ENHANCING BIOFILTRATION TECHNOLOGY FOR THE TREATMENT OF WILDFIRE-IMPACTED RAW WATER

by

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Abstract

Wildfire events significantly contribute to the degradation of surface water quality by introducing ashes and unburnt carbon through runoff, resulting in elevated turbidity, increased levels of total suspended and dissolved solids, and higher concentrations of dissolved organic matter (DOM). Biofiltration has emerged as a sustainable and cost-effective water treatment technology, particularly valued for its environmentally friendly ability to remove DOM. This study evaluated the performance of process intensified biofiltration in removing dissolved organic carbon (DOC) from wildfire-impacted raw water using three bench-scale biofilters composed of different media: sand, granular activated carbon (GAC), and a combination of sand and GAC. The influent was pretreated using aeration and a roughing filter. DOC was tested at three levels: 20, 50, and 100 mg/L. Water quality parameters such as pH, turbidity, temperature, alkalinity, ultraviolet absorbance at 254 nm (UVA_{524}), specific ultraviolet absorbance at 254nm ($SUVA_{524}$) – calculated, and microbial activity – adenosine triphosphate (ATP), extracellular polymeric substances (EPS), and dissolved oxygen (DO) were monitored. Results indicate that at 20 mg/L DOC, the sand + GAC biofilter achieved the highest mean DOC removal efficiency of 70 ± 20%, followed by GAC (68 ± 16%) and sand (68 ± 22%). At 50 mg/L, removal efficiencies declined to 54 ± 16% (sand + GAC), 47 ± 19% (GAC), and 33 ± 19% (sand). At 100 mg/L, performance dropped remarkably, with average removals of only 16 ± 26% (sand + GAC), 16 ± 21% (GAC), and 12 ± 22% (sand). UVA_{524} and $SUVA_{524}$ values remained below 2 L/mg·m, indicating the dominance of hydrophilic and low molecular weight organic matter. ATP concentrations peaked at 14 ± 2 mM in the sand biofilter, confirming its superior microbial activity. EPS analysis revealed higher sugar than protein concentrations, with tightly bound EPS peaking at 296 ± 37 mg/g total suspended solids. Elevated influent DOC concentrations were associated with increased pH (up

to 9.52 ± 0.12), which may have negatively impacted microbial activity and DOC removal. Statistical analysis revealed a strong negative correlation between DOC concentration and DOC removal efficiency. Furthermore, analysis of variance confirmed that DOC concentration significantly influenced removal efficiency, while media type did not. However, Tukey post-hoc test showed that at DOC concentrations of 20 and 50 mg/L, the biofilters significantly reduced DOC levels, while at 100 mg/L, no significant reduction was observed, indicating diminished effectiveness at higher concentrations. The findings of this study underscore the robustness and adaptability of biofiltration systems in treating wildfire-impacted water, particularly in the effective removal of DOC and turbidity. By providing a nature-based, cost-effective treatment solution, biofiltration offers a sustainable and climate-resilient strategy for safeguarding drinking water sources against emerging contaminants.

Table of Contents

List of Tables

List of Figures

List of Equations

Acronyms and Abbreviations

ANOVA	Analysis of Variance
AO	Aesthetic Objectives
AOP	Advanced Oxidative Processes
ATP	Adenosine Triphosphate
BDOC	Biodegradable Dissolved Organic Carbon
BOM	Biodegradable Organic Matter
BSA	Bovine Albumin Serum
COD	Chemical Oxygen Demand
DBP	Disinfection By-Product
DO	Dissolved Oxygen
DOC	Dissolved Organic Carbon
DOM	Dissolved Organic Matter
EBCT	Empty Bed Contact Time
EFL	Enhanced Forestry Laboratory
EPS	Extracellular Polymeric Substances
GAC	Granular Activated Carbon
GCDWQ	Guidelines for Canadian Drinking Water Quality
HLR	Hydraulic Loading Rate
KS	Kolmogorov-Smirnov
MAC	Maximum Acceptable Concentration
N-DBP	Nitrogenous Disinfection By-products
NF	Nanofiltration

NOM	Natural Organic Matter
OG	Operational Guidelines
PPCPs	Pharmaceuticals and Personal Care Products
RO	reverse osmosis
SA	Sand-Anthracite
SSF	Slow Sand Filtration
TOC	Total Organic Carbon
TSS	Total Suspended Solids
$SUVA_{524}$)	Specific Ultraviolet Absorbance at 254 nm
UVA	Ultraviolet Absorbance
UVA_{524}	Ultraviolet Absorbance at 254 nm
UVR	Ultraviolet Radiation
WTP	Water Treatment Plant

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Contributions

The following academic contributions emerged from this MASc work:

1. Ali, A.A., Iorhemen O.T., Thring R.W. (2025). Climate adaptation and resilience of biofiltration as a low-cost technological solution for water treatment – A critical review. *Cleaner Water.* 3: 100062.
2. Ali, A.A., Nabaterega, R., Thring, R.W., Iorhemen, O.T., Process-enhanced water biofiltration for the treatment of wildfire-impacted raw water. *in preparation.*

Details of contributions from the candidate and co-authors are listed below:

1. In this publication, the candidate conducted extensive review of related literature on climate change and water, water treatment technologies that are resilient to climate change, biofiltration as low-cost techno-ecological nature-based solutions for water treatment, and resilience and adaptation of biofiltration to climate change, manuscript writing, and proofreading. Dr. Oliver Iorhemen provided significant contributions in terms of review and editing during manuscript preparation. Dr. Ronald Thring provided significant contributions in terms of technical guidance and review during manuscript preparation.
2. In this article, the candidate designed and performed all the experiments except the ones stated below to have been conducted by others. The candidate also collected and analyzed experimental data, wrote the first draft of the manuscript, revised the manuscript, and wrote the final manuscript. Dr. Resty Nabaterega contributed to writing the first draft of the manuscript, and proof reading, data analysis in Minitab software, research guidance, scientific input and comments during manuscript write up. Dr. Oliver Iorhemen and Dr. Ronald Thring contributed to research guidance, scientific input and comments during manuscript write up, and proof reading.

Chapter 1 Introduction

1.1. Background

The quality and quantity of water are of vital significance for human existence. Both human and environmental health have constantly been threatened by water pollution resulting from increasing population, industrial development, and climate change [1], [2]. Over the years, climate change has shown a direct effect on surface water quality. Extreme events such as wildfires, rising temperatures, and changes in precipitation patterns pose concerning threats to drinking water. Wildfires increase erosion, resulting in increased total suspended solids (TSS) and turbidity in surface waters [3], [4], [5]. This increases the chance of unburnt organic matter transport from the terrestrial ecosystem to nearby water bodies. The presence of unburnt organic matter in water bodies results in increased concentrations of dissolved organic matter (DOM) in surface waters [4]. Additionally, the increase in temperature and precipitation, posed by climate change, has resulted in increased ultraviolet radiation (UVR) exposure to humans [6] and transportation of contaminants into surface waters [3], [7]. The carcinogenic effects of UVR have resulted in the frequent use of sunscreens to reduce the risk of skin cancer [8]. These sunscreens end up in our surface water sources.

Current water treatment plants (WTPs) in low resource contexts struggle to remove these elevated levels of DOM, turbidity, and pollutant load of the raw water sources, thereby increasing energy costs and impairing efficiency of WTPs [9]. Despite the challenges caused by climate change, biofiltration systems have a strong potential to increase resiliency. Biofiltration, a low-cost, nature-based technological solution, has emerged as a viable strategy for improving water quality. This innovative water treatment technology makes use of natural processes to remove pollutants present in various raw water sources. Biofiltration offers a cost-effective and

environmentally friendly approach by utilizing microorganisms. However, climate change threatens the effectiveness of biofiltration, making it necessary to understand the resilience of this sustainable solution to climate change.

Research has demonstrated that biofiltration offers a sustainable solution by effectively removing dissolved organic carbon (DOC) and turbidity in wildfire-impacted water [6], [10], [11], [12], [13], [14]. Blackburn et al. [6] explored the resilience of biofiltration to wildfire ash-associated organic carbon threat to potable water treatment using slow sand filtration (SSF) as a biofilter operated at room temperature and an extended empty bed contact time (EBCT) of 10h. Results showed an average of ~20% DOC removal and turbidity reduced to ≤0.3 NTU. However, there has been limited research on the robustness of biofiltration for high DOC concentrations. Therefore, the current study applied biofiltration to enhance its treatment effectiveness for wildfire-impacted raw water at varying DOC concentrations.

1.2. Research questions

The proposed research sought to address two research questions focused on enhancing the treatment performance of biofiltration in the face of a changing climate. These include:

a) How does media type (adsorbing and non-adsorbing) affect biofiltration performance?

b) What is the impact of varying DOC on the treatment performance of biofiltration?

Answering these questions provided insights into the optimal conditions for the operation of biofiltration as a climate-adaptive and resilient water treatment technology.

1.3. Research objectives

The primary objective of this research was to enhance biofiltration, a nature-based treatment system, for the treatment of wildfire-impacted water. To achieve this, the study focused on two specific objectives:

a) To determine the impact of media type (adsorbing and non-adsorbing) on biofiltration performance.
b) To determine the influence of varying DOC concentrations on the treatment performance of biofiltration

These objectives provided actionable insights for enhancing biofiltration efficiency for the treatment of wildfire-impacted water.

1.4. Organization of the thesis

This thesis is structured into five chapters. Chapter 1, the current chapter, introduces the background, the impacts of climate change on water quality and water treatment technologies, the resilience of biofiltration in treating wildfire-impacted raw water, discusses the research question and highlights the objectives of the research.

Chapter 2 covers an in-depth literature review of the impacts of extreme weather events on water quality and the operations of WTPs. Several conventional water treatment methods were highlighted, and their limitations in treating emerging contaminants were discussed. The mechanisms through which biofiltration removes contaminants as well as the key parameters that influence biofiltration, such as biofilter media, types of microorganisms, temperature, pH, nutrient supply, etc., and the dominant microbes present in biofilters were reviewed. The adaptation and resilience of biofiltration systems to challenges posed by climate change in water treatment were

extensively discussed. Finally, the limitations and opportunities related to the adaptation and resilience of biofiltration were discussed, emphasizing the need for more proactive measures to optimize biofiltration systems

Chapter 3 outlines the experimental procedures for the treatment of wildfire-impacted raw water using biofiltration. The chapter details the use several experimental runs to understand the relationships between variables and their effects on DOC removal. In addition, analytical procedures performed during the experimental period were discussed in full detail. Outlier and normality tests, as well as Spearman's correlation analysis were used to highlight the various correlations between each factor and DOC removal efficiency. This combined approach aimed to identify optimal conditions for wildfire-impacted raw water treatment for further optimization studies.

Chapter 4 presents and interprets the experimental findings from this study. It begins with an analysis of key water quality parameters, including dissolved oxygen, temperature, turbidity, pH, and alkalinity, emphasizing their influence on biofilter performance. Organic matter characteristics through ultraviolet absorbance at 254 nm (UVA_{524}), specific ultraviolet absorbance at 254nm ($SUVA_{524}$), and DOC removal trends were explored, linking these to microbial activity and treatment efficiency. Microbial dynamics are further examined using adenosine triphosphate (ATP) concentrations and extracellular polymeric substances (EPS) composition. The discussion integrates these findings with existing literature to contextualize the observed trends and variations.

Chapter 5 summarizes the research findings on the impact of each experimental factor on treating wildfire-impacted water. This chapter compares the previous studies biofiltration systems

for drinking water with this study, and it concludes with recommendations for future research directions. The references supporting this thesis are listed at the end.

Chapter 2 Literature Review

2.1. Synthesis of literature review on biofiltration application to water treatment

2.1.1. Literature selection

A comprehensive search was conducted to identify peer-reviewed articles, technical reports, conference papers, and relevant books that discuss biofiltration as a water treatment technology and its resilience and adaptation to climate change. A systematic search on reputable academic databases, including Google Scholar, Web of Science, ScienceDirect, Scopus, and Semantic Scholar, was conducted for pertinent peer-reviewed journal publications, with additional data obtained through forward citation elimination. The focus was on journal articles published within the years 2000 – 2023. Keywords such as “bio-filtration”, “climate change”, “resilience of biofiltration”, “water quality”, and “water treatment” were used within the "Topic" field. To narrow the results down, Boolean operators were used in the search queries and the keywords included “biofiltration AND water treatment”, “biofiltration of drinking water” “climate change AND biofiltration” “climate change AND water quality OR water treatment plants”, “removal mechanisms in biofiltration during water treatment”, “factors influencing biofiltration”, “resilience AND biofiltration AND climate change”, “adaptation AND biofiltration AND climate change”, “resilience of biofiltration to climate change”. The search was refined to include only English-language documents, peer-reviewed journal articles and scientific reports. Only journal articles with relevant titles and abstracts were further reviewed if they discussed biofiltration as a water treatment technology with a focus on its resilience and adaptation to climate change. 934 articles

were chosen for further analysis, among which 64 were review articles, 153 were proceeding papers, and the rest were research articles (Figure 2.1). From this subset, 87 articles were specifically cited for their relevance to biofiltration as a water treatment technology in the face of climate change. Figure 2.1 shows the literature selection procedure on Web of Science.

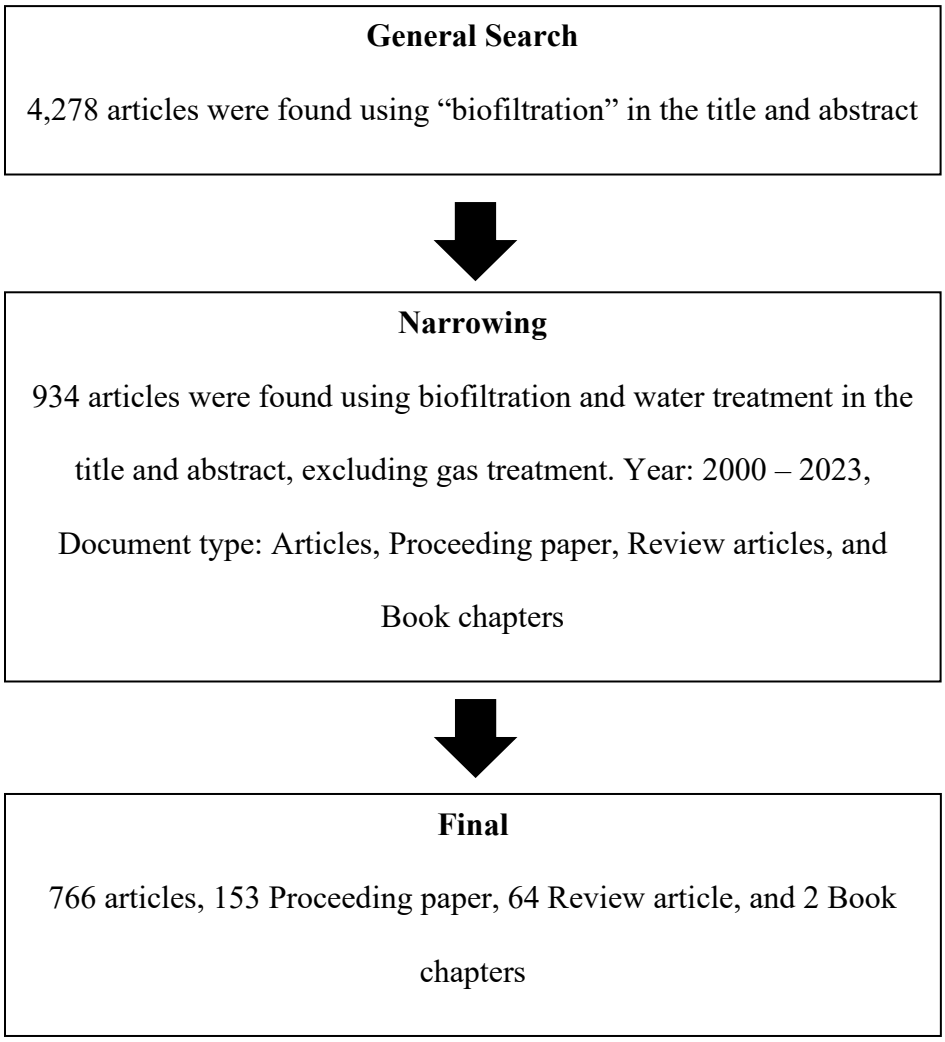

Figure 2.1: Flow chart for the overall procedure in literature selection.

2.1.2. Bibliometric overview

A total of 934 documents were retrieved by focusing on biofiltration in water treatment within 24 years. The data extracted was grouped and arranged in a chronological order to identify the

trends and the annual growth of the publications from 2000 to 2023. Figure 2.2 shows the annual publications of research articles on biofiltration for water treatment indexed in Web of Science in the last 24 years. Based on Figure 2.2, the oldest publication on biofiltration for water treatment began in 2000, and there was a total of 18 publications in that year. It clearly shows that the research trend in the first six years (2000 – 2006) did not receive very high attention, as the annual publications for each year are lower than 20. The total publications within 2000 – 2006 were 99, accounting for only 10.5% of the total cumulative publications from the year 2000 to 2023.

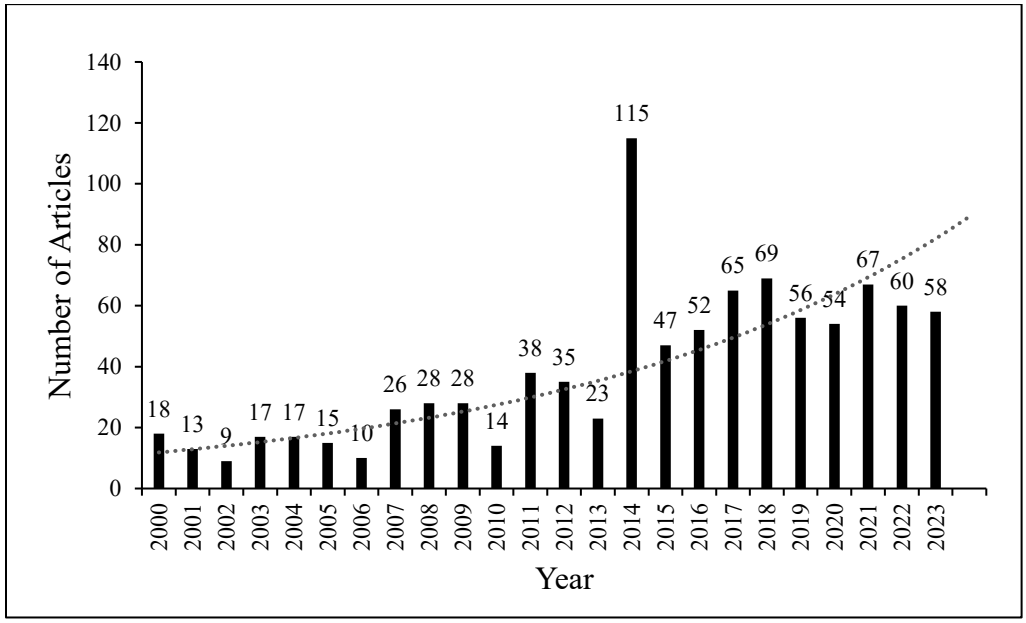

Figure 2.2: Number of peer-reviewed articles on biofiltration of drinking water since year 2000.

The strong interest in biofiltration for water treatment started from 2007, as there was a significant and rapid increase in the total publications from that year onwards. Between 2007 and 2014, the annual publications increased from 26 to 115 in 2014, which resulted in approximately 440% growth, the highest during this period. Moreover, the steady non-linear increase of the

cumulative numbers of publications also indicates that the research trend for the annual publications will continue to rise in the future.

2.1.3. Presentation structure

This review offers an extensive examination of the impact of climate change on water quality and treatment, biofiltration as a treatment technology, and the resilient and adaptive nature of biofiltration to climate change. It also highlights the important factors that affect biofiltration systems. The review procedures and bibliometric analyses are summarized in sections 2.1.1 and 2.1.2. Section 2.2 discusses the impacts of climate change on water quality and WTPs. Section 2.4 explores several water treatment technologies and their limitations in treating emerging contaminants. Section 2.5 delves into biofiltration as a treatment technology, explains the removal mechanism, and highlights the important factors that influence the removal efficiency of biofiltration in water treatment. Section 6 outlines resilience and adaptation of bio- filtration to climate change.

2.2. Climate change and water

2.2.1. Impacts of climate change on water quality

The consequences of climate change on water quality are threatening. The major factors that influence water quality are increased temperature, wildfires, and extreme hydrological events such as floods and droughts [4], [5], [15]. It is no coincidence that climate change and the hydrological cycle are connected [16]. Usually, human activities cause water to be polluted, but climate change, in the form of increased temperature and changes in the precipitation cycle, results in surface water quality degradation. Climate change, primarily through warming and extreme events, contributes to alterations in water quality parameters such as basic physico-chemical properties, DOM, nutrients, and biological parameters [15], [17].

An increase in water temperature can affect the solubility and reactions of substances in water, leading to higher concentrations of dissolved substances but lower concentrations of dissolved gases, notably oxygen [15]. Rivers in Europe, specifically the Rhine and Meuse, experienced an average temperature increase of around 2◦C following the severe drought of 2003, along with pH changes and a decrease in oxygen solubility [18]. It was observed that increased DOC and biodegradable organic matter (BOM) assimilation by microorganisms contributed to the decrease in dissolved oxygen (DO) [19]. Additionally, floods and droughts can result in the dilution or concentration of dissolved substances [15], [16], [20], impacting parameters such as DO and pH, and affecting nutrient loads through runoff.

Water bodies are influenced by weather seasonality, impacting nutrient patterns, and a warmer climate is anticipated to indirectly increase nutrient loads in surface and groundwater, counteracting policies aimed at reducing external nutrient loading [18], [21]. Soil warming may result in elevated nutrient releases, runoff, and erosion, affecting pollutant transport, particularly after drought periods. Mechanisms such as increased mineralization and runoff following droughts and heavy rainfall mobilize nutrients from the soil of farmlands to surface water, thus leading to speedy bacterial growth [15], [16], [22]. For instance, in Lake Taihu, China's third-largest freshwater lake, higher temperatures and lower wind speeds encouraged the growth of algae blooms after nutrients were washed into the lake during tropical cyclones [23]. Cyanobacteria, which can produce harmful toxins, benefit from warmer water temperatures and nutrient influxes, leading to more frequent and intense blooms [15], [17]. Another example is the case of hypoxia in Pamlico Sound, North Carolina, USA. After several hurricanes, many nutrients, freshwater, and organic carbon were delivered into the United States' second-largest estuarine system [24]. The effects of weather on nutrient loading and circulation of water have also been linked to the severity

of summer hypoxia [16]. Higher water temperatures support the survival and spread of certain waterborne diseases and pathogens [15].

The impact of wildfire on water quality was evaluated in the sub-regions of the Boreal ecozone in Canada [25]. There was an increase in total phosphorus, inorganic nitrogen, and algae biomass in the Boreal Plain lakes with burned watersheds. An increase in DOC and water colour was also observed on Boreal Shields. The effects were associated with climate change, lake characteristics, and lake drainage ratio. After a wildfire, an increase in erosion often leads to increased turbidity and TSS in surface water, as well as the concentration of DOM [26]. This creates concern as DOM can react with disinfectants during water treatment and form disinfection by-products (DBPs) [27]. An increase in organic nitrogen in stream water after wildfire may enhance the formation of nitrogenous disinfection by-products (N-DBP) and di-halogenated acetic acid.

Climate models have predicted reduced precipitation and increased temperature for the Mediterranean region by the end of the 21st century[28], [29], [30]. An increase in runoff and precipitation intensity results in an increase in turbidity, thereby elevating the level of organic matter and pollutants in water sources. In an evaluation study of the impact of climate change on drinking WTP operations, the future (2050) water quality of the Ohio River was predicted by modifying its 1998 baseline data [31]. The DOC, total hardness, and alkalinity of the river were estimated to increase or decrease by + 0.036, -0.22, and + 0.03 mg/L, respectively, per year. However, ammonia and temperature were estimated to increase by 25 % and 2°C, respectively. Ultraviolet Absorbance (UVA), pH, turbidity, and calcium were estimated to remain the same by 2050. By the end of the century, climate change is projected to lead to higher concentrations of water quality indicators in raw water in Norway, particularly due to an increase in rainfall [32]. This is expected to cause a deterioration in the quality of drinking water, with the most significant

effects anticipated in the Western and Northern parts of the country. In a situation where climate change continues to alter weather patterns, degradation in water quality will persist. Table 2.1 shows changes in water quality of different sources over the years.

