Czech University of Life Sciences Prague

Faculty of Environmental Sciences

Department of Applied Ecology

Diploma Thesis

Effects of Different Filtration Materials on Removal of Selected Micropollutants from Synthetic Greywater

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Responsible water management in built-up areas in relation to the surrounding landscape

CZECH UNIVERSITY OF LIFE SCIENCES PRAGUE

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DIPLOMA THESIS ASSIGNMENT

Fatma Öykü Cömez

Landscape Engineering
Landscape Planning

Thesis title

Effect of Different Filtration Materials on Removal of Selected Micropollutants from Synthetic Greywater

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Objectives of thesis

The main aim of the thesis is to evaluate the efficiency of different filtration materials to remove selected micropollutants (diclofenac, coffein, metals, etc.) from synthetic greywater and assess their potential use in infiltration trenches.

Methodology

The work will be based on a series of laboratory experiments with columnn test fill up with different filtration materials, the efficieny to remove micropollutants will be evaluated.

The proposed extent of the thesis

70 pages

Keywords

Reuse of Greywater; Batch Test; Column Test; Removal Efficiency; Micropolutants; Diclofenac; Coffein

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Recommended information sources

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Prague on 19. 03. 2021

Declaration

I declare that I have worked on my diploma thesis titled " Effects of Different Filtration Materials on Removal of Selected Micropollutants from Synthetic Greywater" by myself and I have used only the sources mentioned at the end of the thesis. As the author of the diploma thesis, I declare that the thesis does not break copyrights of any their person.

In Prague on _____

Fatma Öykü Çömez

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Abstrakt

Přímé použití neošetřené šedé vody může způsobit významné ohrožení životního prostředí. Především díky rostoucímu používání léčiv a přípravků pro osobní péči (PPCP), které mohou působit ekotoxicky a být škodlivé pro biotu. Výzkum odstraňování těchto znečišťujících látek je proto nezbytný pro bezpečné opětovné použití šedé vody, což je slibné řešení nedostatku pitné vody. Hlavním cílem této práce je identifikovat účinnost odstraňování znečišťujících látek ze syntetické šedé vody (SGW) různými filtračmi materiály. Studie je založena na laboratorním experimentu, který testuje různé filtrační materiály po dobu 5 týdnů kolonovým testem. Kolony byly plněny týdně připravovanou SGW a vzorky byly měřeny denně po 20 resp. 72 (víkend) hodinách kontaktního času. Vybranými mikro znečišťujícími látkami byly PPCP (Diclofenac, Benzotriazol, DEET, Methylparaben, Kofein), aniontové povrchově aktivní látky, celkový dusík (TN), celkový organický uhlík (TOC) a stopové prvky (bór, měď, zinek a nikl). Filtrační lože kolon bylo navrženo ve třech vrstvách; nejprve 3,3 cm štěrku (4–8 mm), poté 13 cm písku (0–4 mm). Horní vrstva (tloušťka 28,6 cm) obsahovala technogenní půdu (kompost, písek a půda) a aditivum. Každá skupina obsahovala jen jedno z aditiv (dřevní štěpka, mykorhiza, drcené cihly, drcený vápenec a biouhel 5 nebo 10% objemu horní vrstvy). V každá testované skupině bylo 5 opakování. Výsledky ukázaly, že kromě benzotriazolu mohou ostatní PPCP a aniontové povrchově aktivní látky podléhat sorpci a biodegradaci. Z materiálů biofiltrů dosáhly vysoké účinnosti odstranění všech organických sloučenin včetně benzotriazolu pouze filtry s biouhlem. Účinnost odstranění kovů byla u všech testovaných materiálů více než 90%. V případě bóru bylo zjištěno jeho uvolňování z filtračních materiálů. Nejlepší výsledky účinnosti sorpce a filtrace byly zjištěny u filtrů s 5% biouhlu ve svrchní vrstvě, kde účinnost odstraňování PPCP a kovů byla vyšší než 90 % (kromě boru). Vzhledem k výsledkům může být biouhel potenciálně účinným a levným řešením pro úpravu šedé vody, pro odstraňování PPCP a těžkých kovů.

Klíčová slova: Infiltrace; Filtrační materiály; Syntetická šedá voda; Kolonový test; Účinnost odstranění; Diklofenak; Benzotriazol; DEET; Methylparaben; Kofein

Abstract

There is a risk of environmental damage if untreated greywater is used directly. Mainly due to the growing use of pharmaceuticals and personal care products (PPCP), which can have an ecotoxic effect and thus be harmful to biota. Research into the removal of these pollutants is therefore essential for the safe reuse of greywater, which is a promising solution to water shortage. The main goal of this study is to identify the role of different filter materials on the efficiency of removing pollutants from synthetic greywater (SGW). This study is based on a laboratory experiment which is testing different filtration materials for 5 weeks by a column test. The columns were filled with weekly prepared SGW and samples were measured daily after 20 resp. 72 (weekend) hours contact time. Selected micro-pollutants were PPCPs (Diclofenac, Benzotriazole, DEET, Methylparaben, Caffeine), anionic surfactants, total nitrogen (TN), total organic carbon (TOC), and trace elements (Boron, Copper, Zinc, and Nickel). The filtration bed of the columns was designed in three layers; first 3.3 cm of gravel (4-8 mm), then 13 cm of sand (0-4 mm). The top layer (28.6 cm thick) contained technogenic soil (compost, sand and soil) and an additive. Each group contained only one of the additives (wood chips, mycorrhiza, crushed bricks, crushed limestone and biochar 5 or 10% of the volume of the top layer). There were 5 replications in each treatment group. The results indicated that except benzotriazole, the other PPCP's and anionic surfactants might undergo sorption and biodegradation. Among the biofilter materials, only biochar filters achieved high removal efficiency for all organic compounds including benzotriazole. The metal removal efficiency of all biofilter materials was more than 90%. However, boron was found to be leached. Finally, the best results of sorption and filtration efficiency was observed for 5 % biochar filters (PPCP and metal removal efficiency) and it was higher than 90 % (excluding boron). Given the results, biochar can be a potentially effective and inexpensive solution for greywater treatment, PPCP and heavy metal removal.

Keywords: Infiltration; Filtration Materials; Synthetic Greywater; Column Test; Removal Efficiency; Diclofenac; Benzotriazole; DEET; Methylparaben; Caffeine

Table of Content

List of Abbreviations

AOPs	Advanced Oxidation Processes
BW	Blackwater
BOD	Biological Oxygen Demand
BTA	Benzotriazole
COD	Chemical Oxygen Demand
DCF	Diclofenac
DEET	Diethyltoluamide
EC	Electrical Conductivity
FOG	Fats, Oils,and Grace
GW	Greywater
HRT	Hydraulic Retention Time
IC	Inorganic Carbon
K	Potassium
MTP	Methylparaben
N	Nitrogen
NBS	Nature-based Solutions
NSAID	Non-steroidal anti-inflammatory drugs
SS	Suspended Solids
PPCPs	Pharmaceuticals and Personal Care Products
RW	Rainwater
TC	Total Carbon
TN	Total Nitrogen
TOC	Total Organic Carcon
TP	Total Phophate
TSS	Total Suspended Solids
P	Phosphorus
XOCs	Xenobiotic Organic Compounds
WW	Wastewater
WWTPs	Wastewater Treatment Plants

1. Introduction

Due to excessive consumption, pollution, climate change, and poor management practices, water is considered as a scarce resource. According to the estimations of the United Nations (UN), today, almost 800 million people live under water stress conditions and by 2025, it will reach 3 billion people because of the growing demand for water consumption at more than twice the rate of population growth through the last century (UN Water, 2014). Even though water is a scarce source, using water for daily household purposes cannot be considered as a luxury. However, domestic water consumption has a considerable impact on water pollution. In this sense, greywater plays a crucial role in wastewater produced by household's activities due to its considerably high potential of reuse. Greywater is a type of wastewater that is generated from household activities such as bathtubs, showers, washing machines, and kitchen sinks (kitchen effluent is in some cases excluded and considered as green water due to the concentration of fat). There are many studies that have been focused on the characteristics and reuse potential of greywater (Dixon, et al., 1999; Eriksson, et al., 2002; Jeffrey and Jefferson, 2003; Anderson, 2003; Bracken, et al., 2007; Ajit, 2016; Al-Gheethi, et al., 2019). The motivation to focus on greywater comes mostly from creating a new freshwater resource, protection of water resources, decreasing pollution, saving energy and cost, and mitigating excessive consumption of water (C. Ramprasad 2016; Arden and Ma 2018; Al-Gheethi, et al. 2019). In addition to that, greywater can be infiltrated into the ground in urban areas where it can help to restore the urban hydrological cycle (Eriksson, et al., 2002). Another important aspect is underlined by the study of Sinclair et al. (2013) made in Melbourne. It discovered that reusing greywater might influence people's choices towards using more environment-friendly products. However, measuring or estimating the acceptance of the community is a strong challenge for the implementation of greywater reuse methods (Jeffrey & Jefferson, 2003).

Although the reuse of greywater promises a considerable solution for water scarcity, direct reuse of untreated greywater could cause irreversible problems for the environment and human health such as contamination of groundwater resources (Turner, et al., 2019). Therefore, the qualification and quantification of greywater for pretreatment conditions are essential concerns as well as after the treatment process to assure the safety of both human and environment. Noticeably, micro-pollutants that are called xenobiotic organic compounds (XOCs) (such as pesticides, detergents, pharmaceuticals, personal care products, metals, and many others), have been detected in greywater due to the increasing intensity of their consumption (Noman, et al., 2019). Especially, during the last two decades, with the growing market of pharmaceuticals and personal care products (PPCPs), concerns about the existence of PPCPs in the environment are increasing due to the detecting of various types of medicinal products (hormones, anticancer drugs and

antidepressants, antibiotics, etc.) in numerous environmental compartments, such as surface water, ground water, soil, air, and biota (Daughton and Ternes, 1999; Daughton, 2001; Ebele, et al., 2017; Žižlavská and Hlavínek, 2020). As a nonsteroidal anti-inflammatory drug, one of the most consumed pharmaceutical compounds, diclofenac (DCF) has been well reported that the long-term existence has an ecotoxicological effect in wildlife and due to constant usage, it starts to become a persistent pollutant (Sathiskumar, et al., 2020). Similarly, as a high production volume chemical benzotriazole (BTA) and its derivatives have been included in different widely used products such as corrosion inhibitors, UV absorbers, dishwasher detergents and antifogging agents for photography (Kowalska, Felis, Sochacki, & Bajkacz, 2019). Therefore, there is a growing attention to improve an efficient method for the removal of such pharmaceuticals from wastewater. Including DCF, most of the PPCPs are not biodegradable, but also not easy to remove from aqueous phases by conventional water treatment plants due to its high polarity and solubility in water. That is why, many alternative methods (chemical treatment, ultraviolet light, etc.) have been developed, however, due to their high prices and energy consumption, additionally, various other obstacles such as ozonation and advanced oxidation processes (AOPs) cause residual toxic by-products, they have not been accepted widely (Bhadra, et al., 2017). In addition to these, household products for cleaning could be a source of phosphorus which is clearly identified as a potential environmental risk (Turner, et al., 2013) and the existence of metals in greywater is reported detailly by Eriksson and Donner (2009).

Especially in urban areas, creating self-sustainable communities has a crucial importance for the future cities, environment, and generation. Growing interest for improvement of nature-based solutions (NBS) drives less demanding, simple, reliable, environment supporting, cost-effective and energy saving solutions which are possible to use for households in both urban and rural areas. Among the NBS, to treat greywater, adsorption techniques provide a good alternative solution. Natural filtration systems (also so called biofilters) are considered as the primary treatment for greywater, mainly targetting suspended solids (SS) and adsorption of organic micro pollutants, and the most used systems for on-site greywater treatment. Therefore, different kinds of natural filtration materials are examined by different researchers (Šabršulová 2020; Perez-Mercado, et al. 2018; Dalahmeh, 2013) to design an efficient filtration bed for households, urban and rural green areas.

Regarding all the mentioned above, information about the presence of various pollutants in different types of greywater (bathroom, laundry, and kitchen) and knowledge about the removal efficiency of pollutants by natural materials are very limiting. In this manner, this thesis aims to evaluate the efficiency of different natural filtration materials to remove selected PPCPs, metals, and other micro-pollutants from a synthetic greywater and contribute to the knowledge of greywater treatment methods existing in literature.

2. Objectives

The main goal of this study is:

- To evaluate the efficiency of natural filtration materials according to the removal of micro-pollutants from synthetic greywater. Therefore, as biofilters; biochar 10%, biochar 5% (the different percentages of biochar represent different volume of biochar within the composite), compost (technogenic soil), woodchips, mycorrhiza, crushed brick, and limestone chippings were tested for their efficiency of pollutant removal. All the materials were mixed with sand, compost, and soil in different ratios.

In this case, the main objectives are:

- To analyze the removal efficiency of organic compounds; diclofenac, benzotriazole, DEET, methylparaben, and caffeine.
- To analyze the removal efficiency of boron and metals such as copper, nickel, and zinc.
- To analyze the removal efficiency of anionic surfactants.
- To analyze the removal efficiency of total organic carbon, total nitrogen, ammonium nitrogen.
- To analyze the removal efficiency of ions, such as fluoride, chloride, nitrite, nitrate, bromide, phosphate, and sulphate.

3. Literature Review

3.1. Origin of Greywater

The origin and characteristics of grey water are important to evaluate for treatment and reuse possibilities. Basically, it depends on three different criteria: quality of the water supply, different types of water distribution system both for drinking water and grey water (leaching from piping, chemical and biological processes in the biofilm on the piping walls), and the activities of the household because the existing compounds in grey water can vary according to the source depending on the lifestyle, customs, installation, and use of chemical products in households (Eriksson, et al., 2002).

Household wastewater are classified mostly into six categories as black, brown, yellow, grey, green, and storm waters. Blackwater (BW) contains both urine (called yellowwater) and faeces (called brownwater); greywater (GW) contains mainly detergents, soups, shampoos, and personal care products which are derived from laundry, baths, or sink; greenwater contains food particles, fats, oils and derived from kitchen sink and dishwashers; and stormwater is rainwater (RW) (Racek, 2020). Greenwater (also so-called kitchen greywater), in some definitions, considered as greywater, however, it highly contains fats, oils, and grease (FOG). The well-known impacts of FOG are high possibility for blocking the collectors due to accumulation of FOG and distasteful odor emission due to oxidation of fatty acids (Dalahmeh, et al., 2011). On the other hand, even though yellowwater is not considered as greywater (due to urine), both baths and laundry can be a source for urine which causes high concentration of phosphorus and nitrogen (Boano, et al., 2020).

The percentage of GW generated from household consumption is between 50-80% of total water usage, which means quantity of grey water highly based on domestic consumption (Al-Gheethi, et al., 2019). The quantity of GW can change according to the number of household members, their age, nature of living, demographics, and level of occupancy, geographical location, social habits, and water usage pattern and time (Al-Gheethi, et al., 2019). Hence, different countries have different GW production rates with different number of usages. For example, according to the study of Mohammed et al. (2016b) in Malaysia, most of the greywater generated from household activities was recorded from showers with an average of 50% of the total GW. This can be said because of hot and humid air conditions.

Additionally, by the separation of BW from wastewater, 80% of organic matter and main content of pollutants are being eliminated initially (Racek, 2020). In this sense, GW can be considered as a big potential of water resources for household consumption such as toilet flushing, laundry, lawn irrigation, windows, and car washing, and

groundwater discharge and fire extinguishing. Furthermore, agricultural irrigation is another vital area to consider the reuse of greywater since it is one of the most water consuming sectors according to the FAO (the FAO, 2021).

3.2. Chemical Composition of Greywater

The characteristics of greywater (GW) are separated as physical, microbiological, and chemical (Al-Gheethi, et al., 2019). Temperature, color, turbidity, suspended solids (SS), and total dissolved solids (TDS) are considered as physical characteristics of GW (Eriksson, et al., 2002). TDS indicates inorganic salts (such as calcium, magnesium, potassium, sodium, bicarbonate, chloride, and sulfates) and small amount of organic matter that is dissolved in water. The WHO suggests that higher TDS concentration than 1200 mg/L is not acceptable for drinking water; however, extremely low concentration of TDS would make the taste of water flat and insipid which is also unacceptable (the WHO, 2006 (a)). Total coliforms, Fecal coliforms, Thermoresistant coliforms, *Escherichia coli*, *Pseudomonas aeruginosa*, and Enterococci are described as microbiological indicators of GW which means, in case of their existence, a highly efficient GW treatment system, or highly disinfection is necessary (Racek, 2020).

Chemical characteristics of GW, which are pH, electrical conductivity (EC), chemical oxygen demand (COD), biochemical oxygen demand (BOD), total nitrogen (TN), and total phosphate (TP), alkalinity, heavy metals, disinfectants, bleach, surfactants, and detergents give more details about organic and inorganic constitutes. pH of greywater can range between 5 and 11 (Abed & Scholz, 2016), where on the other hand, blackwater pH ranges only between 6 and 7.7. Although there is no direct impact of pH on human, it is accepted one of the most important operational water quality parameters and suggested optimum pH range is 6.5 – 9.5 (the WHO, 2006 (a)).

Greywater can be obtained from different sources like laundry, bathroom, and kitchen. Laundry GW is known as the main resource for surfactants. High levels of surfactants present in GW can considerably accumulate in the soil and cause hydrophobic soil phenomena when it is used for irrigation (Dalahmeh, et al., 2011). Greywater (GW) from bathrooms can contain soaps, shampoo, body-fats, hair, fabric fiber, urine, and skin (Wurochekke A. A., 2016). In addition to these, pharmaceutical and personal care products (PPCPs) can be washed out easily during personal hygiene. Therefore, to express the pollution load, it is useful to divide the greywater as low-light (baths, shower, and sink) and high-strength (kitchen, washing machine, and dishwasher) according to the source of origin (Abed & Scholz, 2016). Additionally, even though the quality of organic matter in greywater can be similar with domestic mixed wastewater, it shows differences in terms of concentration (Al-Gheethi, et al., 2019). In the same way, the ratio of nutrient content in greywater considerably differs than in BW. For instance, the nitrogen (N) ratio is distributed as 93% and 7%, respectfully in BW and GW and similarly, the phosphorus (P) ratio is given as 83% in BW and 17% in GW; the potassium (K) ratio is 87% in BW

whereas 13% in GW (Racek, 2020). The COD is accepted as one of the main chemical characteristics which reveals real organic pollutants in grey water and the BOD is an index for the biological oxidation of organic compounds in the presence of molecular oxygen as an oxidizing agent to produce carbon dioxide and water (Al-Gheethi, et al., 2019). For a standard municipal wastewater BOD and COD ratio is described as 2:1 and a higher ratio indicates the presence of high concentrations of biodegradable organic matter (Racek, 2020). In another way, BOD determine the amount of oxygen need for microorganisms to decompose the organic matter present in wastewater. However, in GW, presence of detergents, shower products, and pharmaceutical compounds, and lower presence of faecal matter could result in high COD in comparison to BOD (Al-Gheethi, et al., 2019). In addition to COD and BOD, the total nitrogen (TN) is another important quality indicator of GW. TN concentration in greywater is expected to be less than mixed domestic wastewater due to the absence of urine in greywater. Despite this anticipation, urine has been detected time to time in greywater from bathrooms, as reported by Eriksson, et al. (2002). However, greywater sourced from kitchens has the highest concentration of TN with a range between 40-74 mg/L among the types of greywaters (Eriksson, et al., 2002). In addition to that, in some countries like Malaysia where both kitchen and bathroom wastewater are accepted as greywater, TN values recorded as from 10 mg/L to 38 mg/L (Wurochekke, et al. 2016a). In terms of ammonia (non-ionized (NH_3) and ionized (NH_4)) contamination, natural levels show differences such as in surface water the level usually under 0.2 mg/L, but anaerobic compartments may contain up to 3 mg/L (the WHO, 2006 (a)). Ammonia indicates possibility of bacterial existence, sewage, or waste pollution in water. The other parameter, the level of total phosphorus (TP) might differ as amount quite according to the ingredient of washing detergents.

According to the conducted literature review, it is observed that domestic mixed wastewater and greywater show similar compounds, whereas the concentration is changing, and the pathogen content is much less in greywater than domestic wastewater. Additionally, there is also another important aspect that social acceptance of greywater is higher and easier than mixed wastewater (Nghiem, et al., 2006). In the Table 1, the minimum and maximum values from some of the physical, chemical, and microbiological characteristics of greywater, that are obtained from literature, are shown.

Erikkson et al. (2002) provides a detailed literature review of greywater characteristics based on different sources such as bathroom (from 13 different sources), laundry (from 9 different sources), and kitchen (from 10 different sources) originated greywaters (Table 1). It is shown that kitchen effluent has much higher nutrient concentrations than the other effluents and similarly, biological, and chemical oxygen demand and total organic carbon (TOC) were observed as higher concentrations than the others. Moreover, Nghiem et al. (2006) compared greywater and mixed domestic wastewater values in their paper according to an in-depth literate review. According to that, nutrients (TN and TP) contents are higher in wastewater than greywater. The characteristics of greywater that are given by Hourlier et al., (2010) (shown in the Table 1) were collected from five households from north-west of France in urban and rural areas.

