.3.1. Deposition of wildfire residues enhances methane emissions from wetland sediment

The presence of wildfire residues significantly ($p < 0.05$) increased methane emissions from wetland sediments after 23 days of the experiment (**Figure 5.2**). Within the first 24 hours of the experiment, the emission of methane in the batch containing 5% wildfire residues was 72% lower than in the control (0% residue). However, after the initial 24 hours, batches with wildfire residue constantly emitted higher daily methane than batches without residues. After 5 days of incubation, batches with wildfire residue emitted 1.34 ± 0.19 kg m^{-3} g^{-1} methane, which is more than twice that emitted from the batch without wildfire residue (0.66 ± 0.11 kg m^{-3} g^{-1}). After 23 days and four cycles (e.g., four spikes of acetate), wildfire-containing batches emitted 56% more methane than the control experiment.

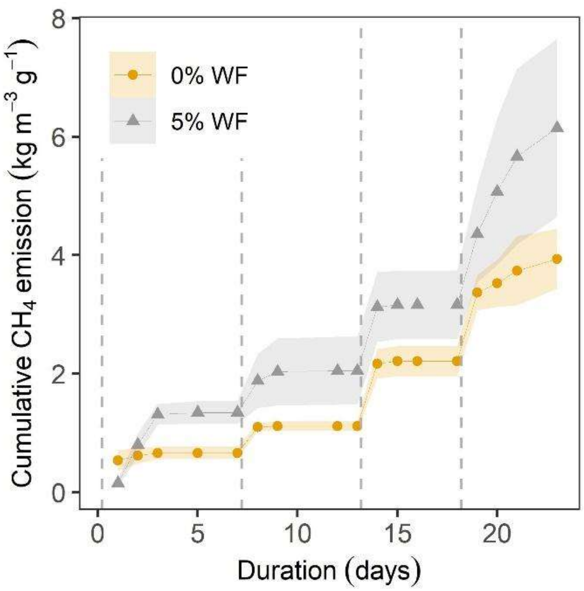


Figure 5.2 - Cumulative emission of methane from batch experiments in the presence and absence of wildfire (WF) residues. Vertical lines indicate a spike of acetate into the solution or starting of one cycle. Triplicated batch experiments were utilized for each experimental condition. Average number is reported, and shaded areas represent the standard deviation over mean of triplicated experiments.

5.3.2. Threshold amount of wildfire residues for enhanced methane emission

Increases in wildfire residues concentration enhanced methane emission from wetland sediments (**Figure 5.3).** For instance, an increase in 10 times wildfire residues concentration increased methane emission from 1.21 ± 0.22 kg m^{-3} g^{-1} to 1.70 ± 0.29 kg m^{-3} g^{-1}, resulting in a mean increase of 41%. However, compared to the control experiments, the increase of methane emission at low wildfire residues concentration is not statistically different ($p > 0.05$). Nonetheless, a high coefficient of determination ($R^2 = 0.89$) was calculated based on the results, which show that the presence of wildfire residues significantly affects the production of methane.

.

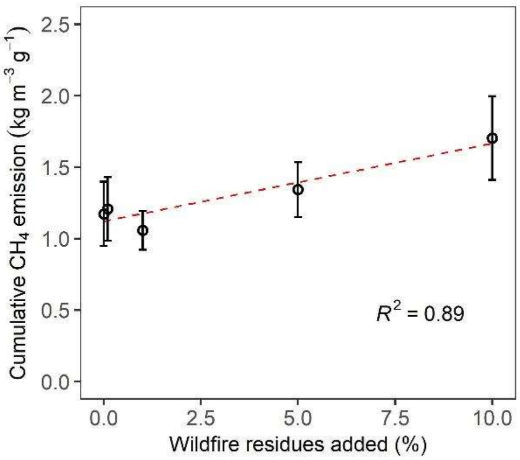


Figure 5.3 – Cumulative methane emission from wetland sediments after 5 days of experiments in the presence of wildfire residues. Triplicated batch experiments were conducted for each condition and the error bars represent the standard deviation. Wildfire residues addition was calculated as weight-by-weight. Dashed line represents the linear correlation and R value represents the Pearson correlation.

5.3.3. Wildfire-induced production of H_2O_2 and silica affects methane emissions

The presence of silica increased methane emission but to a lesser extent compared to the presence of wildfire residues (**Figure 5.4**). After 21 days, sediment with wildfire residues emitted 2 to 3 times more methane than the sediment with silica and control. Although silica batches appear to emit more methane (0.13 ± 0.05 kg CH4 m^{-3} g^{-1}) than control experiment (0.08 ± 0.05 kg CH_4 m^{-3} g^{-1}), the results are not statistically significant ($p > 0.05$). When hydrogen peroxide was added to the solution without the presence of acetate, methane production significantly ($p < 0.05$) increased compared to the control (e.g., without H_2O_2 or acetate). The addition of H_2O_2 increased methane emission by nearly 2-fold compared to control experiments (**Figure 5.5**).

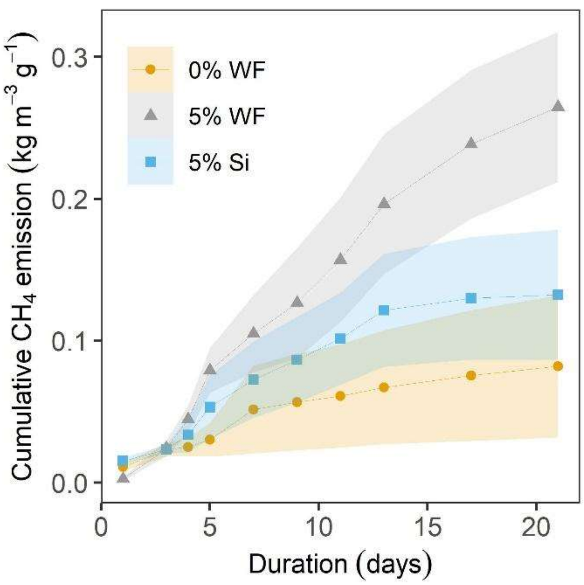


Figure 5.4 – Effect of wildfire residues and silica in the emission of methane from wetland sediments. Triplicated batch experiments were utilized for each experimental condition. Average number is reported, and shaded areas represent the standard deviation over mean of triplicated experiments.

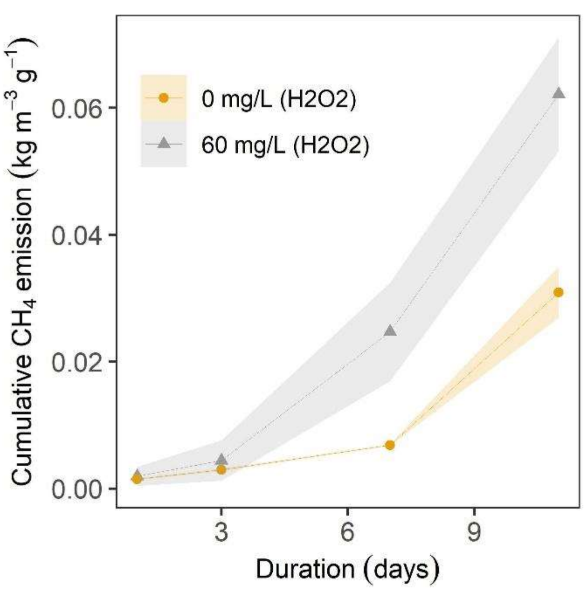


Figure 5.5 – Effect of H_2O_2 on methane production from wetland sediments. Duplicated batch experiments were utilized for each experimental condition. Average number is reported, and shaded areas represent the standard deviation over mean of triplicated experiments. 60 mg/L of H_2O_2 is 1.7 mM or 0.006% of H_2O_2.

5.3.4. Microbiome abundance and expression in field experiments

The microbial community results show that the addition of wildfire residues may affect the abundance and expression of genes in wetland sediments in the field experiment (**Figure 5.6**). The results show that total 16S rRNA increased initially in the presence of wildfire residues, but it was drastically reduced after 21 weeks of experiments. Compared to the control experiments, wetland sediments with presence of wildfire residues presented 2.5-log less 16S rRNA after 21 weeks. While *mcrA* abundance remained the same despite the presence or absence of wildfire residues, *mcrA* expression decreased after 1 week of experiment and remained low when wildfire residues were present. After 21 weeks, the expression of *mcrA* was 2.0-log higher in control experiments than wildfire residues containing experiments.

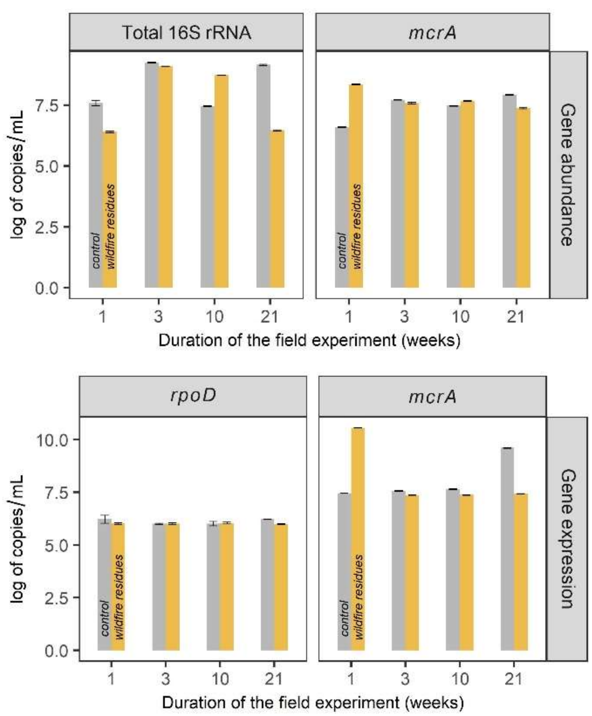


Figure 5.6 – Gene abundance and expression of Total 16S rRNA, *mcrA*, and *rpoD* that was performed along 21 weeks in the Ballona Wetland. Standard deviation represents triplicated measurements from the same sample.

5.4. Discussion

5.4.1. Wildfire residues may induce H_2O_2 formation and increase bioavailable organic carbon due to silica competition to active sites

Our results showed that wildfire residues deposition increased methane emissions from wetland sediments. Methanogens bacteria forms methane through anaerobic respiration using carbon monoxide as the final electron acceptor. This process, also known as methanogenesis, is the final step in the decay of organic matter. Therefore, the production of methane is enhanced by the break down of large and complex organic matter into smaller organic compounds. Our data proved that depositing wildfire residues to wetland sediments increased methane emission by more than 40% depending on the amount of wildfire residues added. We attributed the result to an increase in the concentration of hydrogen peroxide and silica in water, which could accelerate the decomposition of DOC and increases DOC concentration in the pore water, therefore facilitating methane production. In our study, the presence of hydrogen peroxide increased methane production by 3 times, possibly due to the accelerated the decomposition of organic matter. Similarly, our silica results showed that methane production increased by 1.5 times, possibly due to the competition for active sites between silicon, phosphorus, and organic carbon, which results in an increased organic matter in pore water. Although wildfires are known to increase the concentration of organic matter in riverbed (Son et al. 2015), another study showed that wildfires decrease the concentration of organic matter in upland artic streams (Rodríguez-Cardona et al. 2020), showing that local climate may also impact. In addition, a previous study showed that methane production may increase because wildfire residues contain a high amount of silicon (Maksimova and Abakumov 2014). The competition between silicon, phosphorus and DOC for binding sites results in an increased DOC in porewater, therefore favoring methane production (Reithmaier et al. 2017). Moreover, wildfire residues may generate hydrogen peroxide (H_2O_2)

which can break down complex chemical structures and hydrolyze them into simple/soluble compounds, resulting in an increase in soluble chemical oxygen demand which favors the methane emission (Perendeci et al. 2018). Although high concentrations of H_2O_2 may create inhibitory by-products and reduce methane emissions (Perendeci et al. 2018), each gram of our wildfire residues sample was generating nearly 20 ppm of H_2O_2 which is too low for this inhibition to occur.

