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MICROPLASTICS IN BANKFILTRATED DANUBE WATER

Master thesis
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submitted by:
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"Have you also learned that secret from the river, that there is no such thing as time?" That the river is everywhere at the same time, at the source and at the mouth, at the waterfall, at the ferry, at the current, in the ocean and in the mountains, everywhere and that the present only exists for it, not the shadow of the past nor the shadow of the future."

(Hermann Hesse, Siddhartha)

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I dedicate this work with great love to Sofie Vymetal and Hermine Rabitsch.

Table of contents

Abstract

Massive accumulation of plastic and microplastic particles has been reported in marine ecosystems, posing a risk to the biota. Less attention has been given to freshwater ecosystems. Since plastic particles have been found in the Danube, the question arises whether these can pass through the bank filtration into the drinking water.

This research therefore aims to address this question by evaluating microplastic particles > 30 µm in Danube water, bank filtrate and groundwater.

Two sampling methods were applied in this study. One method used three stainless steel filter holders (Stainless Steel Cascade- SSC) and the other system consisted of five cartridge filters (Blue Filter Cascade- BFC), equipped with nylon filters of descending pore sizes (500 - 5 µm) and cellulose nitrate filters (1.2 µm), in order to create a cascade. The methods were used at five sampling sites close to the Danube to extract particles of different size fractions. In addition to sampling, particle counting and turbidity measurements were carried out. In the laboratory, various separation and collection methods such as density separation with a saturated NaCl solution, a chemical separation and an ultrasonic separation were performed. The best results were obtained by density separation. Separated particles > 30 µm were characterized with the Raman spectrometer. It was possible to identify microplastic particles in two Danube water samples. No microplastics > 30 µm could be detected in the bank filtrate or in groundwater. Furthermore, recovery experiments with the SSC with microplastic particles (500 - 20 µm) were executed. The recovery rate was 89 % in average.

Zusammenfassung

In marinen Ökosystemen wird von massiven Plastik- und Mikroplastikanhäufungen berichtet und vor einem Risiko für Biota gewarnt. Den Süßwasserökosystemen wird indes weniger Aufmerksamkeit geschenkt. Seit dem Fund von Kunststoffpartikeln in der Donau stellt sich allerdings die Frage, ob eine Möglichkeit besteht, dass diese durch die Uferfiltration auch in das Trinkwasser gelangen.

Diese Arbeit geht daher der Frage nach, ob sich Mikroplastikpartikel > 30 µm in Donauwasser, Uferfiltrat und Grundwasser befinden.

In dieser Studie wurden zwei Probenahmeverfahren angewendet. Ein Verfahren verwendete drei Edelstahlfilterhalter (Stainless Steel Cascade- SSC) und das andere System bestand aus fünf Patronenfiltern (Blue Filter Cascade- BFC). In beiden Systemen wurden in einer Filterkaskade Nylonfilter mit absteigender Porengröße (500 - 5 µm) und Cellulosenitratefilter (1,2 µm) verwendet. Die Methoden wurden an fünf Probenahmestellen entlang der Donau eingesetzt, um Partikel unterschiedlicher Größenfraktionen abzufiltrieren. Neben den Probenahmen wurden Partikelzählungen sowie Trübungsmessungen durchgeführt. Im Labor wurden verschiedene Trenn- und Sammelverfahren wie die Dichtetrennung, eine chemische Trennung und eine Ultraschall Separation erprobt. Weitere Untersuchungen der separierten Partikel wurden mit einem Raman-Spektrometer durchgeführt. Mit diesem laserbasierten Charakterisierungsverfahren konnten Mikroplastikpartikel in zwei Donauwasserproben identifiziert werden. Es konnte kein Mikroplastik > 30 µm, im Uferfiltrat oder im Grundwasser nachgewiesen werden. Darüber hinaus wurden mit der Edelstahlkaskade Wiederfindungsversuche mit Mikroplastikpartikeln (500 - 20 µm) durchgeführt, um sowohl quantitativen als auch qualitativen Fragestellungen nach zu gehen. Die Wiederfindungsrate betrug im Durchschnitt 89 %.

1 Introduction

The pollution of the world's oceans with plastics, especially microplastics, has achieved great public as well as scientific attention and research examining the occurrence of microplastics in the marine environment has substantially increased. Field and laboratory work regularly provide new evidence regarding the fate of microplastic debris. This debris has been observed within every marine habitat, including the polar regions and the deep sea. In 2013, Ivar do Sul and Costa reviewed 101 papers investigating microplastics pollution, all saying that most of the marine organism groups are at an eminent risk of interacting with microplastics. Microplastics are commonly studied in relation to plankton samples, sandy and muddy sediments, vertebrate and invertebrate ingestion and chemical pollutant interactions.