Samples were collected from baths, showers and wash basins of the households and were mixed before analyses. In the end, the real greywater was composed of adults (80%), children (<15 years old 10%), and babies (<2 years old 10%). The work by Oteng-Peprah, et l., (2018) indicates results from one hundred eighty greywater samples collected from the Central Region of Ghana (in the Table 1, the results only from in-house water source are considered). The samples were collected as mixed greywater of handwash basins, kitchen, and bathrooms from households with children.

In addition to these, Chrispim and Nolasco (2017) have been reported in their study, raw greywater characteristics from a university building in Brazil. The building was used daily by 65 people and only three of them used the shower every day, and nearly 30 people used the washing machine in the building. The characteristics of the greywater sourced from showers, lavatories, and washing machine showed differences. TN (mg/L) measured 50.3 in shower greywater, whereas 5.1 and 4.3 measured for lavatory and washing machine greywaters, respectively. Additionally, sulphate was 111.5 mg/L in washing machine greywater, 10.9 mg/L in shower greywater and 12.3 in lavatory greywater.

The review of the published literature reveals that the concentrations of contaminants in greywater highly depended on the source and locations. This implies that treatment systems for greywater must consider contaminant alterations and influent of peak contaminant loads.

Table 1: Characteristics of Real Greywater in Literature

Characteristics of Greywater	Reference		Erikkson et al. (2002)			Nghiem et al. (2006)		Hourlier et al. (2010)	Oteng et al. (2018)
	Greywater Source		Bathroom GW	Laundry GW	Kitchen GW	Domestic Wastewater	Greywater	Greywater	Greywater
Physical		*Total solids (mg/L)*	250-631	410-1340	1500-2410	200-1200	113-2410		
		TSS (mg/L)	54-200	120-280	235-720	24-608	3.1-330	23-80	192-414
Chemical		*pH*	6.4-8.1	8.1-10	6.3-7.4	5.9-7.7	5-10.9	6.5-7.8	5-8.1
		EC (µS/cm)	82-250	190-1400				331-434	1204-2434
		BOD_5 (mg/l)	76-200	48-380	1040-1460	28-400	33-1460	85-155	87-301
		TOC (mg/L)	30-104	100-280	600-880	42-290	30-880		
		COD (mg/L)	100-633	725	936-1380	45-1000	3.8-1380	176-323	207-1299
		Chloride (mg/L)	9.0-18	9.0-88					18-49
	Ground Elements	*Boron (mg/L)*	<0.1	<0.1-0.5					
		Calcium (mg/L)	3.5-7.9	3.9-14	13-30	1.1-200	3.6-200		9-43
	Nutrient	*NH_4-N (mg/L)*	<0.1-15	0.06-11.3	0.2-23				7.0-22.0
		NO_3-N (mg/L)	0.28-6.3	0.4-2	0.3-5.8				0-5
		Nitrogen (mg/N/L)	5.0-17	6.0-21	40-74	20-117	0.28-74		
		Phophorus (mg/P/L)	0.11-2	0.062-57	68-74	3.2-30	0.0062-74		1-3
		PO_4-P (mg/l)	0.94-48.8	4-171[a]	12.7-32				
	Heavy Metals	*Cu (mg/L)*	0.06-0.12	0.05-0.27	0.068-0.26				
		Ni (mg/L)		<0.028	<0.025				
		Zn (mg/L)	0.2-6.3	0.09-0.44	0.12-1.8				
Microbiological		*Total coliform (per 100 ml)*	70-2.4x10^7	85-3x10^8				1.7x10^8-1.4x10^9	2.5x10^6-4.9x10^6

[a] Detergent contains phophorus

3.2.1. Micro-pollutants in Greywater

Even GW is a type of wastewater which has lower concentrations of infectious pollutants than mixed domestic wastewater, still, it includes many different micro-pollutants. In the literature, micro-pollutant of GW has been studied quite detailly (Eriksson, et al., 2002; Eriksson & Donner, 2009; C. Ramprasad, 2016; Turner, et al., 2019; Al-Gheethi, et al., 2019). Therefore, it is necessary to consider origins of the pollutants and health aspects due to microorganisms and accumulation of xenobiotic organic compounds (XOCs) in the environmental compartments like soil, ground water, and surface water (Eriksson, et al., 2002). XOCs are one of the most common organic compounds and can exist in GW due to pharmaceuticals, personal care products, cleaning agents, pesticides, and many others.

3.2.1.1. PPCPs in Greywater

Pharmaceuticals and personal care products (so called PPCPs) include a wide-ranging compound. Pharmaceuticals are explained as therapeutic drugs used for the prevention and treatment of human and animal diseases, while personal care products aim to increase daily life quality (Ebele, et al., 2017). PPCPs are considered as a unique group of emerging environmental contaminants due to their increased utilization and non-breakable properties. Typical PPCPs detected in GW are generally ingredients of cosmetics, medicines, preservatives, disinfectants, detergents, and many more that are being used in household activities.

There are many different pharmaceuticals that has been detected in GW. Some of the common PPCPs, that are also studied in this thesis, are classified into different categories, as shown in the Table 2.

Table 2: Categories of Some Pharmaceutical and Personal Care Products (PPCPs)

Category / Sub-category	***Compound***	***Mode of Entry***	***Environmental Compartments***	***Reference***
Pharmaceuticals				
Analgesics / Non-steroidal anti-inflammatory drugs				
	Diclofenac	Discharges, disposal, accidental spills, farmland waste, wastewater irrigation	Wastewater treatment plant, rivers, streams, groundwater, agricultural runoff	(Bhadra, et al., 2017), (Arslan, et al., 2017), (Xu, et al, 2009)
Psychoactive Drugs				
	Caffeine	Household, direct disposal of waste	Wastewater treatment plants, terrestrial runoff, freshwater, marine, estuarine environments, and sediment	(Al-Mashaqbeh, et al., 2019)
Personal Care Products				
Insect repellents				
	N, N-diethyltoluamide (DEET)	Shower waste, direct disposal of waste	Wastewater treatment plants, terrestrial runoff, freshwater, marine, estuarine environments, and sediment	(Al-Mashaqbeh, et al., 2019)
Detergents, Surfactants				
	Sodium dodecyl sulphate	Industries, laundries, households,	Wastewater treatment plants	(Ramprasad, 2016)
Preservatives				
	Methylparaben	Kitchen waste, shower waste	Wastewater treatment plant	(Li, et al., 2015)
UV filters and Stabilizers				
	Benzotriazole	Municipal waste, shower waste, discharge from swimming pools	Waste water treatment plants, marine, surface waters, soil, and groundwater	(Montesdeoca-Esponda, et al., 2021) (Kowalska, et al., 2019)
Solvent				
	Glycerin	Shower and sink waste		

Diclofenac belongs to the group of non-steroidal anti-inflammatory drugs (NSAID). In 2017, 248.881 packs of diclofenac were sold in the Czech Republic (Žižlavská & Hlavínek, 2020). It is very commonly detected in surface waters and most commonly, the source is effluents of wastewater treatment plants (WWTPs) (Buser, et al., 1998). However, diclofenac has been found in groundwater, hospital effluents, and drinking water. Although some analgesics, like aspirin, were not identified as problematic for the environment thanks to biological degradation processes, diclofenac has been found problematic due to its high persistence and some negative impacts on some species such as rainbow trout and for some predatory birds (Žižlavská & Hlavínek, 2020). Diclofenac showed the most acute toxic nature with effects being observed at concentrations below 100 mg/L among the NSAID (Santos, et al., 2010). In Europe, as the legislative target, 100 ng/l is recommended for diclofenac approval in water ecosystems (Žižlavská & Hlavínek, 2020). Diclofenac was detected in WWTP effluents at maximum concentrations of 2400 and 1420 mg/L in Switzerland and Belgium respectively, which highlighted that the concentrations are of sufficient magnitude to suspect chronic toxicity in aquatic organisms. Diclofenac has also been found in rivers, groundwater, hospital effluents, and drinking water but at concentrations in the order of ng/L (Ebele, Abdalla, & Harrad, 2017).

Benzotriazole (BTA) is used in wide-ranging of activities from cleaning products to corrosion inhibitors. BTA is an highly water soluble compound and not easily biodegredable (Kowalska, et al., 2019).

N, N-Diethyl-meta-toluamide (DEET) (also called diethyltoluamide) is a very known active ingredient in insect repellents (Sui, et al., 2010).

Methylparaben (MTP) is included in parabens which are commonly used for cosmetics and preservative products such as shampoos, make-up, shaving products, and deodorants. Their level of toxicity is usually low and therefore, it is being used in make-ups safely (Šabršulová, 2020). Additionally, parabens are commonly found in food additives (Nguyen, et al., 2021) such as blueberries and alcohol beverages. Furthermore, like the other preservatives, MTP are used to prevent growth of molds, yeasts, microorganisms, and fungi for cosmetics.

Similarly, like other PPCPs, MTP is commonly found in grey waters due to its application in self-care cosmetics on skin. In addition to this, parabens are detected in effluents of WWTP and untreated wastewaters (Nguyen, et al., 2021). MTP as a high biodegradable compound is reported 100% removal efficiency by *Pseudomonas beteli* and *Enterobacter cloacae* and over 95% removal efficiency by mixed culture and activated sludge (Nguyen, et al., 2021). Similar results also indicated by Li, et al, (2015) that parabens including methylparaben were degrading 91.8% of the initial mass loading and contribution of sorption to this lost was only 7.5%. So, biodegradation is the most significant way for removal of parabens during conventional treatment.

Caffeine (CAF) due to its quite commonly usage is considered as the most representative emerging pollutant among pharmaceuticals and, is a widely used ingredient for food and drinks like coffee, tea, chocolate, etc. CAF is a pharmaceutical compound

which is used to reduce physical fatigue and restore alertness. It is existence are reported in many water compartments including wastewater, surface water, and groundwater. The most effective method of caffeine removal is adsorption due to its high removal efficiency and simplicity (Sui, et al., 2010).

3.2.1.2. Surfactants

The official name “surfactants” is named for surface-active compounds. Its alternative name is “tensides” where comes from the Latin word “tensus” which means “tense” in English. There are different types of surfactants exist in wastewaters such as anionic surfactants, non-ionic surfactants, cationic surfactants, and amphoteric surfactants. As general, surfactants represent the largest group of XOCs in greywater (Noman, et al., 2019). They are also considered under personal care products due to their existence in shampoos and detergents.

Anionic surfactants are the most used group in cleaning products (Steber, 2007). It has been reported that 67.16% of 6.7 million metric tons worldwide produced surfactants were anionic surfactants (Noman, et al., 2019).

3.2.1.3. Trace Elements in Greywater

In domestic greywater, boron is present due to everyday usage of detergents. The constant entrance of boron can be toxic to plants and environment; therefore, it is necessary to reduced boron concentration before reuse of greywater (Ghavanloughajar, 2015). Boron can be sourced from nature or human activities like detergent contained sodium perborate. Although boron limit was decided as 0.3 mg/L by WHO in 1993 guideline for health, WHO (a) (2006) suggests that 0.5 mg/L boron concentration is acceptable for drinking water due to its highly existence in some natural compartments.

There are broad range of possibilities for metals that can exist in GW. The range of potentials sources goes from plumbing materials to cutlery, jewelry, coins, home products for maintenance, products of arts and crafts, and dental fillings (Eriksson & Donner, 2009). Copper is an essential nutrient and contaminant of water. It is used to make pipes, valves, and coatings where can easily be a path to enter to greywater. Also, in developing countries, food and water can be a source for copper. It is stated by WHO (2006) (a) that 2 mg/L is the limit concentration for copper in drinking water. Nickel can leak from certain type of kettles, non-resistant materials of pipes or taps or food. 0.07 mg/L is the value of acceptance for nickel concentration in water (WHO, 2006 (a)). Zinc is an essential trace element found particularly in all food and potable water in the form of salt or organic complexes. Normally, the levels of zinc in surface water and groundwater do not exceed 0.01 and 0.05 mg/L, respectively. However, in tap water, it is possible that the level of zinc can be higher due to dissolution of zinc from pipes. The

WHO quality of drinking water suggests that drinking water containing zinc at levels above 3 mg/L is not acceptable for people (the WHO, 2006 (a)).

3.3. Chemical Composition of Synthetic Greywater (SGW)

Simulation of greywater has gained attention from researchers who work on the treatment and reuse of greywater due to its stable properties (Abed & Scholz, 2016). Therefore, many different recipes have been developed by different researchers such as Nghiem, Oschmann and Schafer (2006), Diaper, Toifl and Storey (2008), and Nazim and Meera (2013).

Several qualifications are described for the synthetic greywater formulation which are considered:

- Mimicking of a real greywater composition,
- Maintaining a condition that micro-organism and pathogen can survive,
- Detectable concentrations of compounds that have a detrimental effect on environment and could be observed in real greywater, and
- Providing reproduceable and consistent quality for different batches and users (Diaper, Toifl, & Storey, 2008).

Nghiem, Oschmann, and Schafer (2006) investigated the treatment approach of submerged ultrafiltration membrane for recycling of greywater. The synthetic greywater recipe that they had used in their study includes kaolin, cellulose, humic acid, calcium chloride, sodium chloride, and sodium bicarbonate. Kaolin is commonly selected for synthetic greywater recipes in order to represent suspended inorganic and organic solids in greywater, which can be originated from kitchen and laundry effluents. Similarly, cellulose is commonly chosen to represent organic fibers in greywater which originates from kitchen effluent. Diaper, Toifl and Storey (2008) developed a synthetic greywater recipe based on products that expected to be found in average Australian households including personal care products, detergents, and some additional laboratory chemicals (sodium dodecyl sulphate, sodium hydro-carbonate, sodium phosphate, boric acid, and lactic acid), and clay. Boric acid is commonly used in synthetic greywater recipes to represent boron ions in greywater. Also, in recipes, iron (III) chloride, manganese (II) chloride, chromium (III) nitrate, zinc sulphate, copper sulphate cadmium oxide, and lead (II) oxide are used in the recipes to provide heavy metal compositions to synthetic greywater (Abed & Scholz, 2016). Different recipes of synthetic greywater that are inspired by many other studies in the literature, are shown in the Table 3. However, it is noted by Abed & Scholz (2016) that accurate reproduction of most recipes are not realistic due to unreported and variable environmental conditions. Additionally, personal care products like shampoo or sun screens, and other ingredients like cow dungs are not always indicated clearly.

Table 3: Reported Recipes of Synthetic Greywater in Literature

(Source: Adapted from Abed & Scholz, 2016)

Reference	*Abed & Scholz (2016)*		*Nazim & Meera (2013)*		*Hourlier et al. (2010)*		*Diaper et al. (2008)*	
Country	Sweden		India		France		Australia	
Study Approach	Testing effects of the storage time to SGW characteristics		Treatment of SGW by Garbage Enzymes after filtration		Testing the performance of direct nano-filtration process for recycle of SGW		Three different testing: 1)Biological with suspended media. 2) Chemical flocculant dosing, UV and four stage filtration. 3) Settling, biological with fixed media	
		Low Concentration (mg/L)	High Concentration (mg/L)	mg/L		mg/L		mg/L
	Kaolin	15	100	Glucose	300	Lactic acid	100	Sunscreen or moisturiser	15 -10
	Cellulose	15	100	Sodium acetate trihydrate	400	Cellulose	100	Toothpaste	32.5
	Humic acid	5	20	Ammonium chloride	225	Sodium dedocyl sulphate	50	Deodorant	10
	Sodium chloride	10	120	Sodium dihydrogen phosphate	150	Glyserol	200	Sodium sulphate	35
	Sodium hydrogen carbonate	10	85	Potassium dihydrogen phosphate	75	Sodium hydrogen carbonate	70	Sodium hydrogen carbonate	25
	Calcium chloride	10	55	Magnesium sulphate	50	Sodium sulphate	50	Sodium phosphate	39
	Potassium nitrate	0	90	Cow dung	225	Septic effluent	10	Clay	50
	Calcium nitrate	0	150					Vegetable oil	0.7
	Magnesium sulphate	2	240					Shampoo/hand wash	720
	Monopotassium phosphate	13	85					Laundry	150
	Iron(III)chloride	0.3	50.0					Boric acid	1.4
	Boric acid	0.6	3.0					Lactic acid	28
	Manganese(II)chloride	0.03	3.20					Secondary treatment effluent	20
	Zinc sulphate	0.25	15.00						
	Copper sulphate	0.025	7.000						
	Ammonium molybdate tetrahydrate	0.35	0.35						
	Cadmium oxide	0.02	12.50						
	Nickel oxide	0.02	0.06						
	Chromium(II)nitrate	0.045	70.000						
	Sodium sulphate	2.6	25.0						
	Sodium phosphate monobasic	0.00	250.00						
	Lead(II)oxide	0.16	1.40						
	Secondary treatment effluent with microbial content (ml/L)	20.00	100.00						

3.4. Removal of Pollutants from Greywater

Segregation of GW from wastewater can have many benefits including saving water and reducing energy consumption and operation cost by decreasing wastewater amount in WWTPs. Considerable amounts of GW are being produced by household activities, e.g., in Europe, 75% of total domestic wastewater is GW (Boano, et al., 2020). Thanks to its lower number of contaminants than wastewater, GW is suitable for onsite recycling which can provide a new water resource, critical in arid regions. In this sense, nature-based solutions (NBS) for GW treatment are attracting increasing attention. On the contrary with conventional systems, NBS supports the reduction of energy and water consumption, and provides aesthetic value to the environment as ideally, also shows high removal efficiency for micropollutants in GW. Therefore, there are some basic parameters that are necessary to be considered for the selection of an onsite GW recycling system for households which have been studied and discussed by different researchers (Shaikh & Ahammed, 2020; James, Surendran, Ifelebuegu, Ganjian, & Kinuthia, 2016; Dixon, et al., 1999). These parameters can be listed as follows:

- Qualitative characteristics of GW

As already discussed previously on the part of chemical composition, GW shows differences in composition according to the source (kitchen, bath, sink) in terms of contamination level.

- Quantitate characteristics of GW

GW production can vary according to culture, age, income level, socio-economic level, education, country (location, climatic region), gender, number, and occupancy of household members (Shaikh & Ahammed, 2020).

- Variation of water flow

GW flow pattern shows variations between weekdays (Monday-Friday) and weekends (Saturday-Sunday) reported by different studies (Shaikh & Ahammed, 2020). In addition to this, during a day, it is notices that the highest GW flow was between 7:00-10:00 h and 17:00-22:00h. Because these hours are representing mostly before and after work and school hours.

- Storage conditions of GW

Another underlined indicator about the quality of greywater is the storage time (Eriksson, et al. 2002) and indicated that 24 hours storage can have an increasing effect on the quality of greywater, whether more than 48 hours can create serious problems due to depletion of dissolved oxygen. Additionally, Dixon et al. (1999) mentions storing greywater 48 hours in the temperature between 19-26^{o}C causes the growth of micro-organisms which is a serious health risk for human.

- Environmental Effects of GW

The effects of organic and inorganic compounds in greywater to soil and plants have been studied intensively, and the issue of removing pharmaceuticals from the water cycle is not an easy task. Conventional wastewater treatment plant is not enough effective or only capable of degrading those substances that are eagerly degradable with high adsorption abilities (Žižlavská & Hlavínek, 2020). The concentration of drugs in wastewater can have differences according to various factors including the behavior of people in seasonal periods, urban sprawl, water changes due to heavy rains, and synergic impact of other pharmaceuticals.

3.4.1. Conventional Technologies for GW Treatment

Conventional wastewater treatment systems are centralized systems that are commonly used in many countries around the world. These systems are connected to a central treatment plant collecting different types of wastewaters as domestic, commercial, industrial, storm, and urban runoff water, and then, discharges after treatment into the nearest surface water. Conventional technologies are classified as chemical, physical, physio-chemical, and biological according to the type of removed contaminants, and adopted process (Boano, et al., 2020). The aim of the conventional wastewater treatment processes are the removal of pathogens and priority pollutants such as COD, BOD, and TSS. Coagulation and flocculation as a chemical GW treatment process are one of the most popular which have succeeded to remove 85-89% BOD_5, 64% COD, 13% TN, >99% TOC and >99% E. coli (Boano, et al., 2020). Physical GW treatment focuses on more removal of turbidity, TSS, colloidal and can achieve up to 93% TSS, 98% BOD5, 94% COD, 98% TN, 100% TP, and 100% E. coli removal efficiencies (Boano, et al., 2020). However, there is not enough contribution that these technologies are sufficient to remove XOC's. For instance, one of the current treatment method advanced oxidation process (AOP), is besides being insufficient and expensive, it has also many toxic by-products. The other technologic treatment processes are ultrasonic irradiation, electrochemical, biodegradation, and ozonation. However, conventional techniques are not being preferential recently due to their high energy requirements and cost of maintenance.