5.4.2. Threshold concentration of wildfire residues

The increased methane emissions depended on the concentration of wildfire residues deposited in the wetland sediments. While the deposition of 0.1% and 1% (w/w) of wildfire residues did not affect methane emissions, our results showed that methane emissions was significantly ($p < 0.05$) higher when 5% or 10% (w/w) of wildfire residues were deposited in the wetland sediments. We explain our results in terms of the generation of chemicals (e.g., reactive oxygen species and silica content) as small amounts of wildfire residues would produce small amounts of these chemicals, therefore reducing the impact of these chemicals on methane production. Elevated levels of suspended solids and turbidity is often reported after wildfires (Hohner et al. 2017), achieving concentrations as high as 11,000 mg/L (Uzun et al. 2020). Assuming that the effective area of the Ballona Wetland is nearly 56,000 m^2 and that our study with 5% wildfire residues was depositing 1.5 g of wildfire residues per 0.002 m^2, it would take nearly 44×10^3 kg of wildfire residues to reach that threshold in the Ballona Wetland. With these assumptions, it would take only 4,400 m^3 of post-wildfire runoff to reach that threshold, which is a very small fraction of the previously recorded flow rate in the Ballona Creek (3.4×10^7 m^3) which is adjacent to the Ballona wetland (McPherson et al. 2005). Thus, the results reported here are realistic and most wetlands near wildfire-risk areas will easily reach that 5% threshold.

5.4.3. Enhanced emission of methane from sediment in the presence of wildfire residues was not explained by the abundance of *mcrA* genes

In the field experiments, it was found that the abundance and expression of *mcrA* was greater than the control in the first week of sampling. At 3, 10, and 21 weeks, *mcrA* abundance was slightly lower, higher, and lower than the control respectively, while expression showed a slight decrease at 3 and 10 weeks and a large drop at 21 weeks. The initial spike in abundance and activity of methanogens can be explained by methanogens being extremophilic archaea, who have historically survived in extreme environments such as those with high temperatures and salinity (Taubner et al. 2015). As such it would be more likely that they would be the initial survivors of environmental stress such as the deposition of wildfire residues, which greatly increases the electric conductivity of water. In addition, alkaliphilic methanogens, generally hydrogenotrophic, can survive in high pH caused by wildfire residues (Wormald et al. 2020). However, rapid microbial succession is primarily driven by Proteobacteria and Bacteroidetes (Ma et al. 2020) which may explain why the relative abundance of methanogens trended down over the course of the field experiment. The variability in the abundance of *mcrA* can be caused by local weather conditions (Ma et al. 2012), biogeochemistry (Freitag Thomas and Prosser James 2009), and/or global warming (Wang et al. 2021). Previous studies have also shown that the abundance and activity of methanogens in peatlands vary seasonally. In acidic bogs, methanogenic archaea are primarily hydrogenotrophs and tend to increase their relative abundance during the winter, while in fens, they use acetate and dominate during the spring (Sun et al. 2012). It was found that the greatest potential for methanogenesis occurred in July in all peatlands, correlating with peak plant growth. From August through October, it was found that organic compounds released by plant roots could be oxidized to acetate, stimulating the presence of methanogens in peatlands where acetate-utilizing methanogens dominate. In addition, methanogens are able to use dead plant matter year-round (Sun et al. 2012). In addition, cold temperatures may disfavor methanogenic activity while methanogenic substrates accumulate, allowing an increase in methanogenesis once temperatures rise (Sun et al. 2012). Our results are in accordance with a previous study that showed that the addition of nitrogen to soil reduces the abundance of *mcrA* and decreases methane uptake by soil due to an imbalance between methanotrophs and methanogens (Hu et al. 2021). Wildfire residues (or wildfire ash) are rich in nutrients and can significantly increase the concentration of N in the soil. However, these results are counter intuitive as the addition of wildfire residues increased methane emissions. According to a previous study, an increase in methane emission has weak correlation with increased *mcrA* genes in soil (Freitag Thomas and Prosser James 2009), which is similar to our results. In contrast, another study showed that biochar – a type of black carbon – can increase the concentration *mcrA* depending on the type of biochar's feedstock (Yuan et al. 2018). In fact, wildfire residues or black ash can enhance the abundance of genes that are involved in the Calvin cycle as ash can reduce dissimilatory nitrate reduction to ammonia (DNRA), nitrate assimilation and nitrification, but increase denitrification (Zhang et al. 2021). Because wildfire residues may promote the growth of other types of bacteria including *E. coli* (Valenca et al. 2020), sulfate reducing bacteria could compete with methanogens for available H_2 and reduce methane production (Zhao and Zhao 2022).

There may also be abiotic or non-microbial factors contributing to elevated methanogenesis from wetlands. It has been observed that elevated temperatures can facilitate the release of methane to the atmosphere via plant release and seepage of thermogenic methane through the soil (Zhang et al. 2020). As such, there may be factors concerning the uptake and release of methane that cause discrepancies in the correlation between observed methane emissions and the activity and abundance of methanogenic archaea.

5.4.4. Implications on global warming

Wetlands are the main natural contributors to methane emission globally, emitting nearly 158.6 Tg of CH_4 per year. In fact, aquatic ecosystems contribute to nearly half of global methane emissions (Rosentreter et al. 2021). Such contribution will be substantially larger if we account for the deposition of wildfire residues in wetlands as wildfires are becoming more frequent and intense (Nzotungicimpaye et al. 2021). Although a previous study showed that methane emissions is lower in burned areas of a wetland because fire removes labile organic material that could be otherwise used by methanogenesis (Davidson et al. 2019), long-term impacts of wildfire will likely increase methane emissions. Wildfire residues can increase nutrient concentration in surface water which can further increase methane emission as urbanized rivers and streams are known to emit more methane than unurbanized ones because of higher nutrient levels (Tang et al. 2021). The effects of wildfire on increased methane emission goes beyond the deposition of wildfire residues in wetlands. For instance, wildfires reduce soil permeability, therefore increasing the likelihood of the ponded areas. This coupled to the fact that climate drivers like tropical storms increase methane emission due to increases in wetland areas (Pandey et al. 2017) further enhances the implications of wildfires on global warming. Furthermore, wildfire residues may increase plant diversity in wetlands which enhances methane emission (Zhang et al. 2012). Similarly, exotic plants may increase methane emission by increasing the contribution of methylotrophic methanogenesis or by decreasing the competitive inhibition by sulfate reducers (Kim et al. 2020), but the implications of wildfire on exotic plants remains unknown.

5.5. Conclusions

The study answered the question of whether and to what extent the deposition of wildfire residues can affect the methane emission from wetland sediments and explained the possible mechanisms that are related to the methane emission variability. Specific conclusions are:

- The deposition of wildfire residues in wetland sediments increases methane emissions, but the extent of the emissions depends on the quantity of wildfire residues deposited.
- Increased methane emissions were partially explained by the generation of hydrogen peroxide from wildfire residues, coupled with the increase in silica content.
- Compared to wetland sediments without the deposition of wildfire residues, wetland sediments with wildfire residues decrease the *mcrA* gene abundance in short-term but maintain higher *mcrA* gene abundance in long-term.

5.6. References

Angle, J.C., Morin, T.H., Solden, L.M., Narrowe, A.B., Smith, G.J., Borton, M.A., Rey-Sanchez, C., Daly, R.A., Mirfenderesgi, G., Hoyt, D.W., Riley, W.J., Miller, C.S., Bohrer, G. and Wrighton, K.C. (2017) Methanogenesis in oxygenated soils is a substantial fraction of wetland methane emissions. Nature Communications 8(1), 1567.

Basso, M., Vieira, D.C.S., Ramos, T.B. and Mateus, M. (2020) Assessing the adequacy of SWAT model to simulate postfire effects on the watershed hydrological regime and water quality. Land Degradation & Development 31(5), 619-631.

Belova, S.E., Kulichevskaya, I.S., Akhmet'eva, N.P. and Dedysh, S.N. (2014) Shifts in a bacterial community composition of a mesotrophic peatland after wildfire. Microbiology 83(6), 813-819.

Burton, C.A., Hoefen, T.M., Plumlee, G.S., Baumberger, K.L., Backlin, A.R., Gallegos, E. and Fisher, R.N. (2016) Trace Elements in Stormflow, Ash, and Burned Soil following the 2009 Station Fire in Southern California. Plos One 11(5), e0153372.

Davidson, S.J., Elmes, M.C., Rogers, H., van Beest, C., Petrone, R., Price, J.S. and Strack, M. (2019) Hydrogeologic setting overrides any influence of wildfire on pore water dissolved organic carbon concentration and quality at a boreal fen. Ecohydrology 12(7), e2141.

Dean, J.F., Middelburg, J.J., Röckmann, T., Aerts, R., Blauw, L.G., Egger, M., Jetten, M.S.M., de Jong, A.E.E., Meisel, O.H., Rasigraf, O., Slomp, C.P., in't Zandt, M.H. and Dolman, A.J. (2018) Methane Feedbacks to the Global Climate System in a Warmer World. Reviews of Geophysics 56(1), 207-250.

Freitag Thomas, E. and Prosser James, I. (2009) Correlation of Methane Production and Functional Gene Transcriptional Activity in a Peat Soil. Applied and Environmental Microbiology 75(21), 6679-6687.

Garcia, L.C., Szabo, J.K., de Oliveira Roque, F., de Matos Martins Pereira, A., Nunes da Cunha, C., Damasceno-Júnior, G.A., Morato, R.G., Tomas, W.M., Libonati, R. and Ribeiro, D.B. (2021) Record-breaking wildfires in the world's largest continuous tropical wetland: Integrative fire management is urgently needed for both biodiversity and humans. Journal of Environmental Management 293, 112870.

Gedalanga, P.B., Pornwongthong, P., Mora, R., Chiang, S.-Y.D., Baldwin, B., Ogles, D., Mahendra, S. and Kivisaar, M. (2014) Identification of Biomarker Genes To Predict Biodegradation of 1,4-Dioxane. Applied and Environmental Microbiology 80(10), 3209-3218.

Hales, B.A., Edwards, C., Ritchie, D.A., Hall, G., Pickup, R.W. and Saunders, J.R. (1996) Isolation and identification of methanogen-specific DNA from blanket bog peat by PCR amplification and sequence analysis. Applied and Environmental Microbiology 62(2), 668-675.

Hohner, A.K., Terry, L.G., Townsend, E.B., Summers, R.S. and Rosario-Ortiz, F.L. (2017) Water treatment process evaluation of wildfire-affected sediment leachates. Environmental Science: Water Research & Technology 3(2), 352-365.

Hu, Y., Jiang, H., Chen, Y., Wang, Z., Yan, Y., Sun, P. and Lu, X. (2021) Nitrogen addition altered the microbial functional potentials of carbon and nitrogen transformation in alpine steppe soils on the Tibetan Plateau. Global Ecology and Conservation 32, e01937.

Kim, J., Lee, J., Yun, J., Yang, Y., Ding, W., Yuan, J. and Kang, H. (2020) Mechanisms of enhanced methane emission due to introduction of Spartina anglica and Phragmites australis in a temperate tidal salt marsh. Ecological Engineering 153, 105905.

Leonard, S.S., Castranova, V., Chen, B.T., Schwegler-Berry, D., Hoover, M., Piacitelli, C. and Gaughan, D.M. (2007) Particle size-dependent radical generation from wildland fire smoke. Toxicology 236(1), 103-113.

Li, X., Song, K. and Liu, G. (2020) Wetland Fire Scar Monitoring and Its Response to Changes of the Pantanal Wetland. Sensors 20(15), 4268.