Table 2.1: Trends of water quality parameters over the years

Water Source	Parameters	Start Year Average	End Year Average	References
Xiangcheng WTP	Temperature (℃)	16.7	18.1	[33]
2012 – 2015	Turbidity (NTU)	25.3	104.5	
	NH_4^+ - N (mg/L)	0.22	0.769	
	COD_{Mn} (mg/L)	2.6	2.6	
Barekese Catchment	Temperature (℃)	26	32	[34]
2000 – 2019	Rainfall (mm)	76	60	
	pH	6.4	9.4	
	Turbidity (NTU)	20	50	
Jinpen Reservoir	TP (mg/L)	0.02	0.09	[35]
Nov 2013 – Oct 2014	TN (mg/L)	1.3	1.5	
	TOC (mg/L)	2.6	2.7	
	Fe^{3+} (mg/L)	0.18	0.3	
	Mn^{7+} (mg/L)	0.07	0.08	
	NH_4^+ - N (mg/L)	0.18	0.46	
	Total Ammonia (mg/L)	n/a	0.05	[36]

Cerro Grande	TP (mg/L)	n/a	0.38	
Northern New Mexico, USA	Total Cyanide (µg/L)	n/a	<2.8	
	Total Calcium (mg/L)	n/a	245	
Silver Creek	Total Ammonia (mg/L)	n/a	<0.005	[36]
British Columbia, Canada	TP (mg/L)	n/a	0.047	
	Total Cyanide (µg/L)	n/a	0.8	
	Total Calcium (mg/L)	n/a	25.8	
Water Source	**Parameters**	**Unburned watershed**	**Burned watershed**	
		Grand Rapids	**Fort McMurray**	
Athabasca River	Total Lead (mg/ha)	8.1	10.3	
Grand Rapids & Fort McMurray, Canada	TN (g/ha.d)	3.8	3.3	[37]
	TP (g/ha.d)	0.54	0.45	
June 2016 – August 2016				

2.2.2. Impacts of climate change on water treatment

Climate change influences the design and operation of WTPs just as it affects raw water quality. The deterioration in raw water quality can lead to the inefficiency of a WTP and cause the treatment plant to fail to satisfy water quality standards [31]. For this reason, modification may be necessary in WTPs [38]. For example, Li et al. [31] used Monte Carlo simulations to show that the Richard Miller plant located in Ohio, USA would be able to treat water using the existing treatment processes by 2050. Although the total organic carbon (TOC) would be increasingly high, it could be managed by adjusting some operational parameters. The research also showed that the expected average operational cost would rapidly increase due to the strict performance in terms of TOC [31], [39]. In addition, increased levels of indicator bacteria, turbidity, and colour in raw water caused by climate change could complicate water treatment processes [32]. The impact assessment applied to Grafham WTP indicates that future scenarios could lead to increased nitrate levels and algal blooms, with potential consequences for WTP operations, including increased energy costs for aeration to prevent stratification and the need for additional treatment processes to adhere to drinking water standards [17].

Warmer temperatures can also cause more drastic weather, leading to hurricanes, heavy winds, cyclones, hail, floods, and thunderstorms, which could potentially cause damage to water treatment infrastructure. The United Nations International Panel on Climate Change predicted a sea level increase of 18 – 58 cm in the 21st century, which does not include future dynamic change in ice flow [40]. This increase in sea level will affect WTPs located close to water bodies influenced by tides. Additionally, wildfires reduce vegetation, allowing more precipitation to reach the land surface [41]. Exposure of the land surface to rainfall increases erosion and runoff. Wildfire-impacted waters are rich in pollutants such as

heavy metals, nutrients and natural organic matter (NOM), and the long-term release into the water column drastically reduces the quality of water and affects the performance of WTPs [26], [27], [42]. Furthermore, the rise in particulate level, NOM and DOC concentration affects the coagulation process in conventional WTPs because the coagulant demand is increased and its effectiveness is compromised [6], [26], [27]. Thus, this increases the costs of infrastructure and operation, disrupts services and causes outages [26], [43]. Table 2.2 highlights past and predicted climate events and their impacts on water quality and WTPs.

Table 2.2: Summary of the impact of climate change on water quality and WTPs

Climate events (past and future predictions)	Impacts on Water Quality and WTPs	References
Series of hurricanes in Pamlico Sound, North Carolina, USA.	Nutritional loading and circulation in water led to extreme cases of summer hypoxia.	[24]
Wildfire in Boreal ecozone in Canada.	Increase in TP, N, and algae biomass in Boreal Plain lakes and increase in DOC and water colour in Boreal shields.	[25]
Increased sea level and tides in the 21st century.	Flood and damaged water treatment facilities located close to water bodies.	[40]
Hurricanes, cyclones, hailstorms, and thunderstorms.	Heavy winds and floods destroy water treatment infrastructure	[44]
Lowered precipitation and increased temperature in the Mediterranean region at the end of the 21st century.	Increase in turbidity, organic matter, and pollutants in water sources	[45]
Future water quality of Ohio River, USA in 2050.	Increase in water hardness, DOC, alkalinity, ammonia, and temperature.	[31]
Warmer temperatures and lower wind speeds in Lake Taihu, China.	Growth of blooms on the lake after nutrients from vegetation were washed into the Lake.	[23]
Warmer temperatures.	Boost biodegradation and enhance pollutant removal efficiency	[46], [47]
High-intensity rainfall or increased precipitation.	More volumes of raw water increase WTP energy cost	[46]
Wildfire	Increases NOM, nutrients, and metals, thus reducing water quality and increasing operational costs, disrupting services, and causing outages.	[26] [43]
Wildfire	Increases erosion and runoff by reducing vegetation	[41]

2.3. Drinking water treatment technologies resilient to climate change

Drinking water treatment methods adopted for treating water depend on the quality of the raw water. This section highlights and compares some of these methods.

2.3.1. Conventional filtration

Filtration using media such as sand is a traditional treatment method that removes fine particles and dissolved organic matter from raw water as it passes through a porous bed of filter medium [48], [49]. Conventional filtration includes screen filters, slow and rapid sand filtration, diatom filters, and charcoal filters [50]. Conventional filtration alone has been demonstrated to be insufficient in treating water to the required effluent quality. To remove diclofenac from potable water, traditional sand filtration was compared to GAC filtration [50], [51]. While sand filtration was insufficient in removing diclofenac, GAC filtration achieved over 99.7 % of removal. GAC filtration works efficiently for micropollutant removal, but its application is expensive, which is concerning in underdeveloped and developing countries [49], [52]. Sabogal-Paz et al. [53] conducted another study examining the impact of flow configuration in a slow sand filter, specifically comparing intermittent and continuous flows in a household system. It was found that the flow configuration of a slow sand filter alone is insufficient for effective treatment, as it fails to adequately remove organic foulants. The limitations of conventional filtration necessitate the need to consider alternative treatment methods.

2.3.2. Adsorption on activated carbon

Adsorption, a cost-effective and efficient advanced water treatment technology, involves the separation of pollutants (adsorbates) in raw water (liquid phase) and their adsorption onto the adsorbents [54], [55]. The removal capacity of adsorbents is dependent on material and bulk densities, porosity, surface area, pore-size distribution, and operational parameters such as

temperature, pH, and contact time [54], [55], [56]. Activated carbon is the most used material because of its high specific surface and porosity, which makes it efficient in removing contaminants [57]. Nano adsorbents and engineered nanomaterials have been developed recently to enhance the removal of emerging pollutants [55]. Research have shown that activated carbon adsorption is efficient in treating emerging pollutants which have been in the increase in the phase of a changing climate [58], [59]. In a study to improve the removal of estrogenic and pharmaceutical compounds from river water polluted by sewage effluent using full-scale GAC, significant reduction in pharmaceutical compounds – mebeverine (84 – 99 %) as well as carbamazepine and propranolol (17 – 23 %) [58] was achieved. In the determination of the adsorption capacity of ciprofloxacin on three types of carbon-based materials: activated carbon, carbon nanotubes, and carbon xerogel, Carabineiro et al. [59] demonstrated that activated carbon selectively removes emerging pollutants present in raw water. However, there is a gap in the current knowledge about the influence of other parameters on the performance of adsorption systems and a lack of understanding of scale-up parameters. Additionally, most of the research studies focused on lab-scale tests, which do not provide insight into the feasibility of full-scale processes.

2.3.3. Coagulation and flocculation

Coagulation and flocculation, which are traditional water treatment methods, when combined, have proven to be highly effective in removing suspended particles [54]. These processes precipitate metal hydroxides, remove suspended particles and inorganic contaminants present in raw water [49], [52], [54]. The primary objective is to neutralize and destabilize small particles, facilitating their collision to form agglomerates [60]. The small particles in water are mostly negatively charged, repelling each other and preventing agglomerate formation. Neutralization,

which is crucial in the removal of these stable particles, is achieved using chemical agents known as coagulants. The most used coagulants are aluminum sulfate and iron salts, such as ferric sulfate or ferric chloride [54]. These coagulants carry a positive charge that attracts the negatively charged particles, leading to the formation of new neutral particles. Snyder and Kim's research removed 30 % of dichlorodiphenyltrichloroethane and 70 % of benzo(a) pyrene in bench-scale coagulation, flocculation and sedimentation experiments [61]. Although coagulation and flocculation are efficient in removing organic compounds, they are less effective in removing polar and semi-polar pharmaceuticals and pesticides [52], as well as organic micropollutants not attached to agglomerated particles [49].

2.3.4. Membrane processes

Membrane processes are effective in the removal of solids, organic, and inorganic particles from raw water using materials that support the separation of contaminants (retentates) from water (permeate) [55], [62]. The materials' characteristics, such as hydrophobicity and pore size, used in the production of membranes determine the type of contaminants that can be retained [55]. Several membranes such as microfiltration, ultrafiltration, nanofiltration (NF and reverse osmosis (RO) can be used for potable water treatment [49], [54], [55], [62]. The type of membrane used for treatment is dependent on the raw water quality, targeted contaminant to be removed and the drinking water treatment requirements [55]. Several researchers have used membrane processes to achieve denitrification of nitrate, ammonia removal, and TOC reduction [63], [64]. In an investigation of nitrate removal from polluted groundwater using two-stage anoxic/oxic biofilm membrane bioreactor, Ravnjak et al. [63] achieved 99 % nitrate conversion at a residence time of the liquid phase equal to 2.5 h. In another study, a laboratory-scale membrane bioreactor using a submerged polyethylene hollow-fibre membrane module was used to treat a raw water source

slightly polluted by domestic sewage [64]. Results showed that there was a significant reduction in the concentrations of TOC, NH_3-N, turbidity, and total coliforms. Although NF and RO are efficient in treating pesticides, pharmaceuticals and personal care products (PPCPs), and toxic metals [55], their major shortcoming is membrane fouling [49], [50], [54]. Membrane fouling reduces the efficiency of membrane and shortens their life span [49]. Membrane fouling requires chemical cleaning which increases operational cost [49], [54]. Additionally, pollutants retained on membranes are transferred, not degraded [55], [65]. Thus, requiring secondary treatment and disposal.

2.3.5. Advanced oxidative processes and disinfection

Advanced oxidative processes (AOPs) are now commonly applied for water treatment. AOPs involve the generation of hydroxyl radicals for continuous oxidation of organic and inorganic pollutants available in raw water [50], [54], [66]. Hydroxyl radicals are potent, non-selective chemical oxidants that oxidize most water-based compounds through two mechanisms [54]. The radical first breaks the double bond between two carbon atoms and adds to them. In the second mechanism, the radical reduces the amount of hydrogen bonded to the carbon. Key AOP methods include ozone, hydrogen peroxide, UV light, combining Fenton's reagent with hydrogen peroxide, combining ozonation and hydrogen peroxide, and using UV irradiation with hydrogen peroxide [54], [55]. AOPs are versatile and can address concerns such as color removal, oxidation of synthetic organic chemicals, and taste and odor control in water treatment [50], [67]. Also, they can degrade emerging contaminants such as pharmaceuticals [54]. However, the complexity of the chemical reactions involved in AOPs makes designing an optimal treatment system challenging. They are cost and energy intensive and not sustainable for low- and middle-income countries [50], [52], [55]. Additionally, AOPs produce toxic secondary effluents and DBPs from the degradation

of contaminants when chemical oxidants are used [49]. Further- more, small concentrations of chemical oxidants can be found in the treated effluents [55].

2.4. Biofiltration as a low-cost techno-ecological nature-based solution for water treatment

2.4.1. Biofiltration as a water treatment technology

Biofiltration is an established, chemical-free, low energy, and efficient biotechnology used in removing pollutants present in water [68]. It is the process by which a filter, with attached microbial biomass on the surface, is used to remove suspended and fine particles and degrade dissolved organic compounds through physicochemical and biological processes [11], [68], [69]. Although it was initially a biological air pollution control technology with a reported removal efficiency of 95–99 % of volatile organic compounds, it is now applied for the purification of surface waters for potable use [68], [70]. The use of biofilters for potable water treatment started after the discovery of the regrowth of microbial mass on the inner surface of pipe- lines used in distributing water [68], [70]. This microbial regrowth results from pollutants such as BOM, ammonium, sulphur, and DBPs in water.

The primary purpose of biofiltration in water treatment is to produce biologically stable and potable water that is clear of microbial growth during distribution. It also controls the formation of DBPs. Research has shown that biofiltration is efficient in reducing the content of BOM and electron donors that cause biological instability in pipelines [70]. The capacity of biofiltration to remove organic compounds that support the growth of microbial mass in distribution systems makes it a salient technology in water treatment. This technology reduces the need and use of chemicals in water treatment, thereby making the application of biofiltration a green and sustainable engineering technology [11], [71]. It is also a cost-effective, and eco-friendly

conventional contaminant removal method, compared to other methods such as chemical coagulation, membrane separation, ion exchange, ozonation, and many more [71].

2.4.2. Pollutant removal mechanism in biofiltration during water treatment

In biofiltration, the primary treatment mechanisms are filtration (physical separation), adsorption, and biodegradation [68], [70], [72]. A nutrient-rich rough layer gradually formed on the filter media by suspended particles attracts microorganisms, which colonize it, forming biofilms on the filter media surface over time [68]. When raw water is made to pass through a biofilter, organic matter which serves as carbon to the microorganisms, is absorbed into the biofilm. Using the absorbed organic matter as a carbon source, the microbes grow well to form colonies that end up degrading the other contaminants [71]. The elimination of inorganic contaminants that are not biodegradable primarily depends on their adsorption onto the filter media or biofilms [14]. GAC has stronger adsorptive capacity compared to non-adsorptive media such as anthracite and sand. Some contaminants can be adsorbed onto the GAC; however, the adsorption capacity is limited and can become exhausted, leading to the breakthrough of adsorbable compounds [72]. Less biodegradable substances may also desorb from the GAC. After a period of acclimation, which can vary in length depending on the contaminant and the source of the filter media, biodegradation and biotransformation become the main removal mechanisms [72], [73], [74]. During acclimation, microorganisms grow on the filter media, forming a biofilm that degrades contaminants. This process occurs on the media surface and sometimes within macropores [72]. Understanding the natural behaviour of microorganisms and their biochemical reactions is vital in designing and operating biofiltration systems for water treatment. Biofiltration has also demonstrated its capability of removing micropollutants through microbial degradation

rather than physical straining. Figure 2.3 provides a visual overview of the operational principles of biofiltration.

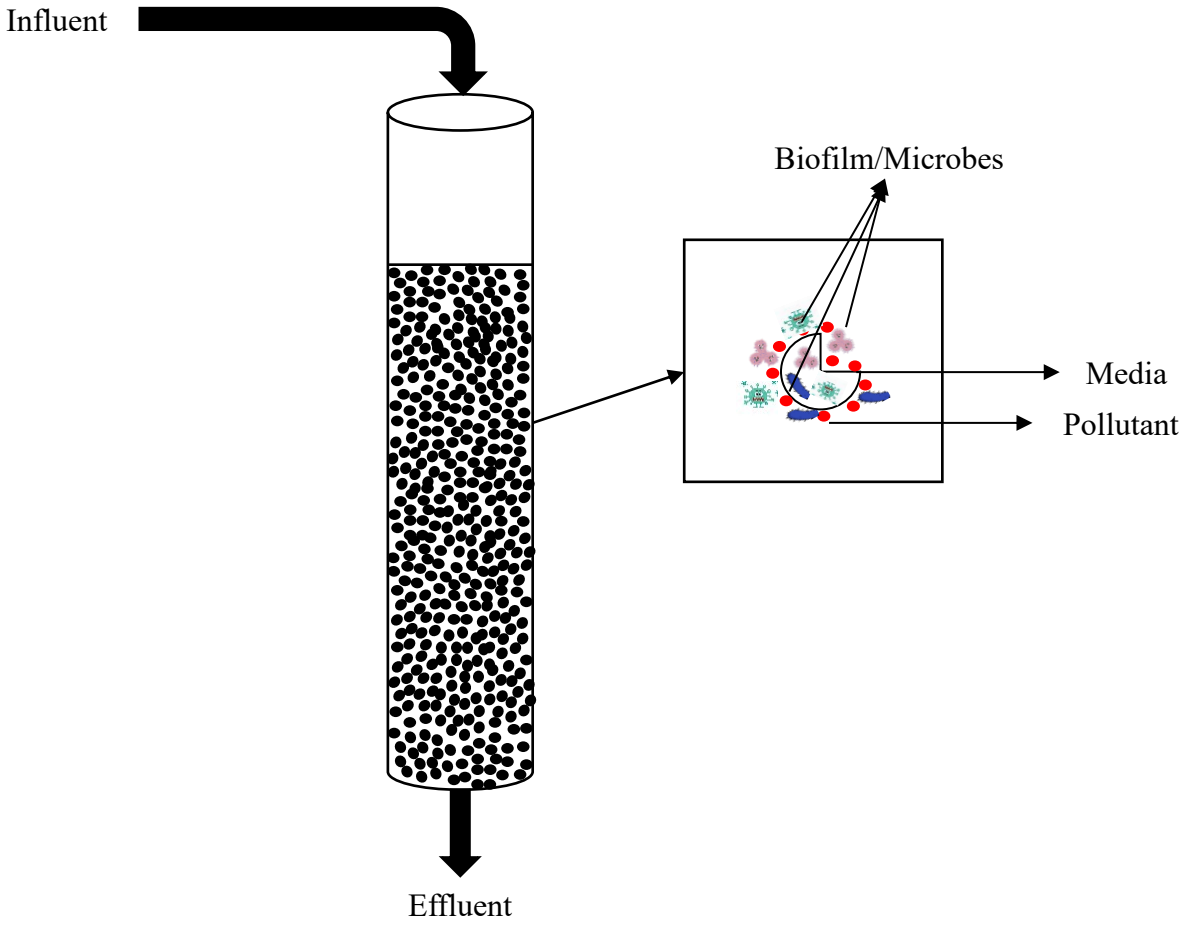

Figure 2.3: Mechanism of biofiltration systems.

2.4.2.1. Attachment of microorganisms

For biofilm development on the filter media, microorganisms are transported to the surface of the filter media through either natural attachment or artificial immobilization [71]. To facilitate the natural attachment process, nutrients are introduced to the filter media to support the accumulation of a microbial community [68]. For artificial immobilization, the movement of the microbes is controlled through diffusion, convection, gravity sedimentation, or the active mobility of the microorganisms [14]. Bacteria by-products, EPS, which also support molecular distribution and

conversion of DOM to smaller molecules, are used by microorganisms to adhere to the filter media [14], [75]. Microbes are immobilized in the biofilter bed to achieve the best production rate of microbial products through improved metabolism and increased loading cells. The attachment of microorganisms to the filter media is the most critical parameter that governs the biofiltration process. It has been suggested that a microorganism's attachment and colonization depend on the bulk fluid, cell structure properties, and substratum or filter media surface properties [70], [76]. The substratum's properties, such as texture, roughness, and hydrophobicity, can enhance attachment by preparing specific cell surfaces to be drawn to the media. Furthermore, bulk fluid conditions, including nutrient levels, flow velocity, pH, and the presence of inhibitors, can encourage cell attachment and microbial colonization of the media due to the availability of food and protection from the surrounding environment [76].

2.4.2.2. Growth of microorganisms

Once attached to the media, the microorganisms slowly develop on the surface of the biofilter media to form a slime layer known as a biofilm. The microbes secret EPS which contains proteins and polysaccharides as the major components as well as other components including uronic acids, lipids, humic acids and nucleic acids [77]. Polysaccharides instigate covalent bonds during colonization and form an active layer on the biofilter surface [71]. This active layer is responsible for capturing the contaminants [71]. The development could take up to several months, depending on the organic concentration of the influent. As explained by Fick's first law, the substrate moves to the outer surface of the biofilm from the bulk liquid, where it diffuses into the biofilm for its metabolism. Factors such as mass transfer of substrate to the biofilm, substrate diffusion into the biofilm, and active utilization within the biofilm affect the substrate utilization rate. Fick's second law then describes the mass transfer of substrate to the biofilm, while Monod's expression

describes the substrate utilization. Biomass growth could also be affected in an unsteady condition [70].

2.4.2.3. Decay and detachment of microorganisms

The success of a biofilter depends on the preservation and detachment of the biomass attached to the biofilter media. Biomass is detached using grazing, erosion, filter backwashing, abrasion, or sloughing. Fluid shear causes biomass erosion, while abrasion is the collision of an outer particle to scrape off the bio cell from the surface. Sloughing detaches biomass in large patches, and some sections of the biomass on the outer shell get lost to protozoa grazing. Most studies have focused on biomass detachment due to shear [70], [78].

2.4.3. Factors influencing biofiltration

Several factors (biological, chemical, and physical) may influence the removal efficiency of biofiltration in water treatment applications. These factors include type and concentration of microorganisms, type of filter media, nutrients supply, operating temperature, empty bed contact time (EBCT), pH, hydraulic loading rate (HLR), frequency of back- washing and filter media replacement [68], [71], [79].

2.4.3.1. Type and concentration of microorganisms

The major component in the biofiltration process are the microorganisms. They are crucial contributors to the removal and degradation of pollutants. Additionally, they play a significant role in the co-metabolism of diverse organic micropollutants [68], [80]. The microbial community formed on the filter media comprises nitrifying bacteria, methane-oxidizing bacteria, iron-oxidizing bacteria, and heterotrophic bacteria [68]. Although these catalytic microorganisms include viruses, fungi, protozoa, and bacteria [71], heterotrophic microbes – fungi and bacteria- are the most common and predominant. They attach and colonize the surface of the biofilter bed

and degrade the contaminants [71]. The most resistant microbial population is selected after acclimatization. The biodegradability level of a pollutant and the nature of the filtering materials affect a biofilter bed inoculation. Thus, this demonstrates the importance of selecting and using the appropriate type and concentration of biological organisms.

The microbial communities in biofilters, primarily shaped by water quality, are complex and contribute to addressing safety challenges in drinking water. However, research may underestimate the active role of these microbiomes in biofilters. Bai et al. [68] indicated that biofilters are composed of diverse microbiomes, with conventional biofilters significantly contributing to water safety. The microbial ecology among rapid sand filtration (RSF), granular activated carbon filtration (GACF), and SSF vary with each process featuring distinct microbial communities that influence the removal of different contaminants [68].

2.4.3.2. Type of biofilter media

The type of biofilter media used significantly influences biofiltration processes. Biofilter media type and pollutant biodegradability affect biofilter bed inoculation. The filter media provides a surface for biofilm growth after the microorganisms have been transferred [81]. Common media used for the construction of the biofilter bed are sand, GAC, and anthracite. These materials are considered due to their pore size distribution, cost effectiveness, versatility, and permeability as well as large and irregular surface area [82], [83], [84], [85], [86]. The mentioned features enable a biofilter to support the homogenous distribution of influent, enhance the metabolism and growth of microbes responsible for biofilm formation, and reduce the compression of biofilters. It was stated that combining organic materials and inert bulking materials increases filter media's useful life. Table 2.3 below shows some characteristics of materials used in constructing biofilter beds.

Table 2.3: Characteristics of biofilter media

Biofilter media	Characteristics	DOC Removal (%)	References
GAC and Anthracite	• Random pore structure and pore size distribution • Strong adsorptive capacity • Higher molecular weight • Cost-effective and versatile. • Provides a constant barrier against water pollutants. • A large mass of carbon provides a large surface area.	> 94%	[82], [83], [87]
Sand	• Affordable and easy to operate. • Good permeability • Reliable and durable. • High pollutant removal efficiency • Good water holding capacity.	41 – 98%	[84], [85]
Anthracite	• Large and irregular surface • Protects microbes from shear loss. • Belongs to the hydrophobic group (C-C and C-H) • Good adsorbent for organic matter removal • Contains low impurities	≤20%	[86], [87], [88]

Sand, GAC, and anthracite are common media used in standard biofilters for water purification. The adsorptive capacity of GAC to remove specific pollutants makes it the most commonly used filter media in potable water treatment processes [11]. GAC is referred to as biological activated carbon (BAC) when its intended use is as a biofilter growth medium for microorganisms. BAC filters, with their rough GAC grain surfaces and the ability to accumulate and sorb substrate within the particles, offer ideal conditions for biofilm growth and are more effective at removing trace organics than other media filters [76]. Additionally, GAC grains can mitigate the impact of inhibitors in the influent through sorption and desorption processes. This raises the question of whether BAC enriches certain specialist microorganisms with specific cell characteristics or whether it provides a suitable environment for generalist microorganisms from the feed water to scavenge low substrate concentrations more effectively.

Compared to sand and anthracite, the porosity, surface area, and roughness of GAC support the growth of microbial communities 4 – 8 times more in biomass per gram [89]. GAC can be regenerated due to its ability to adsorb and retain biodegradable parts of the contaminants. It also has a lesser specific surface area and an irregular surface, which makes biomass attachment easy. Kumar et al. [90] reported that the integration of activated carbon into filter bed materials improved pollutant biodegradation. When comparing the performance of GAC and anthracite as primary dual media filters, Thiel et al. [91] reported that DOC removal was higher with GAC (15 – 20%) compared to anthracite (2 – 7%) at EBCT of 16 min. At an EBCT of 8 mins, DOC removal ranged between 11 – 14% using GAC and 1 – 3% using anthracite. Emelko et al. [92] observed that TOC removal at temperatures in the range 21 - 24℃ was higher using GAC (23%) compared to anthracite (14%). Although several media types can be used in biofiltration, GAC has proven to be a robust media that supports microbial growth and attachment [11].