3.4.2. Nature-based Technologies for GW Treatment

3.4.2.1. Constructed Wetlands

Constructed wetlands (CWs) are one of the most known alternative and nature-based method for conventional treatment systems for greywater reuse and recycle (Arden & Ma, 2018). There are various types of CWs for wastewater treatment explained by Vymazal (2010) as; free water surface (FWS), horizontal subsurface flow (HSSF), vertical subsurface flow (VF), and combination systems (or so called hybrid systems) which consists of different types combination. In addition to these, green roof water cycle

(GROW) and recycling vertical flow (RVF) constructed wetland types are described by Arden & Ma (2018). FWS CWs are effective systems for removal of organic compounds through microbial degradation and settling, and removal of nitrogen through nitrification; however, they are not generally efficienct for phosphorus removal. HSSF CWs contain gravel or rock beds isolated with an impereable layer and wetland plants. In this type of CWs, wastewater enter to the system from an inlet and flows through a porous medium in a more or less horizontal path to the outlet where it is collected and discharged. This systems can use both microbial degradation for the removal of organic compounds and chemical/physical processes for the removal of suspended solids and nitrogen (usually by denitrification). Besides, VF CWs are the ones which have more requirements of maintanence and cost due to periodically pumping system of wastewater to the wetlands surface. In this type of wetlands, the wastewater is fed by large batches. Then, the water percolates down through the sand medium and only after the all water percolate, a new batch is added which makes this sytem more aerobic than HSSF CWs (Vymazal, 2010). CWs are nature-based wastewater treatment technologies which is an alternative cost-efficient technology. They are designed to mimic the same processes occurs in natural wetlands (Vymazal, 2010) and have been extensively studied for the removal of organic matter and nutrients (nitrogen and phosphorus) (Vymazal, 2013), but also for the removal of pharmaceuticals (PhCs) from wastewater (Ramprasad, 2016; Ilyas & Hullebusch, 2019).

Ilyas and Hullebusch (2019) investigated detailly the removal of PhCs by CWs based on the role of design and operational factors, as well as the physicochemical parameters of four types of CWs which are FWS CWs, HF CWs, VF CWs, and hybrid constructed wetlands (HCW). The considered parameters and factors were treatment scale and type, wastewater type, depth, area, hydraulic loading rate (HLR), organic loading rate (OLR), experiment duration, system age, filter media composition, pH, temperature, effluent dissolved oxygen, and oxidation-reduction potential (ORP). In this regard, their paper reviewed information about over 253 CWs from 19 countries. The results showed a significant correlation for removal efficiency of about half of the examined PhCs with two or more design and operation factor of CWs. One of the significant effects to removal efficiency was presence of plants that help the removal of some PhCs. Additionally, using substrate materials that have high adsorption capacity, rich organic matter, and high surface area helped the removal of PhCs like codeine, clarithromycin, erythromycin, ofloxacin, oxytetracycline, carbamazepine, and atenolol.

Many other pollutant removals by constructed wetlands have been studied in the literature. Ramprasad (2016) had studied the performance of HFCW and VFCW in pilot scales for greywater treatment. In the study, specifically fate of surfactants (sodium do decyl) and personal care products were evaluated together with the effects of hydraulic retention time (HRT) and external organic load change. The results show that summer season has a marginal effect on removal of pollutants postively and proposed that in order to achieve the reuse standards by treatment of greywater, 6.8 day of HRT is required within the diamentions of 7.5 m lenght and 90 cm depth.

Regarding boron removal by wetlands, a study from United States was found that tested the ability of fourteen plants common in wetland environments for the region to accumulate selenocyanate, arsenic, and boron from electric utility wastewater. After a necessary time for plant growth, the wetland was irrigated by utility wastewater with boron concentration about 50 mg/L. According to the results over 42 days, removal efficiency of boron was 31% by the wetland and the significant way of removal pathway was through absorption to soil layer. Seven plants out of fourteen were able to accumulate boron in their structure; however, this was only 3% of the total boron removal (Ghavanloughajar, 2015).

3.4.2.2. Bio-filters

The application of biofilters has been increased with the development of green infrastructure around the world. Even though these systems were considered more convenient for treatment and infiltration of stormwater, they are being discussed as a very useful method for extension of application for greywater treatment. To illustrate, an innovative biofiltration (bioretention) system that is designed to establish in a parking lot is given in the Figure 1. These systems are recently improved natural treatment systems that use vegetations and soils action and can be used for residential or urban settings (Hydro International UK, 2021). In general, biofiltration is a multipurpose treatment system used for wastewater treatment. If disinfectants do not pass throught a filter, it can create a good environment for microorganism to grow and form a active biofilm (Greenstein, et al., 2018).

Figure 1: Biofiltration System Designed for Treatment of Stormwater

(Source: taken from Hydro International UK, 2021)

Greywater, as generated from household activities such as cleaning or washing, contains a low concentration of nitrogen, whereas inversely may contain high concentrations of organic compounds, surfactants, salts (Dalahmeh, et al. 2011). On the other hand, greywater from kitchen can contain high concentrations of nitrogen, fats, oil, and grease, and detergent due to dishwashing (Wurochekke, et al., 2016). That is why, pre-treatment is required for greywater before reuse. Even though there are various ways of treating greywater according to its characteristics, there is no globally accepted way of design. In some countries, especially where there is a shortage of potable water like the USA (California, Arizona), Australia, Jordan etc., there are simple greywater treatment systems and standards for the reuse of treated water (Dixon, et al. 1999; Al-Gheethi, et al. 2019). Greywater most preferable treated on-site systems which are detached to households (Dalahmeh, et al. 2014) by separating from central sewage system and decentralized (A possible design example is shown in the Figure 2. On-site treatment systems are expected to be easy to operate with basic skills, cost, and energy efficient; and reliable for affordable usage by private owners (Lens, et al., 1993). Therefore, filtration systems are accepted as quite efficient treatment systems for greywater due to their ability of removing microorganisms and pathogens by physical adsorption or entrapment.

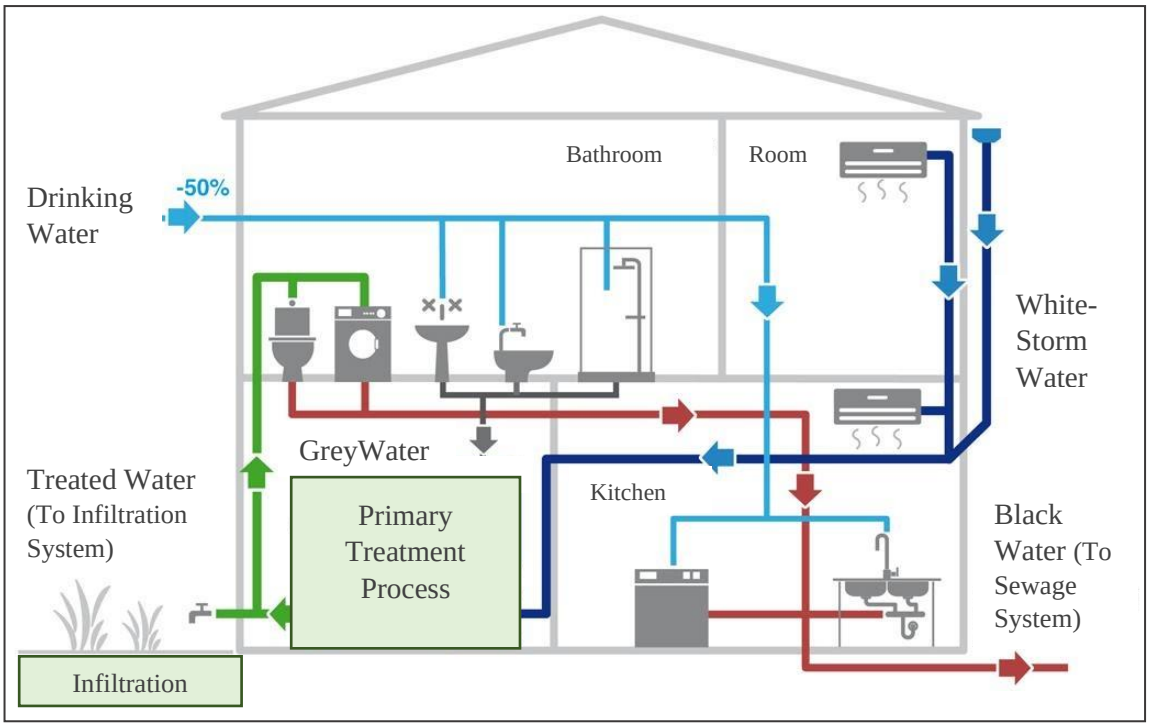

Figure 2: A Possible Hybrid System (Preliminary + Nature-based Treatment) attached to a Household for Separation of Wastewater (Source: taken from REDI, 2021)

As mentioned in the conventional technologies, the other options for greywater treatment (GWT) are physical/chemical GWT system which includes filtration and disinfection methods and biological treatment methods with aeration and membrane bioreactors (MBRs) (Wurochekke, et al., 2016) Although these methods provide high efficiency, they (conventional systems) are not commonly applied due to high cost and energy consumption. In this sense, natural treatment systems which contain preliminary, primary, and secondary processes (a hybrid system) can offer low cost, easy operation, and high efficiency in removal of nutrients, heavy metals, and microorganisms (Wurochekke, et al., 2016).

Regarding the on-site wastewater treatment systems, the filter system is commonly preferred (Dalahmeh, et al. 2014) and sand is widely used natural filtration material around the world (Dalahmeh, et al. 2011). Additionally, many organic by-products have been studied as alternative filtration materials such as bark, compost, peat, corncobs, wheat straw, woodchips, bricks, limestone, etc., and some of their combinations (Dalahmeh, et al. 2011). During the percolation period, purification happens as physical (filtration and adsorption) and biological (degradation) (Lens, et al., 1993). The existing literature on sand and soil filters about the removal performances are extensively describes both for steady and changing conditions (Dalahmeh, et al. 2014) and there is a growing interest in the literature for different low-cost treatment systems for greywater (Wurochekke, et al., 2016).

In their study, Dalahmeh, et al. (2012) evaluated the performance of bark, activated charcoal, polyurethane foam, and sand filters for the reduction of contaminants from synthetic greywater treatment with a laboratory column experiment. According to the results, pine bark and activated charcoal filters gave high efficiency for the removal of BOD_5 as 98 and 97%, and for the removal of TP as 97 and 91%, respectively. Additionally, the effluent of charcoal was offered to use in agriculture instead of chemical fertilizers due to high nutrient concentration.

Properties of Filter Materials

The removal capacity of the pollutant depends on properties of the filter materials such as particle size and distribution, specific surface area, surface chemical composition, adsorption capacity, and porosity (Dalahmeh S. , 2013). Microorganisms can attach to particles, interphases between materials, pores within the materials, cracks which is called retention. Adsorption and straining are the main processes that originates the retention. Adsorption is the process in which an atom, ion, molecule, or particle from other materials, attaches each other or accumulate on the surface of other materials. Hydraulic and organic loading capacities of filter materials are the other important properties to avoid any clogging or preferential ways during the percolation (Dalahmeh, et al., 2014) which could decrease the impact of adsorption and degradation on the treatment efficiency.

Soil and sand are known as the most used filter materials for greywater treatment; however, the soil is not enough to use singly as a filtration material for on-site systems due to limited soil permeability (Lens, et al., 1993) which can cause soil and groundwater pollution. There are a couple of more materials that catch the attention like bark, woodchips, charcoal, and non-activated charcoal (biochar) (Dalahmeh, et al., 2012) that have been studied for filtration material. Recently, biochar studies showed efficient results for using as a filter material for greywater treatment (Perez-Mercado, et al., 2019; Šabršulová, 2020) For instance, biochar scored 95% of TN, and 99% of COD removal which was the best removal rate in comparison with the other filtration materials including perlite (45% TN, and 75% COD removal), river sand (45% TN, and 75% COD removal) and expanded clay (50% TN and 40% COD removal). On the other hand, biochar achieved only 26% of TP removal where the highest TP removal was achieved by vermiculite as 61% (Pradhan, et al., 2019).

Loading Conditions and Treatment Capacity

Grey water flow can show variability characterized by daily and hourly minimum and maximum flows and immediate peak flows that occur during the day. These changes of flow affect onsite systems by potentially causing hydraulic overload of the system during peak flow conditions (US EPA, 2002). The efficiency of treatment processes may vary according to the seasonal (for instance, hot weather of summertime could increase shower frequencies), weekly (weekdays and weekend differences), and daily changes of greywater loading. The amount of greywater can vary according to the daytime. For

example, usually within working hours (between 8 am to 5 pm), household water consumption is expected to be less than during the time just before and after working hours (6 am to 8 am and 5 pm to 8 pm). It is suspicious that these instabilities or constant situations can affect the efficiency of the filter materials, therefore, some studies have been simulating these variations. Dalahmeh, et al., (2014) reported a performance analysis on three different filter materials (pine bark, activated charcoal, and sand) for different hydraulic regimes with synthetic greywater and found out that charcoal filters showed a large capacity for treating of high flows of greywater, on the contrary, bark filters were efficient for treating greywater under low flows and after high grey water flow, they were able to recover back the treatment performance.

Life Span of the Filters

The service life of a filter can be expressed as the period when the filter can perform efficient removal of pollutants. One of the most reported failures of filter materials are caused by clogging. In their study, Lens, et al. (1993) reported that woodchips and bark showed no clogging for a period of 150 days after application of raw domestic wastewater while peat filters were clogged after about 100 days of application. It is important to consider service life especially for woodchips and mycorrhiza due to their organic origins which make them more fragile for degradation than the inert material of biochar and sand. Additionally, the drought period (such as holidays) when there is no wastewater supply to the system affected the filtration efficiency and especially caused an increase in nitrate and nitrite levels in the effluent (Lens, et al., 1993).

Furthermore, the uprising question is the fate of filter materials after the end of usage period. For instance, it has to be carefully decided what to do with filters contaminated with PPCP's or heavy metals or how they can be stored to not cause any pollution to the environment. This issue needs to be more detailly investigated.

3.5. Reuse of Greywater

Reuse of greywater is opening a great chance to mitigation measures of drought and water stress. However, raw, or direct reuse of greywater can cause several problems for the environment and human health due to high content of organic matter, pathogens, and solids (Anderson, 2003). Only few studies about greywater have explored the potential environmental pollutants like pharmaceuticals and personal care products (Eriksson & Donner, 2009). The growing consumption of pharmaceutical products, both human and veterinary have captured many researcher's attention due to its ecotoxicological risk to environment (Cordy, et al. 2004; Ebele, Abdalla and Harrad 2017; Ilyas and Hullebusch 2019), but little is done about their removal from greywater. As the reuse of greywater possibilities are being recognized, different countries or regions start to setup their own quality requirements as regulations and guidelines. Basic aim of these

guidelines is to provide reliable information and encourage the reuse of grey water (GW) and give instruction of reuse to avoid any health risk for people and the environment. Regarding all these mentioned above, the occurrence of drugs in the aquatic environment are being included currently in EU and US legislation on environmental impact assessments and evaluation methods (Santos, et al., 2010).

One of the most known institution is the US Environmental Protection Agency (EPA). In their report (US EPA, 2002), reuse of GW for irrigation and other purposes are being considered. Especially the arid regions of the USA are creating their own GW reuse criteria. In 2001, the Arizona Department of Environmental Quality (ADEQ) issued new regulations for the use of domestic greywater. These rules support public to reuse their own greywater up to approximately 1500 L per day with regulations and some limitations such as no contamination of GW by hazardous chemicals, minimized possibilities of standing GW on surface and avoiding of human contact with GW and soil with irrigated GW.

One of the important aspects of greywater is the regulations about reuse and disposal of grey water. Another leading country, Australia has integrated sensitive water management system which try to control and minimize the public health risk associated with GW. Therefore, the Department of Health and Community Services created a guideline which indicates design and instalment criteria about GW treatment systems and reuse such as GW must be filtrated via a filtration trench or sedimentation, and application system must be colored and market in the house area.

Another considerable issue is the gap in legislation regarding environmental contamination by pharmaceuticals. The most important reason to be listed as first is the insufficiency of current data to quantify the actual profile of contamination. Additionally, it remains unclear if long-term exposure to pharmaceuticals and their metabolites causes chronic toxicity to fauna and flora. In the European Union, for the first time, the directive 92/18/ECC introduced the prerequisite requirement for an environmental risk assessment to obtain marketing authorization.

Human perception and acceptance are one of the challenges of greywater reuse. According to the unpublished results of the survey conducted as online by the project team of SWAMP (2020) about re-use of greywater, the acceptance is growing with increasing education. The survey was responded by totally 113 people from around the world. Among 113 people, 61% had bachelor or higher degree, and 46.9% of responders are not afraid to use treated greywater, 29% of them were hesitating. All respondents expressed positive regard for the reuse og grey water if they use greywater for toilet flushing, watering garden or washing car. 28.3% of the participants did reply to never the reuse greywater for the purpose of laundry or house cleaning. Overall, many people and complained about the lack of knowledge about greywater treatment and lack of incentives from local and central governments to support greywater reuse.

3.6. Impacts of Greywater Discharge on the Environment

The risk of contamination of soil and receiving water bodies from GW while used for infiltration and irrigation is a wondering topic which has been discussed among researchers for the last few decades. Besides of being a potential freshwater resource, direct discharge of untreated greywater can cause serious contamination problems on the environment and water sources (Al-Gheethi, et al., 2019). Consequents of the increase of the world's population, water resources are under serious pressure. Between the years 1960 and 2006, the total consumption of water on Earth has quadrupled (Racek, 2020). The consumption and demand for water in different countries due to climatic conditions, water resources, sewage, and water prices, etc. (Wurochekke A. A., 2016). Onsite separation of wastewaters such as black and grey waters from households can improve water quality and quantity, decrease environmental pollution, and save energy and cost. However, when planning reuse of greywater, it is very essential to be aware of the characteristics of greywater with respect to physical parameters, as well as the content of both chemical compounds and microorganisms. Therefore, an efficient treatment process is necessary to apply (Wezel and Jager 2002; Cordy, et al. 2004; Eriksson and Donner 2009; Turner, et al. 2013).

Pharmaceuticals are described as therapeutical drugs that aims to prevent or treat human and animal diseases, whereas personal care products aim to improve the quality of daily life (Ebele, Abdalla, & Harrad, 2017). During the last few decades, their presence in various natural compartments of the aquatic environment (e.g., surface waters, sediments, and biota) has become a notable concern due to their extensive and increased usage for human and veterinary medicine. Especially, the last two decades have witnessed an increase in the consumption of pharmaceuticals significantly worldwide for the care of human and animal health issues. Continuously enter of PPCPs into environmental compartments possibly can cause persistence, bioaccumulation, and toxicity, and therefore, may create destructive effects on aquatic organisms. The EU Water Framework (WFD) recognized PPCPs such as diclofenac, iopamidol, musks, and carbamazepine as future emerging priority pollutants and ibuprofen, clofibric acid, triclosan, phthalates and bisphenol A were proposed for adding to the list (Ebele, Abdalla, & Harrad, 2017).

Contamination of the environment by PPCPs can arise through various routes, whereas mostly distributed by effluent or/and sludge of wastewater treatment plants (WWTPs). Moreover, conventional WWTPs have been found as inefficient for removal of PPCPs (Hijosa-Valsero, et al., 2010). Santos et al. (2010) and later Ebele et al. (2017) drew the possible pathways of environmental contamination by PPCPs (the Figure 3). In the end, metabolism, diagnostic compounds, household disposal and anthropogenic activities are four obvious creators for the environmental contamination by medicines and personal care products (Santos, et al., 2010). Most often, PPCPs that are applied externally are discharged through shower, bathing, swimming, and washing sinks. Therefore, they can mix easily to greywater that is the concern of this study. For instance, detergents and

personal body care products may persist in nature for long periods, which bring forward the possibility of toxicity and bioaccumulation in organisms (Noman, et al., 2019).

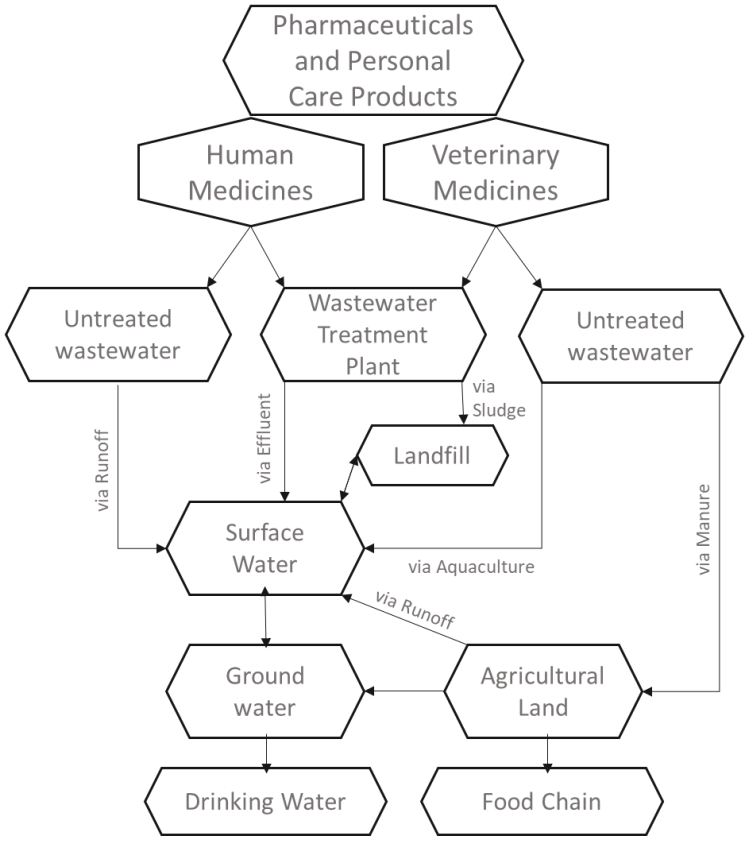

Figure 3: Representative pathways of pharmaceuticals and personal care products to the environment (Source: Adapted from Santos, et al., 2010 and Ebele, et al., 2017)

The reuse of greywater for various purposes has been increasing in many regions around the world such as Florida and Arizona in the US, Jordan, and Australia. Therefore, it is a necessity to concern about the safeness of greywater reuse in terms of both human and environmental health. While reuse of greywater is being a great alternative for potable water and solution for less consumption, at the same time, it can consist of different organic wastewater compounds (OWCs) that could persist on and create ecotoxicological problems for environmental and human health. Even though greywater shows less pollutants than blackwater, it is detected that emerging organic pollutants such as pharmaceuticals and personal care products (PPCPs) exist in the grey water and their fate in the environment has still obscurities (Turner, et al., 2019; Xu, et al., 2009). Due to this concern, the existence of micro-pollutants in greywater and their impacts to natural compartments have attracted considerable attention as recent research topics. Turner, et al. (2019), during their research in Australia about grey water irrigation, found out that 22

different organic micropollutants were in the greywater which are potential pollutants in local hydrosphere such as shallow ground water and nearby surface water. In their carefully designed study, Turner, et al. (2019) provided the potential household resources for organic micro pollutants detected in greywater. For instance, diclofenac exists in nonsteroidal anti-inflammatory drugs and since it is used as a cream for skin, it can easily be washed out during personal hygiene such as shower or hand wash and mix with grey water. Likewise, Triclosan as an antibacterial and antifungal agent exist in consumer products such as soap, detergents, toys, and antibacterial gels that have been increasingly consumed during Covid-19 pandemic.