Ma, K., Conrad, R. and Lu, Y. (2012) Responses of Methanogen mcrA Genes and Their Transcripts to an Alternate Dry/Wet Cycle of Paddy Field Soil. Applied and Environmental Microbiology 78(2), 445-454.

Ma, Y., Huang, S., Gan, Z., Xiong, Y., Cai, R., Liu, Y., Wu, L. and Ge, G. (2020) The succession of bacterial and fungal communities during decomposition of two hygrophytes in a freshwater lake wetland. Ecosphere 11(10), e03242.

Maksimova, E. and Abakumov, E. (2014) Wildfire effects on biological properties of soils in forest-steppe ecosystems of Russia. Solid Earth Discuss. 2014, 71-90.

Mansilha, C., Duarte, C.G., Melo, A., Ribeiro, J., Flores, D. and Marques, J.E. (2019) Impact of wildfire on water quality in Caramulo Mountain ridge (Central Portugal). Sustainable Water Resources Management 5(1), 319-331.

McPherson, T.N., Burian, S.J., Stenstrom, M.K., Turin, H.J., Brown, M.J. and Suffet, I.H. (2005) DRY AND WET WEATHER FLOW NUTRIENT LOADS FROM A LOS ANGELES WATERSHED1. JAWRA Journal of the American Water Resources Association 41(4), 959-969.

Medvedeff, C.A., Inglett, K.S. and Inglett, P.W. (2015) Can Fire Residues (Ash and Char) Affect Microbial Decomposition in Wetland Soils? Wetlands 35(6), 1165-1173.

Mendez, G.O. (2010) Water-quality data from storm runoff after the 2007 fires, San Diego County, California. US Geological Survey, 2010-1234.

Nahlik, A.M. and Fennessy, M.S. (2016) Carbon storage in US wetlands. Nature Communications 7(1), 13835.

Narrowe, A.B., Borton, M.A., Hoyt, D.W., Smith, G.J., Daly, R.A., Angle, J.C., Eder, E.K., Wong, A.R., Wolfe, R.A., Pappas, A., Bohrer, G., Miller, C.S., Wrighton, K.C. and Orsi, W. (2019) Uncovering the Diversity and Activity of Methylotrophic Methanogens in Freshwater Wetland Soils. Msystems 4(6), e00320-00319.

Nazaries, L., Murrell, J.C., Millard, P., Baggs, L. and Singh, B.K. (2013) Methane, microbes and models: fundamental understanding of the soil methane cycle for future predictions. Environmental Microbiology 15(9), 2395-2417.

Nzotungicimpaye, C.M., Zickfeld, K., MacDougall, A.H., Melton, J.R., Treat, C.C., Eby, M. and Lesack, L.F.W. (2021) WETMETH 1.0: a new wetland methane model for implementation in Earth system models. Geosci. Model Dev. 14(10), 6215-6240.

Pandey, S., Houweling, S., Krol, M., Aben, I., Monteil, G., Nechita-Banda, N., Dlugokencky, E.J., Detmers, R., Hasekamp, O., Xu, X., Riley, W.J., Poulter, B., Zhang, Z., McDonald, K.C., White, J.W.C., Bousquet, P. and Röckmann, T. (2017) Enhanced methane emissions from tropical wetlands during the 2011 La Niña. Scientific Reports 7(1), 45759.

Perendeci, N.A., Gökgöl, S. and Orhon, D. (2018) Impact of Alkaline H_2O_2 Pretreatment on Methane Generation Potential of Greenhouse Crop Waste under Anaerobic Conditions. Molecules (Basel, Switzerland) 23(7), 1794.

Perron, M.A.C. and Pick, F.R. (2020) Stormwater ponds as habitat for Odonata in urban areas: the importance of obligate wetland plant species. Biodiversity and Conservation 29(3), 913-931.

Ramsay, J.A., de Lima e Silva, M.R., Tawadrous, M.A.R. and Ramsay, B.A. (2021) Does Addition of Phosphate and Ammonium Nutrients Affect Microbial Activity in Froth Treatment Affected Tailings? Microorganisms 9(11), 2224.

Reithmaier, G.-M.S., Knorr, K.-H., Arnhold, S., Planer-Friedrich, B. and Schaller, J. (2017) Enhanced silicon availability leads to increased methane production, nutrient and toxicant mobility in peatlands. Scientific Reports 7(1), 8728.

Rodríguez-Cardona, B.M., Coble, A.A., Wymore, A.S., Kolosov, R., Podgorski, D.C., Zito, P., Spencer, R.G.M., Prokushkin, A.S. and McDowell, W.H. (2020) Wildfires lead to decreased carbon and increased nitrogen concentrations in upland arctic streams. Scientific Reports 10(1), 8722.

Rosentreter, J.A., Borges, A.V., Deemer, B.R., Holgerson, M.A., Liu, S., Song, C., Melack, J., Raymond, P.A., Duarte, C.M., Allen, G.H., Olefeldt, D., Poulter, B., Battin, T.I. and Eyre, B.D. (2021) Half of global methane emissions come from highly variable aquatic ecosystem sources. Nature Geoscience 14(4), 225-230.

Son, J.-H., Kim, S. and Carlson, K.H. (2015) Effects of Wildfire on River Water Quality and Riverbed Sediment Phosphorus. Water, Air, & Soil Pollution 226(3), 26.

Sun, C., Brauer, S., Cadillo Quiroz, H., Zinder, S. and Yavitt, J. (2012) Seasonal Changes in Methanogenesis and Methanogenic Community in Three Peatlands, New York State. Frontiers in Microbiology 3.

Tang, W., Xu, Y.J., Ma, Y., Maher, D.T. and Li, S. (2021) Hot spot of CH4 production and diffusive flux in rivers with high urbanization. Water Research 204, 117624.

Tanner, P.A. and Wong, A.Y.S. (1998) Spectrophotometric determination of hydrogen peroxide in rainwater. Analytica Chimica Acta 370(2), 279-287.

Taubner, R.-S., Schleper, C., Firneis, M.G. and Rittmann, S.K.M.R. (2015) Assessing the Ecophysiology of Methanogens in the Context of Recent Astrobiological and Planetological Studies. Life (Basel, Switzerland) 5(4), 1652-1686.

Uzun, H., Dahlgren, R.A., Olivares, C., Erdem, C.U., Karanfil, T. and Chow, A.T. (2020) Two years of post-wildfire impacts on dissolved organic matter, nitrogen, and precursors of disinfection by-products in California stream waters. Water Research 181, 115891.

Valenca, R., Ramnath, K., Dittrich, T.M., Taylor, R.E. and Mohanty, S.K. (2020) Microbial quality of surface water and subsurface soil after wildfire. Water Research 175, 115672.

Wang, Y., Hu, Z., Shen, L., Liu, C., Islam, A.R.M.T., Wu, Z., Dang, H. and Chen, S. (2021) The process of methanogenesis in paddy fields under different elevated CO2 concentrations. Science of the Total Environment 773, 145629.

Wenner, B.A., Wagner, B.K., St-Pierre, N.R., Yu, Z.T. and Firkins, J.L. (2020) Inhibition of methanogenesis by nitrate, with or without defaunation, in continuous culture. Journal of Dairy Science 103(8), 7124-7140.

Wormald, R. and Humphreys, P. (2019) Hydrogenotrophic methanogenesis dominates at high pH. Access Microbiology 1(1A).

Wormald, R.M., Rout, S.P., Mayes, W., Gomes, H. and Humphreys, P.N. (2020) Hydrogenotrophic Methanogenesis Under Alkaline Conditions. Frontiers in Microbiology 11.

Ye, R., Jin, Q., Bohannan, B., Keller, J.K., McAllister, S.A. and Bridgham, S.D. (2012) pH controls over anaerobic carbon mineralization, the efficiency of methane production, and methanogenic pathways in peatlands across an ombrotrophic–minerotrophic gradient. Soil biology and biochemistry 54, 36-47.

Yuan, H.-Y., Ding, L.-J., Zama, E.F., Liu, P.-P., Hozzein, W.N. and Zhu, Y.-G. (2018) Biochar Modulates Methanogenesis through Electron Syntrophy of Microorganisms with Ethanol as a Substrate. Environmental Science & Technology 52(21), 12198-12207.

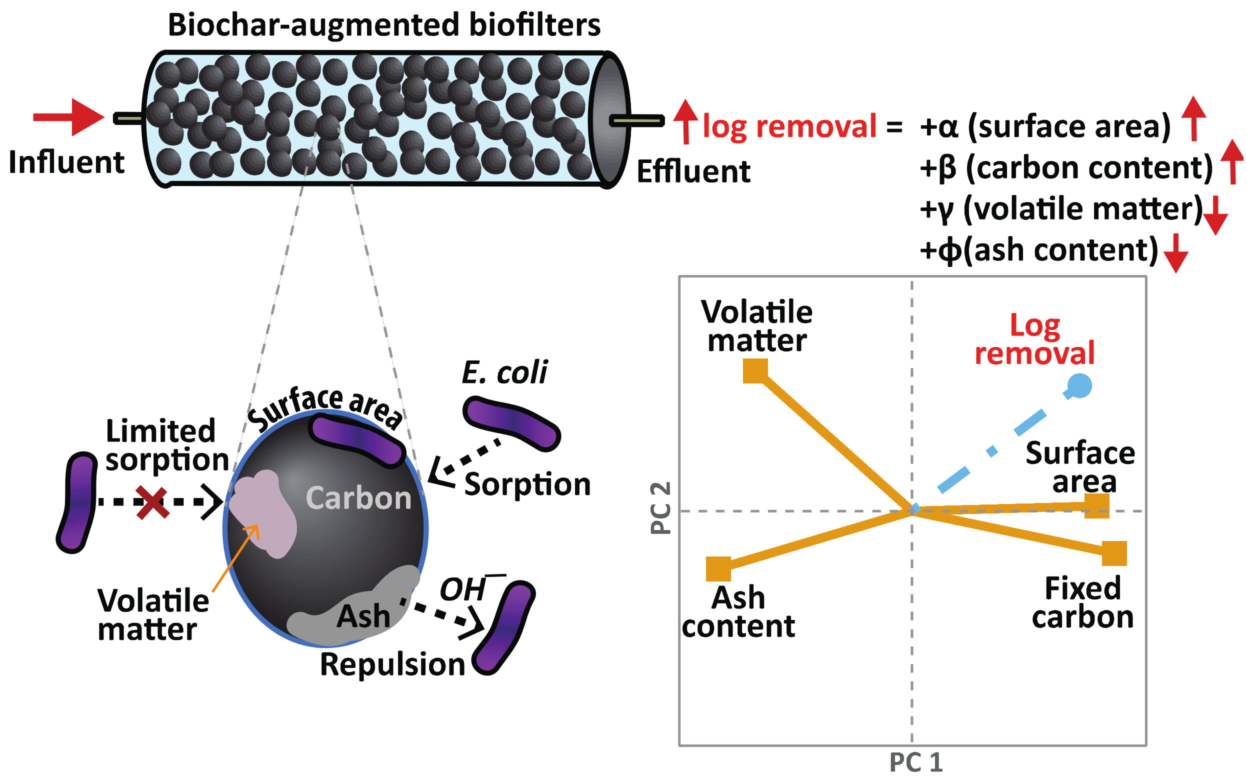
Zhang, C.-B., Sun, H.-Y., Ge, Y., Gu, B.-J., Wang, H. and Chang, J. (2012) Plant species richness enhanced the methane emission in experimental microcosms. Atmospheric Environment 62, 180-183.

Zhang, S., Zhang, F., Shi, Z., Qin, A., Wang, H., Sun, Z., Yang, Z., Zhu, Y., Pang, S. and Wang, P. (2020) Sources of seasonal wetland methane emissions in permafrost regions of the Qinghai-Tibet Plateau. Scientific Reports 10(1), 7520.