2.4.3.3. Operating temperature

The removal of BOM within biofilters is related to temperature. The number of active microorganisms and the degradation of contaminants are dependent on the operating temperature of a biofilter [71]. Temperature changes microbial structure, which in turn affects the substrate metabolism rate [71]. Microbial activities are low at lower temperatures (<10 ℃), thus reducing pollutant removal efficiencies [11]. The removal rate of contaminants is greater at a higher temperature [79]. According to Liu et al. [79], the removal rate of BOM is slightly higher at 20℃ than 5℃ in the absence of chlorine while using a GAC filter. This is different in the presence of chlorine; the removal rate is greater at a higher temperature. Chaudhary et al. [70] showed that biomass adapted at 10℃ but increased with increasing temperature up to 30℃. A temperature increase from < 10℃ to 10℃ - 20℃ to > 20℃ was recorded to have improved the removal efficiency of TOC from 10 % to 12–17 % respectively in a review of the performance of BOM and rapid-rate biofilter [69]. These studies support the notion that an increase in temperature enhances pollutant removal efficiency.

Contrarily, Liu et al. [79] observed that cold temperature (5℃) could favour biofiltration provided that the biofilter was backwashed in the absence of chlorine. Additionally, Emelko et al. [92] also reported that the removal of TOC was not significantly affected by temperature while using anthracite and GAC. It was noted that TOC removals by the GAC filter adsorber were slightly higher during the cold-water season compared to those observed during the warm water season, but these differences were not statistically significant. It was also observed that BOM removal may be affected at low temperatures. Moll et al. [93] observed significantly low removal of NOM and the NOM that reacts with disinfectants when the biofilter was operated at 5℃. Hoyland et al. [94] also noted a decrease in Mn removal as influent temperature decreased. When

the water temperature began to increase, Mn removal increased, eventually returning to greater than 98 % Mn removal.

The biofiltration guidance manual for drinking water facilities highlighted that biofiltration is most efficient at warmer temperatures (> 15◦C) due to the doubling of the reaction kinetics at every 10◦C increase [72], [74], [95]. Additionally, [96] observed that the biofilm formation rate increases significantly at WTPs with temperature greater than 15°C.

2.4.3.4. Empty bed contact time

The EBCT is an important operating parameter that was introduced by Zhang and Huck in 1996. EBCT was incorporated alongside the specific surface area of a biofilter, biodegradation rate, and substrate diffusion rate [70]. Additionally, a change in filter depth and hydraulic loading can lead to an increase in EBCT [70]. Vines and Terry [97] observed that the protein-like component of NOM was degraded efficiently from the lake and river water with increasing EBCT (15 – 30 mins). The study showed that EBCT for more than 5 min aids the removal of additional fractions of fluorescing components. A 30 – 50 % rise in TOC removal was recorded with an increase in EBCT of 5 – 20 min. Although the highest DOC removal (73 ± 6 %) was achieved at an EBCT of 30 min with a biological ion exchange (BIEX) filter, lower EBCTs could improve the NOM removal rate when operated concurrently at higher temperatures [98]. The study concluded that EBCT and temperature should be considered concurrently for an efficient removal of NOM using biofilters. Moona et al. [99] observed that the removal efficiency of DOC, protein-like (F280/340), and microbial humic-like (F290/420) fluorescent organic matter was high with increasing EBCT. The results from this study showed that longer EBCT (> 30 min) improved the operations of biofilters, thus, proving that longer EBCT improves pollutant degradation. Similarly, Terry and Summers [69] observed that biofilters removed 12 % of the influent TOC at an average

EBCT of 12 mins. Ko et al. [100] observed that there was a higher DOC removal with higher EBCT, highest being 20 mins. Although, the typical EBCT of biofilters ranges between 5 and 15 min [101], an improvement in contaminant removal is noted when the EBCT increases by 10 mins at temperatures higher at 15°C [72], [74]. Nevertheless, an EBCT > 10 mins is advantageous for the removal of Trace Organic Compounds when the temperature is < 15°C [72], [74].

2.4.3.5. pH

The role of nutrients in enhancing microbial activity and degrading pollutants is feasible under optimum pH. Although the optimum pH for biological processes in most drinking water sources ranges between 6.5 and 8.5 [72], [95], some microorganisms perform better under extremely acidic conditions. Thus, the ideal pH of the microbial population of interest has to be considered when designing proficient WTPs. Heterotrophic microorganisms that survive on biofilter beds operate at a neutral pH of 7. It has also been demonstrated that the disparity in pH influenced removal performance and microbial activity. Acidification causes pH disparity which in turn impedes a biofilter's performance. In the evaluation of the potential of biological Mn removal at the lower pH conditions (6.2–6.3), four laboratory-scale biofilters were operated over a pH range of 6.3–7.3 [94]. At pH 6.3, Mn was oxidized and over 98 % of Mn was removed. In addition, over 90 % of the simulated organic ozonation by-products was removed in all columns.

In a study to engineer the environment of a biomass structure in biofilm and biofilter on a reverse osmosis membrane, Jeong et al. [102] observed a 2 % increase in the removal of DOC when the pH was adjusted from 7.5 to 6.5. Similarly, Granger et al. [103] noted different responses of nutrient-enhanced biofilters to pH adjustment. For filters with a carbon-phosphorus (C:P) ratio of 100:15, an average 6 % increase was noted when the pH increased from 6.0 to 9.0, while filters with a C:P ratio of 100:3 observed an increase of 14 % on average [103].

2.4.3.6. Hydraulic loading rate

The hydraulic loading rate (HLR) is an essential factor in water treatment efficiency. Increasing the HLR results in the reduction of removal efficiency. A study investigating the removal efficiencies of five trace-level emerging organic contaminants (EOCs) and the microbial response in two identical drinking water treatment residue-based bio-filters under different HLR conditions found that higher HLR boosted the EPS production, increased community diversity, and enhanced denitrification [104]. This improved the removal efficiency of biodegradable EOCs like roxithromycin (80 %) and sulfamethoxazole (76 %). In contrast, lower HLR caused carbon limitation and selective pressures, enriching important species such as *Bacillus*, which play a crucial role in EOC degradation.

A pilot-scale system study was conducted with three dual-media GAC and sand biofilter columns running in parallel [100]. The HLRs for the filters were set at 24 m/h and EBCT of 5 mins for filter 1, 12 m/h and EBCT of 10 min for filter 2, and 6 m/h and EBCT of 20 mins for filter 3. The DOC removal efficiencies were 31 %, 26 %, and 19 % for filters 3, 2, and 1, respectively, suggesting that a higher EBCT and lower HLR are preferable for DOC removal [100]. An investigation on the effects of HLR on semi-volatile organic compounds (SVOC) removal and microbial community structure in drinking water treatment biofilters was conducted using six biofilters [105]. At an optimal HLR of 3.0 m/h, over 85 % of chemical oxygen demand and assimilable organic carbon was removed. Although results showed that increased HLR reduced the removal efficiency of SVOCs, up to 84 % of the main SVOCs were removed (71.2 % of Di-n-butyl phthalate and 84.4 % of bis(2-ethylhexyl) phthalate). At the lowest HLR, nearly 65 % of 2,6-dinitrotoluene and 80 % of isophorone were removed. The study also confirmed that the

dominance of *E. coli* in low-HLR biofilters likely contributed significantly to the high SVOC removal.

In a study that explored biological Mn removal at the lower pH conditions (6.2–6.3) using four laboratory-scale biofilters operated be- tween pH 6.3 and 7, the stress tests showed that well-acclimated manganese oxidizing bacteria could endure variations in hydraulic loading rate (1.36×10^{-3} – 2.72×10^{-3} m/s), Mn concentration (0.1 – 0.2 mg/ L), and temperature (7 – 22°C) commonly found in surface water treatment plants, at least for short periods (1–2 d) [94].

2.4.3.7. Backwashing

Filter backwashing is the periodic cleaning of the filter media bed with water or a combination of air scour and water [106]. This process fluidizes the filter bed to remove deposited material that can clog the filter pores and increase the head loss within the bed [106]. It has been shown that biomass detachment, biomass and accumulated particle removal, media distribution, and oxidant impact on backwash water are all dependent on adequate backwashing [11]. Kim et al. [107] and Qi et al. [108] indicated that regular backwashing, using water or air scour, and water are the most efficient methods for granular rapid filters to maintain the adsorption performance of GAC filters and prevent secondary contamination of the filter effluent until regeneration is required. Liao et al. [109] observed the impact of the backwashing process on the biomass and biofilm structure within a pilot-scale BAC filter to improve the removal of DOC. Results showed that backwashing was found to significantly influence the bacterial diversity and community composition of the BAC biofilm. However, these effects were seen to be gradually restored as the filtration process continued after the backwashing.

Similarly, Kim et al. [107] reported that filter backwashing temporarily disrupted the bacterial diversity, but the community was swiftly re-established during the subsequent operation. In a study

to observe the influence of backwashing on the BAC filter process, with respect to the removal of non-coagulable dissolved organic carbon (NC-DOC) and the composition of the microbial community, a laboratory-scale BAC filter was operated for a duration of up to 5 months, with backwashing conducted every 5 d [110]. It was found that when the BAC filter is backwashed at the optimal frequency, certain advantages, such as the biofilm developing a higher abundance of BOM degrading bacteria and a lower population of opportunistic pathogenic and filter clogging bacteria, as well as additional benefits for the coagulation process, can be realized. In the examination of the impacts of air scour and sub-fluidized backwash on oxalate and TOC removal at warm and cold temperatures, it was observed that the backwash conditions had little to no effect on BOM removal, but oxalate removal was high when air scouring was used under cold temperatures [92].

Ikhlef and Basu [111] investigated the effects of varying backwash methods on DOC reduction, backwash water usage, and effluent water clarity on a lab-scale biofiltration setup. Three dual media biofilters were run concurrently, each with 520 mm of GAC atop 180 mm of sand and 15 mm of synthetic drainer as support. Results indicated that DOC removal improved from 13 – 21 % when collapse pulsing was introduced to the backwash cycle under nutrient-scarce conditions compared to simple water backwashing. Reducing the bed expansion from 30 % to 20 % did not compromise DOC removal (35 %), however, backwash water consumption reduced by about 20 % [111]. For efficient performance, Liu et al. [79] recommended that biofiltration be performed at lower temperatures without chlorine when dealing with easily biodegradable compounds. You-qun [112] also highlighted that backwashing with chlorinated water had negative impact on biofilms.

2.4.4. Dominant microbes in biofiltration

The structure, function and dynamics of the microbial communities significantly contribute to biofiltration process, thus necessitating the need to customize these communities to improve treatment efficacy [113]. Putative keystone taxa that are necessary for maintaining the functional stability of systems were identified with the help of microbial co-occurrence networks using random matrix theory (RMT) [113], [114]. Keystone taxa are interconnected species that help shape microbial community structure and function, regardless of their abundance over time and space [113], [115]. Their removal can lead to significant changes in microbiome function and ecosystem processes. An investigation was conducted to study the interaction of microbial communities in BAC filters. The co-occurrence network indicated that microbes in the BAC filter are likely more collaborative than competitive. Phylum *Proteobacteria*, and *Cyanobacteria* were identified with relative abundance of 0.27 %. In addition, genus *Hyphomicrobium*, *Tabrizicola*, *Phreatobacter*, *Candidatus Obscuribacter*, *Pseudorhodoplanes*, and *Rhodospirillales* with relative abundance of 0.08–0.38 %, 0.27 %, 0.15 %, 0.21 %, 0.05 %, and 0.01 %, respectively, were identified. However, the bacterial community identified in the BAC using metaproteomic and metagenomic analyses are *Sphingomonas*, *Gemmata*, *Belnapia*, *Hyphomicrobium*, and *Crenalkalicoccus*. Despite the relatively low abundance of bacterium *Hyphomicrobium*, it contributed 0.3–1 % more to the most prevalent functions and produced 5–21 more proteins/g GAC compared to the dominant bacterium *Sphingobium*.

In the investigation to identify narrow host range phages infecting essential bacteria in groundwater-fed RSFs, 24 groundwater-fed RSFs were analysed in Denmark [116]. The analysis identified a core phageome commonly found in these systems. This core phageome is linked to dominant microbes essential for water purification processes. In addition, Palomo et al. [116]

found that *Siphoviridae*, *Myoviridae*, and *Podoviridae* were dominant species of the taxonomically classified viral sequences, and they accounted for 57 %, 28 %, and 14 %, respectively. The hosts, predicted using the multiple in silico prediction methods, encompassed 15 bacterial phyla, with the *Hyphomicrobiaceae* family accounting for 21 % of virus-host pairs and the *Nitrospiraceae* (most predicted) family for 15 %. With 77 % of the detected phages being lytic and numerous specific phage-bacterial interactions, it's suggested that phages help control bacterial populations by predation, thus preventing the dominance of any single species and maintaining functional redundancy. In summary, bacteriophages are likely crucial in water treatment within biofilters through their interactions with key bacterial species.

2.5. Resilience and adaptation of biofiltration to climate change

As a nature-based water treatment technology, biofiltration has proven to be remarkably resilient and adaptable to climate change. The resilience of a system refers to its capacity to withstand and recover from disruptions, while adaptation describes a system's ability to respond and adjust to long-term changes [117]. Biofilters house diverse microbial communities that are essential to the treatment process. These communities are flexible and can adjust to changing environmental conditions. As climate change causes variations in temperature, precipitation patterns, and contaminant levels, the microbial communities present in biofilters can adapt and retain their pollutant removal efficiency. This system has consistently shown strong treatment performance under extreme weather conditions such as rainfall, wildfire, drought and many more. These extreme weather events affect the HLRs, but biofilters exhibit stability and maintain their effectiveness in pollutant removal. The ability of microbial communities to self-regulate and recover from climatic disruptions explains the resilience.

Climate change increases the difficulty in the treatment of water and its source quality. Extreme events such as wildfires pose more concerning threats because significant drivers of water treatment technique design and optimization, such as turbidity and DOM are affected [6], [27]. Changes in DOM concentrations challenge the removal capacity of standard treatment technologies because they increase the formation of DBPs. In an experimental research on removal of organic pollutants of drinking water by bio- filtration, results showed that biofiltration could remove trace organic pollutants effectively [112]. The resilience of biofiltration to wildfire ash-associated organic carbon threat to potable water treatment was investigated and results showed that biofiltration offers resilience by softening the altered DOC concentration and improving the consistency of potable water in distribution systems [6]. In a 2-d and 7-d wildfire ash disturbance period, the removal rate of DOC increased in biofilters removing wildfire ash-amended water compared to the control biofilters. Water extractable organic matter of the readily biodegradable low molecular weight neutrals present in the wildfire ash amended biofilter influent makes DOC removal easier versus the control biofilters treating the source water baseline. It was also discovered that the control biofilter removed more humic substances, a driver of UV254 absorbance [118] compared to ash-amended water. The authors concluded that if the balance between the DOM recalcitrant and readily removed fractions changes, the resilience of biofiltration might be compromised. However, it can be attenuated by using coagulation to treat DOM fractions with lower biodegradability before using the biofiltration process.

The response to drinking water treatment after a Colorado, USA wildfire was studied [27]. It was noted that wildfire influences DOM character and conventional processes could still be used to treat postfire source water. To do this, a higher dose of coagulant was needed for water treatment. Samples collected post-rainstorm were unresponsive to conventional water treatment;

lesser DOC was removed and DBP formation was high. NOM removal in potable water treatment was investigated using two-stage biofiltration – GAC and sand-anthracite (SA) [119]. High NOM and turbidity removal efficiencies were demonstrated. Sand anthracite biofilters were used at the treatment's first stage and effectively removed > 74 % turbidity. GAC contactors used in the second stage removed NOM: > 24 % DOC, > 547 % UV254, and > 44 % SUVA254. GAC contactors removal efficiency for UV254 can be improved by pre-oxidation.

In research aimed at removing NOM and emerging contaminants such as pharmaceuticals and personal care products using two-stage biofiltration for drinking water treatment, a combination of sand/ anthracite (SA) biofilters and biologically active GAC post-filters were employed [119], [120]. Results indicated that biofiltration process can effectively remove turbidity, NOM, and an average of 53.4 % of PPCPs, with SA filters removing turbidity and GAC filters contributing significantly by removing 48.1 % of the PPCPs and NOM. It was observed that smaller molecules with less complex structures were more effectively removed. Additionally, a regression model was created to forecast the removability of specific PPCPs. The study also highlighted that biofiltration reduced health risks by an average of 79 %, suggesting that the levels of PPCPs in treated water would not pose significant health risks.

Ongoing research in biofiltration processes has led to improved designs, materials, and operational strategies. These innovative techniques provide improved adaptability to climate change, optimize pollutant removal efficiency, boost system durability, and address some challenges caused by climate change [71]. While biofiltration exhibits resilience and adaptability, it is necessary to observe that the effectiveness of biofiltration systems can be influenced by local factors, maintenance practices, and the specific pollutants being treated. To ensure continued resilience and adaptability in climate change, the following adaptation strategies have been advised

[6], [117]: 1) altering media maintenance and replacement based on humidity distribution and decay rates, and 2) regular monitoring, proper maintenance, and periodic assessment of system performance.

2.6. Challenges of biofiltration

In biofiltration, filter media such as sand and GAC naturally support the growth of diverse microbial communities, thus promoting biofilm formation [50], [121]. As a result, these filters not only act as adsorptive media but also support biomass substrate of microbes that utilize the readily available carbon in the system [49], [122]. Previous lab- and pilot-scale research indicate that sand filter-associated bacterial communities can potentially degrade contaminants and micropollutants [49]. Although Wang et al. [123] considers biological filtration to be the most efficient in producing biologically stable water, there are concerns regarding the appropriate design and implementation of this treatment process, especially regarding the size and type of filter media to be used [50]. Additionally, native microbial communities present in biofiltration usually lack the necessary metabolic capabilities to completely degrade pollutants [49], [124]. Consequently, leaving traces of pollutants or their degradation products being present in treated water [125].

The diversity of microbes present in biofilters is difficult to regulate because of the characteristics of influent raw water. Hwang et al. [126] proved that pathogen strains dominate the Schmutzdecke layer in their assessment of microorganisms and the Schmutzdecke of the bio sand filter for the treatment of river water. Treating water using biological processes creates opportunities for pathogenic contamination of potable water [62]. Furthermore, it is almost impossible to achieve the presence of all effective pollutant removal in a single treatment system. This necessitates the need to select microorganisms towards specific contaminants to improve the effectiveness of biological water treatment.

2.7. Summary of reviewed literature

Although the use of biofiltration to treat water is promising, there are still areas that require further research. There is a lack of detailed understanding of the mechanism of metabolic pathways by which organic pollutants are degraded by microbes [71]. A microbial metabolic pathway is dependent on the nature of microbes and their community in the biofilter bed. Further studies on microorganism growth and degradation mechanisms are required to promote the implementation of efficient biofiltration technology.

The effectiveness of biofiltration relies significantly on the type of microorganisms, their community on the filter bed [70], [71], and microbial activities, which are temperature sensitive. Extreme heat or cold events posed by climate change can harm the microbial communities in the biofilter, thus decreasing the overall performance. Further research is critically needed on the resilience of biofiltration under different conditions such as cold temperatures, sharp temperature changes (e.g., sudden temperature change from warm to cold), and the study of the impact of raw water quality shifts (due to climate change effect) on potable water treatment require further studies.

In addition, the increase in temperature, posed by the changing climate, has led to increased UVR exposure [127]. The carcinogenic effects of the UVR have resulted in the frequent use of sunscreens to reduce the risk of skin cancer. Recently, UV filters, mostly used in cosmetics, are being discharged into the environment in higher quantities [128]. The treatment of these micropollutants has been futile, and they end up being discharged into source water bodies. There is a need to develop advanced WTPs that can achieve high removal efficiency of these UV filters. Research into the capability of biofiltration to remove these emerging contaminants is critically required.

Raw water quality is altered after extreme weather events such as wildfires. This change affects the concentration of pollutants and characteristics. An increase in contaminant levels leads to the development of a thicker biofilm layer, which stops the diffusion of contaminants from reaching the depth of the biofilm, thereby increasing the inactivity of microbial biofilm with different pore sizes [129]. Although biofiltration excels at degrading certain pollutants, it may be less effective in degrading higher concentrations of pollutants or new pollutants. Research on the optimal thickness of the biofilm on the media is imperative to enhancing and maintaining the treatment performance of biofilters.

Furthermore, it is crucial to observe the efficiency of the biofiltration system and develop an operational code if a nature-based solution will be used to treat potable water [6]. To guarantee optimal performance, biofiltration systems need regular maintenance and monitoring. This might be difficult to maintain during severe weather events or when resources are limited. This brings about the need to design and optimize techno-ecological natural-based solutions and operational strategies.

In summary, this chapter has synthesized current knowledge on the impacts of climate change on water quality and WTPs, emphasizing the growing challenges posed by extreme events such as wildfires and rising temperatures. These phenomena increased UVR exposure to humans and contaminants transportation into surface waters. Conventional treatment methods—including coagulation, flocculation, AOPs, and filtration—have demonstrated varying degrees of effectiveness in removing emerging contaminants, though they often entail high operational costs and generate secondary waste. Biofiltration has emerged as a promising, sustainable alternative, leveraging natural microbial processes to treat raw water. Its adaptability to diverse climatic conditions and its ability to support microbial activity under stress highlight its potential as a

resilient treatment strategy. The successful implementation of biofiltration depends on careful system design, appropriate media selection, and consistent operational monitoring. Despite its limitations, biofiltration stands out as one of the most environmentally friendly and cost-effective water treatment technologies. Advancing this approach could significantly enhance the resilience and sustainability of water treatment systems in the face of ongoing climate change.

Chapter 3 Materials and Methods

3.1. Wildfire-impacted raw water preparation

The baseline raw water was reverse osmosis water collected from the University of Northern British Columbia Enhanced Forestry Laboratory (EFL). Subsequently, wildfire-impacted raw water was simulated by amending the baseline raw water using 0.5 g/L of bottom ash collected from the University of Northern British Columbia Bioenergy plant. The DOC levels of the wildfire-impacted raw water were adjusted using 0.938 g/L sodium acetate ($C_2H_3NaO_2$) and 0.208 g/L sodium propionate ($NaC_3H_5O_2$) up to three levels: 20 mg/L, 50 mg/L, and 100 mg/L. The addition of bottom ash, $C_2H_3NaO_2$, and $NaC_3H_5O_2$ represented the severity of the disturbance and the resulting degradation in source water quality [6].

The bottom ash was initially mixed with 10 L of baseline raw water for a period of 24 – 48 h at a rate of 100 m/h to ensure water extractable organic matter was adequately leached from the ash. Following mixing, the mixture was then introduced to the 200 L feed tank. The comprehensive water quality of the influent can be found in Appendix A.

3.2. Pre-treatment

The wildfire-impacted water was pretreated using aeration and roughing filtration (Figure 3.1). Diffused aeration introduces oxygen into the water in a fine bubble form, which promotes the oxidation of organic matter, making them more amenable to removal in the subsequent treatment stages. Diffused aeration replicates cascade aeration which is more cost effective on a large scale [130]. Following aeration, wildfire-impacted raw water passed through rouging filters to reduce the turbidity to <10 NTU. The rough filtration technology is effective in removing the larger particles of ash, charred debris, and other suspended solids that are common in water sources affected by wildfires [6]. These methods are simple, cost effective, and require minimal

maintenance. Removal of altered NOM, measured as DOC, is a common water treatment challenge faced after a wildfire event [6].

Figure 3.1: Roughing filter set-up

3.3. Biofilter design and operation

Three 3-L bench-scale biofilters were set up in the EFL and run for 11 months. The biofilter columns had an inner diameter of 7 cm and an estimated bed depth of 70 cm. For this research, sand was selected as the biofilter media with an effective size of 0.475 mm and a uniformity coefficient of 1.26, along with GAC with an effective size of 0.9 mm and a uniformity coefficient of 1.88. Apart from the fact that both biofilter media are commonly used, they are also well known for their high pollutant removal efficiency, affordability, and versatility [82], [83], [84], [85]. The filters were operated in a downflow mode for 242 d during biofilter acclimation and 93 d during the experimental periods. Each experimental run lasted for 30 d, and the biofilters were backwashed for 24 h after each run. At the early phase of the acclimation period (168 d), the bench-scale biofilters were designed with flowrate of 0.053 L/s (EBCT of 15 mins and HLR: 2.8 m/h). Due to water logging in the biofilters caused by biofilms thickening, the flowrate was reduced to

0.018 L/s (EBCT: 56 min and HLR: 0.75 m/h). Throughout the entire experimental period, the biofilters were maintained at a room temperature of 18 – 23 °C. Figure shows the water treatment system used for this study.