To indicate greywater as a potential reusable water resource for especially outdoor purposes like irrigation of landscapes, or washing cars, it is essential that the recognition of pharmaceutical compounds in grey water, their fate in the environment and consequents to the environment need to be reviewed in accordance with the aim of the thesis. A grey water system has many various of systems that can be called either very simple or very complex, or in between. However, ecotoxicological compound needs more careful attention before reuse. Physiological effects and resistant structure of pharmaceuticals make them a more concerning compound that has the potential of being bioaccumulative and toxic for ecosystems. Instead, there is still little knowledge about the environmental fate and behavior of organic micro-pollutants (Santos, et al., 2010; Turner, et al., 2019).

As showed in the Figure 3, PPCPs usually spread into the environment via wastewater treatment plant's effluent and sludge, untreated wastewater from household consumption, aquaculture and manure, and water run-offs from agriculture. Eventually, some of them enter the surface waters like rivers, lakes, estuaries, and so on. At the end, PPCPs may mix into groundwaters where directly can affect the drinking waters. In their 2019 paper, Turner, et al. conclude that grey water irrigation might be a reason of contamination of shallow groundwaters and surface water by organic micro-pollutants, however, their risk for environment needs further analysis to be more precise. In smaller streams or during dry seasons where the water flow is lower, the exposure risk could be higher. Conversely, heavy rains like seasonal transitions can cause inadequate removal efficiencies for WWTPs (Daughton & Ternes, 1999).

Studies about the environmental fates of organic micro-pollutants indicate that they may be imposed by biological degradation, photodegradation, or other abiotic transformation on the way from soil to groundwater that results in partial loss or mineralization (Ebele, et al., 2017). For instance, photolysis is a degradation process may affect the ones (such as nitrates, humic acids) depends on factors like the intensity of solar irradiation, latitude, seasons of the year, and presence of photosensitizes (Santos, et al., 2010). In their seminal paper, Ebele, et al. (2017) found photolysis as one of the primary degradation types for PPCPs in surface waters. Other than that, they might be adsorbed to sediments, transformed into other compounds by biotic and abiotic process, or/and volatilized (Cordy, et al., 2004). However, with the current knowledge, it is still difficult to draw the full picture of the contamination pattern in final receiving surface waters because of the water dilution, treatment, and discharging processes (Cordy, et al., 2004).

Ecotoxicological effects of pharmaceuticals on non-target organisms could occur as acute or chronic effects. Santos, et al. (2010) provides a detailed review of 94 articles published between 1996 and 2009 about acute and chronic ecotoxicological studies. It is shown that acute toxicity studies have simpler experimental work than chronic toxicity studies, therefore, more data had been found about acute toxicity. On the other hand, acute toxicity data is valuable only in case of accidental discharge of drugs occurs.

4. Methods and Experimental Design

4.1. Methodology

A laboratory-scale column experiment was conducted. To understand the efficiency of different additives material in filtration beds to selected organic and inorganic compounds. Synthetic greywater was used in the experiment.

The experimental part of the thesis is based on the utilization of a physical model (column model) of infiltration trench filled up by different compositions of filtration materials. Water samples prior and after treatment by filtration were collected during weekdays after approximately 20 hours of contact time from Tuesday to Friday, Monday sampling provide information about impact of longer contact time and simulate weekend operation with approx. 72 hours contact time. To observe the efficiency of different filtration materials to remove selected micropollutants as organic compounds of PPCPs (BTA, CFF, DEET, DCF, MTP), TOC, TC, IC, TN, ions (F^-, Cl^-, NO_2^-, Br^-, NO_3^-, PO_4^{3-}, SO^{2-}_4), trace elements and metals (B, Cu, Ni, Zn), anionic surfactants, ammonium nitrogen, pH, conductivity and temperature (Figure 4). Samples were collected and analyzed every weekday.

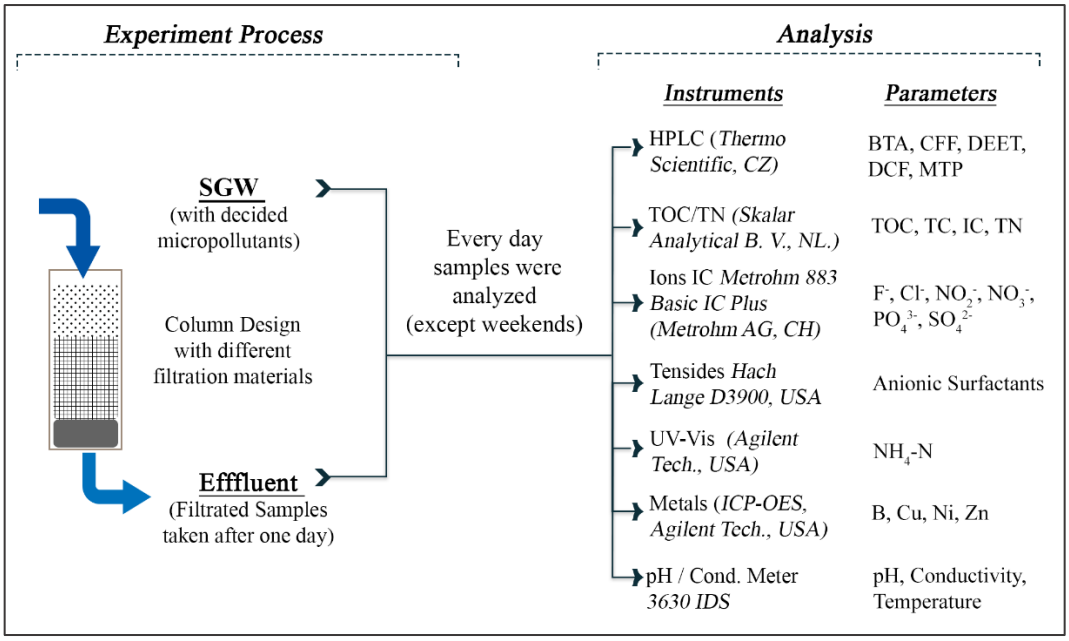

Figure 4: Diagram of the Methodology

The experiment was carried out inside a greenhouse of the Czech University of Life Sciences, Prague. The average temperature of the greenhouse during the experiment was at 22 °C.

4.2. Filter Materials and the Column Design

The physical model of the experiment consists of 30 columns filled with different filtration materials.

For conducting the experiment, 2 liters columns were chosen with the diameter of 5 cm and height of 50 cm. A representative model of the column design is shown in the Figure 5. The columns are divided in three layers. A 3.3 centimeters of gravel media was placed at the bottom and second layer was made of sand (13 cm thick). These layers were identical for each column. Gravel particles size was in the range 4-8 mm and sand particles were 0-4 mm. The upper layer, (28.6 cm thick) was composed of technogenic soil with different additive. The technogenic soil was prepared by mixing soil, sand, and compost. Different additives were added to the technogenic soil for each treatment group. The layers were separated by a circular geotextile to avoid clogging of lower layers by fine material of the upper layer. Additionally, a geotextile was placed to the top of each filter column to prevent any preferential flow way that could occur during irrigation. Furthermore, all columns were covered with aluminum foil to prevent sunlight as a precaution for growth of microalgae and other autotropic organisms. A hole with plastic house was made in the

bottom to allow draining of the columns. The effluent was collected in 1 liter glass flasks. A laboratory pusher was used to keep the house close to guarantee required contact time of SGW with the filtration materials.

Table 4: Composition of the Upper Layer as Proportions of the Used Materials

Type of Media Composite	***Proportions of All Materials***	***Number of Replicates***
Biochar 10%	Sand:5 – Compost:2 – Soil:2 – Biochar:1	5
Biochar 5%	Sand:10 – Compost:5 – Soil:4 – Biochar:1	4
Compost (Default)	Sand:5 – Compost:3 – Soil:2	5
Woodchips	Sand:5 – Compost:2 – Soil:2 – Woodchips:1	5
Mycorrhiza	Sand:4 – Compost:3 – Soil:2 – Mycorrhiza:1	5
Crushed Bricks	Sand:4 – Compost:3 – Soil:2 – Crushed Brick:1	3
Limestone Chipping	Sand:5 – Compost:3 – Soil:2 – Limestone:2	3

Six different biofilter materials were tested as additives to technogenic soil, specifically biochar (as 5 and 10% of upper layer volume), compost, woodchips, mycorrhizas, crushed bricks, and limestone chippings. The proportions of the materials are given in the Table 4 together with the information of how many replicates each treatment had. Each treatment had 5 replicates, due to leakage some of the columns have to be removed from the experiment.

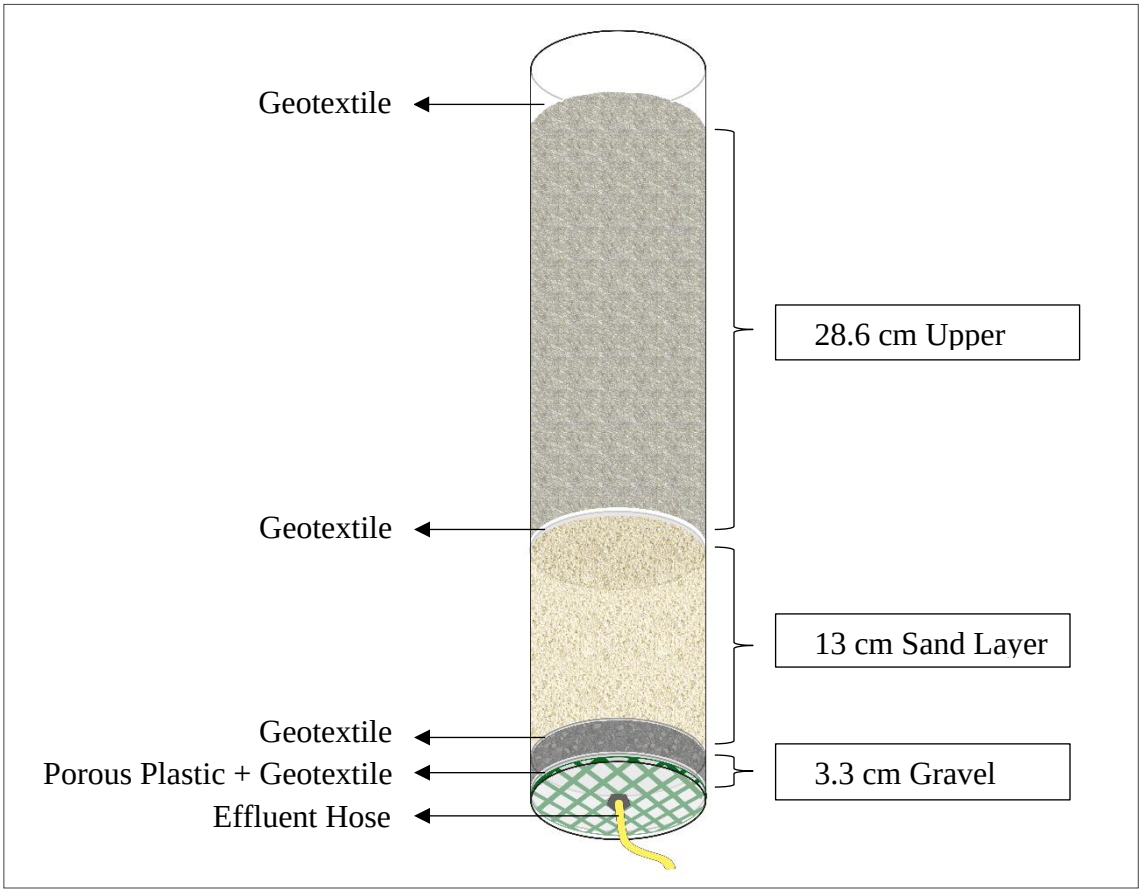

Figure 5: The Representative Model of the Column Design

Further, to understand the water holding capacity of the filter materials, prior to the experiment, when they were dry, a water flow test was conducted. 0.5 liters of tap water were added, and the time watch was started synchronously. The first water flows observed from the pipe and the time was recorded. According to the results, woodchip filters were recognized as has the highest water holding capacity. The first flow came after 5 min, 17 seconds, and after two hours, 0.35 liters of water captured in total. Conversely, limestone was found as has the least water holding capacity due to its particles creating an easy flow way to the water. The first flow was observed less than 1 minute and after two hours more than 0.45 liters of effluent was captured. As the second-highest holding capacity, biochar 5% was detected. Then, compost, crushed bricks, biochar 10%, and mycorrhiza were observed in the given order (the Table 5).

Table 5: Retention Time of the Biofilters

Type of Media Composite	Volume of Inflow (L)	Retention Time (min: second)	Volume of Outflow (L) (2 hours after the inflow)
Woodchips	0.5	5:17	0.35
Biochar 5%	0.5	1:40	0.45
Compost	0.5	2:22	0.47
Crushed Brick	0.5	2:02	0.48
Biochar 10%	0.5	1:40	0.36
Mycorrhiza	0.5	1:07	0.36
Limestone Chippings	0.5	1:11	0.45

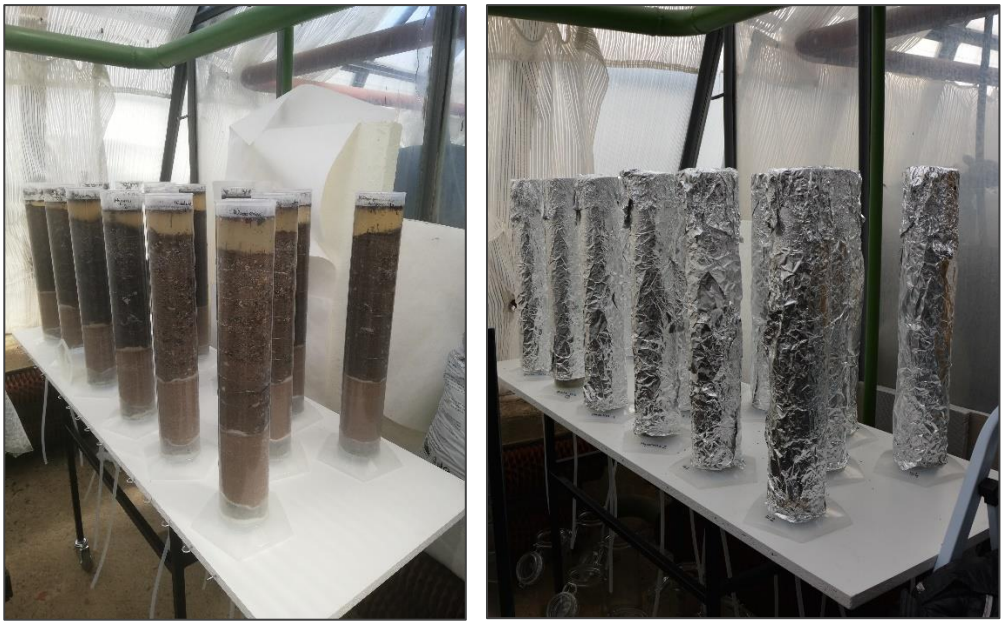

Figure 6: The picture of the columns in the greenhouse. On the right, the columns after covered with aluminum foil.

4.3. Synthetic Greywater (SGW)

In this study, a synthetic greywater has been prepared based on a modified version of the synthetic greywater recipe from Abed and Scholz (2016). The synthetic greywater solution was prepared by mixing inorganic compounds; Ammonium chloride (NH_4Cl), Monopotassium phosphate (KH_2PO_4), and Glycerin ($C_3H_8O_3$), trace elements; Copper (II) sulfate pentahydrate ($CuSO_{4.5}H_2O$), Boric acid (H_3BO_3), Zinc sulfate ($ZnSO_4.7H_2O$), and

Nickel sulfate ($NiSO_4.6H_2O$), and as organic compounds, DEET ($C_{12}H_{17}NO$), Benzotriazole ($C_6H_5N_3$), Caffeine ($C_8H_{10}N_4O_2$), Diclofenac sodium salt ($C_{14}H_{10}Cl_2NNaO_2$), Methylparaben ($C_8H_8O_3$), and Sodium decane-1-sulfonate ($C_{10}H_{21}NaO_3S$) were selected in this study as representative of compounds that commonly occur in GW (the Table 6). At the end, all compounds were mixed within tap water. To prepare the SGW, stock solutions were prepared from metals; 0.157 g of $CuSO_4.5H_2O$ dissolved into 100 ml deionized water for stock solution of Copper; 1.144 g of H_3BO_3 dissolved into 100 ml deionized water for stock solution of Boron; 0.175 g of $ZnSO_4.7H_2O$ dissolved into 100 ml deionized water for stock solution of Zinc; and 0.179 g of $NiSO_4.6H_2O$ dissolved into 100 ml deionized water for stock solution of Nickel.

Table 6: Concentrations of Compounds Present in Used Synthetic Greywater

Compounds	***Formula of Salt and Compound***	***Molecular Weight of the Salt (g/mol)***	***Molecular Weight of the Compound (g/mol)***	***Concentration in SGW (mg/L)***
Nitrogen Ammonium (N-NH_4)	NH_4Cl	53.4	14	25
Phosphate (PO_4^{3-})	KH_2PO_4	136.1	95	5
Glycerin	$C_3H_8O_3$	92.1		25
Boron (B)	H_3BO_3	61.8	10.8	1
Copper (Cu)	$CuSO_4.5H_2O$	249.7	63.5	0.2
Nickel (Ni)	$NiSO_4.6H_2O$	262.8	58.7	0.2
Zinc (Zn)	$ZnSO_4.7H_2O$	287.6	65.4	0.2
DEET	$C_{12}H_{17}NO$		191.3	2.5
Benzotriazole	$C_6H_5N_3$		119.1	2.5
Caffeine	$C_8H_{10}N_4O_2$		194.2	2.5
Diclofenac Sodium Salt	$C_{14}H_{10}Cl_2NNaO_2$	318.1	296.1	2.5
Methylparaben	$C_8H_8O_3$		152.1	2.5
Sodium 1-Decanesulfonate	$C_{10}H_{21}NaO_3S$	244.3		1

The advantage of using synthetic greywater is constant composition, while the composition of real greywater is changing over the time and makes comparison of removal efficiency difficult through the experiment.

The irrigation of the columns took place during all weekdays, allowing approximately 20 hours contact time. Samples collected on Monday were simulating weekend operation of the system, hence the contact time was approximately 72 hours. The amount of greywater dose to the column's changes, while during the first period 0.5 L (full saturation) were added to each column, during the following weeks only 0.4 L of SGW were added as the columns hold part of the water and never emptied completely.

SGW was prepared once a week on Monday and stored in a dark plastic barrel in the greenhouse at average 22 °C. This approach allowed to simulate real situation, when the greywater is stored prior the treatment, and changes in the chemical composition of SGW can be observed. Total 80 L of SGW was prepared for the period 1. To prepare 80 L SGW, 7.6142 g of NH_4Cl and 0.5730 g of KH_2PO_4 were dissolved in 1 L of tap water. In another 1 L of volumetric flask, 1.6 ml of Glycerin, 40 ml of stock solutions of B, Cu, Ni, and Zn were mixed. Lastly, 200 ml of DEET, Benzotriazole, Caffeine, Diclofenac sodium salt, and Methylparaben and 80 ml of Sodium 1-decanusulfonate stock solutions were added. All these compounds were mixed in a barrel with tap water to 80L. To prepare 70 L of SGW for the following weeks, same procedure adjusted accordingly.

4.4. Irrigation Process and Sampling

The experiment was divided to four phases. The first phase was flushing the columns with tap water, to properly rinse the filtration materials and to remove impurities naturally present in the materials. The second phase, inoculation of the columns by real greywater sludge collected from greywater treatment unit (located in the area of campus), took one week. The third and fourth phases are both irrigation with synthetic greywater, however, the volume of influent had changed (the hydraulic loading). As this project is a part of the SWAMP project, in this thesis, the result data from the first 23 days of SGW irrigation process were analyzed. The synthetic greywater (SGW) irrigation process, which is evaluated for this thesis, is divided into two period are called period 1 (day 1 to day 9) and period 2 (day 10 to day 23). In the Table 7, the periods are being explained with the dates, analyses that were done, and type of influent and volume as the activity of the period.