Zhang, Y., Yan, C., Liu, H., Pu, S., Chen, H., Zhou, B., Yuan, R. and Wang, F. (2021) Bacterial response to soil property changes caused by wood ash from wildfire in forest soils around mining areas: Relevance of bacterial community composition, carbon and nitrogen cycling. Journal of Hazardous Materials 412, 125264.

Zhao, Y. and Zhao, G. (2022) Decreasing ruminal methane production through enhancing the sulfate reduction pathway. Animal Nutrition.

6. CHAPTER 6 – BIOCHAR SELECTION FOR ESCHERICHIA COLI REMOVAL IN STORMWATER BIOFILTERS



Citation: Valenca, R., Borthakur, A., Zu, Y., Matthiesen, E. A., Stenstrom, M. K., & Mohanty, S. K. (2021). Biochar Selection for *Escherichia coli* Removal in Stormwater Biofilters. *Journal of Environmental Engineering*, 147(2), 06020005. https://doi.org/10.1061/(ASCE)EE.1943-7870.0001843

Abstract

Biochar's capacity to remove pathogens from stormwater can vary by orders of magnitude, which makes it challenging for the stormwater manager to select specific biochar from suppliers. We tested the removal of *Escherichia coli* (*E. coli*) in model biofilters packed with sand and biochar from four suppliers and developed correlation equations that link short-term and long-term bacterial removal capacities of biochar with its commonly reported properties: surface area, carbon content, ash content, and volatile organic carbon content. The *E. coli* removal capacity of biochar was positively correlated with its surface area and carbon content and negatively correlated with ash content and volatile organic matter. Despite the presence of nutrients in stormwater, *E. coli* in pore water in biofilter did not grow between infiltration events, indicating biochar may continue to remove pathogens after rainfall. Overall, the results could help the selection of biochar from suppliers for the treatment of stormwater and inform the suppliers to tailor biochar production conditions to enrich specific biochar properties.

6.1. Introduction

Exposure to water contaminated with pathogens can cause infectious gastrointestinal illnesses, triggering outbreaks of waterborne diseases worldwide (Fewtrell et al. 2005, Harper et al. 2011, Ma et al. 2010). To minimize the health risk related to contaminated water, it is crucial to develop reliable point-of-use water treatment technology to remove pathogens from stormwater and wastewater, which have been increasingly used to offset water demand in water-stressed cities (Zhang 2015). To remove pathogens and other pollutants, biofilters have been used where contaminated water are passed through a packed sand and amendments (Cohen 2001, Tan et al. 2015, Velten et al. 2011). Among different amendments, biochar is readily available and affordable in many locations (Mohanty et al. 2018) because they can be produced by pyrolyzing available plant biomass (Inyang and Dickenson 2015).

One of the challenges of selecting biochar is its removal uncertainty. For instance, biochar's capacity to remove pathogens or pathogen indicators varies by orders of magnitude (Boehm et al. 2020), which has been attributed to different biochar properties and water chemistry (Afrooz et al. 2018, Guan et al. 2020, Mohanty et al. 2014, Suliman et al. 2017). Unlike activated carbon, biochar properties can vary widely based on preparation conditions and feedstock types (Xiao et al. 2018). Generally, it is recommended to use wood-based biochar prepared at high pyrolysis temperature (Abit et al. 2012, Bolster and Abit 2012) without removing fine particle size (Guan et al. 2020, Mohanty and Boehm 2014, Sasidharan et al. 2016). Despite constraining these conditions, bacterial removal has been showed to vary widely based on published literature, indicating competing effects of different properties (**Figure 6.1**). For instance, an increase in removal has been attributed to an increase in hydrophobicity (Afrooz and Boehm 2016, Lau et al. 2017, Mohanty et al. 2014) and surface area (Afrooz and Boehm 2016, Lau et al. 2017) of biochar, whereas a decrease in removal has been attributed to an increase in oxidation of biochar (Suliman et al. 2017) and volatile carbon content (Mohanty et al. 2014). These surface properties are influenced by bulk chemical properties of biochar including carbon content, ash content, volatile carbon content, and physical property such as surface area (Manya 2012). The relative importance of these commonly measured properties bacterial removal is unknown. This makes it challenging for the selection of biochar from different vendors for field application (Boehm et al. 2020).

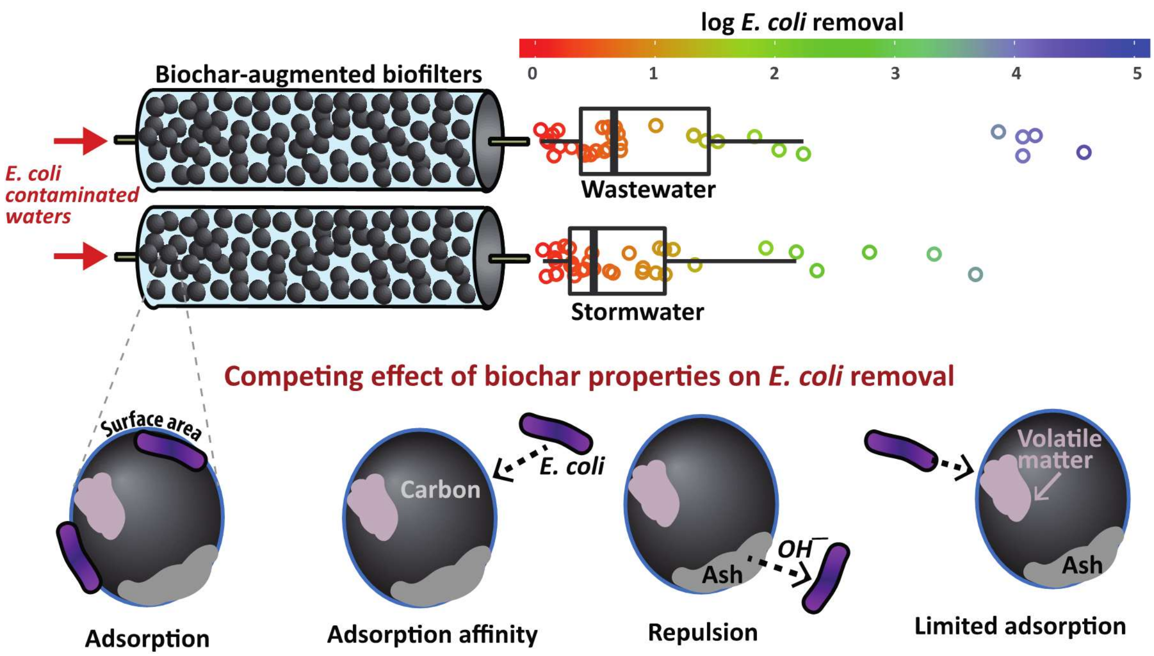


Figure 6.1 – The removal of *E. coli* in biochar-augmented biofilters varies by orders of magnitude, potentially due to competing effects of different properties of biochar. The wide variation of biochar capacity to remove pathogen is one of the main hurdles in selecting biochar for water treatment.

Biochar is traditionally selected based on their short-term laboratory performance. Most of the studies, listed in the review paper (Mohanty et al. 2018), have estimated the removal capacity of biochar based on the clean-bed breakthrough data. With increased exposure to contaminated water, attachment sites may become exhausted (Kranner et al. 2019, Lau et al. 2017, Nabiul Afrooz and Boehm 2017), which may result in lower removal capacity. The long-term removal could also decrease if removed *E. coli* start growing in the biofilters during period between infiltration events utilizing the nutrients in pore water near biochar surface (Berger et al. 2019) or adsorbed on biochar (Huggins et al. 2016, Velten et al. 2011). Biochar aging under typical weathering conditions could influence biochar physical and surface properties (Hale et al. 2011, Mohanty and Boehm 2015), thereby affecting its contaminant removal capacity. If growth is a factor, the increase in duration could increase *E. coli* concentration in pore water assuming growth limiting conditions (drought, nutrient depletion) are absent. Biochar could also increase the removal of *E. coli* from pore water due to bacterial die off and/or inactivation (Gurtler et al. 2014) or adsorption (Mohanty et al. 2014). Increasing duration between infiltration events has been shown to have no effect (Nabiul Afrooz and Boehm 2017, Rahman et al. 2020) or positive effect on bacterial removal (Mohanty et al. 2014). The cause of the inconsistent effect of the duration between infiltration event is unknown. In particular, it is not clear if the fate of attached bacteria between infiltration events is related to biochar properties.

This study aims to identify the biochar properties that have greater influence on the removal of *E. coli* in biochar-sand filters. We hypothesized that the long-term removal would differ from clean-bed removal, and the removal during and in between infiltration events can be predicted based on a commonly measured bulk biochar properties such as surface area, carbon content, ash content, and volatile matter. The results will help develop strategy for selecting the best-performing biochar from different vendors for the treatment of waters contaminated with bacterial pathogens.

6.2. Experimental methods

6.2.1. Filter media

Biofilter media for each biofilter consisted of a mixture of coarse Ottawa sand (0.6 – 0.85 mm) and one of the commercially available wood-based biochar: Terra Char (BioEnergy Innovations Global, Inc., MO, USA), Agricultural Carbon (National Carbon Technologies, LLC, MN, USA), Naked Char (American BioChar Company, MI, USA), and Rogue Biochar (Oregon Biochar Solutions, LLC, OR, USA). Large biochar particles (> 2.0 mm) were removed by sieving to minimize preferential water flow through the filters. To create a homogenous media mixture before packing, sand and biochar (30% v/v) were mixed manually using a sterilized 4-L bucket for 5 minutes.

Each biochar was characterized using ultimate (ASTM 3176) and proximate analysis (ASTM D 3172)—the most commonly used methods to characterize biomass— to estimate the carbon content, ash content, volatile carbon, and elemental composition (C, H, O, N and S). The ratios of O+N and C was used as an indicator for the polarity of the biochar (Chen et al. 2008). The surface area was measured based on the adsorption of nitrogen gas (Peterson et al. 2012). Between biochar types, ash content was varied by a factor of 4.4, the surface area was varied by a factor of 2.8, volatile carbon was varied by a factor of 2.8, and fixed carbon was varied by a factor of 1.3. The (O+N)/C, an indicator of polarity, and O/C, an indicator of oxidized biochar, was varied by a factor of 2 (**Table 6.1**).

Table 6.1 – Preparation condition and properties of four types of biochar used in this study

Parameters	Terra Char	Agricultural Carbon	Naked Char	Rogue Biochar
Vendor	BioEnergy Innovations Global, Inc., MO	National Carbon Technologies, LLC, MN	American BioChar Co., MI	Oregon Biochar Solutions, LLC, OR
Feedstock	Oak Hardwood Sawdust	Wood-based	Southern Yellow Pine Species	80% softwood, 15% hardwood, and 5% nutshells
Pyrolysis temperature (°C)	540	>550	550 – 990	>900
Surface area (m^2 g^{-1})	207	339	283	475
S, %	0.003	0.002	0.005	0.041
C, %	70.16	85.03	80.96	84.66
H, %	1.89	2.77	0.59	0.83
N, %	0.62	0.31	0.53	0.81
O, %	9.36	7.78	5.67	5.43
Polarity Index, (O+N)/C	0.142	0.095	0.077	0.074
Ash, %	17.97	4.11	12.24	8.23
Volatile Matter, %	18.55	12.19	6.66	7.86
Fixed Carbon, %	63.48	83.7	81.1	83.91

6.2.2. Contaminated stormwater preparation

Synthetic stormwater was used throughout the experiment to maintain a constant concentration of nutrients in stormwater and to prevent interference of other constituents such as dissolved organic carbon and natural microorganisms on the interaction of the selected *Escherichia coli* (*E. coli*) with biochar surfaces. The stormwater was prepared following the method described elsewhere (Mohanty and Boehm 2014). Briefly, salts containing major cations, anions and nutrients — 0.75 mM of $CaCl_2$, 0.075mM of $MgCl_2$, 0.33 mM Na_2SO_4, 1.0 mM of $NaHCO_3$, 0.072 mM of $NaNO_3$, 0.072mM of NH_4Cl, and 0.016 mM of Na_2HPO_4 — were mixed in ultrapure water (18 Ω), and the solution was autoclaved at 121 °C and 100kPa for 40 minutes before storing at 4°C for use in experiments. Before usage in the experiments, a suspension containing *E. coli* K-12 with resistance to kanamycin was added to the stormwater to achieve a final concentration of ~10^5 CFU mL^{-1} (Valenca et al. 2020). The *E. coli*-laden stormwater solution was used within 2 hours of the preparation to minimize any growth.