Figure 3.2: Full biofiltration-based water treatment setup

3.4. Experimental design

Nine experimental runs with three varying levels of filter media (non-numerical factor) and three varying levels of DOC (numerical factor) were conducted to examine the individual and combined effects of the filter media type and DOC concentration in the raw water on DOC removal. Specifically, these experimental runs allowed the investigation of every possible combination of the independent variables of media type and DOC concentration measured as chemical oxygen demand (COD) at room temperature (18 – 23°C). Three levels of DOC concentration (20 mg/L, 50 mg/L, and 100 mg/L) and biofilter media (sand, GAC, and the

combination of sand and GAC) were observed. The dependent variable was the DOC removal efficiency, enabling a comprehensive analysis of how these variables interact and influence the DOC removal process. Table 3.1 outlines the experimental factors and their combinations, allowing for a thorough analysis of each condition.

Table 3.1: The experimental runs based on biofilter media type and DOC concentration as the factors

RUN	BIOFILTER MEDIA	DOC LEVEL (mg/L)
R1	Sand	20
R2	GAC	20
R3	GAC + Sand	20
R4	Sand	50
R5	GAC	50
R6	GAC + Sand	50
R7	Sand	100
R8	GAC	100
R9	GAC + Sand	100

3.5. Analytical methods

Performance testing was conducted on parameters such as pH, DO, UVA_{254}, temperature, COD, and alkalinity every 3 d (except for alkalinity) to determine the performance of each biofilter. ATP and EPS were tested every other week to examine the presence and quantity of microbes present in each biofilter. In addition, biodegradable dissolved organic carbon (BDOC) of the feed water was measured to determine the quantity of DOC that can be degraded. All tests

were done in triplicate. Influent and pretreated water samples were collected from the top of the tanks, while biofilter effluents were collected from the bottom of the biofilters. Additionally, media samples for EPS and ATP were collected from the top of the biofilters. The procedures are outlined below.

3.5.1. Dissolved oxygen (DO) and temperature

DO and temperature of feed water, pretreatment and biofilter effluents were measured using the Thermo Scientific Orion^{TM} Star A213 benchtop RDO/DO meter (Beverly MA). The RDO optical dissolved oxygen probe was rinsed and wiped dry with a lint-free tissue and placed into the collected sample. DO and temperature measurements were recorded upon stabilization of the stirrer probe and display showing 'ready'.

3.5.2. Turbidity

Turbidity was determined using Orion™ AQUAfast AQ3010 turbidity meter (Thermo-scientific, Beverly, MA), The sample vial was rinsed and filled with the samples, wiped dry with lint-free tissue and placed in the sample well of the turbidity meter. Readings that appeared on the display of the turbidity meter were recorded in NTU.

3.5.3. pH

The pH of the samples was determined using DPH 7011 Digital pH Meter (General Tools and Instruments, New York, USA). Prior to measurement, the pH meter probe was rinsed and dried with a lint-free tissue. The probe was then placed in a beaker containing the samples. To ensure uniformity, the samples were stirred continuously until a stable reading appeared on the pH meter. The pH value was recorded once the reading stabilized.

3.5.4. Alkalinity

Alkalinity was measured using TNT870 test kits using Method 10239 (Hach, Canada). 2.0 mL of solution A from the reagent set is added to the test vial. Subsequently, 0.5 mL of each sample was added to the test vial. The vial was mixed thoroughly and left to react for 5 min. After reaction, the vial was cleaned using a lint-free tissue and placed into the cell reader. Results were taken in mg/L using a spectrophotometer.

3.5.5. Ultraviolet absorbance at 254 nm (UVA_{524})

The UV-absorbing (UV-254) organics compounds present in the feed and effluent water was measured every 3 d using standard method 5910 [131]. Samples were filtered using a 0.45-µm filter and a vacuum filtration pump. The filtered samples were poured into a sample cell (2624410), and UV absorbance was measured using a Hach DR 6000 spectrophotometer (Loveland, CO) at a wavelength of 254 nm. The results were taken in cm^{-1}. Then $SUVA_{524}$ was calculated from Equation 3.1 [132].

$SUVA_{524}$ was measured using equation

$$\boldsymbol{SUVA} = \frac{\textbf{UV absorbance } (\boldsymbol{cm^{-1}})}{\textbf{DOC (mg/L)}} * 100 \quad \text{Equation 3.1}$$

3.5.6. Extracellular polymeric substances (EPS)

For EPS extraction [133], [134], 10 mL of each biofilter media was collected and put inside a 50 mL vials. Reverse osmosis (RO) water was filtered through a 0.45- µm filter and added to the 50 mL vials up till the 50 mL mark. 0.3 mL of filtered 37 % formamide was added to the vials. The vials were placed in an orbital shaker at a speed of 30 rpm for 1 h. After 1 h, the sample was centrifuged at 5000 g for 15 min at 4 °C. The supernatant was first filtered through a 0.45- µm filter, then 0.22- µm filter. The filtrate was labelled loosely bond EPS (LB-EPS).

To extract tightly bond EPS (TB-EPS), the sediments from LB-EPS was resuspended by adding filtered extraction buffer to 50 mL mark. The pH of the suspension was adjusted to approximately 11 by adding 0.4 mL of 1mol/L of NaOH and placed in an orbital shaker for 3 h. After 3 h, the sample was centrifuged at 10000 g for 15 min at 4 °C. The supernatant was first filtered through a 0.45- µm filter, then 0.22- µm filter. The filtrate was labelled tightly bond EPS (TB-EPS).

To quantify EPS sugar, the colorimetric method for determination of sugars and related substances was used [135]. In a 250 mL amber glassware, 5g of phenol crystals was added to 100 mL of RO water. Four concentrations of glucose solutions were prepared by diluting 5.6 g/L of glucose stock solution. In a 15 mL test tubes, 1 mL of the different concentrations of glucose solution were added followed by 1 mL of 5% phenol, and 5 mL of H_2SO_4. A blank was prepared by adding 1 mL of RO water, 1 mL of 5% phenol, and 5 mL H_2SO_4. The test tubes were sealed and left to react for 10 min. After 10 min, the test tubes were placed on the INTTLAB vortex mixer VM370 for 10–15 s before placed into the Labfish digital water bath LFUS034 for 20 min at 30°C. The test tubes were wiped, and results were recorded using a spectrophotometer at a single wavelength of 490 nm. A calibration curve was obtained from the results and is attached in Appendix A.

For the quantification of LB-EPS, the same procedure was repeated except that 1 mL of sample was mixed with 1 mL of 5% phenol, and 5 mL H_2SO_4 while the sample for TB-EPS was diluted with dilution water (DW) to get a factor of 5. The final sugar concentrations were calculated using the line equation from the calibration curve obtained.

For the quantification of protein, the modified Lowry method was used. Solution A was prepared by dissolving 20g of Na_2CO_3 and 4g of NaOH into 800 mL RO water. More RO water

was added to bring the solution to 1000 mL. Another solution B1 was prepared by dissolving 1g of $CuSO_4.5H_2O$ into 60 mL of RO water, and more RO water was added to bring the solution to 100 mL. Following this step, solution B2 was prepared by adding 2g of $C_4H_4KNaO_6.4H_2O$ in 60 mL RO water. More RO water was added to bring the solution to 100 mL. To prepare solution C, 50 mL of solution A, 0.5 mL of solution B1, and 0.5 mL of solution B2 were mixed. This solution was prepared on the day of analysis and used while still fresh. A final solution E was prepared in an amber glassware by mixing Folin-Ciocalteu reagent with RO water in a 1:1 ratio.

For calibration, Bovine Albumin Serum (BSA) standard solution of concentrations 0, 20, 40, 60, 80, and 100 mg/L were prepared. A 200 mL BSA stock solution was made by adding 0.2g BSA and 1.8g NaCl to 100 mL RO water. The volume was completed by adding RO water until a 200 mL mark. For concentrations 20, 40, 60, 80, and 100 mg/L, 0.5, 1.0, 1.5, 2.0, and 2.5 mL of BSA stock solutions were added and placed into 50 mL vials, respectively. RO water was added to each vial up to the 25 mL mark. For 0 mg/L (blank) standard, RO water was used. In 15 mL test tubes, 1 mL of each concentration standard was mixed with 3 mL of solution C and 0.3 mL of solution E. The test tubes were closed, and the tubes were vortexed for about 10 s. Samples were placed in a light-protected foam box and left to react for 2 h. After the reaction, results were recorded using a spectrophotometer at a single wavelength of 750 nm. A calibration curve was obtained from the results and is attached in Appendix A. The calibration procedure was repeated for the extracted EPS samples. For the quantification of LB-EPS, 1 mL of raw samples was used while the samples for TB-EPS were diluted with DW to get a factor of 5. The final protein concentrations were calculated using the linear equation from the calibration curve obtained.

3.5.7. Adenosine triphosphate (ATP)

ATP was analyzed using the Invitrogen ATP determination kit [136]. To prepare the reagents, the following steps were followed. 50 µL of 20X reaction buffer (component E) was added to 950 µL of dH_2O to make 1 mL of 1X reaction buffer. To make 1 mL of 10 mM D-Luciferin stock solution, 1X reaction buffer was then added to one vial of D-Luciferin (component B, MW 302, blue cap). Following this step, 100 mM Dithiothreitol (DTT) stock solution was made by adding 1.62 mL of dH_2O to 25 mg of DTT (component C, MW 154, black cap). The DTT stock solution was aliquoted into ten 160 µL volumes and frozen at ≤ -20°C. Finally, 10 mL of several concentrations of ATP standard solutions (1 µM, 2 µM, 5 µM, and 10 µM) were prepared by diluting the 5 mM ATP solution (component D, green cap) in dH_2O. The diluted ATP solutions were aliquoted into 2 mL and stored frozen at ≤ -20°C.

In a 15 mL vial, 10 mL of the standard reaction solution was made by adding 8.9 mL of dH_2O, 0.5 mL of 20X reaction buffer, 0.1 mL of 100 mM DTT, 0.5 mL of 10 mM D-Luciferin, and 2.5 µL of firefly luciferase 5 mg/mL stock solution (component B, red cap). The vial was inverted to mix the solution and kept away from light.

In a 96-well plate, 100 µL of the standard reaction solution was added to wells A1, B1, and C1. In wells A2 – C5, the reaction was started by adding 90 µL of the standard rection solution and 10 µL of the different concentrations of ATP standard solutions and biofilter media samples. On the Gen 5 protocol software, an experiment was created. 96 well plate type and luminescence detection method were selected. On the plate layout, each well was labelled according to their content. Following this procedure, the plate was placed in the BioTek Synergy 2 microplate reader (Edmonton, Canada) and the plate was read. A standard curve for the series of ATP concentrations

was generated and used to estimate the ATP concentration of microbes present in the biofilter media. Standards were run with each experiment due to the decreasing sensitivity of the assay.

3.5.8. Biodegradable dissolved organic carbon (BDOC)

BDOC was analyzed using the method developed by Khan et al. [137] and the standard method for BOD [131]. Sample was collected and analyzed within 2 h. Prior to analysis, sample's temperature was adjusted to 20 ± 3 °C by placing it in the incubator and filtered through a 0.45-µm filter. The DW was prepared immediately before use by adding 1 mL of each phosphate buffer, $MgSO_4$, $CaCl_2$, and $FeCl_3$ to 1 L of dH_2O of DO ≥ 7.5 mg/L. In addition, one capsule of Polyseed (InterLab, Texas, USA) was added for every 500 mL of dH_2O. The mixture was thoroughly mixed using a JLT6 Flocculator Tester (Usmate, Italy) at a speed of 200 rpm for 60 mins. Following this step, the mixture was left to settle for 15 mins, supernatant was extracted, and temperature was adjusted to 20 ± 3 °C.

Using DW, three dilutions of samples i.e., 25:75 (% DW: % sample), 50:50 and 75:25 were prepared. This was estimated to produce at least one dilution with DO ≥ 1 mg/L and DO uptake of 2 mg/L or more at the end of a 5-d incubation period. Following dilution, 300 mL of each dilution were poured into BOD bottles. A seed control (sample s) was prepared using the same method except that it was 100% DW. All prepared samples were analyzed for DO and recorded. After DO measurement, BOD bottles were incubated in a Fisherbrand Isotemp BOD refrigerated incubator at a temperature of 20 ± 1 °C for 5 d. Additionally, samples from each dilution were collected for COD analysis.

After the 5-d period, BOD bottles were removed from the incubator. The samples were analysed for DO_f DO_{sf}, DOC_{sf}, and DOC_f. Of all the dilutions, only the dilution 50-50 had DO ≥

1 mg/L and DO uptake of 2 mg/L, therefore the dilution ratio was used to calculate the dilution factor, F, using the equations 3.5-1 to 3.5-3:

$$\boldsymbol{F = \frac{volume\ of\ dilution\ water\ (mL) + volume\ of\ sample (mL)}{volume\ of\ sample\ (mL)}} \quad \text{…….. Equation 3.2}$$

BOD_5 and BDOC were calculated using the following equations:

$$\boldsymbol{BDOC = \left[\left(DOC_i - DOC_f\right) - \left(DOC_{si} - DOC_{sf}\right)\right] * F} \text{…….. Equation 3.3}$$

$$\boldsymbol{BOD_5 = \left[\left(DO_i - DO_f\right) - \left(DO_{si} - DO_{sf}\right)\right] * F} \text{…….. Equation 3.4}$$

Observing the criteria of $DO_f \geq 1$ mg/L and $(DO_i - DO_f) \geq 2$ mg/L [131].

3.5.9. Dissolved organic carbon (DOC)

The DOC concentration was measured in terms of chemical oxygen demand (COD). The COD was determined according Standard Methods 5220D using low range (≤ 90 mg/L) COD solution [131] . This standard COD solution contained 1.2 mL/tube of digestion solution and 2.8 mL/tube of sulfuric acid reagent. To prepare the digestion solution, 1.022 g of potassium dichromate ($K_2Cr_2O_7$) was added to 500 mL of deionized water (dH_2O). 167 mL of concentrated sulfuric acid (H_2SO_4) and 33.3 g of mercuric sulfate ($HgSO_4$) were added to the $K_2Cr_2O_7$ solution and allowed to cool to room temperature. After cooling, the solution was diluted to 1000 mL by adding dH_2O. This digestion solution was stored in a 2L pyrex storage glass bottle. The dichromate ion ($Cr_2O_7^{2-}$) from $K_2Cr_2O_7$ oxidizes the COD present in the sample. To prepare the sulfuric acid reagent, 10.1 g of silver sulfate (Ag_2SO_4) was added to 1000 mL of H_2SO_4 and allowed to dissolve for 24–48 h before mixing. Ag_2SO_4 ensures thorough oxidation of organic matter, while H_2SO_4 helps suppress

interference from compounds like bromine, chloride, and iodide, maintaining accurate processing and results.

After mixing, 1.2 mL of the digestion solution and 2.8 mL of the sulfuric acid reagent were pipetted into 15 mL test tubes. The test tubes were capped and thoroughly mixed for homogeneity. To evaluate the standard solutions, different volumes of potassium hydrogen phthalate stock and dH_2O were added to the prepared COD solution. The test tubes were capped and thoroughly mixed to achieve a homogeneous mix before placing into a pre-heated (150°C) Hach DRB 200 COD digester. The test tubes were left in the digester for 2 h at a temperature of 150°C. They were left to cool to 120°C before taken out of the digester and left to cool again at room temperature (20 – 25°C). Results were recorded using a spectrophotometer at a single wavelength of 420 nm. A calibration curve was obtained from the results and is attached in Appendix A.

To determine the total COD concentration of the samples (influent and effluents), 2 mL of filtered sample (through a 0.45- µm filter) was added to the prepared COD solution and capped. The test tubes were capped and thoroughly mixed to achieve a homogeneous mix before placing into a pre-heated (150°C) Hach DRB 200 COD digester (Loveland, CO). The test tubes were left in the digester for 2 h at a temperature of 150°C. They were left to cool to 120°C before taken out of the digester and left to cool again at room temperature. Results were recorded using a Hach DR 6000 spectrophotometer (Loveland, CO) at a single wavelength of 420 nm. The final COD concentration was calculated using the line equation from the calibration curve obtained. For influent COD concentration of 150 mg/L (DOC = 50 mg/L), 1 mL of sample was diluted with 1 mL of dH_2O to obtain a dilution factor of 2. Similarly for an influent COD concentration of 300 mg/L (DOC = 100 mg/L), 0.5 mL of sample was diluted with 1.5 mL of dH_2O to obtain a dilution factor of 4.

3.6. Statistical analyses

To evaluate the influence of DOC concentration and media type on DOC removal, a comprehensive statistical analysis was conducted. Initially, an outlier test was performed to identify and address any extreme values that could skew the results. This was followed by a normality test to assess the distribution of the data, ensuring the appropriateness of subsequent parametric or non-parametric analyses. Given the nature of the data, a Spearman correlation test was employed to examine the monotonic relationships between DOC removal and the predictor variables. To further explore the predictive capacity of the variables, a fit model regression analysis was conducted, incorporating media types and initial DOC concentration as predictors. Finally, an analysis of variance (ANOVA) was used to determine the statistical significance of the differences in DOC removal across the different media types. A significance level of $p \leq 0.05$ was used to determine the rejection of the null hypothesis, indicating a statistical difference between the analysed data groups. This multi-faceted approach provided a robust framework for understanding the key factors affecting DOC removal efficiency.

Chapter 4 Results and Discussion

4.1. Influent water quality

Detailed influent water quality analysis is presented in Appendix B. The water sample was generally within the acceptable limits set by the Guidelines for Canadian Drinking Water Quality (GCDWQ) for most parameters; however, several exceeded the recommended operational guidelines (OG) or aesthetic objectives (AO). Notably, the concentration of aluminum was measured at 0.23 mg/L, exceeding the OG of 0.10 mg/L, and manganese's concentration was 0.24 mg/L, surpassing the AO of 0.02 mg/L. Iron had a concentration of 0.44 mg/L, above its AO limit of 0.30 mg/L. The pH level of 9.32 exceeded the recommended range of 6.50 to 8.50, potentially affecting disinfection efficiency and corrosion control. Additionally, turbidity at 14 NTU and color at 80 TCU were significantly above their respective AO of ≤1.0 NTU and ≤15 TCU, which may impact taste, appearance, and consumer acceptability.

In contrast, total dissolved solids at 293 ppm and sodium at 77 mg/L were within acceptable ranges, with the AO for sodium being ≤200 mg/L. Microbiological analysis showed no presence of E. coli or total coliforms, indicating no immediate microbial risk. However, the heterotrophic plate count (HPC) was high >2420 MPN/mL, exceeding the general guidance level of 500 MPN/mL, which may suggest the presence of non-pathogenic microbial activity. Overall, while the water is microbiologically safe, several physical and chemical parameters exceed OG or AO, indicating a need for further treatment or monitoring to ensure consumer acceptability and system performance.

4.2. Temperature

The mean temperature across runs and treatment systems ranged between 19°C - 21°C (**Figure 4.1**). Moll et al. [138] noted a higher removal performance (42%) when biofilters operated at higher

temperatures (20 and 35°C), compared to lower temperatures (5°C). Similarly, in a study investigating the removal of assimilable organic carbon (AOC) using a dual media, Price et al. [139] found no AOC removal at temperatures less than 16°C. However, when operated above 16°C, the system achieved an average AOC removal that was 44% higher. Although the present study did not include operations at lower temperatures, the observed DOC removal rate aligns with findings from studies conducted at comparable temperatures.

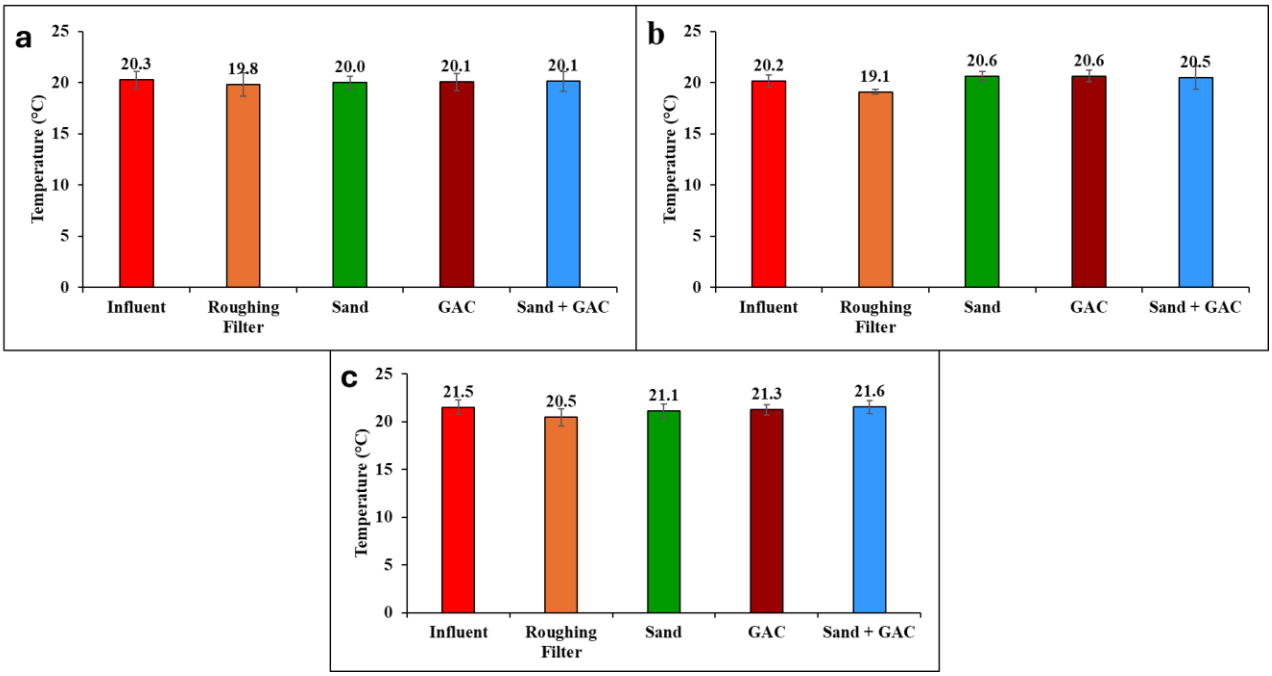

Figure 4.1: Mean temperature of influents and effluents at DOC = 20 mg/L (a) 50 mg/L (b), and 100 mg/L (c)

4.3. Turbidity

Turbidity varied throughout the experimental period. At influent DOC concentration of 20 mg/L (**Figure 0a**), the turbidity of the influent water remained relatively high with noticeable peaks around days 14 and 28, indicating inconsistent water quality before treatment. The effluent from pretreatment system and biofilters all maintained consistently low turbidity levels. An

average of 76 ± 13% (roughing filter), 78 ± 19% (sand), 82 ± 13% (GAC), and 83 ± 9% (sand + GAC) turbidity removal demonstrated effective and stable filtration performance. Among the treatment systems, sand + GAC biofilter appears to provide the most consistently low effluent turbidity, indicating that it is the most effective at reducing particulate matter.

At the influent DOC concentration of 50 mg/L (**Figure 4.2b**), influent water turbidity starts with a sharp spike in turbidity, peaking on day 6 at (85 NTU), indicating a high level of suspended particles in untreated water. After day 6, turbidity drops sharply, due to natural settling. The treatment system maintained consistently low turbidity levels, averaging 74 ± 21% (roughing filter), 80 ± 15% (sand), 77 ± 20% (GAC), and 78 ± 18% (sand + GAC) turbidity removal throughout the 30-d period. This suggests that the filtration methods are highly effective at removing particulates and maintaining water clarity. Similar to influent DOC concentration of 20 mg/L (Figure 4.2a), sand + GAC biofilter is the most efficient due to its dual-stage filtration.

The untreated influent for influent DOC concentration of 100 mg/L (**Figure 0c**), consistently showed the highest turbidity, serving as a baseline. The roughing filter moderately reduces turbidity, while sand filtration performs better with a gradual decline. GAC shows a more significant reduction, and the Sand + GAC biofilter achieved the lowest turbidity levels throughout the period. Throughout the 30 d, the treatment system consistently achieved high turbidity removal efficiencies, averaging 69 ± 16% with the roughing filter, 72 ± 26% with sand filtration, 71 ± 23% with GAC, and 75 ± 22% with the sand + GAC biofilter. This indicates that the Sand + GAC biofilter is the most effective in improving water clarity, with all treatments showing consistent trends supported by error bars indicating measurement variability.