Table 7: Description of Each Period of the Experiment

Periods of Experiment	*Date of start and end*	*Taken analyses during the period*	*Activity*
Flushing	14/01/2021 – 16/01/2021	Ions, TOC, TC, IC, and TN	Each filter columns were irrigated with 0.5 L of tap water
Inoculation	18/01/2021 – 25/01/2021	Ions, TOC, TC, IC, and TN	Each filter columns inoculated with 0.5 L of real GW sludge
Period-1	26/01/2021 – 4/02/2021	Ions, TOC, IC, TN, Anionic Tensides, Ammonium-N, Metals, organic compounds, pH	Each filter columns irrigated with 0.5 L of SGW every day
Period-2	4/02/2021 – 18/02/2021	Ions, TOC, IC, TN, Anionic Tensides, Ammonium-N, Metals, organic compounds, pH, Conductivity	Each filter columns irrigated with 0.4 L of SGW every day

4.4.1. Irrigation Prior Synthetic Greywater

The process carried out, first, adding tap water to each of the columns until reaching the saturation point. Then, when the water started to accumulate on the surface of materials, the inflow water was stopped, and the volume recorded. This process had been applied to each column and, in this way, the amount of saturation volume for each column was calculated as approximately 0.5 L for each. Prior to start of the experiment, each filter system was fed with 0.5 L tap water day^{-1} for 2 days to wash the filter materials and then, the columns were left to rest during the weekend. Furthermore, samples from the outflow were collected to see the preconditions of the filter settings for TOC, TC, IC, TN, and ions. The basic information about columns such as used abbreviation, numbers, materials, and saturation volume are shown in the Table 8.

Table 8: Description of the Columns

Treatment Abbreviation	Column Number	Additive to Filter Material	Water volume for 100% saturation (L)	Water volume for 80% saturation (L)
B10	1, 2, 3, 4, 5	Biochar 10 %	~ 0.5	0.4
B5	1, 2, 3,4	Biochar 5%	~ 0.5	0.4
C	1, 2, 3, 4, 5	Compost	~ 0.5	0.4
W	1, 2, 3, 4, 5	Woodchips	~ 0.5	0.4
M	1, 2, 3, 4, 5	Mycorrhiza	~ 0.5	0.4
Br	1, 2, 3	Crushed Brick	~ 0.5	0.4
L	1, 2, 3	Limestone chipping	~ 0.5	0.4

Additionally, one week before the start of the synthetic greywater irrigation, each column was inoculated with real greywater sludge taken from the building of Czech University of Life Sciences, Faculty of Economics and Management on 18th of January 2021. The sludge was taken to a 25-liter tank and kept with an air pumping. The following day, the columns were inoculated by with 0.5 L of the sludge. Every day, the columns were emptied, the sludge was naturally aerated during the manipulation and put back to the columns (only during the weekend the sludge was left for 2 days). To understand the preconditions and changes during the storage time, also samples were taken from remained sludge. So, during the process, samples were taken two times. The samples were analyzed for TOC, TN, IC, and ions.

4.4.2. Irrigation with Synthetic Greywater

The experiment started on January 26th, 2021 (period 1) by adding the synthetic greywater prepared on January 25th. During the period 1, the SGW was prepared weekly (each Monday) and every day 0.5L of SGW were added to the columns between 1:30 and 2:30 pm, and outflows were taken the next day in the morning between 9:30 and 10:30 am. Influent samples (synthetic greywater) and effluent samples of greywater from each filter were collected at regular intervals during the experimental period. During weekends, the inflow was kept inside the columns to simulate possible real conditions and understand the changes during longer retention.

On February 4th, 2021, the inflow volume was changed to 0.4 L due to observation of clogging in the columns. The second period with lower hydraulic load started. While changing the volume of SGW to 0.4 L, the hose was kept open till the time that outflow was seen to let aeration of the columns.

During the experiment, the effluent samples were collected to plastic 100 mL bottles and immediately were taken to the laboratory for filtration. Before the analyses, all the samples were filtered through PEF 0,22 µm syringe filters (polyethetsulfonate syringe filters Rotilabo (Carl Roth). In addition to these, SGW samples (influent) were also analyzed every day for each parameter to compare influent and effluent and to observe the storage conditions and characteristic changes in the SGW during the stage period.

4.5. Analysis of the Samples

4.5.1. Analysis of Standard Parameters

The physio-chemical parameters (pH, and conductivity) of all samples and the synthetic greywater were determined in the laboratory every day by using WTW pH-meter 3630 IDS with an electrode IDS pH Electrode SenTix® 940. Additionally, the temperature of the greenhouse was recorded each day.

Regarding total organic carbon (TOC), total carbon (TC), inorganic carbon (IC) and total nitrogen (TN) analyses, samples of the effluents and the influents were filtered into 15 ml glass test tubes immediately after sampling. The tubes were placed to *FORMACS$^{HT/TN}$ TOC/TN ANALYZER HTAccess version 3* (Skalar) to conduct the determination of organic and inorganic bound carbon and nitrogen in the samples. The instrument utilizes a high temperature catalytic combustion with infrared detection (NDIR). By the definition, TC represents all carbon in the sample (TC = TOC + IC). IC is the inorganic carbon in the sample that after acidification, turns into carbon dioxide and includes all carbonates, bicarbonate, and dissolved carbon dioxide. Lastly, TOC is the organic carbon that is converted into carbon dioxide after oxidation and TN is all nitrogen in the sample which is organic and inorganic nitrogen.

Figure 7: The FORMACS$^{HT/TN}$ TOC/TN ANALYZER HTAccess version 3

To analyze anions, all samples were filtered by PEF 0,22 µm polyether-sulfone syringe filters Rotilabo (Carl Roth) into 15 ml plastic test tubes, immediately after collected. The tubes were placed into a ionic chromatograph 883 Basic IC Plus Metrohm *MagIC NET*^{TM} with a column Metrosep A Supp 5, 15 cm x 4 mm, 5 µm particles (Metrohm 6.1006.520) to determine the concentration of fluoride (F^-), chloride (Cl^-), nitrite (NO_2^-), bromide (Br^-), nitrate (NO_3^-), phosphate (PO_4^{3-}), and sulphate (SO_4^{2-}). The mobile phase was a mixture of 3.2 mM sodium carbonate and 1.0 mM sodium bicarbonate. The flow rate was 0.7 ml/min, and the injection volume was 20 µl within the instrument.

Figure 8: Ionic chromatography system MagIC NET^{TM}

To measure ammonium nitrogen ($N\text{-}NH_4$), 4 ml of filtered samples, immediately after collection and filtering, were mixed first with 0.4 ml of coloring agent (the coloring agent was prepared by dissolving 65 g of sodium salicylate, 65 g of sodium citrate tribasic dihydrate, and 0.475 g of sodium nitroprusside dihydrate in a 500 ml volumetric flask and stored in a dark bottle in a refrigerator). Then, 0.4 ml of alkaline solution (the alkaline solution was prepared by dissolving 16 g of NaOH with 1 g of sodium dichloroisocyanurate dihydrate in a 500 ml volumetric flask and stored in a dark bottle in a refrigerator) into 10 ml glass test tubes and then, lastly 0.2 ml of deionized water were added and mixed well. The prepared 5 ml samples were left minimum 1 hour waiting period. The measurement was performed photometrically by using indophenol blue method (ISO 7150-1:1984) by measuring the absorbance by Cary 60 UV-Vis spectrometer (Agilent Technologies Inc., USA). The applicable range for the instrument is between 0.01-1 mg/L NH4, therefore, higher concentrations were determined by sample dilution (the Figure 9a).

4.5.2. Analysis of Anionic Surfactants

For analyses of anionic surfactants, LCK 332 anionic surfactant cuvette test (Hach-Lange, USA) 0.05-2.0 mg/L was used and methylene blue method (ISO 7875-1-2-1984) was performed spectrophotometrically. Then, to measure the concentration, the DR3900 Spectrophotometer (Hach-Lange, USA) was used (the Figure 9b).

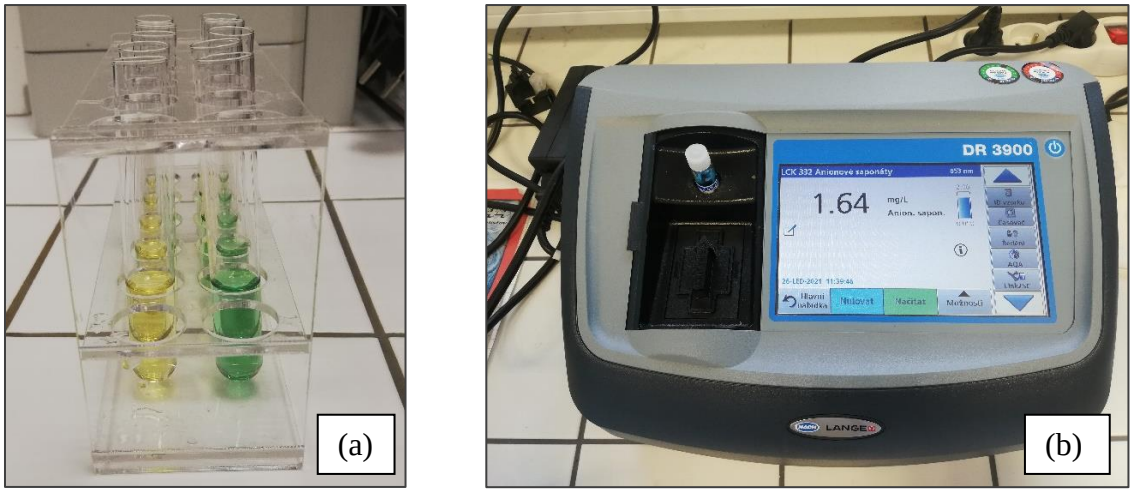

Figure 9: a) The samples that are prepared for NH_4 measurement b) Measurement of anionic surfactants.

4.5.3. Analysis of Boron and Metals

To prepare the samples for metal and boron analysis, firstly, all replicates of each group were mixed equally to get final volume of 10 ml so called mixed sample into 15 ml centrifuge tubes and 0.25 ml nitric acid were added to prevent sorption of the metals on the tube's walls. The process repeated both for filtered and unfiltered samples of effluents and synthetic greywater, to identify dissolved metals and metals bind to suspended solids. Prepared samples were analyzed by an inductively coupled plasma optical emission spectrometry (ICP-OES, Agilent 730, Agilent Technologies, USA) in the laboratory of the Department of Environmental Geosciences.

4.5.4. Analysis of Organic Micro-pollutants

The collected samples were analyzed every day in the analytical instrument, high-performance liquid chromatography (HPLC) coupled with a diode array detector system Ultimate 3000 (Thermo Scientific, Pragolab, Czech Republic) for quantification of caffeine, benzotriazole, methylparaben, DEET, and diclofenac. LC separations were achieved by using C18 Hypersil^{TM} Gold column (250 mm x 4.6 mm; pore size: 5 µm) (Thermo Scientific, Pragolab, Czech Republic) (the Figure 10) with a compatible precolumn (Thermo Scientific, Pragolab, Czech Republic). Injection volume was 20 µl for all type of samples. The compounds were quantified using linear calibration computed using at least 7 calibration levels. The retention times (RT) of the analyzed compounds were: 4.9 min for caffeine, 6.8 min for benzotriazole, 9.3 min for methylparaben, 11.6 min for DEET and 13.8 min for diclofenac. The compounds were qualified at the following

wavelengths: 220 nm for DEET, 255 nm for methylparaben and benzotriazole, and 275 nm for diclofenac and caffeine. The limit of detection (LOD) was 0.01 mg/L, and the limit of qualification (LOQ) was 0.05 mg/L and for benzotriazole, diclofenac and methylparaben, and for DEET and caffeine the LOD and LOQ were respectively 0.05 mg/L and 0.1 mg/L. The LOD values were defined for S/N (signal to noise) at the level of 3 and the LOQ values at the level of 10. The data was evaluated by means of Dionex Chromeleon^{TM} 7.2 software.

The samples after filtration with PES 0,22 µm syringe filters, were injected into the HPLC in the same day or kept in the fridge and in the dark to decrease the possibility of degradation (for example diclofenac can easily be degraded by day-light). During the storage time, MTB recognized as degrading in the barrel during the first period. This may appear due to growing microorganisms on the walls of the barrel. Therefore, each week on Friday, the barrel was cleaned by disinfectant cleaners to avoid such growth of microorganisms.

Figure 10: High-Performance Liquid Chromatography (HPLC) Coupled with a Diode Array Detector System Ultimate 3000 (Thermo Scientific, Pragolab, Czech Republic)

4.6. Statistical Analysis

All the data obtained from the analyses were gathered into an excel file. The calculations for the mean, median, and removal efficiency for each pollutant by each filter media were performed.

The efficiency of reduction for the various parameters analyzed was calculated by using Eq. 1:

$$E = (Cin - Cout)/Cin \times 100$$

Equation 1: Equation of The Efficiency of Reduction as Percentage

Where *E* is the efficiency (percent), C_{in} the influent concentration (mg/L), and C_{out} the effluent concentration (mg/L).

For statistical analysis of the significant differences between groups was used analyses of variance (ANOVA). If the data did not meet the assumptions for using parametric test (ANOVA), the non-parametric equivalent test, the Kruskal-Wallis test were used. Later, for the post-HOC testing was performed by using Turkey's HSD or Dunn's test depending on parametric or non-parametric method, respectively. Statistical analysis was performed in RStudio Version 1.3.1093. Additionally, for the visualization of the results and removal efficiency, Power BI Version 2.90.782.0 were used.

5. Results

5.1. Influent Characteristics

The average pH of synthetic greywater (influent) was 7.13. Also, it is observed that, during the storage time of the synthetic water, the first day of the prepared SGW sample showed more than 7.30 pH values whereas, through the end of the week the value decreased to an average of 6.88. Therefore, a marginal acidification is observed for SGW through the storage period. On the other hand, the conductivity of the SGW was average 576.50 µs/cm.

The freshly prepared synthetic greywater (SGW) contained in average 17.81 mg/L TOC (n=4) and 29.51 mg/L TN (n=4). After storing it in the barrel in the greenhouse for 5 days, the measured concentrations changed and resulted in average values of 12.68 mg/L TOC (n=4) and 28.1 mg/L TN (n=4). The inorganic carbon level (IC) did not significantly change. Overall, within the barrel, reduction of TOC was 28.7% and reduction of TN was 4.5%. In the Table 9, the characteristics of SGW for the whole period of experiment are presented, for fresh SGW and for last day of storage as their means and standard deviations.

Table 9: Influent Characteristics (mean ± standard deviation)

Parameters	***Unit***	***All Samples***	***Fresh Prepared SGW***	***Last Day of Prepared SGW***
pH		7.13±0.3	7.41±0.1	6.83±0.1
Electronical conductivity	*µs/cm*	576.5±11.85	590.5±12.02	571.66±11.01
Total carbon (TC)	*mg/L*	28.8±5.7	32.7±5.4	24.8±7.4
Total organic carbon (TOC)	*mg/L*	15.45±4.3	17.81±4.4	12.68±4.9
Total inorganic carbon (IC)	*mg/L*	14.03±1.5	14.28±1.4	14.19±0.9
Total nitrogen (TN)	*mg/L*	28.31±3.7	29.51±2.0	28.16±3.4
Ammonium ions	*mg/L*	20.9±1.01	20.6±0.70	20.2±2.10
Nitrite	*mg/L*	0.10±0.15	0.06±0.04	0.18±0.18
Nitrate	*mg/L*	15.68±4.08	13.94±6.82	16.00±0.55
Phosphate	*mg/L*	3.31±0.72	3.40±1.27	3.17±0.50

5.2. Filter Performance

The average pH of effluents of all biofilters were 7.33 for biochar 10 %, 7.36 for biochar 5%, 7.20 for compost (default), 7.09 for woodchips, 7.19 for mycorrhiza, 7.38 for crushed bricks, and 7.18 for limestone chippings. From the results, only woodchip media effluents have shown pH values between 6.8 and 7.0 four times over the twelve measurements. Regarding to the electrical conductivity (EC) of the effluents, biochar 5% was significantly higher with an average of 970.53 µs/cm and a maximum 1080.75 µs/cm. Additionally, even though the average of biochar 10% particularly were lower than biochar 5%, one of the replicates of biochar 10% was giving results significantly different than the other replicates. Therefore, when the outlier column is excluded, the results are similar with biochar 5% with an average of 939.22 µs/cm and a maximum of 1055.66 µs/cm.

The analyzed chemical parameters for the samples were TOC (total organic carbon), TC (total carbon), IC (inorganic carbon), TN (total nitrogen), inorganic ions (fluoride, chloride, bromide, nitrite, nitrate, phosphate, and sulphate), and ammonium ions (NH_4^+). According to the complementary results of the biofilters, in general, both biochar 5% and 10% showed lowest average TOC and simultaneously higher nutrition concentrations (total nitrogen, nitrite, nitrate and phosphate) which could be interpreted as lower microbiological activities and high nitrification process within the biofilters. Some of the characteristics of effluents from each filter media are shown in Table 10.

Table 10: Characteristics of the effluents of each filter medias (median ± standard deviation)

Parameters	***Unit***	***SGW***	***Biochar 10%***	***Biochar 5%***	***Compost***	***Woodchip***	***Mycorrhiza***	***Crushed Bricks***	***Limestone Chippings***
pH		*7.13±0.3*	7.3±0.1	7.3±0.1	7.2±0.1	7.0±0.1	7.1±0.1	7.3±0.1	7.1±0.1
Electronical conductivity	*µs/cm*	576.5±11.85	921.7±64.7	970.5±77.2	905.6±50.4	910.1±39.7	904.6±48.1	929.7±72.5	883.2±67.4
Total carbon (TC)	*mg/L*	28.8±5.7	72.3±14.5	72.0±15.9	92.6±22.0	90.5±16.2	91.7±24.0	92.9±26.2	82.8±20.2
Total organic carbon (TOC)	*mg/L*	15.45±4.3	13.4±6.8	12.9±7.3	27.2±13.1	19.94±10.0	26.6±14.6	24.2±15.0	23.0±12.3
Total inorganic carbon (IC)	*mg/L*	14.03±1.5	59.8±10.2	60.2±10.0	65.5±11.1	70.8±8.6	65.2±11.9	68.6±14.3	59.9±10.0
Total nitrogen (TN)	*mg/L*	28.31±3.7	12.8±7.6	14.9±10.3	9.4±5.3	7.7±5.3	8.1±3.9	9.1±4.9	11.6±6.3
Ammonium ions	*mg/L*	20.9±1.01	0.05±0.06	0.04±0.02	0.36±0.46	0.94±0.94	0.12±0.08	0.11±0.13	0.98±1.08
Nitrite	*mg/L*	0.10±0.15	1.83±2.64	0.20±0.44	0.07±0.06	0.08±0.03	0.05±0.03	0.05±0.04	0.08±0.04
Nitrate	*mg/L*	15.68±4.08	47.7±31.9	66.02±45.7	24.4±15.6	10.6±6.67	27.9±17.9	33.3±19.1	32.02±18.9
Phosphate	*mg/L*	3.31±0.72	3.51±1.07	2.97±1.08	3.84±1.47	1.88±0.35	5.70±1.76	2.22±0.54	4.05±1.39

TOC is an important indication to understand the bacterial growth within the column. Therefore, TOC was daily measured (working days only). The highest TOC concentration on average was measured for effluents of compost (default) and mycorrhiza biofilters with 27.25 mg/L and 26.60 mg/L (n=15), respectively. For the first day of the experiment, mycorrhiza, compost, and crushed bricks biofilters performed identically without showing any statistically significant differences (T-test, $p>0.9$). However, the last day the results shown that crushed brick and limestone became identical (T-test, $p=1$) and significantly different from mycorrhiza and compost (T-test, $p<0.05$). Overall measurement is given in the Figure 11 for the effluent of biofilters and the influent.

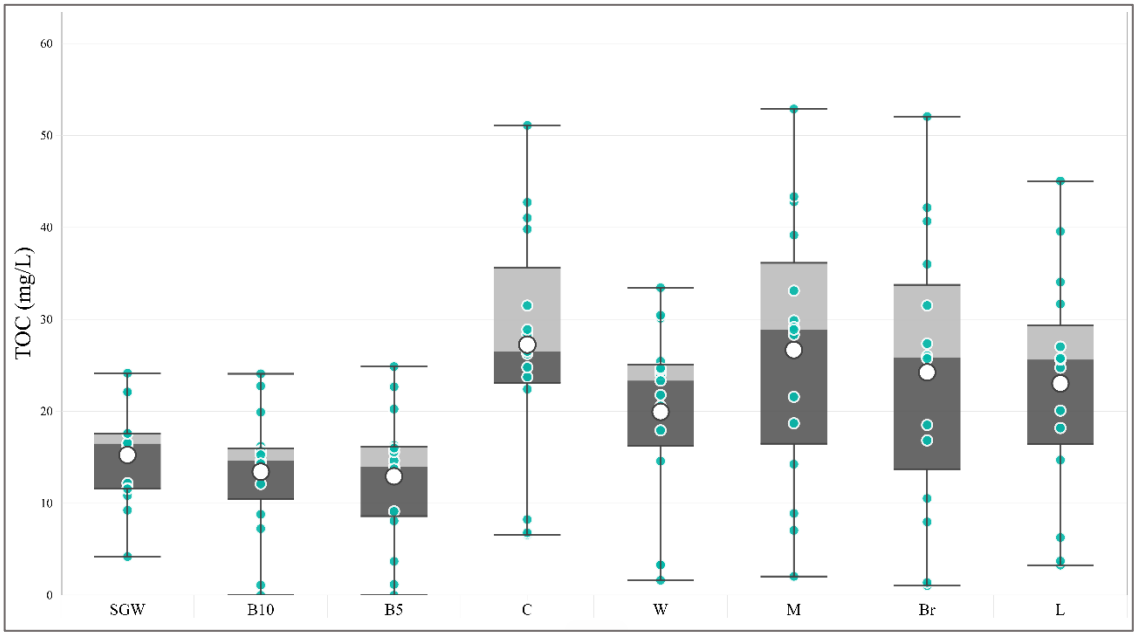

Figure 11: TOC concentration (mg/L) of the last day (day 23) of the experiment in comparison for the each biofilters

The removal efficiency of TOC has shown quite instabilities within all biofilters. The first days of inefficiency might be the result of inoculation period which had been applied prior to the experiment. Then, the TOC removal efficiency was increasing for all biofilter till the day 9 which is the day when hydraulic load has changed. It is detected that the trend of removal efficiency has recovered after the day 20 (Figure 12).