6.2.3. Filter design

Each biochar and sand mixture was packed in polypropylene columns (2.54 cm diameter, 30 cm height). 10 cm^3 of each mixture was poured into the columns to create a small layer, and the layer was packed by gently tapping 20 times on top using a steel rod. The procedure was repeated until the entire column was filled. Glass wool was used at both ends to prevent leakage of filter media. After packing, the columns were saturated by injecting the uncontaminated stormwater at a slow flow rate of 0.05 mL min^{-1} from the bottom of the columns. To condition the filter media with the synthetic stormwater, the uncontaminated stormwater was injected for 24 hours at 2 mL min^{-1}. Based on the weight of the columns at different stages, the bulk density (ρ_b= 1.17 ± 0.04 g cm^{-3}), porosity (η = 0.33 ± 0.01), and pore volume (PV = 50.3 ± 2.8 mL) of each media mixture were determined (**Table 6.2**).

Table 6.2 – Biofilter design parameters. Geomedia added as a mix of sand and biochar (70:30 by volume). Pore volume is calculated as difference between dry and saturated media. Bold values represent average from triplicated columns. Values between parenthesis represent standard deviation of triplicated columns.

Biochar type	Empty Column (g)	Column + dry media (g)	Dry density (g cm^{-3})	Column + saturated media (g)	Pore volume (mL)	Porosity (%)
Terra Char	204.1	376.6	1.13	425.0	48.4	0.32
	(1.8)	(6.1)	(0.03)	(6.3)	(0.4)	(0.00)
Agricultural Carbon	203.2	387.9	1.21	441.6	53.7	0.35
	(0.4)	(1.9)	(0.01)	(4.0)	(3.3)	(0.02)
Naked Char	203.3	382.2	1.18	431.1	48.9	0.32
	(0.6)	(4.8)	(0.03)	(3.4)	(1.7)	(0.01)
Rogue Biochar	203.7	379.3	1.16	429.5	50.2	0.33
	(1.7)	(4.7)	(0.03)	(4.5)	(0.3)	(0.00)

6.2.4. Removal of *E. coli* in sand-biochar filters during intermittent infiltration events

Experiments were conducted with triplicated filters to examine *E. coli* removal in short term (clean-bed removal) and with duplicated filters to examine *E. coli* removal in long term (Figure 6.2). The stormwater containing *E. coli* (~ 10^5 CFU mL^{-1}) was injected through the columns in upward direction at 2.0 mL min^{-1} using a peristaltic pump (Masterflex L/S Digital Drive, Cole Parmer), and effluent sample fractions were collected using 15-mL centrifuge tubes. The clean-bed removal was estimated by injecting the contaminated stormwater for 5 hours (~ 10 PV) and comparing *E. coli* concentration in the last 0.5 PV of effluent. To measure the long-term removal, the experiment was repeated by injecting the contaminated stormwater for 4 hours (~8 PV) after a flow interruption period of 24–96 hours. The experiment was repeated for 10 times, resulting an injection of 75+ pore volume of stormwater. The long-term removal capacity was calculated as the average log-removal during last three infiltration events that occurred between 60 and 75 PV. During each injection, the effluent samples containing the first 0.5 PV and last 0.5

PV were collected, and the *E. coli* concentration was analyzed using spread-plate counting technique.

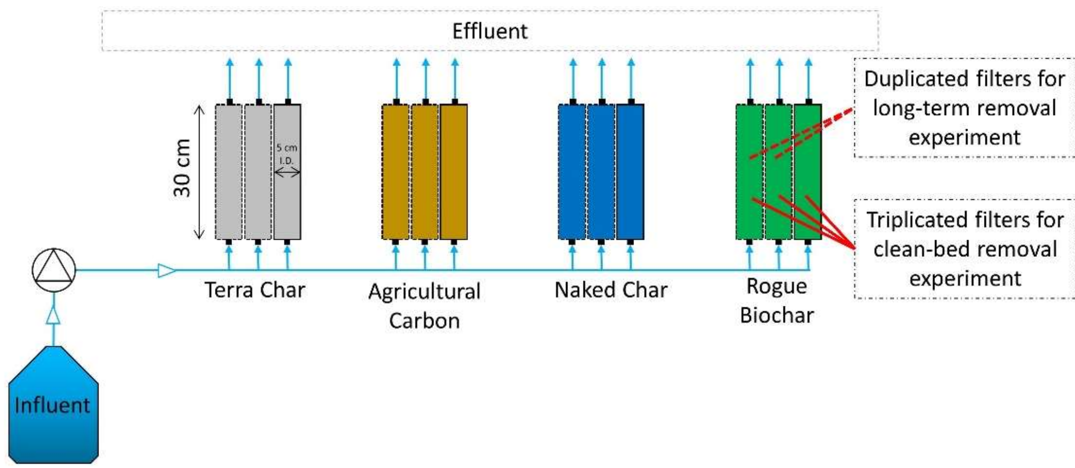


Figure 6.2 – Schematic of filters setup used for both clean-bed and long-term removal experiments.

The effect of flow interruption on the fate of *E. coli* in the filter was analyzed by comparing the concentration of *E. coli* in the effluent fractions before and after the flow interruption lasting between 24 to 96 h. The growth-decay index (GDI), the ratio of *E. coli* concentration after (C_a) and before (C_b) the flow interruption, was estimated to determine if *E. coli* trapped in biofilters grows between infiltration events. The net-growth of *E. coli* in between infiltration events is assumed if log GDI is positive (or GDI > 1) and a net-decay of *E. coli* is assumed if log GDI is negative (or GDI < 1).

6.2.5. Statistical analysis

To identify statistically significant differences between the clean-bed removal and long-term removal capacities, analysis of variance (ANOVA) was performed using Turkey's HSD test. Pearson correlations between biochar's properties and removal capacities and GDI values were estimated. Differences were considered significant at p-value < 0.05. Statistical analysis was conducted using R (version 3.5.3). Principal Component Analysis and Partial Linear Squares (PLS) regression models were analyzed using XLSTAT (version 2020.2.3) to examine how the removal is dependent on selected biochar properties.

6.3. Results

6.3.1. Clean-bed removal capacity varied by more than an order of magnitude based on biochar types

The clean-bed *E. coli* removal was determined based on the breakthrough plateau concentration after an injection of 2 PV of contaminated stormwater. The clean-bed removal of *E. coli* depended on biochar types (Figure 6.3) and varied by more than one order of magnitude between different biochar types (Table 6.3). Biofilters with Rogue Biochar (log removal of 3.20±0.48) or Agricultural Carbon (3.64±0.74) removed considerably more bacteria than biofilters with Terra Char (1.98±0.38) or Naked Char (1.90±0.50).

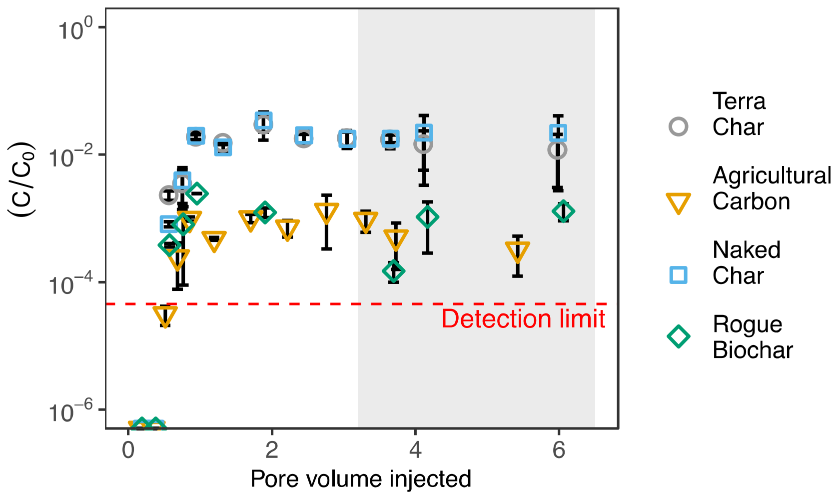


Figure 6.3 – Relative concentration of *E. coli* in the effluent (C) of previously uncontaminated (clean bed) biochar-sand filters during the injection of synthetic stormwater containing *E. coli* at a concentration (C_0) of 4.0±0.7 × 10^5 CFU mL^{-1}. Shaded area represents breakthrough plateau where log removal for each biochar type was calculated. Error bars represent standard deviation of triplicated columns and duplicated agar plates per sample. Dashed horizontal red line indicates the detection limit of 1 colony per plate (20 CFU mL^{-1}) in the effluent samples.

Table 6.3 – Clean-bed and long-term log-removal of applied *E. coli* in columns packed with sand and different types of biochar during injection of contaminated stormwater. Log-removal indicates the average ± one standard deviation. [a]log removal is calculated by taking negative logarithm of the ratio of effluent (at plateau) and influent concentration.

	log *E. coli* removal[a]	
Biochar type	**Clean-bed**	**Long-term**
Terra Char	1.98 ± 0.38	2.71 ± 0.48
Agricultural Carbon	3.64 ± 0.74	2.51 ± 0.49
Naked Char	1.90 ± 0.50	2.12 ± 0.34
Rogue Biochar	3.20 ± 0.48	5.02 ± 0.13

6.3.2. Long-term removal capacity differed from the clean bed removal capacity

The clean-bed removal capacity of each biofilters differed from that of the long-term removal capacity and the extent of difference varied with biochar types (Figure 6.4 – A). Compared to the clean-bed removal capacity, the long-term removal capacity either significantly ($p< 0.05$)

increased in the biofilters containing Terra Char or Rouge Biochar, decreased ($p < 0.05$) in the filters containing Agricultural Carbon, and remained similar ($p > 0.05$) in the filters containing Naked Char. Compared with the clean-bed removal (Figure 6.4 – B), increasing exposure to 70+ pore volume contaminated stormwater increased ($p < 0.05$) the median log removal by 71% and 62% in biofilters containing Rouge Biochar and Terra Char, respectively, and decreased ($p < 0.05$) the median log removal by 20% in the biofilters containing Agricultural Carbon. No significant difference ($p > 0.05$) between the short-term and the long-term removal was observed for the biofilters packed with Naked Char.