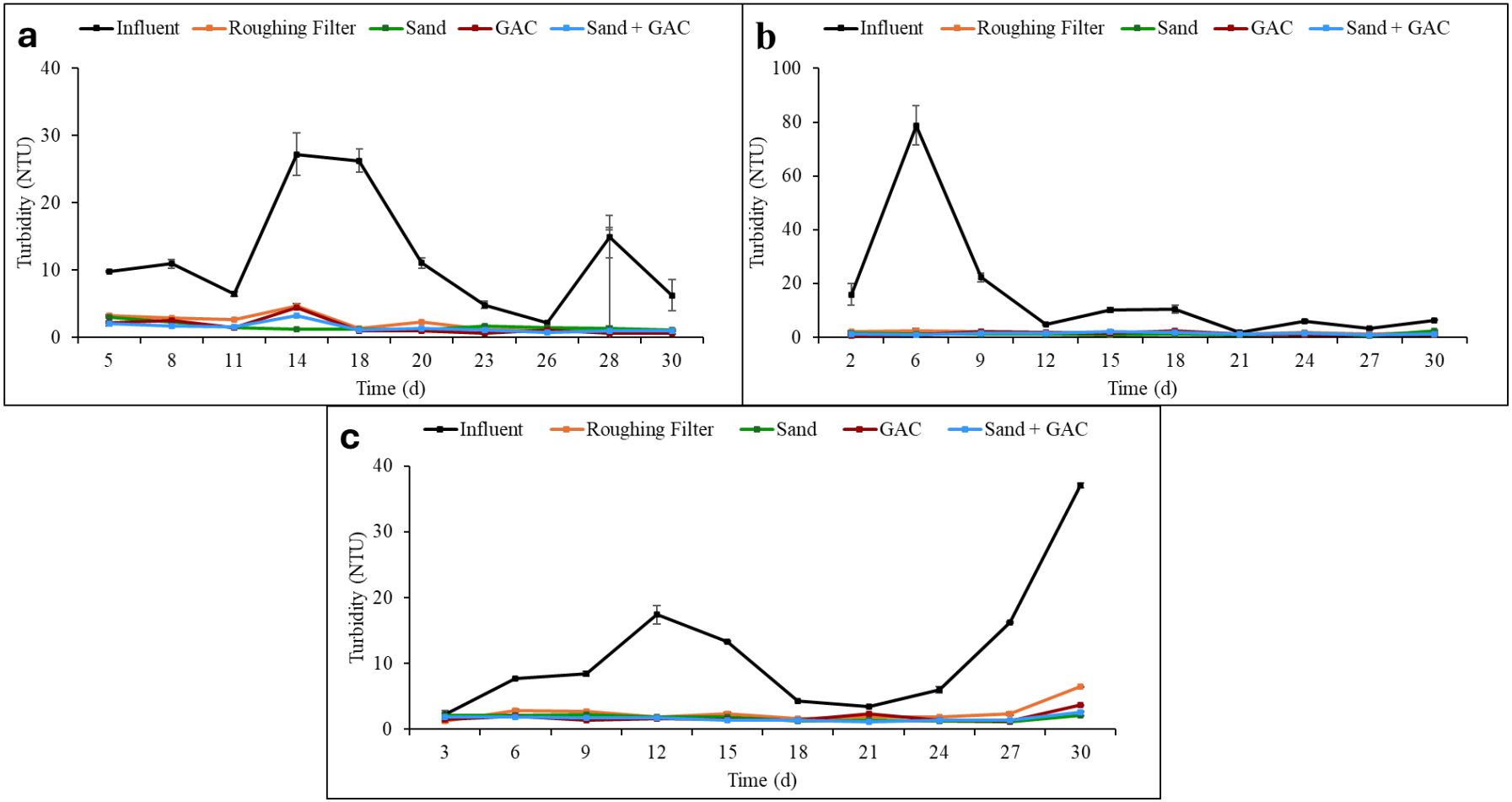

Figure 0: Trends of turbidity reduction at DOC = 20 mg/L (a), 50 mg/L (b), and **100 mg/L (c)**

The turbidity result observed in this study is consistent with previous literature. Blackburn et al. [6] observed effluent turbidity reduced to ≤ 0.3 NTU in 93% of biofilter effluent samples analyzed. Similarly, the filtrate turbidity of biofilters used as pretreatment to membrane-based desalination had a turbidity value of 0.2 – 0.3 NTU and 0.28 – 0.31 NTU for anthracite and GAC biofilters, respectively [140]. Although the turbidity levels in the biofilters' effluent are higher than those reported in previous studies and exceed the GCDWQ operational guideline of ≤ 1 NTU, the results still highlight the effectiveness of biofiltration in reducing turbidity.

4.4. pH

The pH results of the influent and the biofilters at all concentrations are shown in **Figure 0**. The average influent pH was 9.38 ± 0.25, 9.40 ± 0.25, and 9.52 ± 0.12 for influent DOC concentrations of 20 mg/L, 50 mg/L, and 100 mg/L, respectively. At 20 mg/L influent DOC concentration (**Figure 0a**), there was a drop in pH across the treatment system. For the first 14 d, the biofilter effluent had pH ranging from 7.42 – 8.83. The pH of the biofilter effluent increased (8.95 – 9.53) for the next 12 d before finally dropping to a range 8.37 – 8.83. At 50 mg/L influent DOC concentration (**Figure 0b**), the pH showed a slight decline during the first 12 d, followed by a sharp drop over the next 12 d. During the final 6 d, pH levels remained relatively stable with minimal change. For 100 mg/L influent DOC concentration (**Figure 0c**), the starting pH for the GAC and sand + GAC biofilters were below 8.5 for the first 6 d. In the subsequent days, the pH of all biofilters increased to > 9. Generally, the average pH of effluents at 20 and 50 mg/L influent DOC concentration aligns with the ideal pH range of biofiltration systems [1], [28], [29]. However, the effluents from runs experimental runs at 100 mg/L influent DOC concentration exceeded the ideal pH range of biofiltration systems.

Sodium acetate and sodium propionate added to the influent water contributed to its alkaline properties. Sodium acetate dissociates into sodium ions (Na^+) and acetate ions (CH_3COO^-) when it dissolves in water. The CH_3COO^- then reacts with water to form acetic acid and hydroxide ions (OH^-). The resulting OH^- makes the pH more basic by increasing the solution's pH. Similarly, sodium propionate dissociates into Na^+ and propionate ions ($CH_3CH_2COO^-$) when it dissolves in water. The $CH_3CH_2COO^-$ then react with water, producing propionic acid and OH^-. The OH^- that is produced raises the pH.

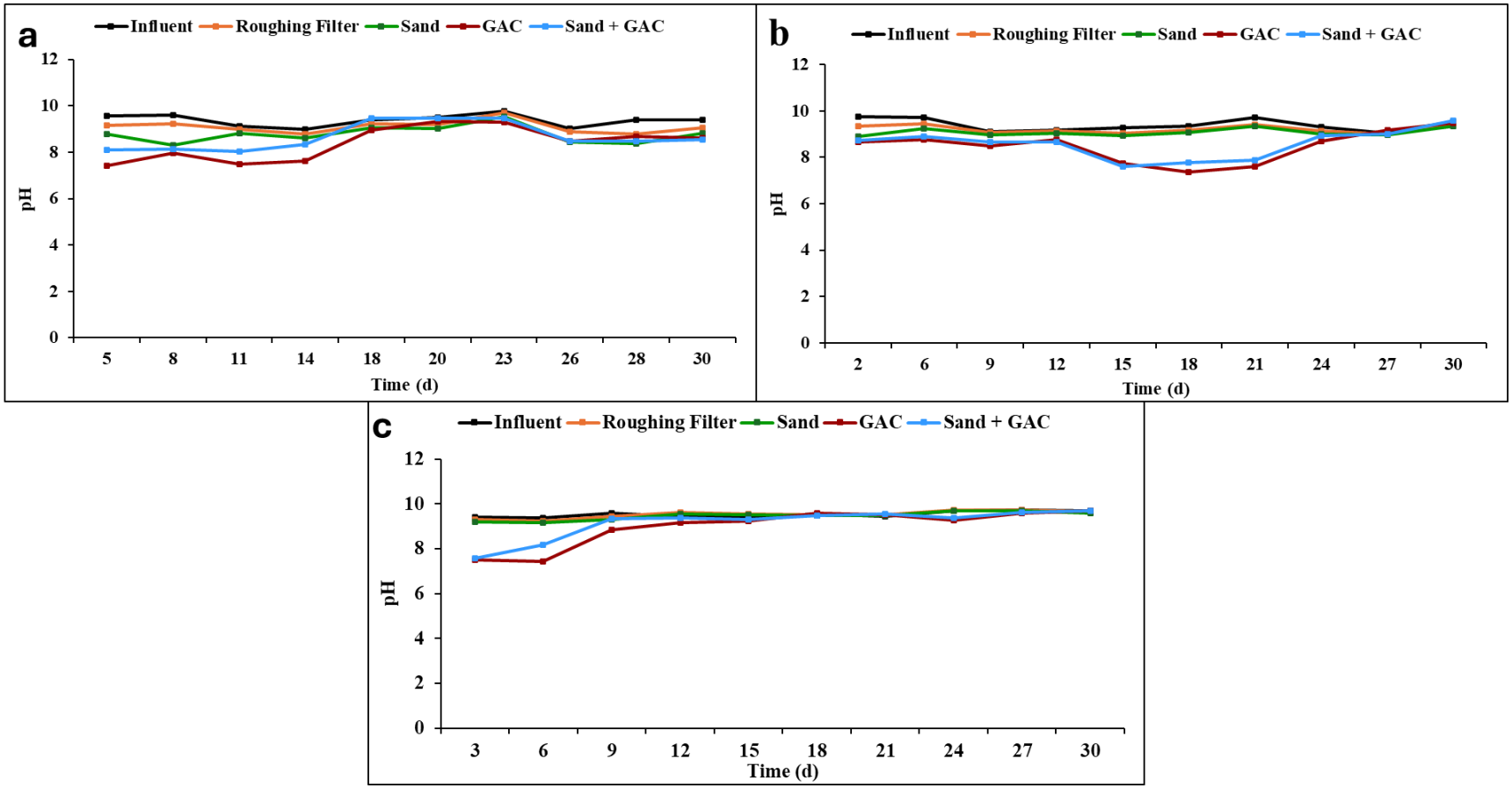

Figure 0: Trends of pH of influents and effluents at DOC = 20 mg/L (a) 50 mg/L (b), and **100 mg/L (c)**

4.5. Alkalinity

All treatments at different DOC concentrations show non-linear, fluctuating trends in alkalinity concentration over time (**Figure 0**). The values range from 259 to 1470 mg/L, indicating remarkable variability. Alkalinity levels of runs at 20 mg/L influent DOC concentration (**Figure 0a**) dropped sharply after day 20 before rising again. In contrast, runs at influent DOC concentration of 50 mg/L (**Figure 0b**) showed a continuous increase in alkalinity up to day 30. Runs at 100 mg/L influent DOC concentration (**Figure 0c**) exhibited significant fluctuations, with pronounced peaks and dips. The elevated alkalinity in the treatment effluents suggests a buffering effect, which may explain the resistance to changes in pH. Hamidi et al. [141] reported that changes in alkalinity had a modest impact on biofilter performance in removing TOC and ammonia, as well as ATP levels. Additionally, the GCDWQ have no aesthetic objective (AO) or maximum acceptable concentration (MAC) for alkalinity. Thus, proving that treated water with high alkalinity is consumable.

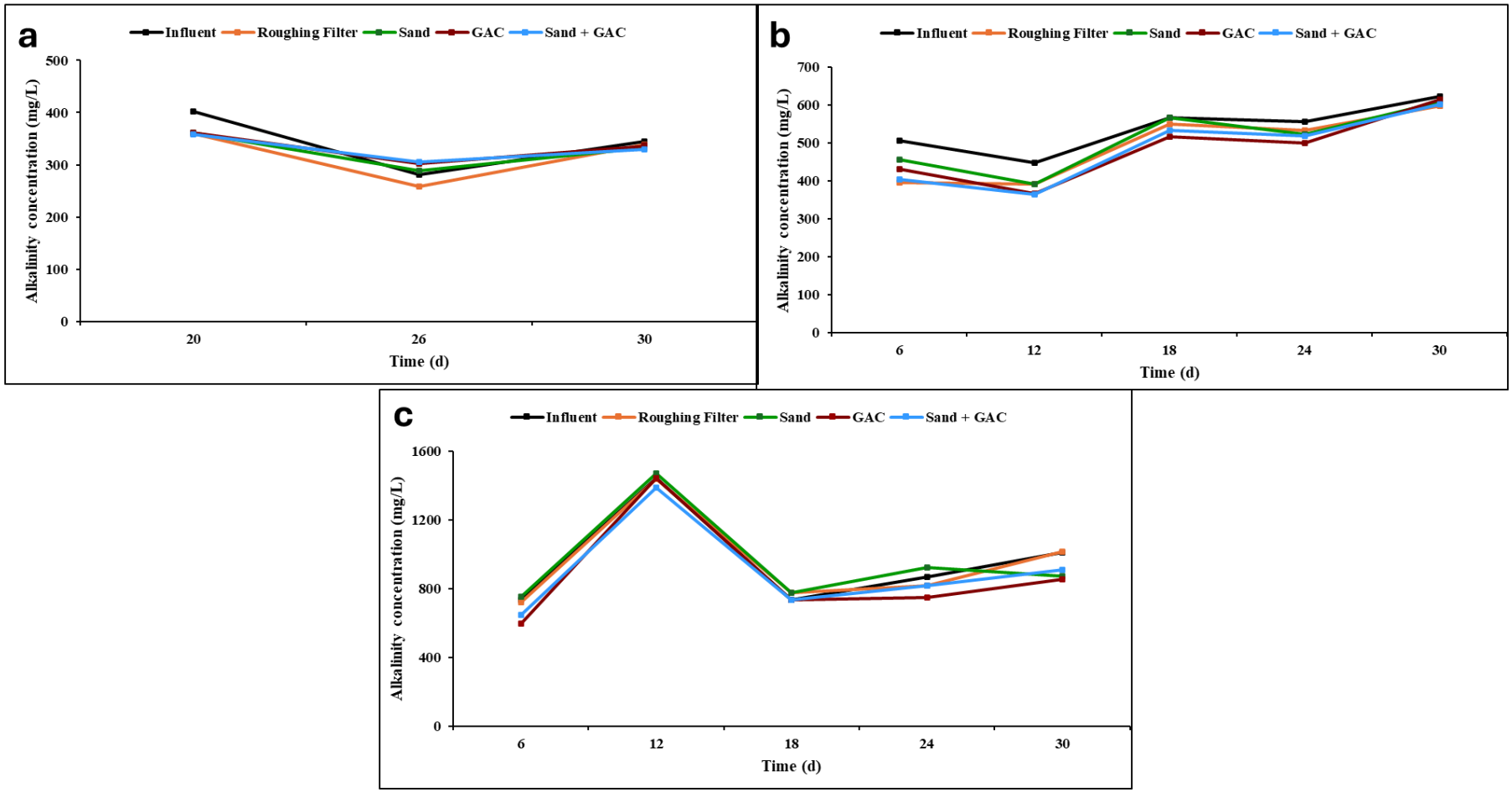

Figure 0: Trends of alkalinity of influents and effluents at DOC = 20 mg/L (a) 50 mg/L (b), and **100 mg/L (c)**

4.6. Ultraviolet absorbance at 254 nm (UVA_{524}) and specific ultraviolet absorbance at 254nm ($SUVA_{524}$)

UVA_{524} results showed notable variation across the different treatment runs. In runs at influent DOC concentration of 20 mg/L (**Figure 3.0a**), the influent exhibited a consistent and steep increase in UV absorbance, particularly after day 10, indicating a rising concentration of organic matter. The roughing filter showed a gradual increase, while the sand biofilter started with low absorbance and steadily increased, peaking between days 20 and 30. The GAC biofilter began with high absorbance and remained relatively stable, while the Sand + GAC biofilter showed a moderate increase, especially after day 10. Runs with influent DOC concentration of 50 mg/L (**Figure 3.0b**) followed similar patterns: the influent maintained a clear upward trend with less fluctuation, the roughing filter showed a gradual rise, and the GAC biofilter remained stable with minor variations. Both the sand and Sand + GAC biofilters displayed increasing trends, with the latter showing slightly more variability.

In runs with influent DOC concentration of 100 mg/L (**Figure 3.0c**), the influent consistently had the highest UV absorbance, reflecting a high organic load. The roughing filter and sand biofilter achieved moderate reductions, with sand biofilter performing slightly better over time. Sand biofilter significantly lowers UV absorbance and maintains stable performance. GAC demonstrated a significant and sustained increase in UV absorbance, while the Sand + GAC biofilter achieved the lowest values from day 12 – 21, indicating the most effective removal. Overall, the sand biofilter proved most effective in reducing UV-absorbing organic compounds. However, a notable increase in UV absorbance is observed across all biofilters after day 18, suggesting a potential decline in filtration efficiency or a change in influent water quality during that period.

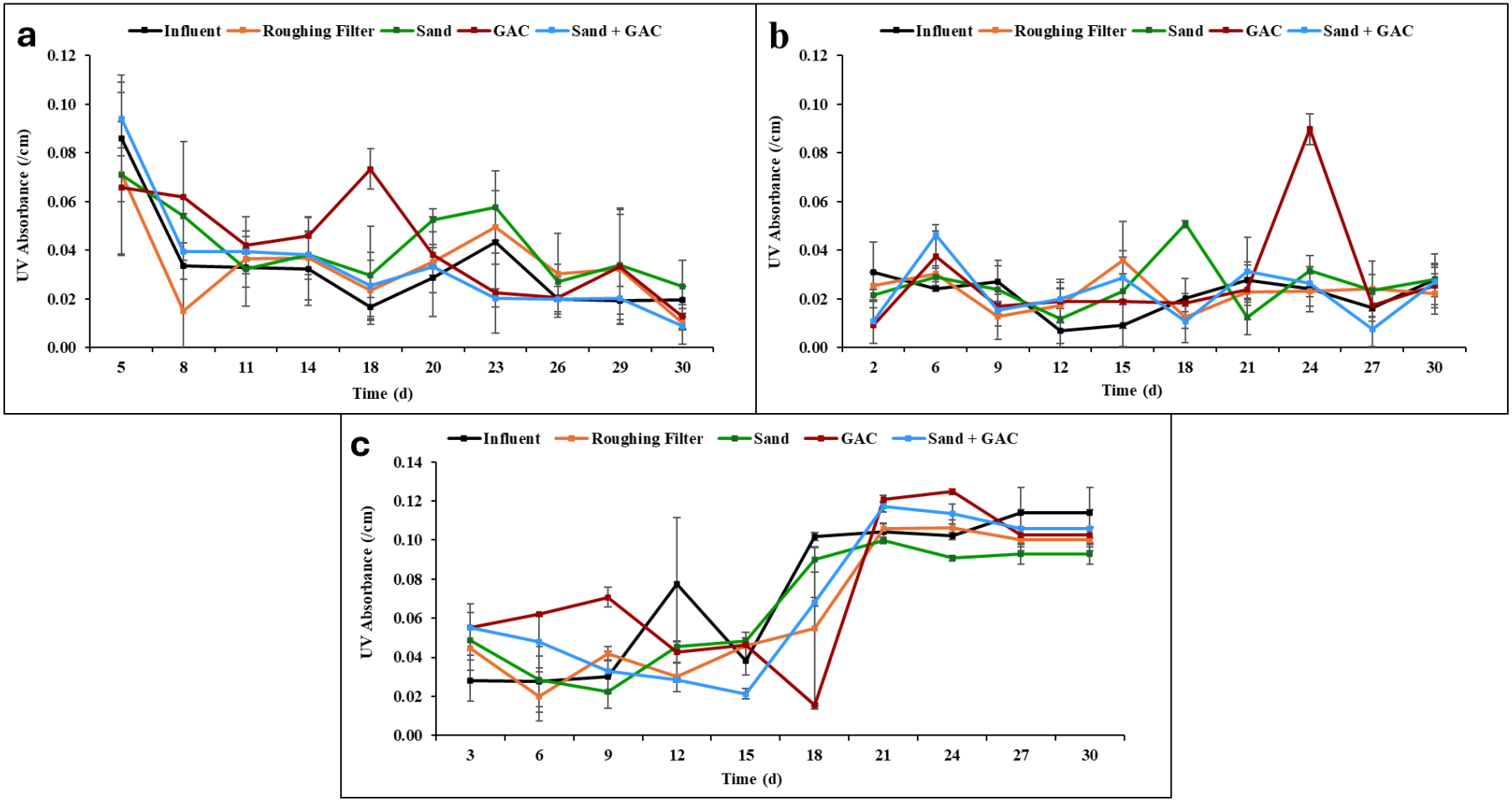

Figure 3.0: UVA_{254} trends for DOC at 20 mg/L (a), 50 mg/L (b), and 100 mg/L (c)

Blackburn et al. [6] reported a difficulty in reducing UVA_{254} which was consistent with the findings of this study and other previous research [72], [97], [142]. Due to the treatment mechanism of biological systems, a significant reduction in UVA_{254} was not anticipated. This is associated with the fact that UVA, especially at the wavelength of 254nm, is directly proportional to the concentration of organic matter in water. It indicates the presence of DOC [72], thus proving the presence of lower DOC concentration in the biofilter effluent.

$SUVA_{254}$ was calculated by dividing UVA_{254} by the relative DOC concentration. $SUVA_{254}$ characterizes the aromatic nature of organic matter in water [143]. A higher $SUVA_{254}$ value indicates a higher proportion of aromatic compounds such as humic substances, which indicate a greater potential for the formation of disinfection byproducts (DBPs) when treated with chlorine. The influent and effluents at all DOC concentrations had $SUVA_{254}$ values < 2 L/mg.m throughout the experimental period (**Figure 4.6**). This indicates that the influent water, roughing filter, and biofilters effluents had mostly hydrophilic and low molecular weight compounds in its NOM composition [132]. Additionally, the potential TOC removal for $SUVA_{254}$ < 2 L/mg.m is 0 – 40%, 2 – 4 L/mg.m is 40 – 60%, and > 4 L/mg.m is 40 – 60%.

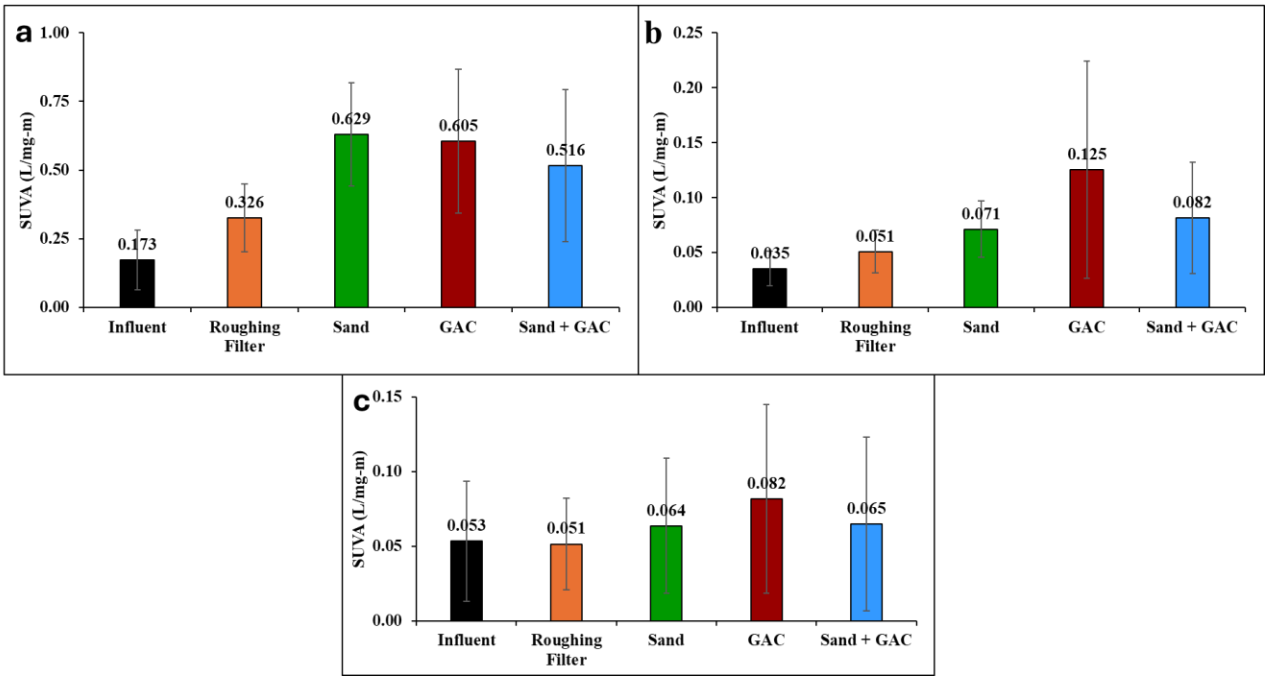

Figure 4.6: Average SUVA of influents and effluents at DOC = 20 mg/L (a) 50 mg/L (b), and **100 mg/L (c)**

4.7. Dissolved Oxygen (DO)

The average DO of the influent water, pre-treated and biofilter effluents, measured at different runs is illustrated in **Figure 4.7**. A decrease in DO was observed across the treatment system with the influent water having the highest average DO level (6 ± 1 mg/L; 7 ± 3 mg/L ; and 7 ± 0 mg/L) while the roughing filter effluent had the lowest (2 ± 1 mg/L; 3 ± 1 mg/L ; and 4 ± 1 mg/L) at DOC concentrations 20 mg/L, 50 mg/L, and 100 mg/L. These findings are consistent with Nemani et al. [144], who recorded influent DO ranging from 7 mg/L to 8 mg/L while evaluating the impact of biofilter operation on microbial community structure and performance, which correspond to the expected saturation levels of dissolved oxygen in water.