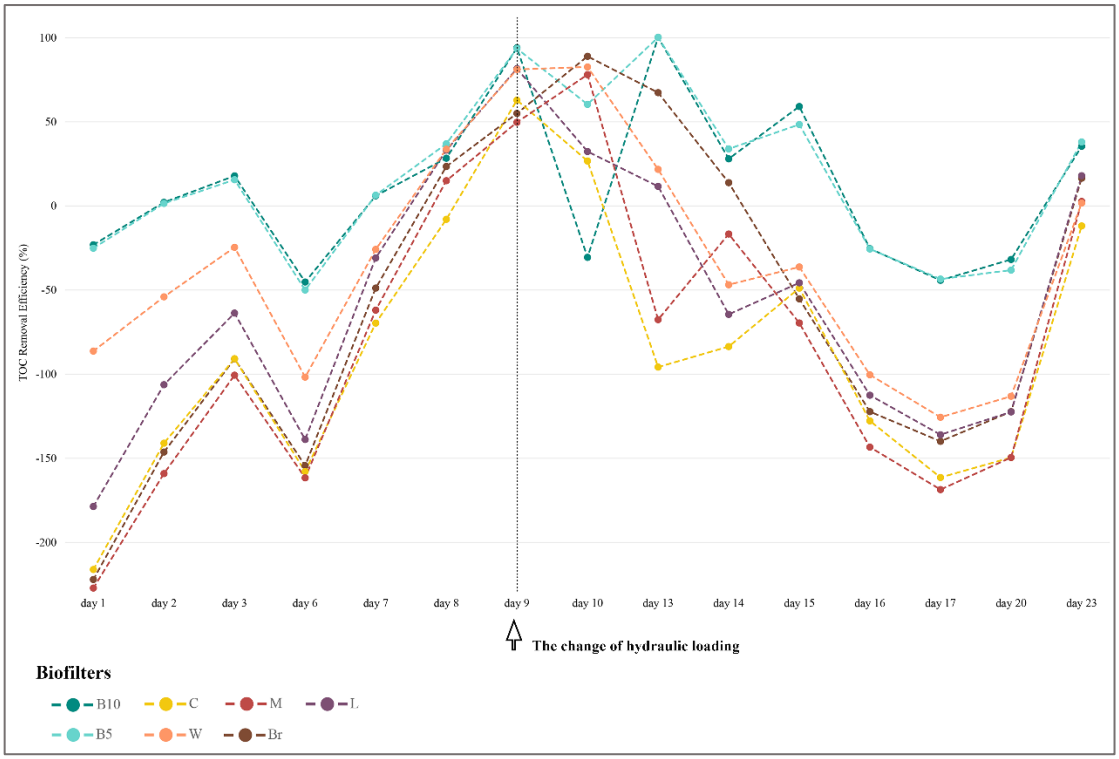

Figure 12: TOC Removal Efficiency of all Biofilters

The other important parameter was total nitrogen removal within the biofilters. The highest TN concentration in an average was recorded from biochar 5% and biochar 10% as 14.90 mg/L (n=15) and 12.88 mg/L (n=15), respectively. According to the last day measurements of the experiment (Figure 13), biochar 5% and mycorrhiza showed statistically significant differences from the other biofilters (T-test, $p<0.05$). The results show compost and limestone chippings are as almost statistically identical biofilters (T-test, $p>0.9$).

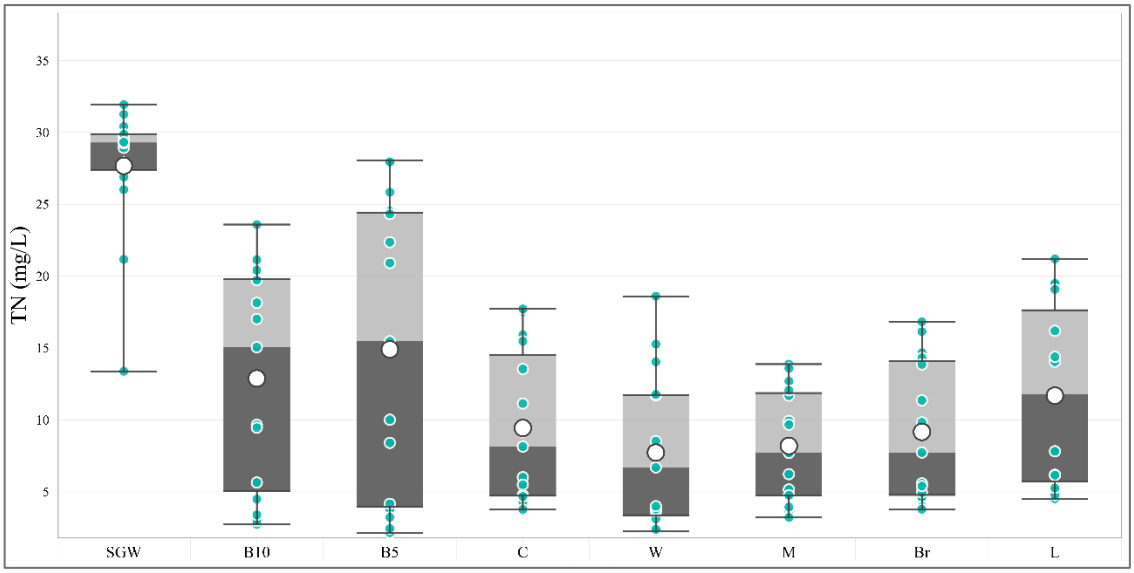

Figure 13: Total Nitrogen (TN) Concentrations of the Biofilter Effluents

The removal efficiency of TN for the biofilters is given in the Figure 14. The removal of TN was more efficient in woodchips biofilters with an average of 70.9% in total. However, the change of hydraulic loading has affected the removal efficiency negatively. In all biofilters, a rapid decrease in TN removal efficiency has been recorded after day 9 (the day the hydraulic loading had been changed). The TN removal efficiency of Biochar 10% and biochar 5% were 64.8% and 62.7% respectively during day 9. On the first day of hydraulic change (day 10), the results indicated that there is growing nitrogen concentration within the biofilters that means the effluent TN concentration is higher than in the influent. After the change of hydraulic load, the TN removal efficiency of the other biofilters has decreased as; crushed bricks from 85.8% to 42.2%, woodchips from 85.1% to 42.3, mycorrhiza from 82.2% to 12.6%, limestone chippings from 80.4% to 12%, and compost from 79.5% to 31.3%. With an approximately 70% decrease, mycorrhiza and limestone chippings were the ones which lost their efficiency at the highest. On the day 23, the highest TN removal efficiency was obtained from mycorrhiza (62.2%), and respectively crushed bricks (47.3%), compost (44.5%), biochar 10% (43.1%), woodchips (41.7%), limestone chippings (40.2%), and as the lowest efficiency from biochar 5% (12.5%).

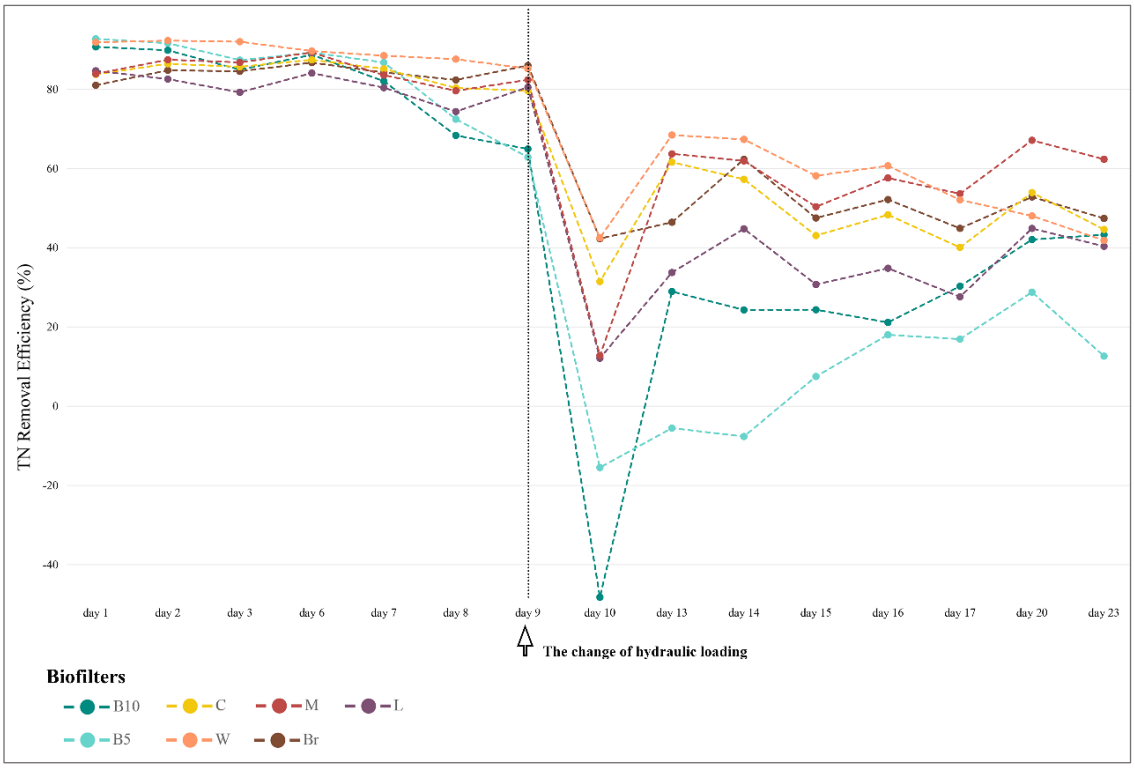

Figure 14: Removal Efficiency of TN for each Biofilters

The ammonium ions (NH_4^+) concentrations in the effluents of biofilters were in the range of 0.1 to 1 mg/L. In general, the removal efficiency of NH_4^+ was high; however, the limestone chipping filter was recognized as losing its efficiency constantly from 99.6% to 85.9%.

Nitrite and nitrate concentration of biofilters showed a considerable difference between biochar filters and the others. Nitrite and nitrate concentrations of influent were 0.10 mg/L and 15.68 mg/L on average, respectively. Nevertheless, with regards to nitrate, only the woodchip filter effluent showed less concentration than influent with 10.66 mg/L on average and 0.08 mg/L for nitrite. The highest nitrite and nitrate concentrations were detected in the effluents of biochar filters, both for 10% and 5 %, 1.83 and 0.20 mg/L for nitrite, and 47.79 and 66.02 mg/L for nitrate, respectively. The effluents of compost, mycorrhiza, crushed bricks, and limestone chipping biofilters had 0.07, 0.05, 0.05, and 0.08 mg/L nitrite on average and 24.40, 27.9 33.3, and 32.02 mg/L for nitrate, respectively. Results of ANOVA test indicated that the removal efficiency at the beginning of the experiment is significantly different than at the end of the experiment for all biofilters (T-test, $p<0.05$) except woodchip filters (T-test, $p>0.05$).

The change of the nitrate concentration during the experiment together with the nitrate concentrations of the influent is shown in the Figure 15, which clearly shows an

increase in the nitrate concentrations of effluents of all biofilters by days except woodchips. The SGW indicates the observed changes on nitrate concentration during the storage time.

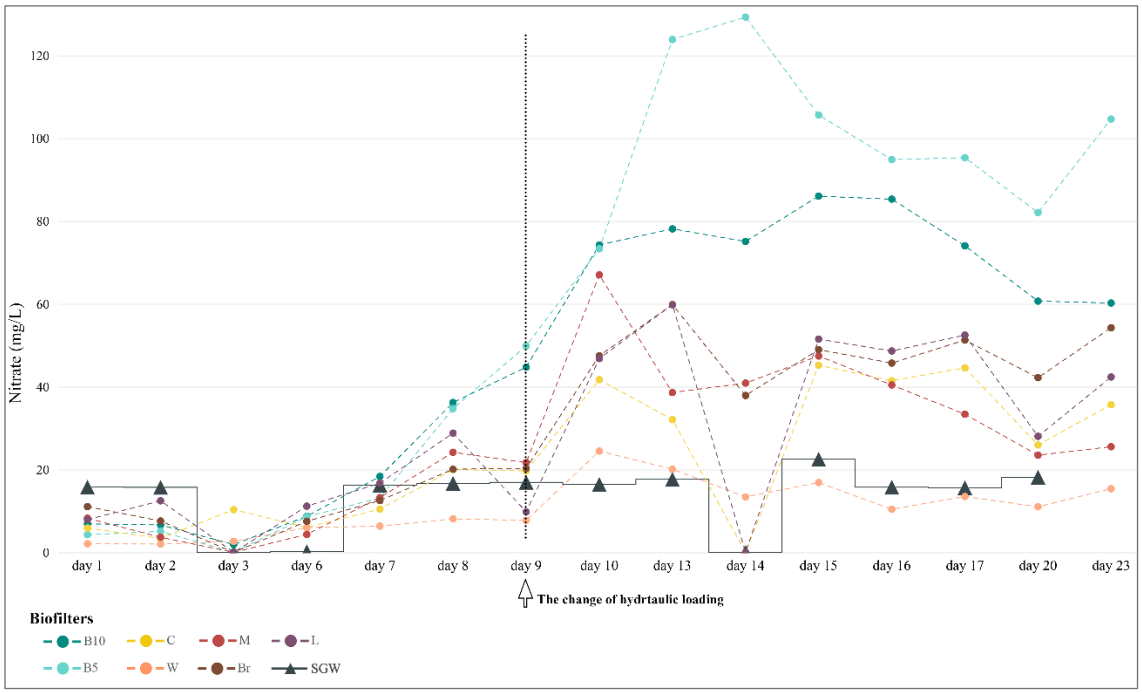

Figure 15: Nitrate Concentration (mg/L) of the Effluents of Biofilters and the Influent (SGW) by days

The change of the nitrite concentration during the experiment together with the nitrite concentrations of the influent are shown in the Figure 16Figure 15 which clearly shows an increase in the nitrite concentrations of biochar 10% effluent from day 6 to day 13, however, after day 20, nitrite concentration of the effluents from all biofilters show approximately equal level of nitrite concentration with the influent's concentration.

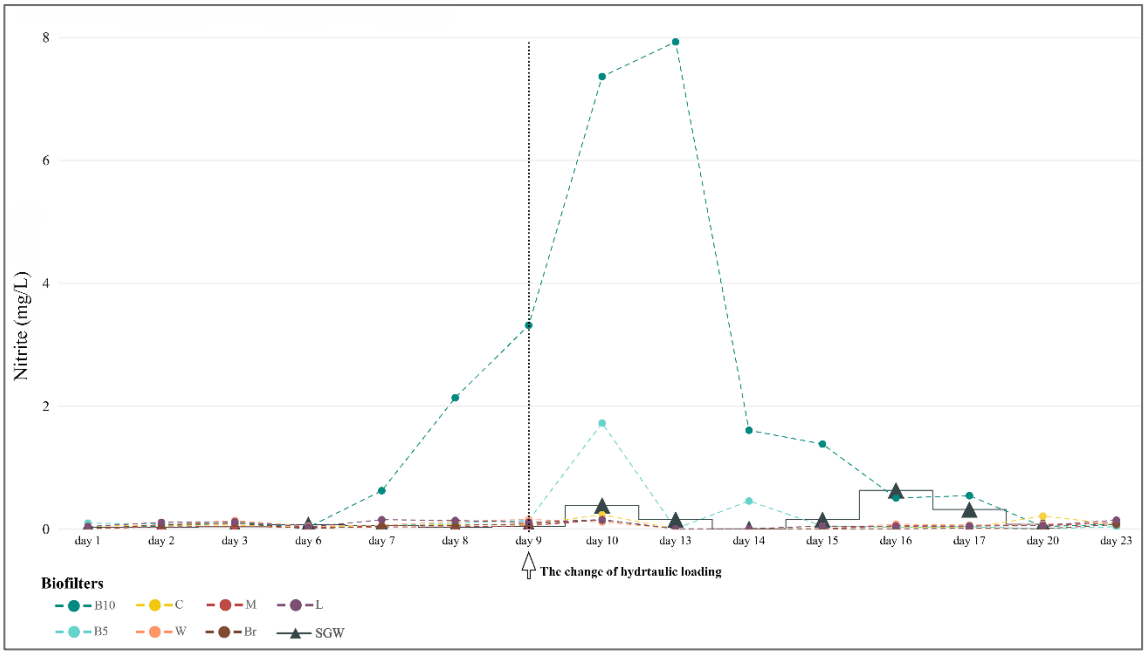

Figure 16: Nitrite Concentration (mg/L) of the Effluents of Biofilters and the Influent (SGW) by days

The phosphate concentration of influent (SGW) was 3.31 mg/L. The most efficient phosphate removal was obtained from woodchip biofilter with 27.9% which had 1.88 mg/L phosphate concentration on average. Then, crushed brick and biochar 5% filters were the ones which shown a slight removal efficiency with 27.9% and 2.11% for phosphate, respectively. Effluents of mycorrhiza biofilters had the highest phosphate concentration with 5.70 mg/L on average among the other biofilters. On the Figure 17, distributions of daily (15 days of 23 days were measured) phosphate concentrations for the influent (SGW) and each biofilter effluent are shown. As can be seen from the box plots (Figure 17), the narrowest distribution was detected for woodchips, which is also supported by the ANOVA test. The test indicated that only woodchip's phosphate removal efficiency did not show any significant difference between the start and the end of the experiment (T-test, $p>0.05$), whereas the other biofilters' removal efficiency at the beginning were significantly different (T-test, $p<0.05$) than at the end of the experiment.

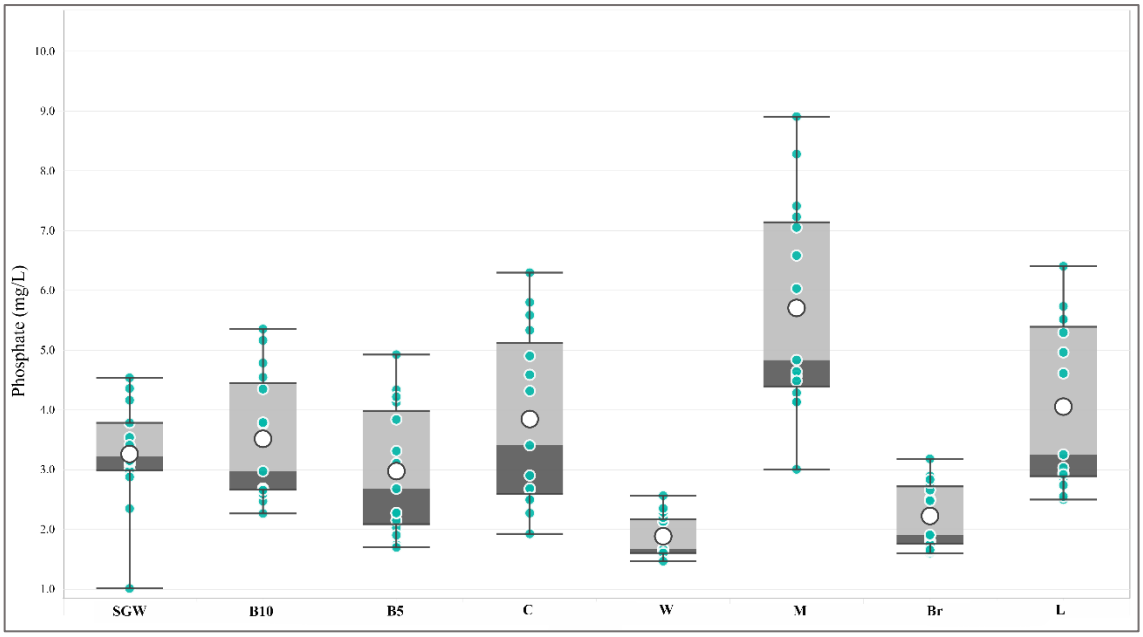

Figure 17: Distribution of Phosphate Concentrations Obtained Daily During the Experiment for Influent (SGW) and Effluent of Biofilters (n=15) (mean values represented by bigger white circle and other values are represented by blue circles)

The removal efficiency of inorganic compounds by biofilters was not found as efficient during the measurement of 23 days. Additionally, any meaningful effect of both hydraulic loading and retention time was not detected. Fluoride concentration was measured in average 0.09 mg/L for the influent (SGW); however, the effluent averages were 0.29 mg/L for biochar 10%, 0.26 mg/L for biochar 5%, 0.23 mg/L for compost, 0.28 mg/L for woodchips, 0.26 mg/L for mycorrhiza, 0.24 mg/L for limestone chippings, and 0.77 mg/L for crushed bricks. The observed higher fluoride concentrations in crushed bricks effluent than influent and other effluents could be interpreted as a leakage from the filtration materials. Regarding chloride concentration, there was a slight removal for all biofilters. The mean value of chloride for influent was 93.28 mg/L whereas, on average, it was 86.7 mg/L for the effluent of all biofilters. No significant difference was detected among biofilters for the efficiency of chlorine removal. Similarly, the amount of sulfate was not removed by the biofilters, instead, the concentration of sulfate was higher in the effluents than the influents (SGW).