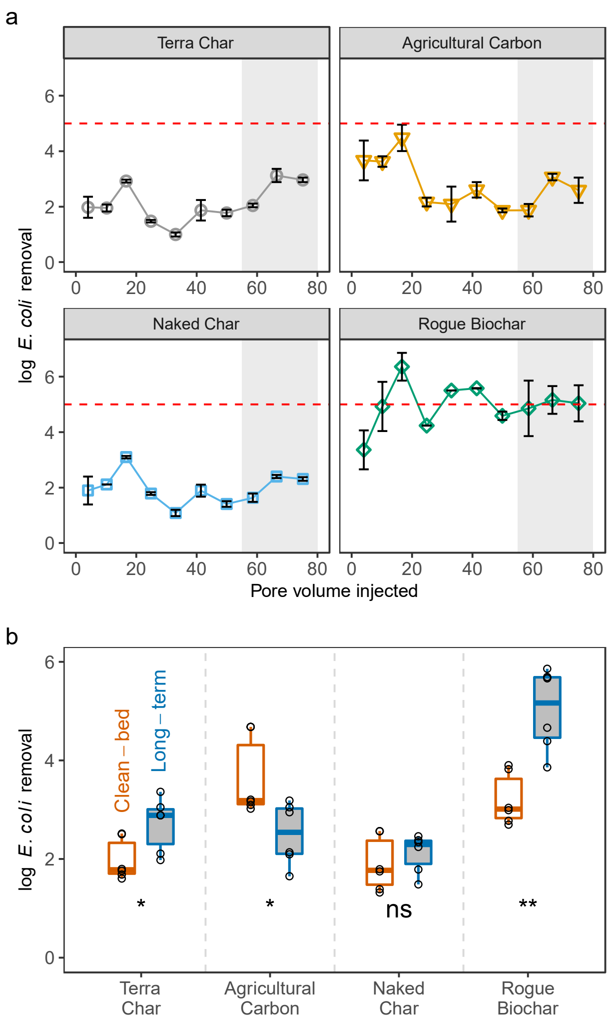


Figure 6.4 – (A) Removal capacity of biochar-augmented filters varied with increased in exposure to contaminated stormwater during 10 infiltration events. Shaded area includes the removal data used to estimate long-term removal. Horizontal solid red line represents detection limit of 1 CFU per plate, which is equivalent to 5 logs removal). The error bars indicate one standard deviation of duplicated columns and duplicated agar plates per sample (n = 4). (B) Comparison between clean-bed removal capacity of filters (empty box plots) and the long-term removal capacity (filled box plot). *p-value < 0.05, **p-value < 0.005, ns = no significance difference.

6.3.3. Fate of *E. coli* in filters between infiltration events

The *E. coli* concentration in pore water of sand-biochar filters mostly decreased (with few exceptions) during intervals between infiltration events, resulting in the growth-die off index (GDI) becoming less than 1 or the log GDI below zero (Figure 6.5). The log GDI values appears to be independent of the duration between infiltration events. The mean (geometric) log GDI values varied with biochar types: 0.097, 0.413, 0.129, and 0.383 for filters with Terra Char, Agricultural Carbon, Naked Char, and Rogue Biochar, respectively.

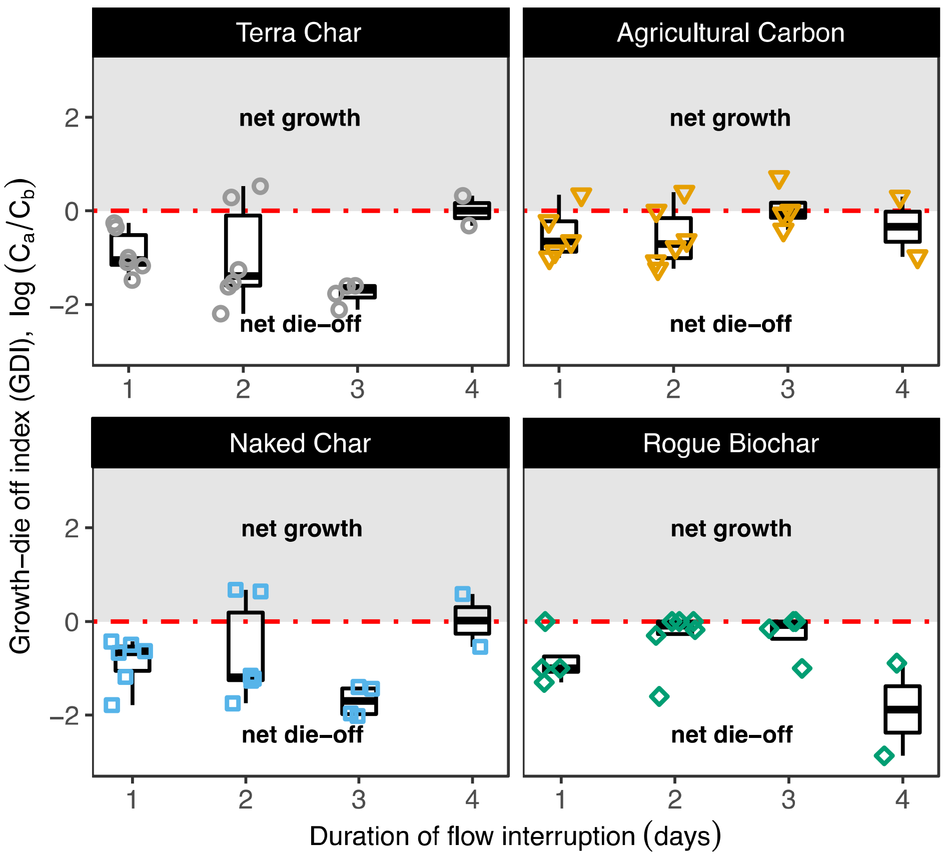


Figure 6.5 – Growth-die off Index (GDI) calculated based on the ratio of *E. coli* concentration after (C_a) and before (C_b) the flow interruption between two consecutive infiltration events. Horizontal dashed red lines are the boundary between GDI values corresponding to net growth and die off or removal of *E. coli* during the flow interruption. Positive GDI values indicate net growth of bacteria in biofilters between infiltration events (shaded area), while negative GDI values indicate net removal by die-off and adsorption.

6.3.4. Effect of biochar's properties on *E. coli* removal during and in between rainfall events

Among all biochar properties (Figure 6.6), surface area positively correlated with the long-term removal capacity (Pearson coefficient, r = 0.79) and the clean-bed removal capacity (r = 0.74), whereas polarity of biochar negatively correlated with the clean-bed (r = -0.38) and the long-term removal capacity (r = -0.29), although the correlation was weaker. The volatile matter had weak and negative correlations with both the clean-bed removal capacity (r= -0.2) and the long-term removal capacity (r = -0.22). Ash content was negatively correlated with the clean bed-removal capacity (r = -0.89), but the correlation significantly decreased for the long-term removal capacity (p = -0.16). Fixed carbon content was strongly correlated (r = 0.64) with the clean-bed removal capacity, but weakly correlated with the long-term removal capacity of biochar (r = 0.22). Growth-die off index was negatively correlated with the biochar polarity (r = -0.5) and ash content (r = -0.92), but it was positively correlated to biochar surface area (0.81) and the clean-bed removal capacity (r = 0.99).

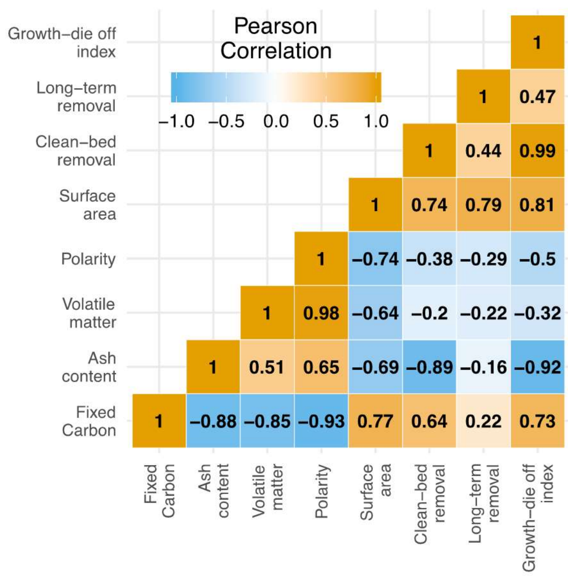


Figure 6.6 – Correlation of clean-bed removal capacity, long-term removal capacity, and growth-die off index (GDI) with specific biochar properties including fixed carbon, ash, volatile matter, polarity, and surface area.

To differentiate the properties of biochar that can concurrently affect the removal capacity and growth-die off index, principal component analysis (PCA) was performed. PCA was used to increase the interpretability of the results by creating new uncorrelated variables through reducing the dimension of the results with little to no information loss (Jolliffe and Cadima 2016). The results (Figure 6.7) showed that the two components presented (PC1 and PC2) characterizes more than 93% of the results. While component PC1 has a relative similar importance of fixed carbon (23.8%), volatile matter (19.4%) and polarity (22.7%), component PC2 is mostly impacted by ash content (45.5%) and volatile matter (34.3%). The PCA analysis shows that while surface area and fixed carbon positively affect the growth-die off index and the clean-bed removal, polarity and ash content negative affect growth-die off index and the clean-bed removal capacity. Long-term removal appears to be uncorrelated or weakly correlated to most variables due to its position near the origin.

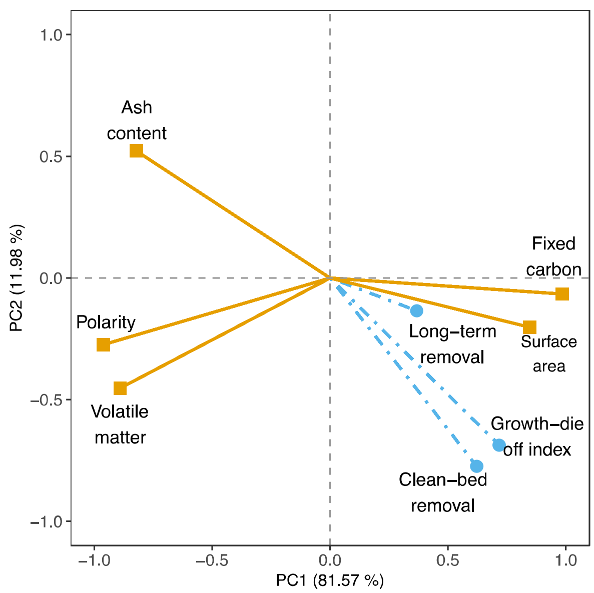


Figure 6.7 – Principal Component Analysis (PCA) between biochar properties, removal capacities and growth-die off index (GDI). The contribution for each factor is as follows: PC1 (23.76% fixed carbon, 16.57% ash content, 19.42% volatile matter, 22.66% polarity, and 17.48% surface area) and PC2 (0.73% fixed carbon, 45.53% ash content, 34.33% volatile matter, 12.50% polarity, and 6.91% surface area). Narrow angle between parameters indicates positive correlation, obtuse angle indicates negative correlation, and a right angle indicates no or weak correlation.

Based on the partial least squares regression, a model was developed to predict clean-bed removal (R_S, equation 6.1) and growth-die off index (GDI, equation 6.2) based on surface area (SA), fixed carbon (FC), ash content (AC), and volatile matter (VM).

$$R_S = 0.0045 \times SA + 0.0097 \times FC - 0.113 \times AC + 0.104 \times VM + 0.531 \qquad \text{Equation 6.1}$$

$$GDI = 0.0008 \times SA + 0.0023 \times FC - 0.019 \times AC + 0.015 \times VM - 0.157 \qquad \text{Equation 6.2}$$

These regression models indicate that biochar surface area had limited positive impact in both clean-bed removal (0.0045) and growth-die off index (0.0008). Fixed carbon content also had a positive impact on the clean-bed removal capacity and growth-die off index, but its impact was 4 times higher in clean-bed removal capacity than on GDI. While volatile matter had positive impact on clean-bed removal and GDI, ash content had a negative impact on both GDI (-0.019) and the clean-bed removal capacity (-0.113). Among all properties, ash content had more influence on GDI than surface area, carbon content, and volatile matter.