Reduction in influent DO water during biofiltration systems is primarily due to the biological activity within the filter. Biofiltration systems designed for the removal of BOM operates under

aerobic conditions, with oxygen serving as the terminal electron acceptor for heterotrophic microorganisms [145]. The growth of biofilms on the filter media involves microbial activity such as respiration and it consumes oxygen. As these biofilms develop and mature, they continue to use oxygen from the water [13], [146]. Therefore, the rate of oxygen consumption is a direct indicator of microbial activity related to DOC removal [145]. Gomez et al. [147] observed that DO affects biofilm development, with excessive DO levels inhibiting biofilm growth. Thus, the decrease in DO indicates microbial activity and biofilm formation within the roughing filter and biofilters. Furthermore, the interdependence of oxygenation and pollutant removal (manganese and ammonium) in drinking water biofilters was established [148].

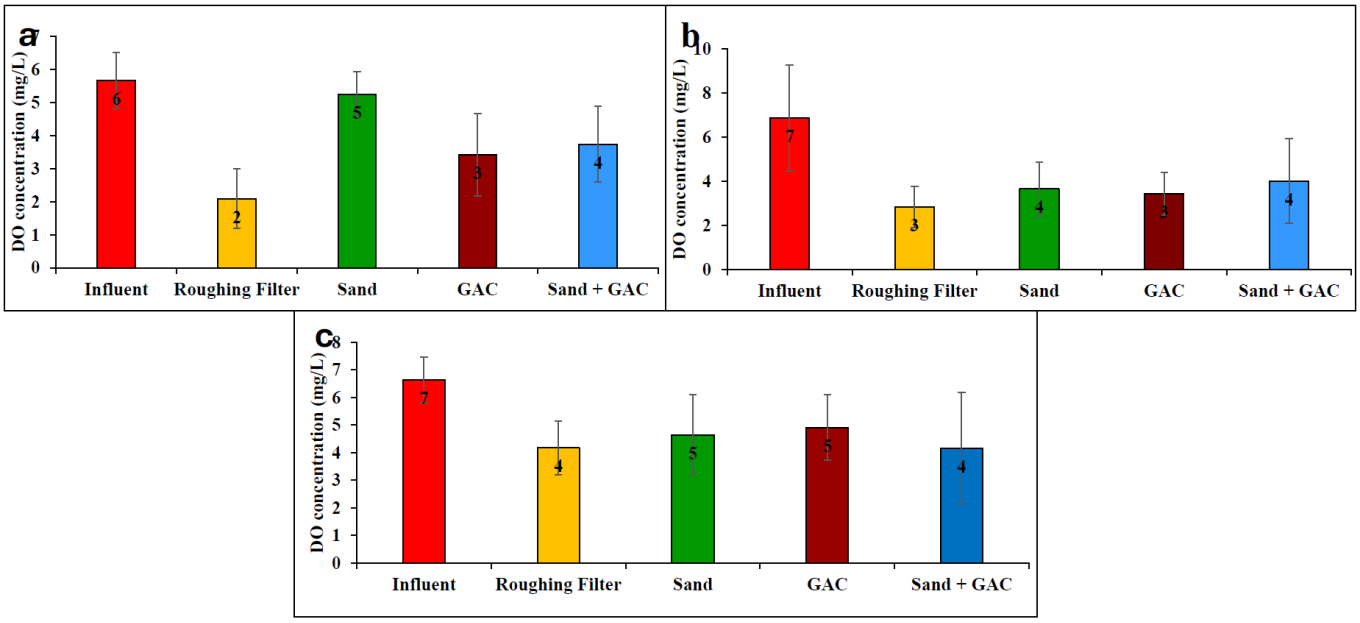

Figure 4.7: Mean DO levels of influents and effluents at DOC = 20 mg/L (a) 50 mg/L (b), and 100 mg/L (c)

4.8. Adenosine triphosphate (ATP)

In runs at influent DOC concentration of 20 mg/L at day 30 **(Figure 4.8a)**, the GAC biofilter shows the highest ATP concentration (1.1 ± 0.1 mM), indicating the most active microbial community among the three media. The sand biofilter follows with a moderate ATP level (1.0 ±

0.1 mM), while the sand + GAC biofilter has the lowest ATP concentration (0.8 ± 0.1 mM), suggesting limited microbial activity. Runs at influent DOC concentration of 50 mg/L shows a different trend. On both day 17 and day 26 **(Figure 4.8a)**, the sand biofilter consistently exhibits the highest ATP concentrations (14.4 ± 1.8 mM, 6.5 ± 1.3 mM), reinforcing its effectiveness in sustaining microbial growth. GAC biofilter maintains moderate levels (4.5 ± 0.7 mM, 2.6 ± 0.5 mM), while sand + GAC biofilter remains the least biologically active (2.2 ± 0.6 mM, 1.5 ± 0.4 mM). The trend suggests that microbial activity decrease over time which could result to decrease in pollutant removal.

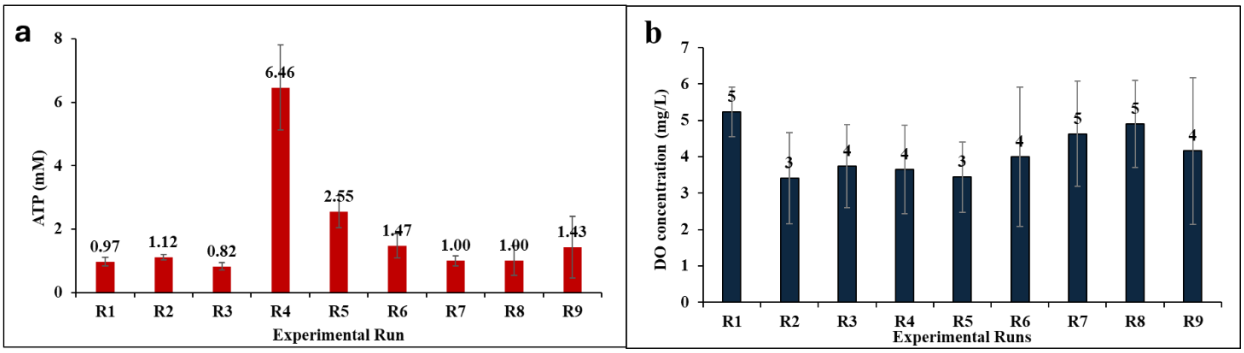

Figure 4.8 ATP concentration at steady state (a) and Average DO concentration (b) of all runs

In run 7 – 9 with influent DOC concentration of 100 mg/L, a significant drop in ATP concentration was observed after day 10 and slightly peaked on day 26. On day 10, the highest ATP concentrations of the sand (4.5 ± 0.4 mM) and GAC (1.9 ± 0.1 mM) biofilters were observed. The highest ATP concentration of the GAC biofilters (1.4 ± 1.0 mM) was observed on day 26 **(Figure 4.8a)**. The lowest ATP concentrations of all biofilters was observed on day 17; 0.8 ± 0.1 mM (sand), 0.8 ± 0.2 mM (GAC), and 0.6 ± 0.1 Mm (sand + GAC). Across all time points, sand biofilter leads in microbial activity, with steadily increasing ATP levels. GAC shows a similar upward trend but at lower concentrations, while sand + GAC remains consistently low except for

day 26. The data confirm that sand biofilter enhances and sustains microbial growth more effectively than other biofilters.

Previous research observed that source water quality, EBCT, media type, and temperature do not affect the ATP concentration at the acclimated biofilters surface [149], [150]. However, an increase in influent DOC concentration and HLR influences ATP concentration [123], [150]. Additionally, this study identified a general trend of a negative correlation between DO and ATP concentrations (**Figure 4.8a & 4.8b**). At each DOC concentration, the lowest DO levels typically corresponded with the highest ATP concentrations, and vice versa – except in Runs 4 (sand at 50 mg/L) and 5 (GAC at 50 mg/L), which deviated from this pattern.

4.9. Extracellular polymeric substances (EPS)

The EPS proteins and sugar concentrations across experimental R1 – R9, distinguishing between loosely-bound EPS(LB–EPS) and tightly-bound (TB–EPS) are shown in **Figure 4.9**. In terms of protein concentration (**Figure 4.9a**), TB-EPS exhibited higher values compared to LB-EPS in runs R1, R2, R3, and R6, while LB-EPS slightly surpassed TB-EPS in runs R4, R5, R7, R8, and R9. The highest protein concentration in TB-EPS is observed in Run R6 (sand + GAC at 50 mg/L), suggesting a peak in microbial activity or biofilm development during that phase. Throughout the experimental period, the highest concentrations of LB–EPS and TB–EPS proteins were produced in R2 (GAC at 20 mg/L) of 6.6 ± 1.5 mg/g TSS and 21.0 ± 7.5 mg/g TSS, respectively, suggesting a peak in microbial activity or biofilm development during that phase. The lowest concentration of LB–EPS proteins observed was in R5 (1.5 ± 0.4 mg/g TSS).

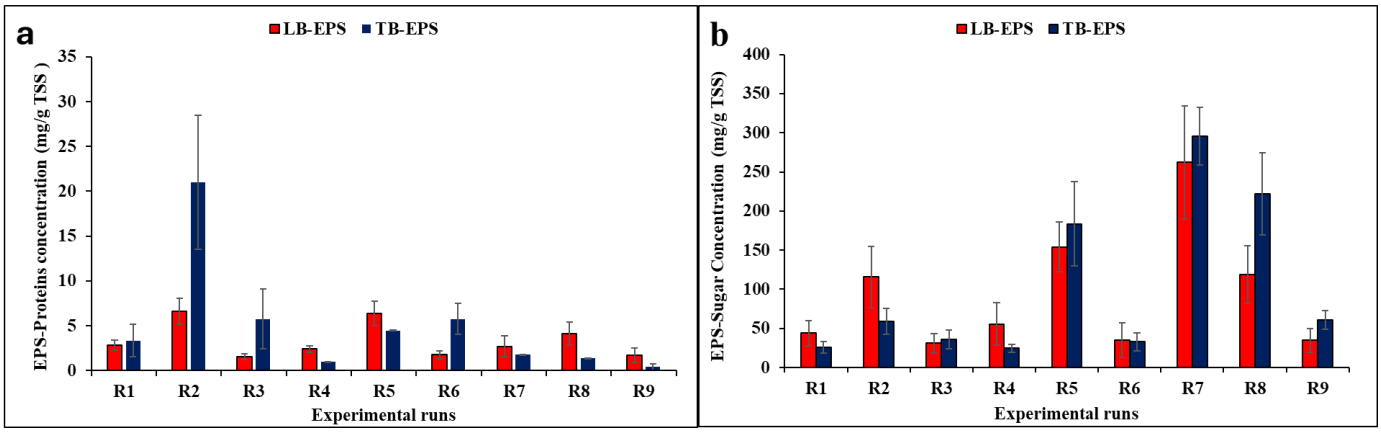

Figure 4.9: EPS proteins (a), and sugar (b) concentration at each run

Similarly, in **Figure 4.9b**, which displays EPS sugar concentrations, LB-EPS dominates runs R1, R2, R4 and R6, showing significantly higher values than TB-EPS. The sugar concentration in TB-EPS also peaks in runs R3, R5, R7, R8, and R9. The highest concentration of LB-EPS and TB-EPS sugar were produced in R7 (sand at 100 mg/L) of 262.3 ± 72.3 mg/g TSS and 295.6 ± 36.9 mg/g TSS, respectively. It is also worth noting that higher concentrations of sugar were produced compared to proteins. The results highlight the critical role of TB-EPS in biofilm structure and function, as it not only contributes more substantially to the EPS matrix but also reflects the intensity of microbial processes within the biofilters.

The variation in EPS protein and sugar concentrations across different experimental runs can be largely attributed to pH differences. The optimal pH range for EPS production is reported to be between 5.50 and 6.50 [151], [152]. Although the pH in Run 2 (GAC at 20 mg/L) ranged from 7.42 to 8.62 – slightly above the optimal range – it was the lowest among all runs, which likely contributed to the highest observed EPS protein production. In contrast, Shu and Lung [153] reported that higher pH levels favored the production of low molecular weight exopolysaccharides with higher yields. This aligns with the findings of this study, where Run 7 (sand at 100 mg/L)

exhibited the highest average pH (9.46 ± 0.17) and correspondingly produced the highest concentration of EPS sugars.

EPS are produced by microbes in biofilters aiding in bacterial attachment, shielding them from harsh environmental conditions [154]. Although, it is hard to numerically compare EPS due to different extraction methods, however, **Table 4.1** summarizes extracted EPS concentration from previous literature and this study.

Table 4.1: Previously reported EPS concentration extracted from water biofilters

Media type	Extraction buffer	EPS Concentration	Units	References
Anthracite and sand	Cation Exchange Resin with 6 mM PBS	Proteins: 34 – 166 Sugar: 11 – 102	μg BSA/g wet media μg D-glucose/cm^3 media	[155]
GAC	Cation Exchange Resin with 6 mM PBS	Proteins: 86.5 – 102.7	μg BSA/cm^3 media	[154]
GAC, anthracite, and sand	Formaldehyde (36.5%) and NaOH (1 N)	Proteins: 1 – 5 Sugar: 0.5 – 3	μg BSA/cm^3 media μg D-glucose/cm^3 media	[156]
GAC, anthracite, and sand	10 mM Tris, 10 mM EDTA, 2.5% NaCl, pH 8	Proteins: 0.27 – 3.38 Sugar: 0.02 – 0.6	mg BSA/g TS mg glucose/g TS	[157]
Sand and GAC	Formaldehyde (37%) and NaOH (1 M)	Proteins: 3.38 – 27.63 Sugar: 67.69 – 557.86	mg BSA/g TSS mg glucose/g TSS	This study

4.10. Biodegradable dissolved organic carbon (BDOC) and dissolved organic carbon (DOC) profile

BDOC, the average biodegradable fraction of DOC, of the influent water at all DOC concentrations, ranged from 18 % to 72 %, and is summarized in **Table 4.2**.

Table 4.2: BDOC concentration of influent water

Experimental Runs	DOC Concentration (mg/L)	Average BDOC Concentration (mg/L)	BDOC (%)
R1 –R 3	20	11 ± 0	55
R4 – R6	50	26 ± 14	72
R7 – R9	100	18 ± 2	18

Figure 4.10 shows the DOC concentration of both influent and effluent at DOC = 20 mg/L (a), 50 mg/L (b), and 100 mg/L (c). Runs 1 – 3 (**Error! Reference source not found. 4.10a**) DOC c oncentration reached steady state after 8 d. The biofilters achieved DOC removal efficiencies of up to 16% (roughing filter), 80% (sand), 85% (GAC), and 83% (sand + GAC), averaging 51 ± 15% (roughing filter), 68 ± 22% (sand), 68 ± 16% (GAC), and 70 ± 20% (sand) at steady stage (day 8 – 30) (**Figure 4.1a**).

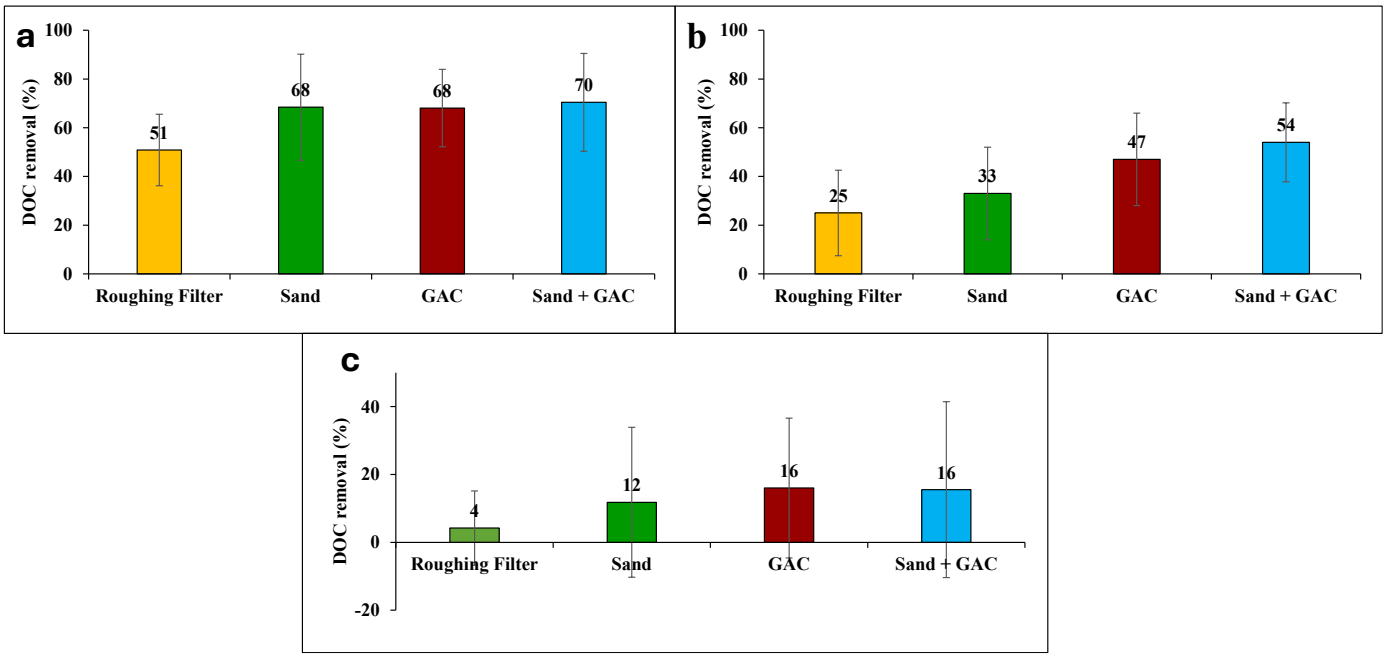

Figure 4.1:Average DOC removal (%) of influents and effluents at DOC = 20 mg/L (a) 50 mg/L (b), and 100 mg/L (c)

In runs with influent DOC concentration of 50 mg/L (Figure 4.9**b**), DOC removal efficiencies of up to 59% (roughing filter), 65% (sand), 78% (GAC), and 69% (sand + GAC), averaging 25 ± 18% (roughing filter), 33 ± 19% (sand), 47 ± 19% (GAC), and 54 ± 16% (sand) (**Figure 4.1b**). The highest DOC removal was achieved on day 12 after which the removal efficiency dropped significantly. This decrease was also noted in the ATP concentration (after day 17). Studies have observed that ATP levels at the surface of the biofilter are not necessarily related to biofilter performance in terms of DOC removal [79], [92], [150], [158]. However, a decrease in primary energy carriers that break down organic matter across the experimental period aligns with the reduction in DOC removal efficiency.

DOC removal efficiencies achieved in runs with influent DOC concentration of 100 mg/L (**Error! Reference source not found.c**) were up to 27% (roughing filter), 58% (sand), 51% (GAC), and 59% (sand + GAC). The average removal efficiencies as shown in **Figure 4.1c,** were 4 ± 11% (roughing filter), 12 ± 22% (sand), 16 ± 21% (GAC), and 16 ± 26% (sand). Between days 3 – 9, the GAC and sand + GAC biofilters had the lowest effluent DOC concentrations, indicating higher removal efficiency compared to the sand biofilter. However, from day 9 to day 30 (except for day 27), effluent DOC concentrations from all biofilters were extremely high, in some cases exceeding the influent DOC levels. This resulted in low or even negative removal efficiencies, ranging from –22% to 59%. These trends align with pH observations: from day 2 to 9, the pH of the GAC and sand + GAC biofilters remained within the ideal range (6.5 – 7.5), supporting effective microbial activity. After day 9, however, pH levels in all biofilters rose above 9, likely inhibiting biological processes and contributing to the reduced DOC removal performance.

The reduction in DOC concentrations from influent to effluent was substantial across all biofilters, aligning with findings from various biological filtration studies. Collins et al. [37]

observed 12–33% removal of DOC in different full-scale slow sand filtration (SSF) plants with EBCTs ranging from 3.8 to 21.9 h. These findings are consistent with the sand biofilter, whose removal efficiency ranged from 12 – 68% across all DOC concentrations. The DOC removals of a biological activated carbon filtration, preceded by coagulation/flocculation/clarification and pre-ozonation was reported to be between 12% and 38% [38]. Additionally, full-scale classical biofiltration treating Grand River water achieved mean TOC removals of 14% with anthracite filter media and 23% with granular activated carbon filter media [39]. In this study, the GAC biofilter achieved DOC removal of 12 – 91%. Although the removal efficiency achieved in this study was significantly higher than that of the previous research, this could be attributed to the pretreatment methods applied. The diffused aeration oxidized certain organic matter, making them more amenable to removal in the biofilters. Overall, the average DOC removal efficiency indicates that sand + GAC biofilter provides the most effective DOC removal across all DOC concentrations.

4.11. Statistical Analyses

4.11.1. Outlier test

Figure 4.1 shows a Grubbs outlier test at the 0.05 level of significance for DOC removal among the biofiltration systems. In these results, the null hypothesis states that all data values come from the same normal population. Because the p-value is 1 for DOC removal, which is greater than the significance level of 0.05, the decision is to fail to reject the null hypothesis because there is not enough evidence to conclude that an outlier exists. Another Grubbs outlier test at a significance level of 0.1 was also tested and the decision was to fail to reject the null hypothesis. Additional Dixon's Q test at significance levels of 0.05 and 0.1 were also test and the results were similar to the previous tests. Therefore, there are no outliers at the 5% and 10% level of significance.

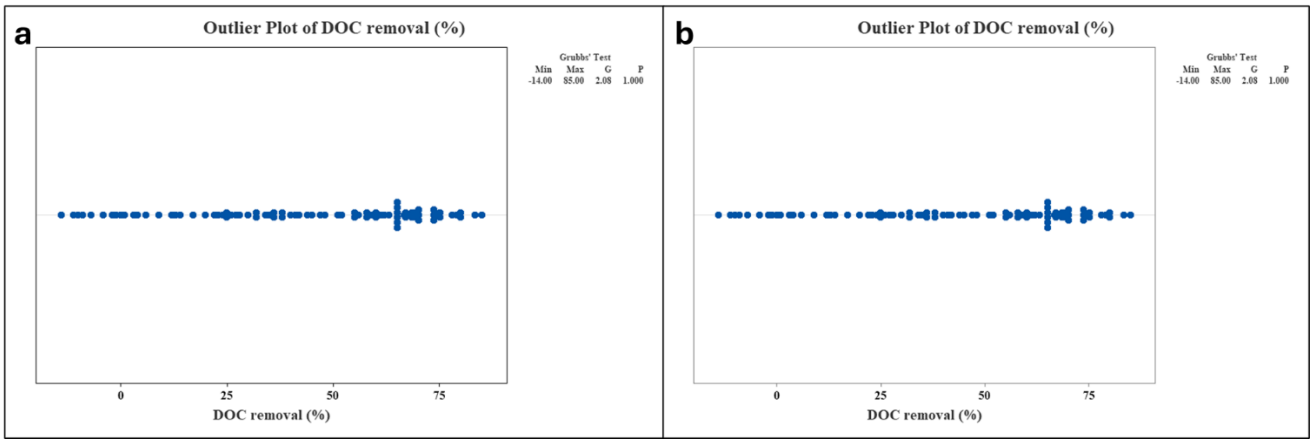

Figure 4.1: Outlier plot for DOC removal at α = 0.05 (a) 0.1 (b)

4.11.2. Normality test

Figure 4.1 shows the normality test results at the 0.05 level of significance for all three biofiltration systems regarding DOC removal efficiency. The mean DOC removal is 43.14%, with a standard deviation of 27.54%, indicating considerable variability in removal performance. The Kolmogorov-Smirnov (KS) test was employed due to the sample size exceeding 50 (n = 85), making it suitable for assessing the normality of large datasets. KS statistic of 0.139 and a p-value less than 0.010 suggest that the data significantly deviate from a normal distribution, implying that DOC removal efficiencies are not normally distributed. This deviation may be due to operational inconsistencies, varying influent water quality, or differences in biofilter media performance.