5.2.1. Removal Efficiency of PPCPs

The obtained average (mean), maximum (max) and minimum (min) concentrations of the compounds of each effluent from biofilters and of the influent

(SGW) are given in theTable 1 Table 11. The data reflects the results from 14 measurements over 23 days.

Table 11**:** Concentration of the Compounds in the Influent and the Effluent of Filter Medias with the Range of Min, Mean and Max Values (mg/L/d) over experimental period

Compounds		*Influent*	*Filter Medias' Effluent*						
		SGW	**B10**	**B5**	**C**	**W**	**M**	**Br**	**L**
CAF	*Min*	2.41	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Mean	2.47	0.01	0.00	0.01	0.01	0.01	0.02	0.02
	Max	2.55	0.03	0.00	0.14	0.03	0.09	0.21	0.26
BTA	*Min*	1.87	0.00	0.00	0.00	0.02	0.00	0.00	0.02
	Mean	1.98	0.22	0.00	0.65	0.64	0.35	0.60	0.89
	Max	2.12	1.26	0.01	1.37	1.33	0.96	1.27	1.56
MTP	*Min*	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Mean	2.02	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Max	2.49	0.00	0.00	0.00	0.01	0.00	0.00	0.00
DEET	*Min*	0.98	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Mean	2.12	0.00	0.00	0.28	0.56	0.57	0.18	0.32
	Max	2.97	0.07	0.09	1.06	1.54	1.37	0.84	1.25
DCF	*Min*	2.32	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Mean	2.42	0.00	0.00	0.04	0.05	0.00	0.06	0.16
	Max	2.52	0.04	0.00	0.14	0.24	0.04	0.21	0.32

According to the results, caffeine (CAF) and methylparaben (MTP) are recognized as highly removal compounds by all biofilters. Methylparaben was removal through all biofilters with 100% efficiency during the experiment. Caffeine removal was also quite high through all biofilters (Figure 18). Although during the first period of irrigation with 0.5 L SGW all the biofilters showed 100% efficiency. However, the reason for this could be their easy degradable compound structure. In addition to that, although an initial decrease of removal efficiency is observed for DEET and diclofenac (DCF), after the first week of the experiment, an increase is noticed for the removal efficiency for each biofilters. This could be explained by probably adaptation of the microbiota. In this sense, among the xenobiotic organic compounds (XOC's), benzotriazole has been indicated as a poorly biodegradable compound.

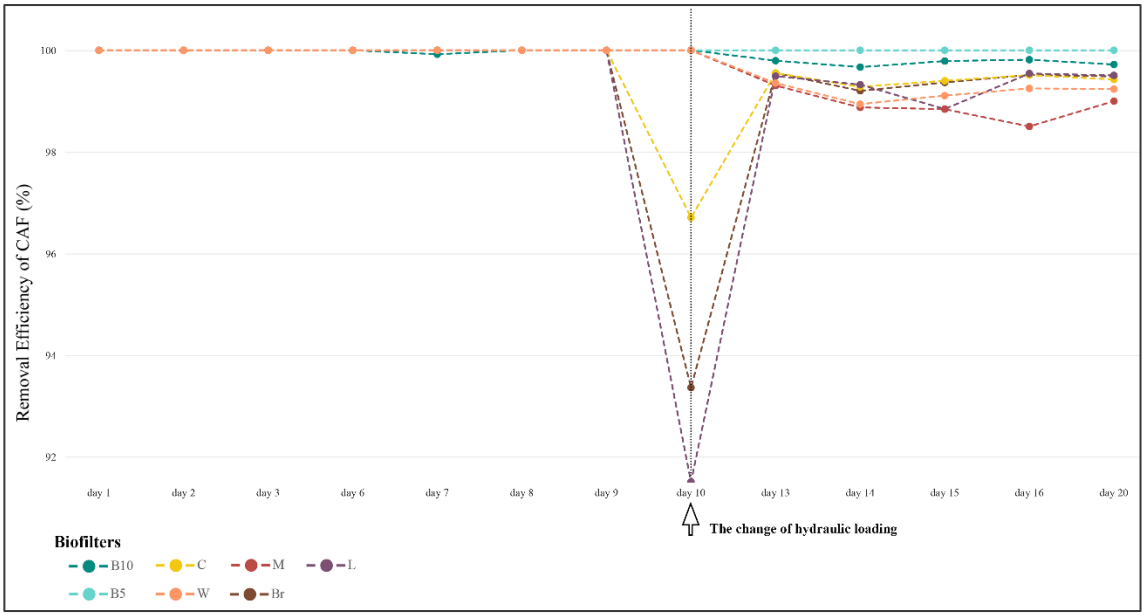

Figure 18: Removal Efficiency of Caffeine during the Experiment

The removal efficiency of DEET is shown on the Figure 19 by the days of the experiment. It is possible to see during the first seven days of the experiment, there is a constant decrease of DEET removal efficiency for compost, mycorrhiza, woodchips, crushed bricks, and limestone chippings biofilters. Biochar filters are shown higher and stable efficiency from the beginning although biochar 10% showed minor fluctuations at the beginning, it performed quite high removal efficiency as a result. The day 10 is the day the influent volume has changed from 0.5 liters to 0.4 liters. Later, all the filter medias showed an increasing trend for the DEET removal efficiency and at the day 20, all medias achieved over 90% of removal efficiency. This situation could be that the biofilters achieved an equilibrium stage over one week and microbiota within the filters had adapted. However, due to lack of knowledge about microorganism growth within the columns, the exact effect of biodegradation is not known.

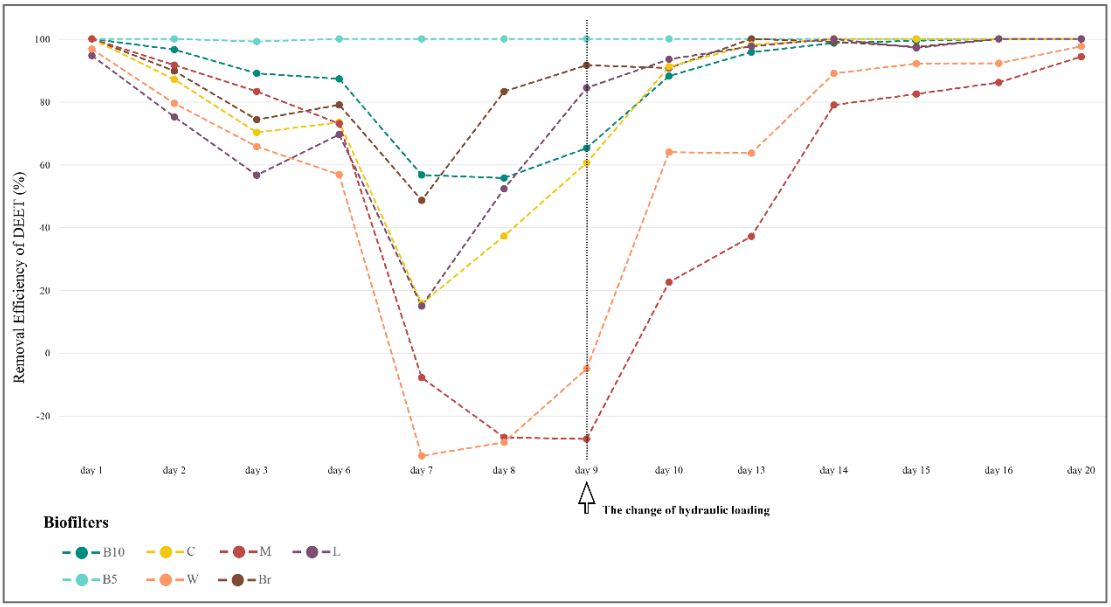

Figure 19: Removal Efficiency of DEET over 20 days. The arrow on the day 9 indicates the change of influent volume from 0.5 L to 0.4 L.

DCF removal efficiency of the biofilters has shown on the Figure 20. Similar with the other compounds, biochar biofilters both 5% and 10% performed 100% removal for diclofenac. In addition to them, mycorrhiza has performed 100% removal efficiency for diclofenac. Regarding to the other biofilters, limestone chippings has performed the lowest removal efficiency for diclofenac. The change of hydraulic loading has made an effect on the removal efficiency of compost, woodchips, and limestone chippings as can be seen from the Figure 20. The day 10 is the first day of measurement of the results from 0.5 liters after the change of hydraulic loading to 0.4 liters. After the hydraulic load change, only crushed bricks efficiency decrease, however, it turned to increase and stayed over 95% efficiency later. After the day 10, all the biofilters except limestone chippings had over 98% removal efficiency whereas limestone chippings also had removal efficiency between 93% and 96%.

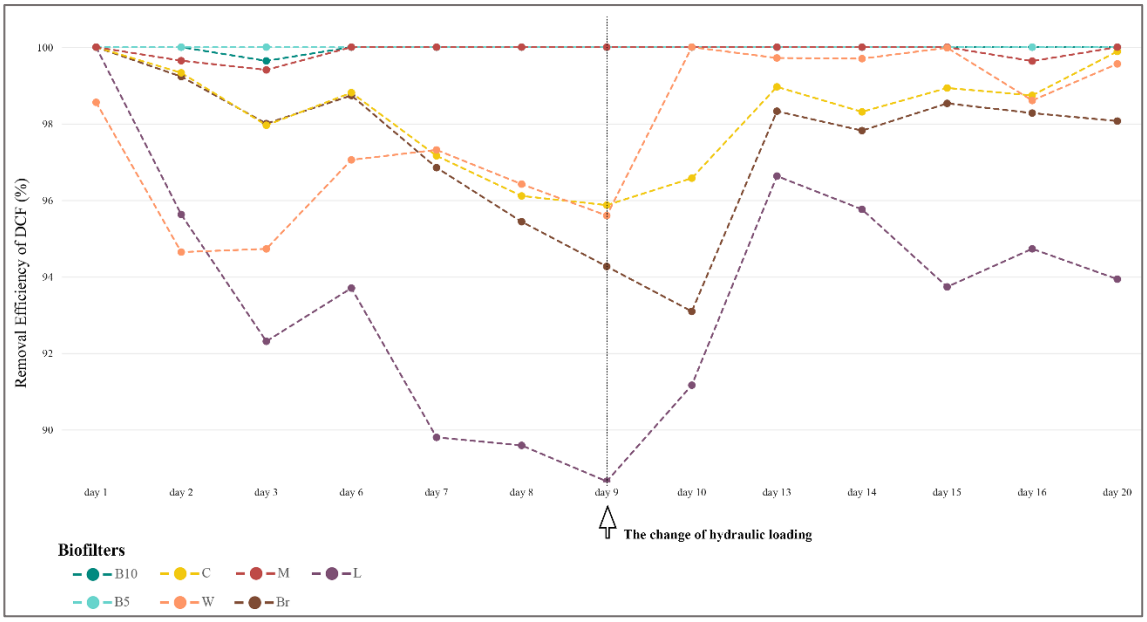

Figure 20: Removal Efficiency of the Biofilters for Diclofenac (DCF)

The overall trend of removal efficiency of benzotriazole (BTA) is shown in the Figure 21. The removal efficiency of benzotriazole of all biofilters has shown a consistent decrease except for both biochar 5 % and 10%. Despite the increase of the efficiency of limestone chipping biofilter after the change of hydraulic loading to 0.4, the decrease for the efficiency has been continued after day 10. The removal efficiency of benzotriazole for mycorrhiza decreased from 100% efficiency to 64% efficiency whereas, compost, woodchips, crushed bricks, and limestone chippings biofilters decreased under 50% at the end from over 90% efficiency at the beginning. The least efficient biofilter was limestone chippings with 35% efficiency on day 20.

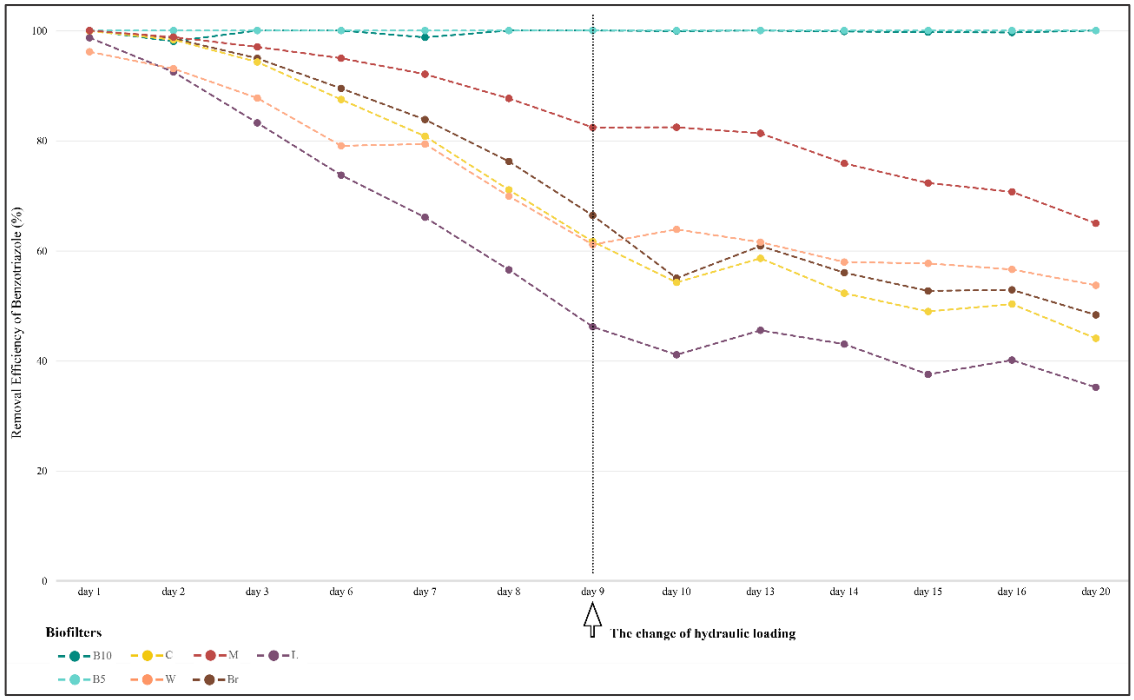

Figure 21: Removal Efficiency of Benzotriazole (BTA) as Percentage

5.2.2. Removal Efficiency of Anionic Surfactants

The average concentrations of the anionic surfactants for the effluent of biofilters and influent (SGW) are given in the Table 12. The average concentrations are obtained from seven measurements for effluents and eight measurements for influent over 23 days. The average concentration of anionic surfactants in freshly prepared SGW was 1.68 mg/L (n=3). The degradation of anionic surfactants within the barrel over 5 days was 9.2%±4.9 under the average temperature of 22^{o}C.

Table 12: The Concentration of Anionic Surfactants in Average of the Experiment

	Biofilters	***Anionic Surfactants Concentration (mg/L)***
Influent	**SGW**	1.57±0.1
Effluent	**B10**	0.26±0.1
	B5	0.17±0.01
	C	0.20±0.03
	W	0.23±0.08
	M	0.22±0.07
	Br	0.23±0.03
	L	0.28±0.05

Removal efficiency of biofilters is shown in the Figure 22. Anionic surfactants removal efficiency of biofilters did not show high variability during the experiment. However, in general, the removal efficiency decreased after the change of hydraulic load. The biggest decrease was obtained from biochar 10% from 95.9% to 64.6%. Any reliable reason is found for this rapid decrease specifically for biochar 10% however, the general reduction in removal efficiency of biofilters might occur due to aeration resulting from the lower water saturation of the columns.

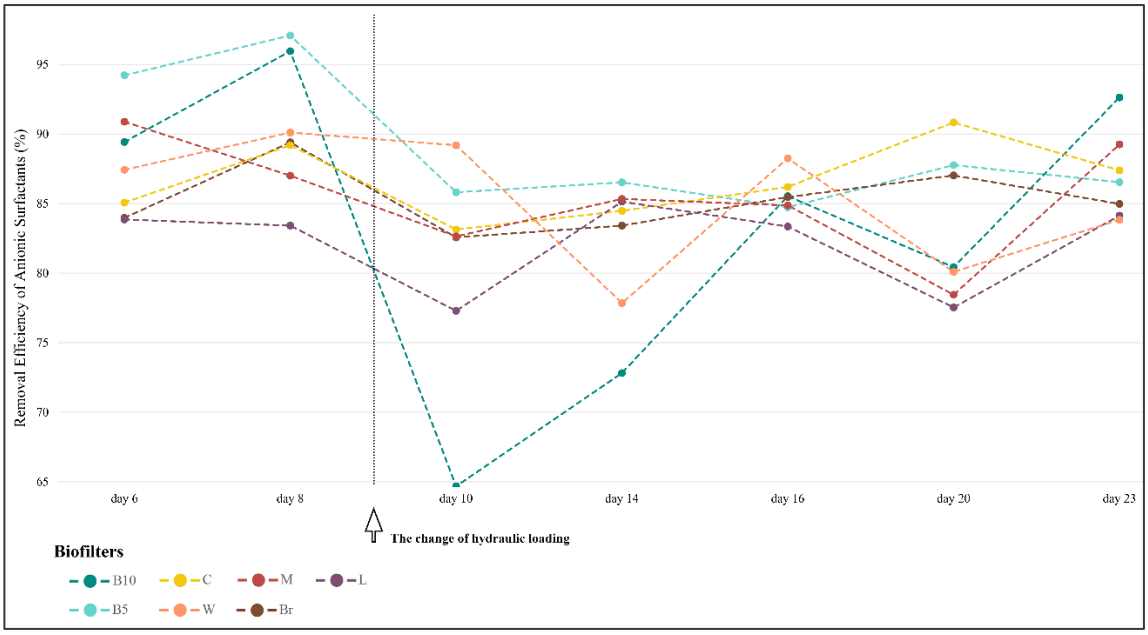

Figure 22: The Removal Efficiency of Anionic Surfactants as Percentage

5.2.3. Removal Efficiency of Boron and Metals

Table 13 gives the minimum (min), mean and maximum (max) concentrations of boron, copper, nickel, and zinc for influent (SGW) and effluents of biofilters. The measurement of metals and boron were done over 16 days. According to the results, metals showed high removal efficiency by biofilters. Copper was removed by 88% in average during the experiment. Although any significant effect of hydraulic load change was recorded on biofilters' removal efficiency of copper, a slightly positive effect of longer retention time (during weekends approx. 72 hours) was observed for limestone chipping. Nickel was removed by 96.54% in average during the experiment with an increasing trend. The most efficient biofilters were biochar 5%, biochar 10% and compost with over 98% efficiency for all at the end. Nevertheless, the other biofilters were performed over 97% of nickel removal efficiency. It is detected that both hydraulic load change and longer retention positively affected the removal efficiency of nickel. Zinc was removed by 90.11% in average during the experiment. During the irrigation with 0.5 liters of SGW (period 1), except compost, the other biofilters showed a decrease on the removal efficiency of zinc. However, after the change of hydraulic loading, an increase on the removal efficiency is observed over most of the biofilters and all the biofilters achieved over 90% removal efficiency.

Table 13: The Boron and Metal Concentrations of influent and effluents (unfiltered samples)

Metals		*Influent*	*Effluent of Biofilters*						
		SGW	**B10**	**B5**	**C**	**W**	**M**	**Br**	**L**
	Min	0.45	0.23	0.33	0.50	0.26	0.30	1.02	0.49
Boron	*Mean*	1.02	0.88	0.91	0.97	0.91	0.88	1.16	0.98
	Max	1.58	1.19	1.28	1.19	1.14	1.14	1.36	1.20
	Min	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Copper	*Mean*	0.16	0.01	0.02	0.02	0.01	0.02	0.02	0.02
	Max	0.21	0.03	0.06	0.05	0.04	0.06	0.05	0.05
	Min	0.07	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Nickel	*Mean*	0.18	0.00	0.00	0.00	0.01	0.01	0.01	0.01
	Max	0.21	0.01	0.01	0.02	0.02	0.02	0.02	0.02
	Min	0.08	0.00	0.00	0.00	0.00	0.00	0.00	0.01
Zinc	*Mean*	0.22	0.01	0.01	0.01	0.01	0.01	0.04	0.03
	Max	0.29	0.05	0.04	0.02	0.03	0.03	0.28	0.11

According to the boron concentration of effluents, biofilters did not show any efficient removal of boron. Effluent of crushed bricks biofilter has given always higher boron concentration than influent concentration whereas, after day 9, the other biofilters effluent had higher concentration than influent. This situation is not related to the change of hydraulic load due to the decreasing trend of efficiency already prior the change in hydraulic load. This inefficiency of boron removal is likely to be caused by leakage of boron from filter materials since boron consists in natural elements in different forms. Therefore, regarding the boron removal, the least efficient filter media have been found as crushed brick biofilters. The composite of crushed brick's effluent always gave higher concentration than initial concentration. The Figure 23 shows the concentration of boron for each effluent of biofilters by days with the average boron concentration of influent.