6.4. Discussion

6.4.1. Clean-bed removal does not predict the long-term removal capacity

Most studies examined removal capacity of biochar-augmented filters based on the breakthrough concentration of *E. coli* in previously uncontaminated filters (Afrooz et al. 2018, Mohanty and Boehm 2014, Phillips 2020), although several studies have shown that long-term removal could decrease (Kranner et al. 2019, Lau et al. 2017, Nabiul Afrooz and Boehm 2017). We showed that, although the clean-bed removal capacity is useful to quickly compare the removal capacity between biochar types, it weakly (Pearson coefficient, r = 0.44) predicted the long-term removal capacity of the biochar. In clean-bed, the *E. coli* is removed mostly by adsorption on pristine or uncontaminated biochar surface. In long term, attachment sites on biochar surface could become exhausted by increasing loading of *E. coli* (Kranner et al. 2019, Lau et al. 2017) and formation of biofilm on biochar (Afrooz and Boehm 2016). With aging, biochar surface properties could become oxidized, which can reduce *E. coli* adsorption (Lau et al. 2017, Suliman et al. 2017). In our case, *E. coli* removal decreased for one biochar type, increased for 2 biochar types, and did not change for one biochar. A consistently high removal or no net reduction in removal with exposure to more polluted stormwater was attributed to difference in composition of synthetic stormwater with natural stormwater. Compared to other studies that examined long-term *E. coli* removal by biochar, our study used stormwater without native bacteria and dissolved organic carbon (DOC). Both can compete with indicator bacteria for sorption sites on biochar (Mohanty and Boehm 2014, Ulrich et al. 2017). As a result, the exhaustion rate could be a lot less in our study compared to other studies that used a natural stormwater. Thus, our results overestimate the capacity of biochar in removing E. coli. It is important to note that same water chemistry and bacteria strain should be used for comparison between biochars as bacterial removal by biochar varies with bacteria strain (Abit et al. 2014, Suliman et al. 2017) and water chemistry (Mohanty et al. 2014). Nevertheless, our results help compare the removal capacities of biochars.

6.4.2. Biochar prevents growth of *E. coli* in pore water in between infiltration events

In contrast to our hypothesis that *E. coli* may grow in biochar-sand filters by utilizing the available nutrients, our results showed that *E. coli* concentration typically decreased during the period between infiltration events. The lack of *E. coli* growth indicates that nutrients retained in biofilters were not bioavailable or sufficient for *E. coli* growth (Valenca et al. 2020). Biofilm was unlikely to be formed in our experiment because the *E. coli* lack extracellular polymeric substance for the formation of biofilm compared with other microorganisms (Afrooz and Boehm 2016). In between the infiltration events, biochar could adsorb more *E. coli* due to increase in residence time (Mohanty et al. 2014) or help inactivate *E. coli* (Sun et al. 2019). Biochar could also adsorb metabolites produced by *E. coli* (Hill et al. 2019), thereby limiting bacterial growth. Thus, unlike other amendment, biochar could continue to remove or inactivate *E. coli* from pore water in between infiltration events, thereby replenishing filter media for the removal of more contaminants in the following infiltration events. This is particularly significant for the treatment of stormwater in low-intensity rainfall events. These rainfall events often yield no outflow, thereby trapping very high concentration of *E. coli* in biofilters, where they can grow (Hill et al. 2019). Excess bacterial growth (Mohanty et al. 2014) and mobilization of bacteria during intermittent infiltration events (Mohanty and Boehm 2015, Mohanty et al. 2013a, b) can result in negative removal or net export of indicator bacteria from biofilters (Boehm et al. 2020). An addition of biochar in those filters would decrease the growth or kill *E. coli* between rainfall events, thereby potentially preventing biofilters to become a source of *E. coli*. Thus, net mass removal in biochar-augmented filters could be higher than predicted in the previous studies that typically did not account for the removal of bacteria in between infiltration events.

We developed a simple growth-die off index (GDI) to compare the biochar impacts on bacterial fate in biofilters in between infiltration events. In general, it is expected that longer duration between rainfall events would allow the bacteria to grow on carbon adsorbent utilizing nutrient in the infiltrating water (Huggins et al. 2016, Velten et al. 2011, Wilcox et al. 1983). In contrast, our results showed that *E. coli* concentration did not increase but decreased, indicating a lack of growth or a net removal of *E. coli*. However, no correlation was observed between the changes in *E. coli* concentration and the duration between infiltration events. We attribute these results to the biochar ability to continue to remove bacteria by inactivation (Gurtler et al. 2014) and adsorption (Mohanty et al. 2014) or to reduce the availability of growth metabolites (Hill et al. 2019). It should be noted that the *E. coli* concentration in some experiments was close to the detection limit, which contributes to high uncertainty in the estimation of GDI values. Nevertheless, our study confirmed that biochar prevents the growth of *E. coli* and remove them in the period between infiltration events.

6.4.3. Removal depends on specific biochar properties: ash content and surface area

Choosing biochar with high capacity is essential in meeting the design goal of removal of bacterial pollutants from contaminated waters. Understanding which properties are related to removal would be critical in the selection of appropriate biochar. Correlating the clean-bed, long-term removal and growth-die off index with specific biochar properties, we showed that an increase in biochar surface area increased both clean-bed and long-term removal capacities, potentially because larger surface area provides more adsorption sites. Our results are in accordance with the previous studies (Afrooz and Boehm 2016, Liu et al. 2020). However, some other studies have shown that biochar with higher surface areas had similar (Guan et al. 2020) or lower (Mohanty et al. 2014) removal capacity. In these cases, other biochar properties may have disproportionally greater impact on adsorption that that of surface area. PLS-model validation showed that our model was able to predict the log removal of another commercially available biochar (Sonoma Biochar) used in a previous published study (Mohanty et al. 2014). This shows that our properties-based model could be used to indicate the *E. coli* removal capacity of commercially available biochars, although validating the model with more biochars can improve the model. It should be noted that water chemistry and bacteria type should be kept consistent to minimize their impacts on the removal capacity of biochar.

We showed that the ash content is the most important indicator of bacterial removal in biochar. The clean-bed removal was negatively correlated with the ash content. A high ash content increases pH of pore water, which can increase electrostatic repulsion between bacteria and biochar owing to a higher negative surface charge of both surfaces at higher pH (Oh et al. 2012). Similarly, biochar's polarity is, albeit weakly, but negatively correlated with the removal capacity. A high polarity indicates more negative surface charge (Tumiran et al. 1996), which can increase electrostatic repulsion and decrease *E. coli* attachment on biochar (Suliman et al. 2017). A decrease in ash content has been shown to improve the adsorption of organic pollutants by hydrophobic interaction (Shimabuku et al. 2016). As *E. coli* adsorb on biochar by hydrophobic interaction, this explains why ash content has a strong influence on biochar removal capacity. This has an important implication on selecting the type of biochar with low ash content as it can vary up to 83% (Shimabuku et al. 2016). The vendors could lower ash content in by biochar by optimizing the production condition such as feedstock type and pyrolysis temperature (Ahmed et al. 2016). If the biochar has high ash content, they can also be washed with strong acids to dissolve and remove the ash, which has been shown to improve removal of organic pollutant (Sun et al. 2013). Thus, lowering the ash content and increasing the carbon content can improve pathogen removal in biochar.

6.5. Conclusions

The study used four commercially available biochar to examine the link between their *E. coli* removal capacity and their bulk properties. Specific conclusions are:

- Clean-bed capacity is unrelated to long-term capacity, although clean-bed capacity can be used as a predictor of *E. coli* fate between infiltration events.
- Between infiltration events biochar limited the growth and increased the removal of *E. coli*, but the removal was independent of the duration between infiltration events.
- The *E. coli* removal capacities of sand-biochar filters were positively correlated with the surface area and organic carbon content of biochar, and negatively correlated with ash content and volatile matter.

- A model relating biochar removal capacity with these commonly measured biochar properties was developed based on partial least squares regression, which has the potential to predict the *E. coli* removal capacity of commercially available biochar. Thus, the model can help the selection of biochar for water treatment application.

6.6. References

Abit, S.M., Bolster, C.H., Cai, P. and Walker, S.L. (2012) Influence of feedstock and pyrolysis temperature of biochar amendments on transport of Escherichia coli in saturated and unsaturated soil. Environmental Science and Technology 46(15), 8097-8105.

Abit, S.M., Bolster, C.H., Cantrell, K.B., Flores, J.Q. and Walker, S.L. (2014) Transport of Escherichia coli, Salmonella typhimurium, and Microspheres in Biochar-Amended Soils with Different Textures. Journal of Environmental Quality 43(1), 371-388.

Afrooz, A. and Boehm, A.B. (2016) Escherichia coli Removal in Biochar-Modified Biofilters: Effects of Biofilm. Plos One 11(12), https://doi.org/10.1371/journal.pone.0167489

Afrooz, A., Pitol, A.K., Kitt, D. and Boehm, A.B. (2018) Role of microbial cell properties on bacterial pathogen and coliphage removal in biochar-modified stormwater biofilters. Environmental Science-Water Research & Technology 4(12), 2160-2169.

Ahmed, M.B., Zhou, J.L., Ngo, H.H. and Guo, W.S. (2016) Insight into biochar properties and its cost analysis. Biomass & Bioenergy 84, 76-86.

Berger, A.W., Valenca, R., Miao, Y., Ravi, S., Mahendra, S. and Mohanty, S.K. (2019) Biochar increases nitrate removal capacity of woodchip biofilters during high-intensity rainfall. Water Research 165, 115008.

Boehm, A.B., Bell, C.D., Fitzgerald, N.J.M., Gallo, E., Higgins, C.P., Hogue, T.S., Luthy, R.G., Portmann, A.C., Ulrich, B.A. and Wolfand, J.M. (2020) Biochar-augmented biofilters to improve pollutant removal from stormwater – can they improve receiving water quality? Environmental Science: Water Research & Technology, https://doi.org/10.1039/d1030ew00027b

Bolster, C.H. and Abit, S.M. (2012) Biochar Pyrolyzed at Two Temperatures Affects Escherichia coli Transport through a Sandy Soil. Journal of Environmental Quality 41(1), 124-133.

Chen, B., Zhou, D. and Zhu, L. (2008) Transitional adsorption and partition of nonpolar and polar aromatic contaminants by biochars of pine needles with different pyrolytic temperatures. Environmental Science and Technology 42(14), 5137-5143.

Cohen, Y. (2001) Biofiltration - the treatment of fluids by microorganisms immobilized into the filter bedding material: a review. Bioresource Technology 77(3), 257-274.

Fewtrell, L., Kaufmann, R.B., Kay, D., Enanoria, W., Haller, L. and Colford, J.M. (2005) Water, sanitation, and hygiene interventions to reduce diarrhoea in less developed countries: a systematic review and meta-analysis. The Lancet Infectious Diseases 5(1), 42-52.

Guan, P., Prasher, S.O., Afzal, M.T., George, S., Ronholm, J., Dhiman, J. and Patel, R.M. (2020) Removal of Escherichia coli from lake water in a biochar-amended biosand filtering system. Ecological Engineering 150, https://doi.org/10.1016/j.ecoleng.2020.105819.

Gurtler, J.B., Boateng, A.A., Han, Y.X. and Douds, D.D. (2014) Inactivation of E. coli O157:H7 in Cultivable Soil by Fast and Slow Pyrolysis-Generated Biochar. Foodborne Pathog Dis 11(3), 215-223.

Hale, S., Hanley, K., Lehmann, J., Zimmerman, A. and Cornelissen, G. (2011) Effects of chemical, biological, and physical aging as well as soil addition on the sorption of pyrene to activated carbon and biochar. Environmental Science and Technology 45(24), 10445-10453.

Harper, S.L., Edge, V.L., Schuster-Wallace, C.J., Berke, O. and McEwen, S.A. (2011) Weather, water quality and infectious gastrointestinal illness in two Inuit communities in Nunatsiavut, Canada: potential implications for climate change. EcoHealth 8(1), 93-108.

Hill, R.A., Hunt, J., Sanders, E., Tran, M., Burk, G.A., Mlsna, T.E. and Fitzkee, N.C. (2019) Effect of Biochar on Microbial Growth: A Metabolomics and Bacteriological Investigation in E. coli. Environmental Science & Technology 53(5), 2635-2646.