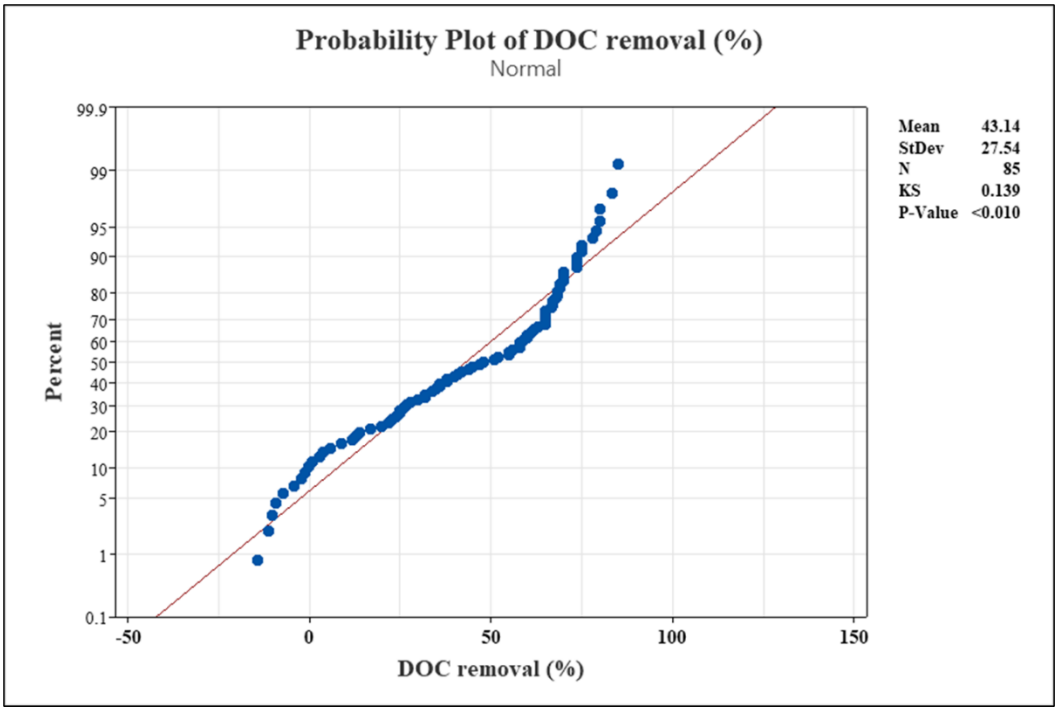

Figure 4.1: Normality test results for the DOC removal efficiency among the biofiltration systems

4.11.3. Residual plots

An assessment was conducted to verify that the residuals are adequate and meets the necessary analytical assumptions using the residual plots (**Figure 4.1**). The residuals versus fits plot proves that that residuals are randomly distributed about zero, and the variance of residuals is constant across the range of fitted values. This supports the assumption of homoscedasticity, further proving the normality test. The residuals versus order plot shows residuals scattered randomly without a trend, indicating that the residuals are independent from one another. Additionally, normal probability plot indicate that the residuals are approximately normally distributed because it follows a straight line. This supports the assumption of normality.

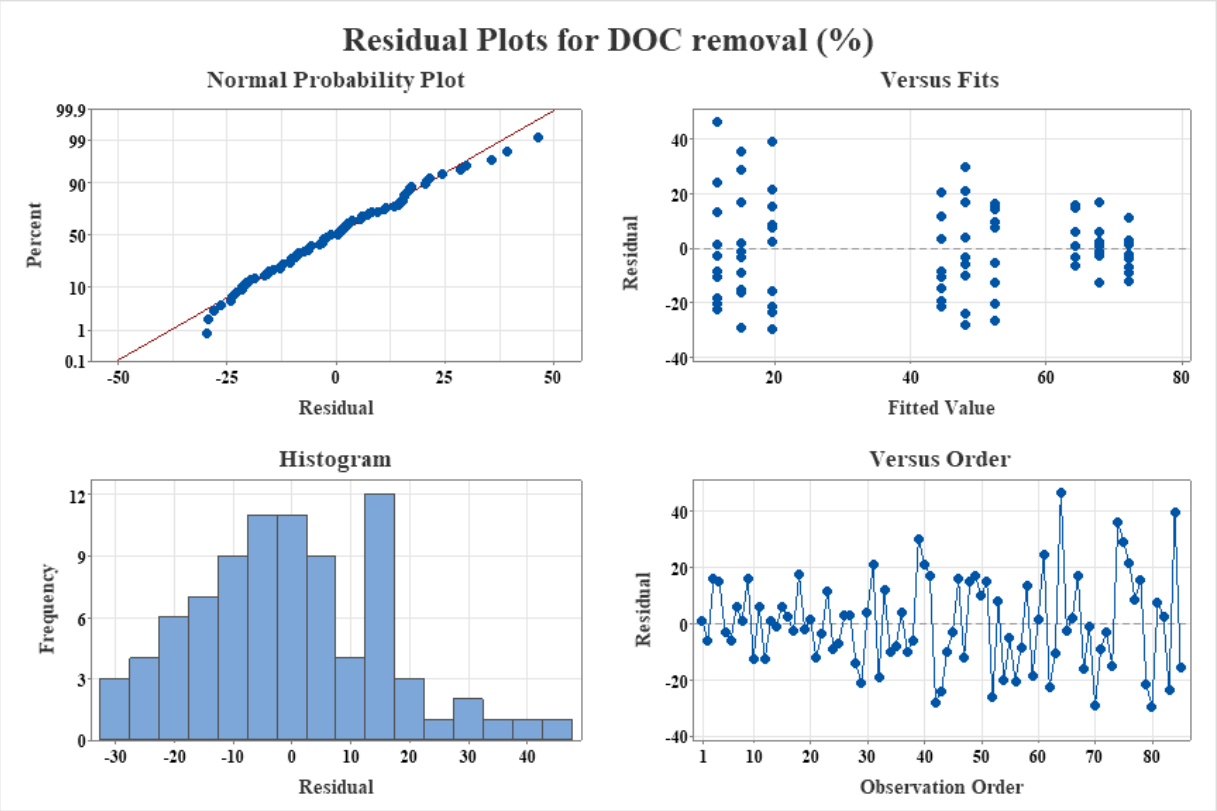

Figure 4.1: Residual plots for DOC removal

4.11.4. Correlation test

A Spearman correlation test was preformed to determine the correlation between influent DOC concentration and the DOC removal efficiency (**Table 4.3** and **Figure 4.1**). This non-parametric test was selected due to the non-normal distribution of the dataset, as indicated in prior analysis [159]. In these results, the Spearman correlation between influent DOC concentration and DOC removal efficiency is -0.798, which indicates that there is a strong negative relationship between the variables. The confidence interval for rho is from -0.872 to -0.688. The p-value is 0.000, which indicates that the relationship is statistically significant at the $\alpha = 0.05$ level. The relationship between these variables is negative, which indicates that, as DOC concentration increases, DOC removal decreases, and vice versa.

Table 4.3: Pairwise spearman correlations

Sample 1	Sample 2	N	Correlation	95% CI for ρ	P-Value
DOC removal (%)	DOC (mg/L)	85	-0.798	(-0.872, -0.688)	0.000

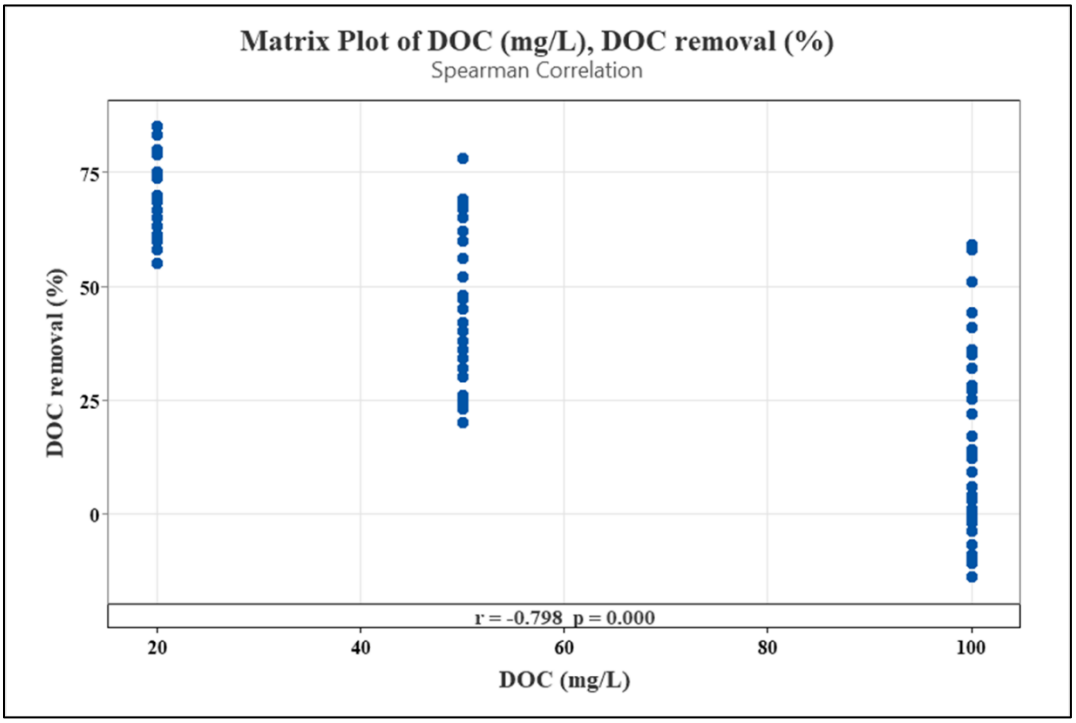

Figure 4.1: Matrix plot of DOC concentration and DOC removal

4.11.5. Pareto chart

A Pareto chart of the effects was used to determine whether media type or DOC concentration contributes the most to the variability in the DOC removal (**Figure 4.1**). The results show that only DOC concentration is statistically significant at the 0.05 level of significance because the bar extends past the red line.

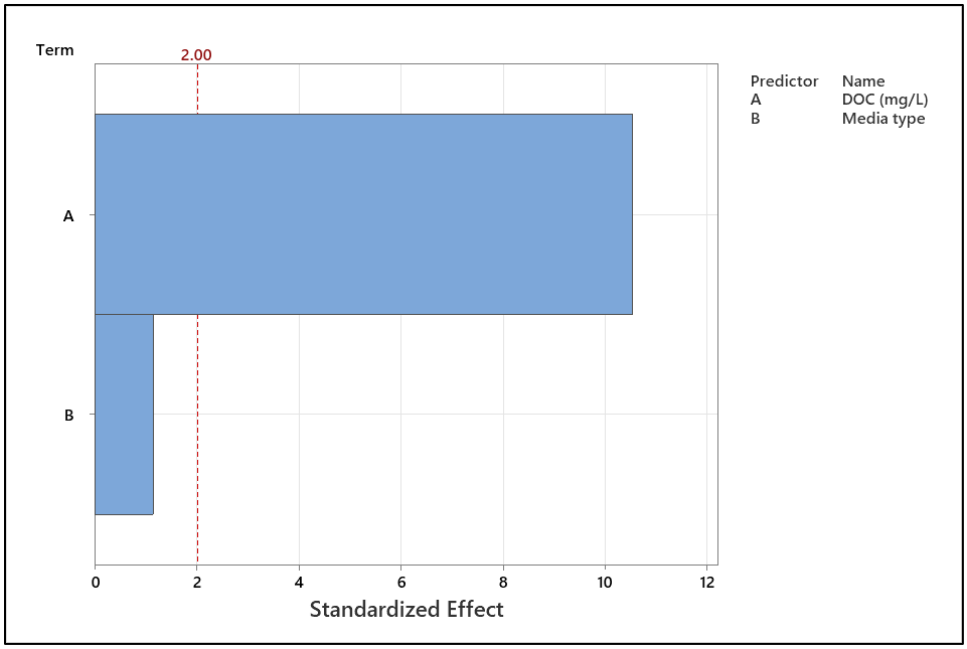

Figure 4.1: Pareto chart of the standardized effects

4.11.6. Analysis of variance (ANOVA)

An analysis of variance was conducted to determine whether any of the differences between the means are statistically significant (**Table 4.1**). A significance level of $p \leq 0.05$ was used to assess the null hypothesis that states that the population means are all equal. The results indicate that the null hypothesis – stating that the mean DOC removal values are equal across all DOC concentrations and media types – can be rejected. This conclusion is based on a p-value is less than the significance level of 0.05, suggesting that at least one group differs significantly in mean DOC removal. However, the p-value for media type (0.201) exceeds the significance level of 0.05, indicating that media type is not a statistically significant factor in influencing DOC removal efficiency. This finding aligns with the trends observed in the Pareto chart, further supporting the conclusion that while DOC concentration significantly affects removal efficiency, the type of media used does not have a statistically distinguishable impact under the tested conditions.

Table 4.1: Analysis of variance

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Regression	3	41492.2	13830.7	50.47	0.000
DOC (mg/L)	1	40509.0	40509.0	147.83	0.000
Media type	2	897.9	449.0	1.64	0.201
Error	81	22195.3	274.0		
Lack-of-fit	5	415.8	83.2	0.29	0.917
Pure Error	76	21779.5	286.6		
Total	84	63687.5			

4.11.7. Tukey post-hoc test

Based on the ANOVA results, the type of media used did not exhibit a statistically significant effect under the tested conditions. To further investigate differences among the biofilters, a Tukey post-hoc test was performed to identify which specific biofilters showed significant differences in their mean DOC removal performance (**Figure 4.15**). The test was conducted using a significance level of $p \leq 0.05$, evaluating the null hypothesis that the difference in means is zero. The results showed that at influent DOC concentrations of 20 mg/L and 50 mg/L, the null hypothesis could be rejected, indicating statistically significant differences among the biofilters. In contrast, at 100 mg/L DOC, the null hypothesis could not be rejected, as the p-value exceeded the 0.05 threshold. These findings suggest that at lower DOC levels (20 and 50 mg/L), the biofilters were effective in reducing DOC concentrations. However, at the higher influent concentration of 100 mg/L, DOC levels increased rather than decreased, indicating diminished or adverse performance under those conditions.

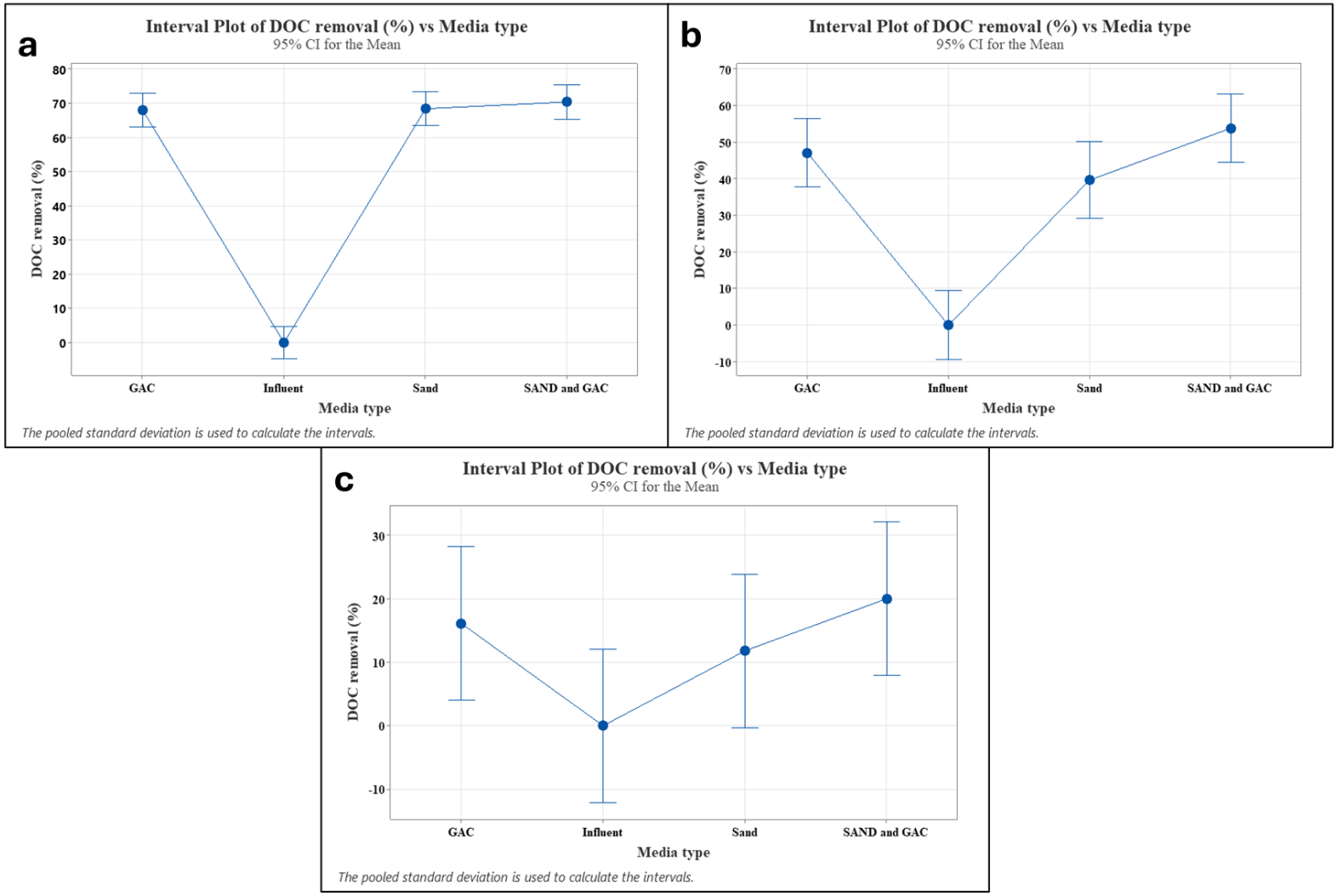

Figure 4.1: Interval plots of DOC removal vs media type at DOC = 20 mg/L (a) 50 mg/L (b), and 100 mg/L (c)

Chapter 5 Conclusions and Recommendations

5.1. Conclusions

The study investigated the impact of media type and varying DOC concentrations on the treatment performance of biofiltration. A consistent decline in dissolved oxygen (DO) across the treatment units confirmed active microbial respiration, a key mechanism in DOC removal. The system operated within an optimal temperature range (19–21 °C), supporting effective biological activity. Despite fluctuations in influent turbidity, all biofilters achieved high removal efficiencies, with the sand + GAC filter producing the clearest effluent.

UVA_{524} measurements showed that organic matter was highest in influent water, with the sand biofilter demonstrating the most stable reduction. pH levels varied due to the addition of alkaline compounds, occasionally exceeding the ideal range, particularly at higher DOC concentrations. Alkalinity fluctuated between 259 and 1470 mg/L, contributing to pH buffering but showing minimal impact on performance.

ATP analysis revealed that the sand biofilter supported the highest microbial activity over time, while EPS analysis indicated that TB-EPS was protein-rich and structurally stable, whereas LB-EPS was more sugar-dominant and dynamic. These findings highlight the role of EPS in biofilm development and resilience.

DOC removal efficiency varied with concentration and time. At 20 mg/L, steady state was reached by day 8, with GAC and sand + GAC filters achieving the highest removal. At 50 and 100 mg/L, performance declined, likely due to reduced microbial activity and elevated pH. ANOVA confirmed that DOC concentration significantly influenced removal efficiency, while media type did not. However, Tukey post-hoc test showed that at DOC concentrations of 20 and 50 mg/L,

biofilters significantly reduced DOC levels, while at 100 mg/L, no significant reduction was observed, indicating diminished effectiveness at higher concentrations.

These results emphasize the importance of pretreatment and operational control in optimizing biofiltration under variable conditions.

5.2. Limitations of the study

This research had some limitations:

- This study was limited to the use of synthetic wildfire-impacted water in a laboratory-controlled environment.
- This study was constrained by the deliberate control of only two operational parameters—temperature, EBCT and HLR. The exclusion of other potentially influential factors may limit the generalizability of the findings to systems operating under different conditions.
- A significant challenge encountered in this study was use of easily biodegradable chemicals to increase the DOC concentration. The DOC concentrations of the influent water decrease after a couple of days thereby causing inconsistency in the influent DOC concentration.
- Several factors such as fluctuating pollutant loads, biofilm development, and external environmental changes made it challenging to accurately characterize microbial behavior.
- The study did not examine biofiltration system performance under colder temperature conditions, which are known to influence microbial activity and treatment efficiency. This limitation may reduce the applicability of findings to colder climates.

5.3. Recommendations

Based on the results obtained from this thesis, it is necessary to further investigate the following research areas:

a. Future studies should consider using lake water as a test matrix to better simulate the complex and variable conditions of wildfire-impacted source water. Unlike synthetic or controlled laboratory water, lake water contains a diverse mixture of natural organic matter, particulates, and microbial communities, which more accurately reflect the challenges faced in real-world treatment scenarios following wildfire events.
b. To enhance the robustness and applicability of future studies, it is recommended that additional environmental factors–such as pH be systematically controlled and analyzed. Expanding the scope of controlled environmental parameters will provide a more comprehensive understanding of their interactions and overall impact on system performance.
c. To address the inconsistency in influent DOC concentrations caused by the rapid degradation of easily biodegradable compounds, future studies should consider using a blend of biodegradable and more recalcitrant organic matter. This approach would help maintain more stable DOC levels over time, better simulating real-world conditions and allowing for more consistent evaluation of biofiltration performance.
d. A comprehensive genomic analysis of microbial communities within biofilters can provide deeper insights into the specific environmental conditions required for optimal microbial growth and performance. Such understanding is essential for tailoring biofiltration systems to enhance treatment efficiency.
e. For future optimization studies, it is essential to investigate the performance of biofiltration systems under colder temperature conditions, as temperature significantly influences microbial activity and treatment efficiency.

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Appendices

Appendix A – Calibration curves

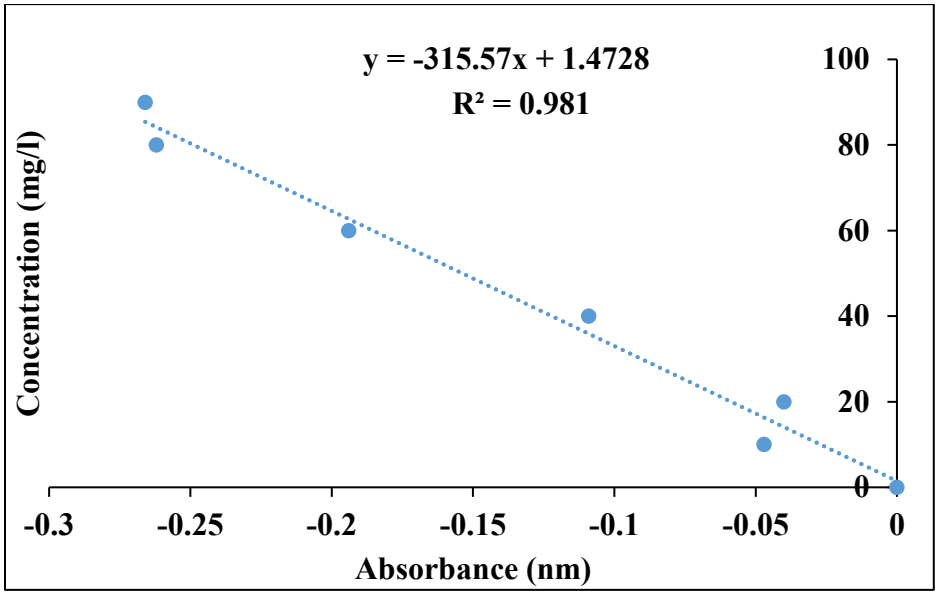


Figure A.1: Calibration curve for COD solution

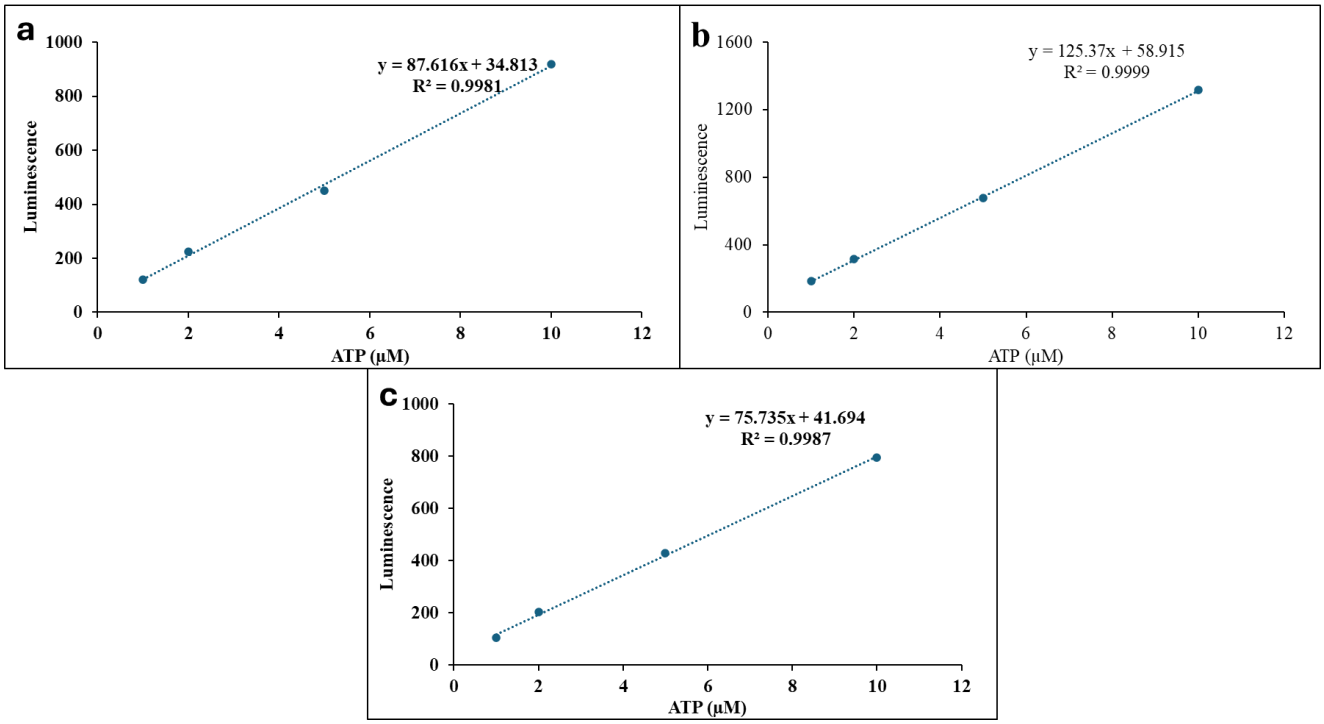


Figure A.2 ATP calibration curve for DOC at 20 mg/L (a), 50 mg/L (b), and 100 mg/L (c)