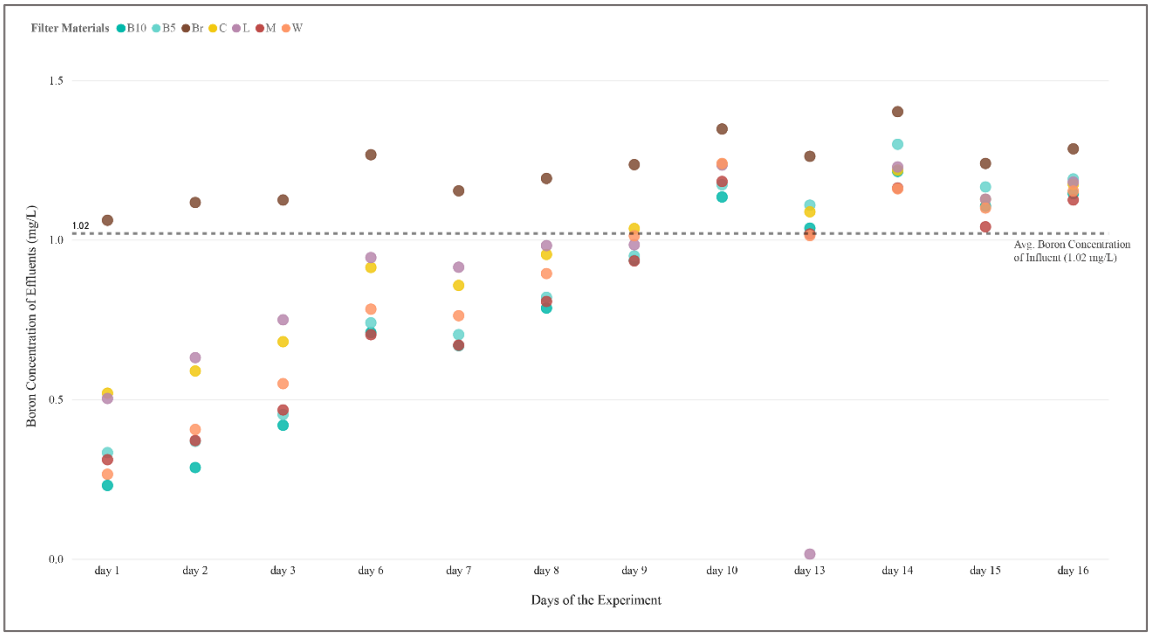

Figure 23: The concentration of Boron in the effluent of each filter medias with the average line of influent boron concentration.

Additionally, for the removal of metals and boron, the samples were measured also after they were filtered with PEF 0,22 µm syringe filters. This was done to understand if the metals and boron are present in the effluent as in dissolved from or bound to the suspended solids. According to the results, metals concentration did not show any significant difference from unfiltered sample concentrations. Overall, copper removal was 87.7%, nickel removal was 96.9%, and zinc removal was 93.7% on average for filtered samples. Therefore, it is possible to note that the metals which exist in effluents were in dissolved form mainly. On the other hand, boron efficiency showed some slight differences between unfiltered and filtered samples, especially at the beginning of the experiment. The point to consider, as can be seen in the Figure 24, during the first period, the day 6 (Monday), which means after weekend (72 hours of contact time) had an impact on the form of boron and helped dissolving the compound almost totally. Therefore the day 6, both measured removal efficiencies of filtered and unfiltered samples were equalized.

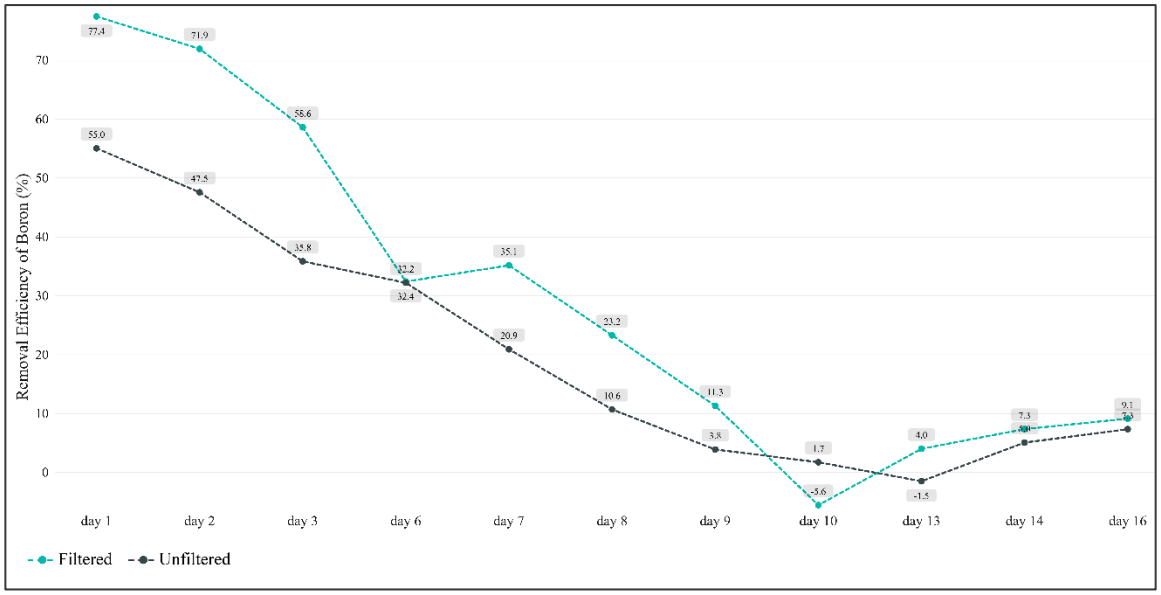

Figure 24: Average Boron Removal Efficiency of all Biofilters in Comparison with Unfiltered and Filtered Samples

6. Discussion

6.1. Nitrogen Removal of the Biofilters

Total nitrogen (TN), nitrate, nitrite, and ammonium ions were analyzed in this study which can give an idea about nitrogen presence and nitrification/denitrification processes within the columns. Nitrogen in the forms of nitrite, nitrate, and ammonium, is a nutrient which is necessary for plant growth; however, overabundance causes several adverse health and ecological effects.

According to the results, the TN removal efficiency of biofilters significantly decreased from the period 1 to the period 2. Thus, the nitrate concentration of biofilters increased from the period 1 to the period 2. The nitrite concentration increase was observed significantly only for biochar 10 and 5 % biofilters. Peaks of nitrate and nitrite were observed between the days 10 and 15 which is approx. the first week of the period 2. It is possible to recognize in the Figure 15 and the Figure 16, an increase both for the concentrations of nitrate and nitrite (nitrite only for biochar's) in the effluents after the day 6; however, especially for nitrate concentration after the day 9 showed an unstable trend. On the contrary, there was no significant change on the removal efficiency of NH_4^+ during the experiment and generally the removal rate was over 95% for all biofilters.

On average, woodchip biofilters achieved the highest TN removal rate with 70.9%. The removal efficiency of the first period was 89.4% and of the second period was 54.7%. Similarly, in comparison with the other biofilter materials that analyzed in this study, the lowest nitrate concentration was in the woodchip's effluent, although there was not any removal of nitrate. The results are in line with the study of Saliling, et al., (2007). Their study about woodchips as an alternative biofilter media to remove nitrate, indicated woodchips as successful biofilter media. In the study, woodchips achieved approx. 100% nitrate removal even though with an increasing nitrate loading rate. Furthermore, it is possible to observe from the Figure 15 and the Figure 16 that woodchips, nitrite and nitrate concentrations were not affected by the hydraulic change as much as the others. This may happen due to woodchips structure which does not keep oxygen and provide a good anaerobic environment for denitrification. However, at the same time their natural structure is vulnerable to degradation, therefore, estimated lifetime of woodchips was 1.2 years by Saliling, et al., (2007).

Effluents of biochar biofilters showed the highest concentrations of TN, nitrate, and nitrite and the lowest concentrations of NH_4^+. Additionally, only biochar biofilters achieved total organic carbon (TOC) reduction among the other biofilters. The removal efficiency of TN of biochar 10 and 5 % was 48.9% and 42.4%, respectively. The biochar 10% biofilters performed 81% removal efficiency through the first period and 20%

removal efficiency through the second period. Similarly, the biochar 5% showed 83% removal efficiency of TN though the first period whereas 6% through the second period. The removal efficiency of NH_4^+ of biochar 10 and 5 % was 99.7% and 99.8% during the experiment, respectively. These results are in line with the indications of the study of Dalahmeh, et al. (2019). In their study, biochar filters both as vertical and horizontal flow filters demonstrated 93% of NH_4-N removal and 71% of TN removal. Similarly, as stated in their study, biochar has a high capacity of NH_4-N adsorption and the condition of changing hydraulic load could provide the system an aerobic condition which drives the adsorbed NH_4-N oxidation to nitrate by a nitrification process within the biochar biofilters. This could be the reason for high nitrite and nitrate concentration after the hydraulic change. As discussed by Zhou, et al. (2019) to enhance the denitrification process through biochar filters, the main contribution can be provided by adsorption-desorption of influent organics on biochar. On the other hand, as mentioned in the study of Dalahmeh, et al. (2012), the high concentration of nitrate in the effluent of biochar biofilters can be replaced with chemical fertilizers and used for irrigation.

Mycorrhiza is generally used in biofilters to enhance the nutrient and metal uptake of plants. Although there is not greywater treatment, studies on mycorrhiza, have found, there are studies focusing on stormwater treatment with mycorrhiza fungi. As stated by the study of Altenstedt (2020) which was comparison of the impacts of biochar and mycorrhiza on the removal of nitrogen, mycorrhiza did not have considerable effect on reducing nutrients. The results from this thesis study make a similar indication due to quite similar performance were observed both for mycorrhiza and compost biofilters. The removal of TN (68% and 64.4%, respectively) and removal of NH_4^+ (99.4% and 98.1%, respectively). Similar results were obtained also for effluent concentration of nitrate and nitrite.

In this study, the chemical composition of the materials have not been considered. Therefore, further studies will be necessary; however, limestone contains calcium carbonate ($CaCO_3$) and it is known as its use for fertilizer. Limestone chippings and crushed bricks can contain alkaline agents (Wurochekke, et al., 2016). This was reported by Wurochekke, et al., (2016) as an considerable aspect for reduction of nitrogen and phosphorus. The change of hydraulic loading may have caused some air accumulation within the hole that limestone chippings provide (like biochar filter medias). Therefore, this situation can decrease the bacterial activity within the biofilters.

6.2. XOC's Removal of Biofilters

XOC's measured in this study were benzotriazole (BTA), diclofenac (DCF), methylparaben (MTB), DEET, caffeine (CAF), and anionic surfactants (AS). All these compounds are a wide representative of PPCPs used in daily life of a household. As the removal of PPCPs in the environment, biodegradation plays a major role (Xu, et al., 2009), however, sorption is reported as main way for the removal of DEET and CAF (Greenstein,

et al., 2018). The comparative experiments conducted with different filter materials showed that XOC's might undergo sorption and biodegradation after the adaptation period achieved and biochar can be a promising material to use for on-site biofilters.

For the removal of MTB, all biofilters achieved approx. 100% removal efficiency. As resulted in the study of Li et al., (2015), the anaerobic tank was the main part where parabens were degraded, and the sorption contribution was not significant. Therefore, the results convince that anaerobic conditions of the biofilters could be fairly good environment for biodegradation of MTB. However, degradation was detected also in the stored synthetic greywater during the third week . For this reason, further studies will be necessary for MTB removal. Similary, CAF removal efficiency was quite high through biofilters. During the first period (0.5 L of SGW irrigation), all biofilters achieved 100% removal efficiency. However, the hydraulic change had an visible effect on the removal efficiency of compost, crushed brick, and limestone chipping biofilters (Figure 18). During the second period, the best CAF removal efficiency was obtained from biochar 5% (as 100%) and then biochar 10% (as 99.8%) biofilters. Luján-Facundo, et al., (2019) studied the adsorption of caffeine through granular activated carbon in a glass column and found out that the adsorption rate increases with increasing column height and but not with contact time. Therefore, rich carbon content helps the adsortion of CAF which also supported by the findings of Greenstein, et al., 2018. However, most probably due to easy degradable structure of caffeine (Sui, et al., 2010), all biofilters achieved more than 98% removal efficiency.

The removal of DCF showed differences among the biofilters. Biochar 5%, 10%, and mycorrhiza did not show any unefficiency removal during the experiment. Their removal efficiency was almost 100%. On the other hand, compost, woodchips, crushed bricks, and limestone chipping biofilters showed a declining efficiency till the day hydraulic loading changed. Then, an increasing trend was observed for the removal efficiency of DCF. In all biofilters, the general observation after the change of hydraulic load, was a long-term increase of TOC concentration in the effluents and a rapid increase and then stabilization of TN concentration in the effluents. However, hard to find a correlation between TN, TOC changes and DCF removal. This is contrary to the findings of Sochacki, et al. (2018) who reported a strong correlation between increasing TN value caused a decrease of diclofenac removal. On the other hand, TOC might have an affect on the removal since the biochar filters had the highest TOC removal efficiency. However, although mycorrhiza gave good results of DCF removal like biochar, TOC vallue of mycorrhiza was the highest which means, it needs further microbiological analyses. In addition to these, DCF is reported as high photodegradable compound (Buser, et al., 1998), in this experiment, all columns of biofilters were covered by alumium foil which prevented sunlight, therefore, the impact of photodegredation on the removal efficiency is limited.

The removal efficiency of DEET, except biochar 5% biofilter, showed a considerable decline during the first week. After the day 7, the removal efficiency of crushed brick, limestone chippings and compost biofilters increased, whereas biochar

10%, woodchips and mycorrhiza biofilters stabilized the removal. After the hydraulic load change, all biofilters performed an increasing removal trend which they achieved at the end more than 90% removal efficiency and showed steady state. Whereas Sui, et al., (2010) found 69% removal efficiency for DEET from a conventional treatment plant, the results of this study indicated better removal rate under green house conditions. As indicated by the results of Turner, et al., (2019) that DEET is found frequently in shallow groundwater and adjecent surface water where the untreated greywater is used for sub-surface irrigation, which can lead to the environmental contamination by organic micro-pollutant. Therefore, as this was a small-scale study, results may not be conclusive; however, it can be a good base for further studies.

Regarding BTA, the removal efficiency of all biofilters was significiantly higher for the first day, which may be the impact of used sludge from a real greywater prior to the start of the experiment (inoculation period). As reported by Kowalska, et al., (2019) that the biodegradation of BTA is negligible unless activated sludge is used. However, a constant decrease of removal efficiency was observed during the experiment except biochar biofilters both 10% and 5%. This might indicate that the adsorption capacity of biochar is higher than the other materials since the biodegradation magnitute for BTA is reported as low in the literature (Kowalska, et al., 2019). After the day 20, the removal efficiency of woodchips,crushed brick, compost, and limestone chipping biofilters was lower than 60% whereas, mycorrhiza biofilters performed slightly better (70%) and both biochar biofilters performed >99% removal efficiency. However, there was not a steady condition and the decrease was constant. Therefore, further studies are required for BTA removal efficiency.

As a surfactant, sodium dodecyl sulphate was added in an average 1.57 mg/L to the biofilters, and high removal efficiency was obtained from all biofilters. It was proven by Ramprasad (2016) that higher temperature is better for biodegradation of sodium dodecyl sulphate. Under the greenhouse conditions, at average 22 ^{o}C, the removal efficiency of anionic surfactants was calculated as 85.21%. Therefore, obtained high removal efficiency of biofilters needs to be supported by a further study under field conditions.

6.3. Boron and Metals Removal of Biofilters

Regarding boron and metal analyses, the effluent samples of biofilters were analyzed as both filtered and unfiltered to understand the form (dissolved or bound to suspended solids) of the compounds in the treated greywater.

Results of metal analyses showed that the concentration of metals (copper, nickel, and zinc) did not give significant disparity between filtered and unfiltered samples and considerably high removal efficiency (>85%) was obtained. Total average of copper, nickel, and zinc was measured as 91.4%, 96.9%, and 93.7% eliminated for filtered

samples and 88.5%, 96.5%, and 90.1% eliminated for unfiltered samples, respectively. Mycorrhiza achieved 89% of copper removal, 96% of nickel removal, and 93% of zinc removal during the experiment for the filtered results. For unfiltered results, mycorrhiza achieved 86 % of copper removal, 95.7% nickel removal, and 95% zinc removal. Although, these results are in accordance with the results of Poor, et al., (2018) that indicates mycorrhiza fungi may increase the uptake of copper and zinc by plants, mycorrhiza performed high removal efficiency without uptake by plants. However, since the other biofilters achieved similar removal efficiency with mycorrhiza, and this study cannot indicate specific effects of different materials for the removal.

Moreover, according to the WHO water quality criteria (the WHO, 2006 (a)) for drinking water existing of nickel and zinc in drinking water should not exceed 0.07 mg/L. The biofilters within this study achieved to remove both nickel and zinc concentration from on average 0.21 and 0.22 mg/L in the influent to lower than the acceptance level. On average 0.02 mg/L from compost, woodchips, mycorrhiza, crushed bricks, and limestone chippings, and 0.01 mg/L from biochar biofilters of nickel concentration in effluents were obtained. Similarly, on average, 0.04 mg/L, 0.03 mg/L, and 0.01 mg/L of zinc concentrations were obtained from the effluents of crushed bricks, limestone chippings, and the rest, respectively. Additionally, copper concentration in water more than 5 mg/L is not recommended due to the change of taste and color. The achievement of biofilters removed the copper concentration from 0.21 mg/L to <0.06 mg/L. Therefore, the biofilters were significantly successful to remove metals from synthetic greywater.

Boron removals in the biofilters were shown decreasing removal efficiency during the experiment. The measured removal efficiency of unfiltered samples was 54.9% on the day 1 and showed a constant decrease until the day 13 (the removal efficiency was -1.5%). On the first day, the highest boron removal was obtained from biochar 10% biofilters (77%), then respectively, woodchips (74%), mycorrhiza (69.3%), biochar 5% (67.2), limestone chippings (50.5%), compost (49.7%), and crushed bricks (-3%). Crushed bricks biofilters were not able to remove boron during the experiment. Moreover, boron was leached from the crushed bricks as the overall boron removal was -10.5% from the crushed brick biofilters. During the experiment,the highest boron removal was observed for bichar 10% with 32.7% removal efficiency. After the day 13, a slight increase was observed on the removal of boron from all biofilters. In the literature, higher removal (around 50%) was reported by Gross, et al., (2007) through a recycled vertical flow constructed wetland.

7. Conclusion

Greywater as one of the possible freshwater resource, is a thrilling topic for sustainable and resilient future cities and societies. Regarding integrated urban and rural water management, greywater treatment and reuse have a crucial role to improve urban water infrastructure. Seperation of wastewater itself and implementing on-site treatment systems can decrease water consumption, treatment load and cost, and save energy. Together with all these, it can improve water reservoirs and their quality. Although wastewater seperation is a topic of 2000's which Australia and Germany played the leading role, some countries like Turkey have the topic recently in their agenda (Giresunlu & Baykal, 2016). Therefore, ongoing researches is searching for new discoveries and complementary knowledge on better wastewater treatment techniques.

In this study, to provide complementary data for on-site treatment methods, removal efficiencies of different biofiltration materials (biochar, compost, woodchips, mycorrhiza, crushed bricks, and limestone chippings) were investigated specifically for nutrients, organic and inorganic compounds, surfactants, boron, and metals. The biofilter materials were composited in different ratios with sand, soil, and compost (technogenic soil). All these materials were designed within 2 liters of column with 3 up to 5 replicates (totally 30 columns) and irrigated with synthetic greywater for 23 days.

The removal efficiency of biofilter materials indicated different results for different compounds under the same room conditions. According to the nitrogen removal of the biofilters, woodchips gave the best removal efficiency for TN, nitrate and nitrite, therefore could be a suitable material for on-site infiltration trenches for greywater treatment. On the other hand, although the removal of TN through biochar biofilters were not efficient, as reported by Dalahmeh, et al., (2012), biochar can be used in fields to replace the chemical fertilizers. Regarding the reuse applications, it is also necessary to consider lifetime of the filter materials. Therefore, a further study also should be conducted to investigate the quality of materials after the treatment process and urge some areas where they can be reused or stored. To illustrate, adsorption of heavy metals is successfully achieved by the filter materials; however, as underlined also by Dalahmeh (2013) that it can create a great risk of contamination in case of reuse.

According to the results of xenobiotic organic compounds, biochar was the most promising filter material both for its larger (10%) and smaller (5%) volume. Among the tested XOC's, benzotriazole was the most persistent compound in terms of concentration in effluents. Finally, except biochar biofilters, the other biofilters were able to eliminate benzotriazole less than 70% with a decreasing trend. On the other hand, it can be concluded that adsorption process achieved the best performance after the adaptation period of biofilters for all compounds. Furthermore, to understand the full capacity of the

six filter materials, exploring their biological activities is necessary to do microbiological analyses.

This study provide a general perspective on the treatment efficiency of the selected biofilter materials for the removal of indicated organic and inorganic compounds. However, different types and characters of the same materials or design of the experiment can perform different results, therefore more research is necessary to do in the future to optimize the potential of these materials for biofilters. The experiment of this study was conducted under laboratory conditions, thus, to determine the long-term treatment capacity of biochar, compost, woodchips, mycorrhiza, crushed bricks, and limestone chippings biofilters, a further study under field conditions is necessary.

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9. List of Figures

10.List of Table