Huggins, T.M., Haeger, A., Biffinger, J.C. and Ren, Z.J. (2016) Granular biochar compared with activated carbon for wastewater treatment and resource recovery. Water Research 94, 225-232.

Inyang, M. and Dickenson, E. (2015) The potential role of biochar in the removal of organic and microbial contaminants from potable and reuse water: A review. Chemosphere 134, 232-240.

Jolliffe, I.T. and Cadima, J. (2016) Principal component analysis: a review and recent developments. Philosophical Transactions of the Royal Society A: Mathematical, Physical and Engineering Sciences 374(2065), https://doi.org/10.1098/rsta.2015.0202

Kranner, B.P., Afrooz, A., Fitzgerald, N.J.M. and Boehm, A.B. (2019) Fecal indicator bacteria and virus removal in stormwater biofilters: Effects of biochar, media saturation, and field conditioning. Plos One 14(9), https://doi.org/10.1371/journal.pone.0222719

Lau, A.Y.T., Tsang, D.C.W., Graham, N.J.D., Ok, Y.S., Yang, X. and Li, X.D. (2017) Surface-modified biochar in a bioretention system for Escherichia coli removal from storrnwater. Chemosphere 169, 89-98.

Liu, J., Yue, P., He, Y. and Zhao, M. (2020) Removal of E. coli from stormwater by bioretention system: parameter optimization and mechanism. Water Science and Technology, https://doi.org/10.2166/wst.2020.2207

Ma, L., Gao, Y., Wang, Z. and Ran, L. (2010) Analysis of the reported infectious diarrhea in China in 2009. Zhongguo Weishengtaxixue Zazhi/Chinese Journal of Microecology 22(7), 658-665.

Manya, J.J. (2012) Pyrolysis for Biochar Purposes: A Review to Establish Current Knowledge Gaps and Research Needs. Environmental Science and Technology 46(15), 7939-7954.

Mohanty, S.K. and Boehm, A.B. (2014) Escherichia coli Removal in Biochar-Augmented Biofilter: Effect of Infiltration Rate, Initial Bacterial Concentration, Biochar Particle Size, and Presence of Compost. Environmental Science & Technology 48(19), 11535-11542.

Mohanty, S.K. and Boehm, A.B. (2015) Effect of weathering on mobilization of biochar particles and bacterial removal in a stormwater biofilter. Water Research 85, 208-215.

Mohanty, S.K., Cantrell, K.B., Nelson, K.L. and Boehm, A.B. (2014) Efficacy of biochar to remove Escherichia coli from stormwater under steady and intermittent flow. Water Research 61, 288-296.

Mohanty, S.K., Torkelson, A.A., Dodd, H., Nelson, K.L. and Boehm, A.B. (2013a) Engineering Solutions to Improve the Removal of Fecal Indicator Bacteria by Bioinfiltration Systems during Intermittent Flow of Stormwater. Environmental Science & Technology 47(19), 10791-10798.

Mohanty, S.K., Torkelson, A.A., Dodd, H., Nelson, K.L. and Boehm, A.B. (2013b) Engineering solutions to improve the removal of fecal indicator bacteria by bioinfiltration systems during intermittent flow of stormwater. Environmental Science and Technology 47(19), 10791-10798.

Mohanty, S.K., Valenca, R., Berger, A.W., Yu, I.K.M., Xiong, X.N., Saunders, T.M. and Tsang, D.C.W. (2018) Plenty of room for carbon on the ground: Potential applications of biochar for stormwater treatment. Science of the Total Environment 625, 1644-1658.

Nabiul Afrooz, A.R.M. and Boehm, A.B. (2017) Effects of submerged zone, media aging, and antecedent dry period on the performance of biochar-amended biofilters in removing fecal indicators and nutrients from natural stormwater. Ecological Engineering 102, 320-330.

Oh, T.K., Choi, B., Shinogi, Y. and Chikushi, J. (2012) Effect of pH Conditions on Actual and Apparent Fluoride Adsorption by Biochar in Aqueous Phase. Water Air and Soil Pollution 223(7), 3729-3738.

Peterson, S.C., Jackson, M.A., Kim, S. and Palmquist, D.E. (2012) Increasing biochar surface area: Optimization of ball milling parameters. Powder Technology 228, 115-120.

Phillips, J.A.S., S.J. (2020) Modeling Improved Performance of Reduced-Height Biosand Water Filter Designs. Water 12, https://doi.org/10.3390/w12051337

Rahman, M.Y.A., Nachabe, M.H. and Ergas, S.J. (2020) Biochar amendment of stormwater bioretention systems for nitrogen and Escherichia coli removal: Effect of hydraulic loading rates and antecedent dry periods. Bioresource Technology 310, https://doi.org/10.1016/j.biortech.2020.123428

Sasidharan, S., Torkzaban, S., Bradford, S.A., Kookana, R., Page, D. and Cook, P.G. (2016) Transport and retention of bacteria and viruses in biochar-amended sand. Science of the Total Environment 548, 100-109.

Shimabuku, K.K., Kearns, J.P., Martinez, J.E., Mahoney, R.B., Moreno-Vasquez, L. and Summers, R.S. (2016) Biochar sorbents for sulfamethoxazole removal from surface water, stormwater, and wastewater effluent. Water Research 96, 236-245.

Suliman, W., Harsh, J.B., Fortuna, A.-M., Garcia-Pérez, M. and Abu-Lail, N.I. (2017) Quantitative Effects of Biochar Oxidation and Pyrolysis Temperature on the Transport of Pathogenic and Nonpathogenic Escherichia coli in Biochar-Amended Sand Columns. Environmental Science and Technology 51(9), 5071-5081.

Sun, K., Kang, M., Zhang, Z., Jin, J., Wang, Z., Pan, Z., Xu, D., Wu, F. and Xing, B. (2013) Impact of Deashing Treatment on Biochar Structural Properties and Potential Sorption Mechanisms of Phenanthrene. Environmental Science and Technology 47(20), 11473-11481.

Sun, M.M., Ye, M., Zhang, Z.Y., Zhang, S.T., Zhao, Y.C., Deng, S.P., Kong, L.Y., Ying, R.G., Xia, B., Jiao, W.T., Cheng, J.Q., Feng, Y.F., Liu, M.Q. and Hu, F. (2019) Biochar combined with polyvalent phage therapy to mitigate antibiotic resistance pathogenic bacteria vertical transfer risk in an undisturbed soil column system. Journal of Hazardous Materials 365, 1-8.

Tan, X.F., Liu, Y.G., Zeng, G.M., Wang, X., Hu, X.J., Gu, Y.L. and Yang, Z.Z. (2015) Application of biochar for the removal of pollutants from aqueous solutions. Chemosphere 125, 70-85.

Tumiran, Kobayashi, S., Maeyama, M., Imada, H. and Saito, Y. (1996) The correlation between surface charge distribution and flashover properties of alumina surface in vacuum. Applied Surface Science 100-101, 238-242.

Ulrich, B.A., Loehnert, M. and Higgins, C.P. (2017) Improved contaminant removal in vegetated stormwater biofilters amended with biochar. Environmental Science: Water Research & Technology 3(4), 726-734.

Valenca, R., Ramnath, K., Dittrich, T.M., Taylor, R.E. and Mohanty, S.K. (2020) Microbial quality of surface water and subsurface soil after wildfire. Water Research, https://doi.org/10.1016/j.watres.2020.115672

Velten, S., Boller, M., Koster, O., Helbing, J., Weilenmann, H.U. and Hammes, F. (2011) Development of biomass in a drinking water granular active carbon (GAC) filter. Water Research 45(19), 6347-6354.

Wilcox, D.P., Chang, E., Dickson, K.L. and Johansson, K.R. (1983) Microbial growth associated with granular activated carbon in a pilot water treatment facility. Applied and Environmental Microbiology 46(2), 406-416.

Xiao, X., Chen, B., Chen, Z., Zhu, L. and Schnoor, J.L. (2018) Insight into Multiple and Multilevel Structures of Biochars and Their Potential Environmental Applications: A Critical Review. Environmental Science & Technology 52(9), 5027-5047.

Zhang, X.D. (2015) Conjunctive surface water and groundwater management under climate change. Frontiers in Environmental Science 3, https://doi.org/10.3389/fenvs.2015.00059

7. CONCLUSION AND RECOMMENDATIONS

7.1. Summary

This dissertation advanced the scientific understanding of how climate could affect pollutant removal in stormwater control measures (SCM). The results inform how to design climate-resilient SCM. Specific conclusions are:

- Climate and design both affect the nitrate removal capacity of SCM, but the extent to which they are important varies with SCM types. Among different SCM, retention ponds provide the best nitrate removal partially because of the long residence time, and their performance is less sensitive to climate. Although bioretention systems remove less nitrate, optimizing their design could significantly improve their nitrate removal capacity and make them more resilient in different climates or seasons.
- Changes in local weather conditions such as an increase in rainfall intensity and temperature affect FIB removal, but the correlation is weak possibly, making any process-based modeling unfeasible to predict the performance of retention ponds. The application of machine learning models utilizing weather conditions as input variables could predict FIB removal in retention ponds with an accuracy of 65%.
- The presence of wildfire residues in surface water reduces the growth of indicator bacteria and accelerates their die-off when compared to unburned soil, suggesting microbial risk post-wildfire is minimal. Thus, wildfire residue may not have any negative impact on microbial water quality because of decrease in subsurface transport and viability of indicator bacteria in surface water relative to natural soil particles. However, the result may have wide implications on other natural processes in nature where native soil microorganisms could be affected by the addition of wildfire residues.

- The deposition of wildfire residues in wetland sediments increases methane emissions depending on the quantity of wildfire residues deposited. The increased methane emissions were partially explained by the generation of hydrogen peroxide in the presence of wildfire residues, coupled with the increase in silica content. Both factors could accelerate the breaking down of complex organic matter and from organic acid precursors for methane production.
- A model relating biochar removal capacity with commonly measured biochar properties was developed based on partial least squares regression, which has the potential to predict the *E. coli* removal capacity of commercially available biochar. The *E. coli* removal capacities of sand-biochar filters were positively correlated with the surface area and organic carbon content of biochar, and negatively correlated with ash content and volatile matter. Thus, the model can help the selection of biochar for stormwater treatment and make the SCM more resilient during changing climates.

7.2. Recommendations

This dissertation showed that stormwater treatment systems are effective in removing a range of contaminants, but their performance can vary with changing climate. Thus, their resilience during changing local climate could be improved using different design features. Their performance is difficult to predict when the weather varies widely. In this case, machine learning techniques should be improved to predict the performance of these systems that account for variability in local weather conditions and stormwater composition. The accuracy percentage of machine learning models would likely improve with an increase in data availability including data related to water composition, accurate water temperature, and UV-light intensity. Thus, a better field data collection protocol and the creation of an extensive, integrated, and multi-functional stormwater database could assist the development of machine learning models and help predict their performance.

The results proved that wildfire residues could limit the growth of bacteria and/or increase the emission of methane from wetlands. Thus, local and federal government agencies should consider the implementation of local stormwater treatment systems that are able to trap wildfire residues and reduce the impact of these particles in downstream water resources. Future studies should examine the effect of wildfire characteristics such as burn intensity, ash types generated, and soil mineralogy on other biochemical functions in SCM.

Finally, this study described a promising model that can predict the performance of biochar in removing pathogens using commonly reported properties of biochar as input variables. The model helps the selection of biochar for water treatment application by the end users and informs the vendors to optimize the production condition such as feedstock type and pyrolysis temperature to produce biochar with the desired quality. The model database should be expanded to increase the accuracy of the model and include other target contaminants such as nutrients and heavy metals.