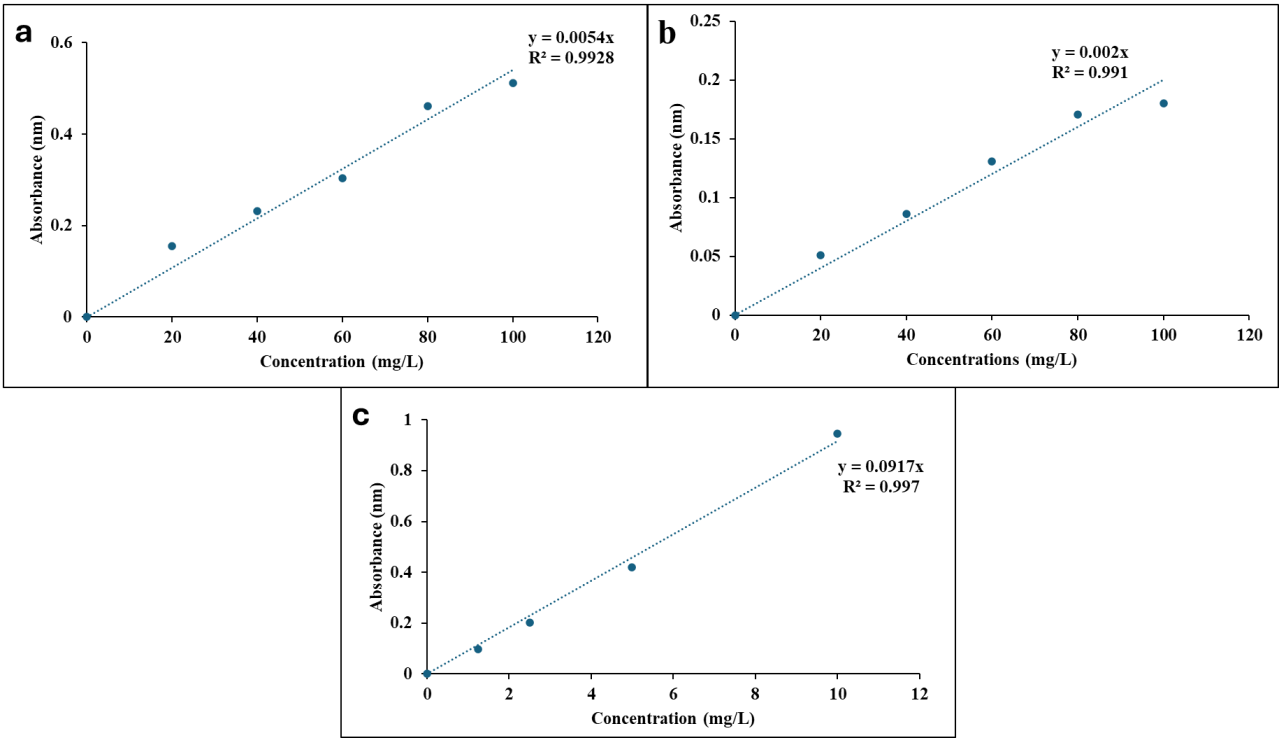


Figure A.3: Calibration curve for EPS - Proteins (a & b) and EPS - Sugar (c)

Appendix B – Influent water quality

Total Metals	Units	CDWQ[a] Guidelines	Ali Water Sample
Aluminum	mg/L	2.9 (MAC); 0.1 (OG)	0.230
Arsenic	µg/L	10	3.0
* Boron	mg/L	5	0.174
* Barium	mg/L	2	0.353
Calcium	mg/L	n/a	28.6
* Cadmium	mg/L	0.007	<0.0007
* Cobalt	mg/L	-	<0.0009
* Chromium	mg/L	0.05	0.005
Copper	mg/L	2 (MAC); 1 (AO)	0.0409
* Iron	mg/L	0.3 (AO)	0.444
Potassium	mg/L	-	39.1
Magnesium	mg/L	n/a	4.90
* Manganese	mg/L	0.12	0.235
Molybdenum	mg/L	-	0.0090
Sodium	mg/L	200 (AO)	77.4
Nickel	mg/L	-	0.0015
* Phosphorus	mg/L	-	1.19
* Lead	mg/L	0.005	<0.005
Sulfur	mg/L	-	1.14
Antimony	mg/L	0.006	<0.011
Selenium	mg/L	0.05	<0.018
* Tin	mg/L	-	<0.010
Uranium	mg/L	0.02	<0.012
Vanadium	mg/L	-	0.0078
* Zinc	mg/L	5 (AO)	0.0138

Physical Tests	Units	CDWQ[a] Guidelines	Ali Water Sample
pH	-	n/a	9.32
Electrical Conductivity	µS/cm	-	602
Hardness	mg $CaCO_3$/L	n/a	91.6
Turbidity	NTU	1	13.98
Total Dissolved Solids	ppm	500 (AO)	293
Total Suspended Solids	ppm	n/a	8
Langelier Saturation $Index_{20C}$	-	n/a	1.84
Colour	TCU	15 (AO)	80.1
Biological Tests	**Units**	**CDWQ[a] Guidelines**	**Ali Water Sample**
**E. coli*	MPN/100mL	0	0
*Total Coliforms	MPN/100mL	0	0
Heterotrophic Plate Count (HPC)	MPN/1mL	n/a	>2419.6

Figure A.4: Influent water analysis report

Anion Scan	Units	CDWQ[a] Guidelines	Ali Water Sample
Fluoride	mg/L	1.5	<0.1
Chloride	mg/L	n/a	6.2
Nitrite	mg/L	3	<0.1
Sulphate	mg/L	500 (AO)	3.0
Bromide	mg/L	-	<0.1
Nitrate	mg/L	45	<0.1
Phosphate	mg/L	-	2.6
Additional Parameters	**Units**	**CDWQa Guidelines**	**Ali Water Sample**
Total Nitrogen-N	mg/L	n/a	0.2
Total Kjeldahl Nitrogen-N	mg/L	n/a	0.2
Organic Nitrogen-N	mg/L	n/a	0.2
Oxidized Nitrogen-N	mg/L	n/a	<0.1
Ammonium-N	mg/L	n/a	<0.05
Total Inorganic Carbon-C	mg/L	n/a	83
Total Organic Carbon-C	mg/L	n/a	11
Odor	-	Inoffensive	Below threshold
Alkalinity	mg/L	n/a	309.6

Figure A.5: Influent water analysis report

Appendix C – Experimental results

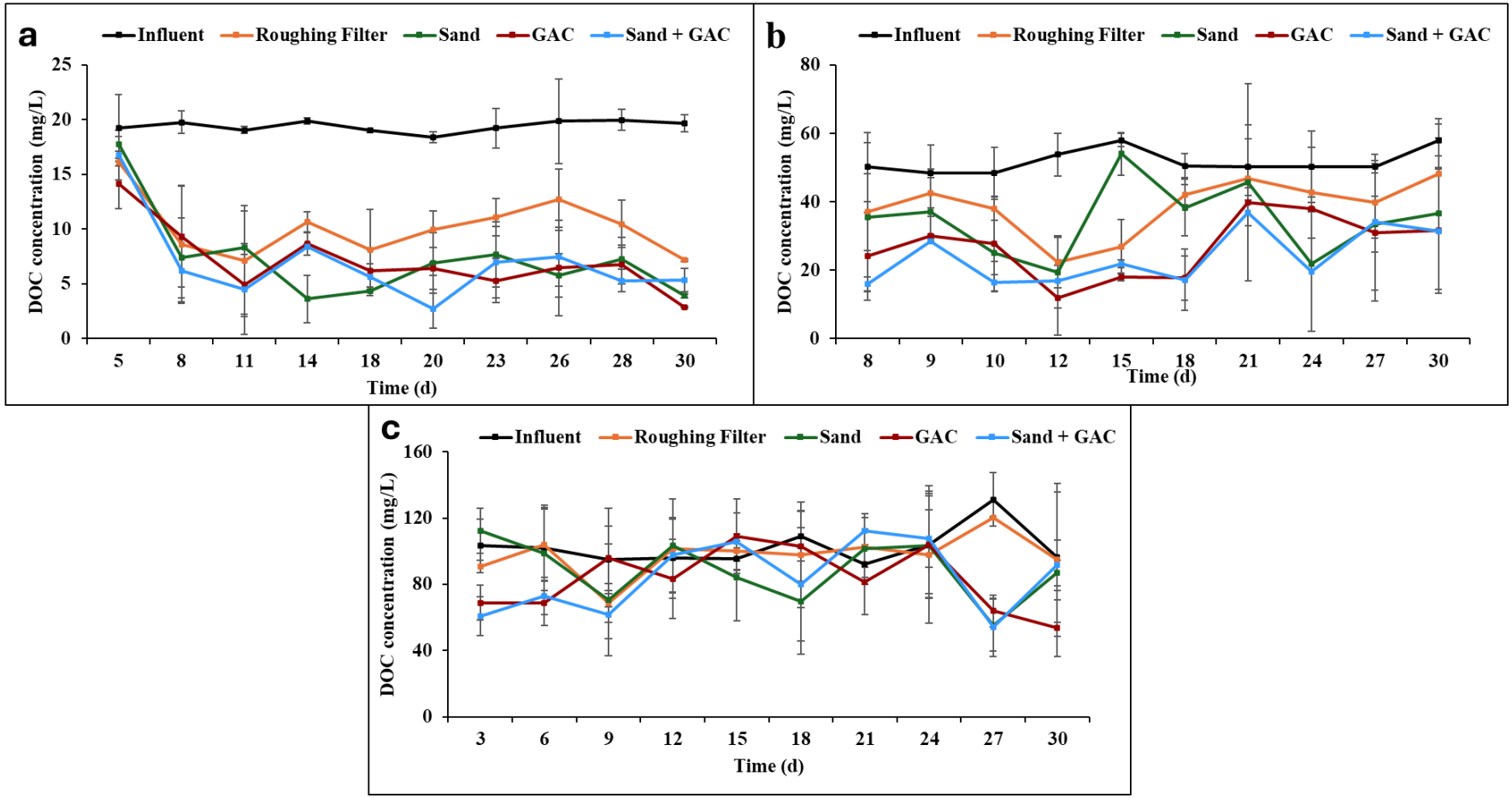


Figure A.6:DOC concentration of influents and effluents at DOC = 20 mg/L (a) 50 mg/L (b), and 100 mg/L (c)

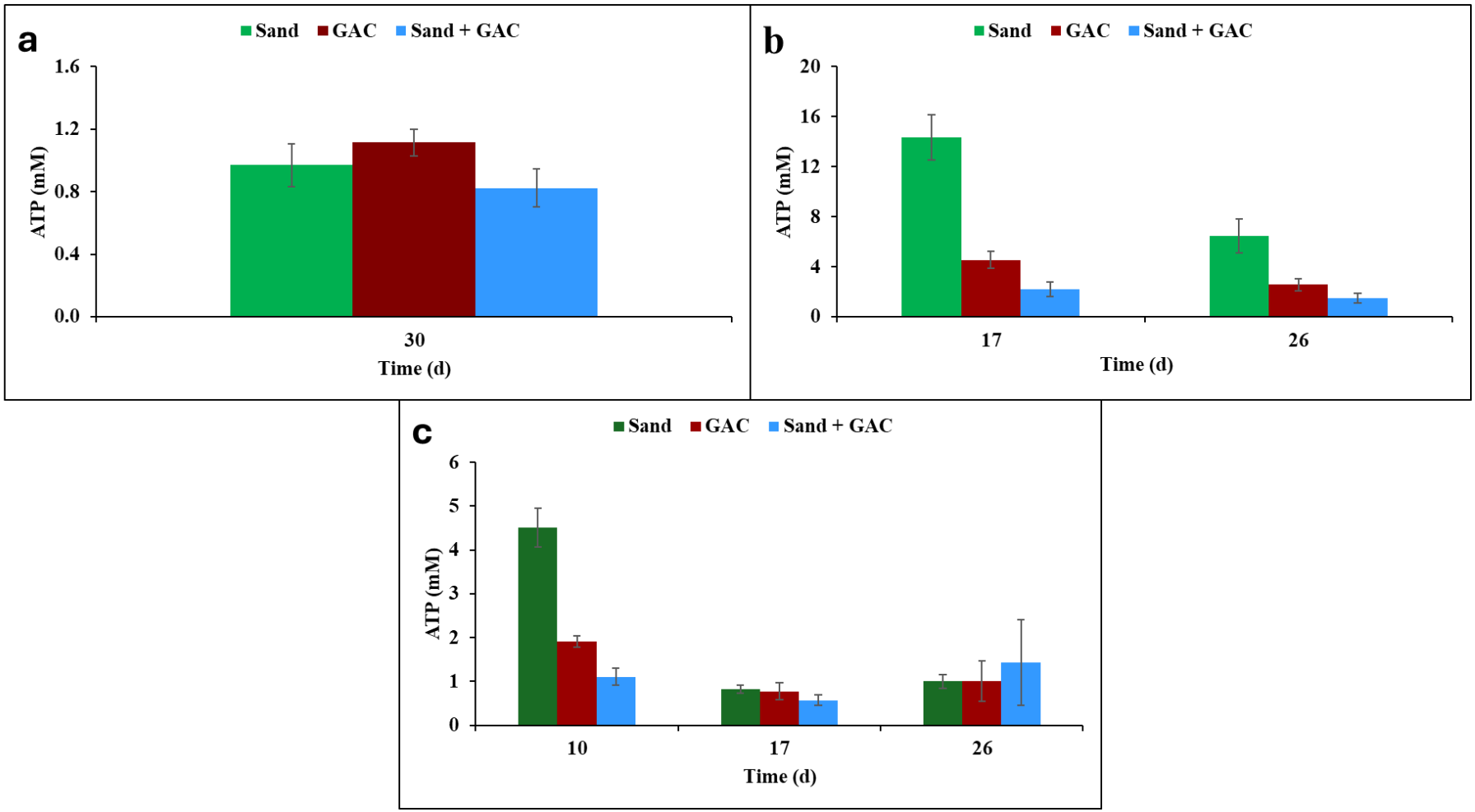


Figure A.7: ATP concentration of biofilter media at DOC = 20 mg/L (a), 50 mg/L (b), and 100 mg/L (c)

Table A.1 Comparison between DO and ATP concentrations of all runs

Experimental run	DO concentration (mg/L)	ATP concentration (mg/L)
R1	5 ± 1	1.0 ± 0.1
R2	3 ± 1	1.1 ± 0.1
R3	4 ± 1	0.8 ± 0.1
R4	4 ± 1	6.5 ± 1.3
R5	3 ± 1	2.6 ± 0.5
R6	4 ± 2	1.5 ± 0.4
R7	5 ± 1	1.0 ± 0.2
R8	5 ± 1	1.0 ± 0.5
R9	4 ± 2	1.4 ± 1.0

Appendix D – Statistical Analyses

Table A.2: Regression equation

Media type	
Sand	DOC removal (%) = 77.40 – 0.6575 DOC (mg/L)
GAC	DOC removal (%) = 82.00 – 0.6575 DOC (mg/L)
Sand + GAC	DOC removal (%) = 85.39 – 0.6575 DOC (mg/L)

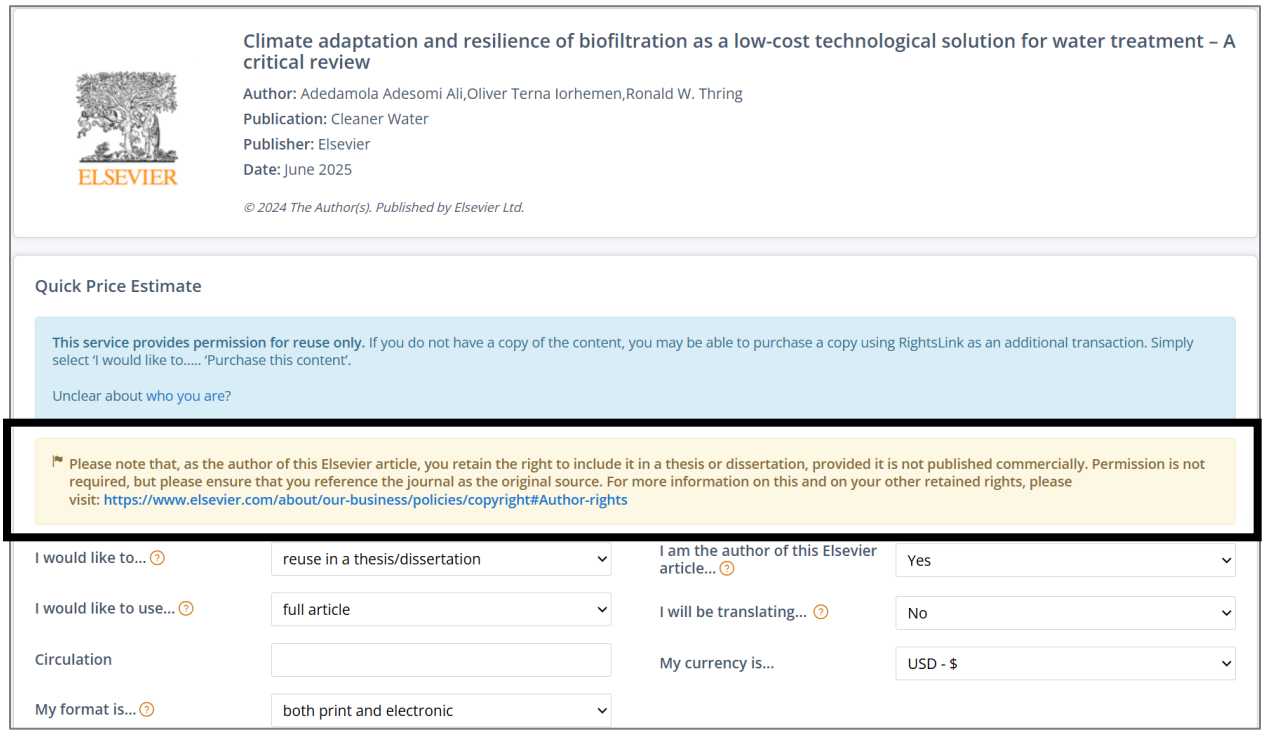
Table A.3: Regression coefficients and model summary

Coefficients					
Variables	**Coef**	**SE Coef**	**T-Value**	**P-Value**	**VIF**
Constant	81.26	3.62	22.44	0.000	
DOC (mg/L)	-0.6575	0.0541	-12.16	0.000	1.00
GAC	0.26	2.53	-0.10	0.918	1.35
Sand	-3.87	2.57	-1.50	0.137	1.35
Model summary					
S	**R-sq**	**R-sq (adj)**	**R-sq (pred)**	**10-fold S**	**10-fold R-sq**
16.5534	65.15%	63.86%	61.56%	16.8767	61.99%

Table A.4: Fits and Diagnostics for Unusual Observation

Obs	DOC removal (%)	Fit	Resid	Std Resid
64	58.00	11.64	46.36	2.88 R
74	51.00	15.25	35.75	2.22 R
84	59.00	19.63	39.37	2.44 R

Appendix E – Copyrights permissions



Climate adaptation and resilience of biofiltration as a low-cost technological solution for water treatment – A critical review

Author: Adedamola Adesomi Ali,Oliver Terna Iorhemen,Ronald W. Thring
Publication: Cleaner Water
Publisher: Elsevier
Date: June 2025

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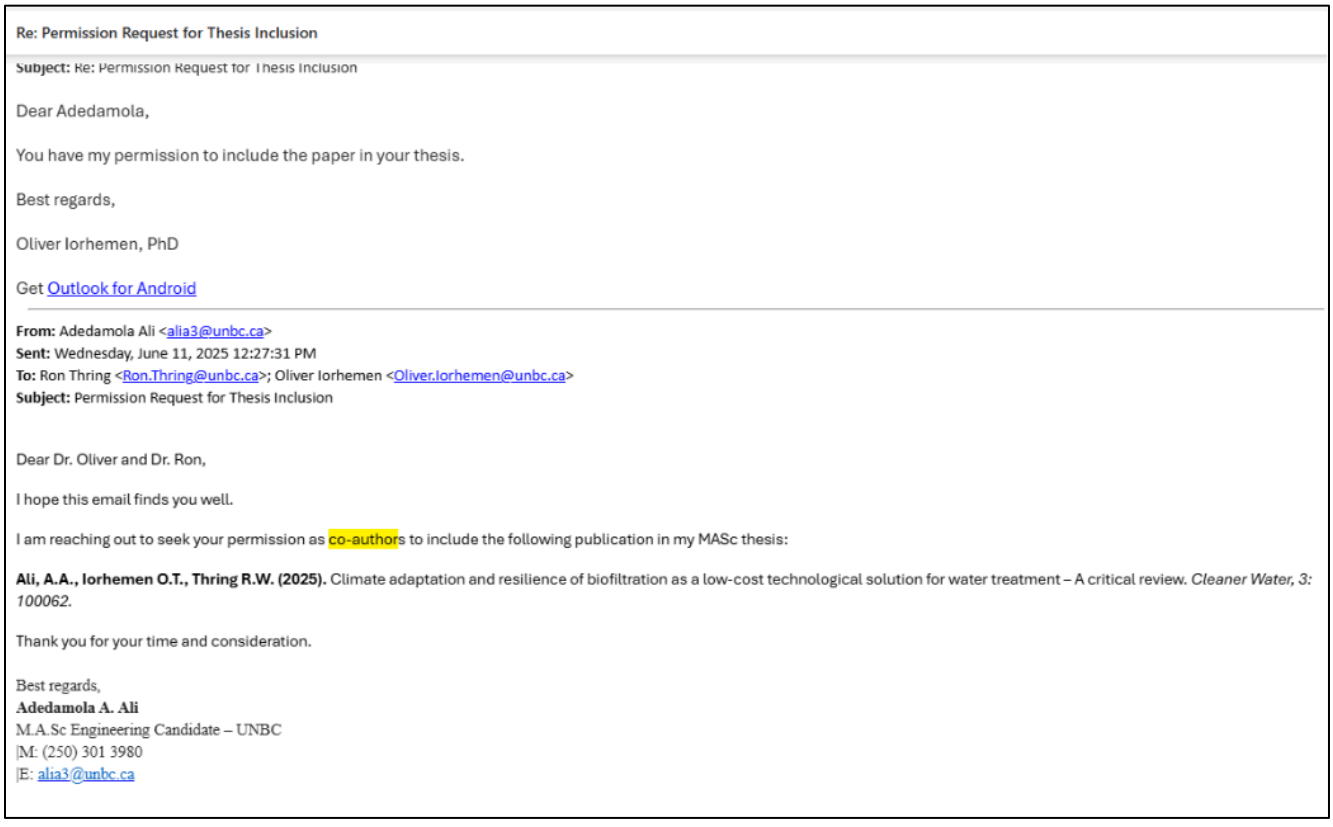
Figure A.8: Journal permission



Re: Permission Request for Thesis Inclusion

Subject: Re: Permission Request for Thesis Inclusion

Dear Adedamola,

You have my permission to include the paper in your thesis.

Best regards,

Oliver Iorhemen, PhD

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From: Adedamola Ali <alia3@unbc.ca>
Sent: Wednesday, June 11, 2025 12:27:31 PM
To: Ron Thring <Ron.Thring@unbc.ca>; Oliver Iorhemen <Oliver.Iorhemen@unbc.ca>
Subject: Permission Request for Thesis Inclusion

Dear Dr. Oliver and Dr. Ron,

I hope this email finds you well.

I am reaching out to seek your permission as co-authors to include the following publication in my MASc thesis:

Ali, A.A., Iorhemen O.T., Thring R.W. (2025). Climate adaptation and resilience of biofiltration as a low-cost technological solution for water treatment – A critical review. *Cleaner Water, 3: 100062.*

Thank you for your time and consideration.

Best regards,
Adedamola A. Ali
M.A.Sc Engineering Candidate – UNBC
|M: (250) 301 3980
|E: alia3@unbc.ca

Figure A.9: Co-author's permissions

Re: Permission Request for Thesis Inclusion

E: alia3@unbc.ca

From: Ron Thring <Ron.Thring@unbc.ca>
Sent: Wednesday, June 11, 2025 12:33
To: Oliver Iorhemen <Oliver.Iorhemen@unbc.ca>; Adedamola Ali <alia3@unbc.ca>
Subject: RE: Permission Request for Thesis Inclusion

Hello Adedamola,

You have my permission as well.

Cheers,

Ron

From: Oliver Iorhemen <Oliver.Iorhemen@unbc.ca>
Sent: Wednesday, June 11, 2025 12:29 PM
To: Adedamola Ali <alia3@unbc.ca>; Ron Thring <Ron.Thring@unbc.ca>
Subject: Re: Permission Request for Thesis Inclusion

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You have my permission to include the paper in your thesis.

Best regards,

Oliver Iorhemen, PhD

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Sent: Wednesday, June 11, 2025 12:27:31 PM
To: Ron Thring <Ron.Thring@unbc.ca>; Oliver Iorhemen <Oliver.Iorhemen@unbc.ca>

Figure A.10: Co-author's permission