In 1972, Carpenter and Smith were the first researchers to ring an alarm on the presence of plastic pellets on the surface of the North Atlantic Ocean, and up to now nothing has changed in the brisance of the subject.

Several million tonnes of plastics have been produced since the middle of the last century (Barnes et al., 2009; Thompson et al., 2009; Andrady, 2011). Worldwide plastic production (thermoplastics, polyurethanes and other plastics excluding PET, PA, PP and polyacrylic fibers) increased from 230 to 322 million tonnes/year between 2005 and 2015, however the European Union share (EU 28 plus Norway and Switzerland) has remained the same (Plastic Europe, 2016) at approximately 60 million tonnes/year.

Microplastics are tiny plastic particles up to 5 millimeters in diameter (UNEP, 2016), although the discussion of size is not yet complete. In the GESAMP study (GESAMP, 2015), particles between 1 nm and < 5 mm are referred to as microplastics, but sampling in surface water is usually carried out with planktonic nets with mesh sizes of 330 µm. In the ISO Working- Draft "Plastics" microplastics (ISO, 2016) are divided into large microplastics between 1 mm and 5 mm and small microplastics < 1 mm (usually within the range of 200 - 800 µm).

The literature distinguishes between primary and secondary microplastics. Primary microplastics include plastic pellets such as granules intended for further industrial processing, or plastic particles used directly or as product additives. Sources of microplastics include cosmetics and personal care products, textiles and clothing (synthetic fibers), traffic (dust from tires), cleaning agents or compressed air cleaning additives and the emissions from plastic producers and processors (plastic resin pellets). A variable proportion of microplastics is released by sewage treatment plants, whereby, depending on the purification procedure, the differences are significant (UNEP, 2016). The secondary microplastics designate minute plastic fragments, which are formed by physical, e.g. UV or mechanical abrasion, biological or microbiological and chemical (e.g., oxidation) processes of larger plastic waste fractions. Over the time, the structural integrity of plastic waste can be lost and microplastics could become nanoplastics.

The smallest microparticles investigated in the oceans are 1.6 µm in diameter (Cole et al., 2011).

The marine studies led to the question if the microplastic debris is a freshwater problem, too. In Austria, microplastics have gained attention due to the study of Lechner et al. (2014). The researchers discovered that the number of microplastic particles in the plankton nets exceeded those of fish larvae during sampling in the Danube. The authors state that, every day, 4.2 tonnes of plastics arrive at the Black Sea through the Danube. A study commissioned by BMLFUW (Hohenblum et al., 2015) showed that plastic particles (> 500 µm to 5 mm) were found in the water of the Danube in Austria. Hohenblum et al. (2015) state, 0.3 plastic particles per m^3. However, no plastic particles could be identified in the examined fish.

Since plastic particles have been found in the Danube, the question arises whether these can also pass through the bank filtration and into the drinking water.

Up to this date, no study exists that has investigated particles smaller than 0.5 mm or microplastics in bank filtrate.

While the research on marine microplastics is more advanced, there are immense knowledge gaps regarding freshwater microplastics. Data on their abundance is incomplete for large and absent for small surface waters. Likewise, relevant sources and their fate in the environment remain to be investigated. Data on the biological effects of microplastic debris in freshwater species is completely lacking. The accumulation of other freshwater contaminants on microplastic particles is of special interest because ingestion might increase exposure to chemicals. Again, data is unavailable on this important issue (Wagner et al., 2014). In this context, method harmonization is needed in order to obtain comparable data from different environmental compartments and sites. This includes sampling strategies, sample treatment and reliable analytical methods to identify microplastics (Dris et al., 2015).

Greater knowledge of the extent and impact of microplastics in marine waters versus fresh waters is reflected in more policy and management interest in marine systems, though even these are still under development (Medrano et al., 2015).

This research has been carried out to clarify the question whether microplastics can be found in bank filtrate. It is important to find out if microplastic particles would make the path through the bank filtration into the drinking water and thus their way into our everyday life and into the human body.

2 Objectives

No study on microplastics in bank filtrate is available in literature yet. As there is no internationally recognized methodology for the measurement of plastic particles in bank filtrate, however two different sampling methods have been developed, tested and applied within this study.

A further concern of this work was to identify size fractions > 30 µm in Danube water, bank filtrate and groundwater samples and reconcile the particle content of the samples with the turbidity.

This research about microplastics aims to:

- evaluate the microplastics content in Danube water, bank filtrate and groundwater samples, identify the particles with the Raman spectroscopy and determines their size;
- determine the particle quantity, size distribution and the turbidity of the samples. Compare the results of the particle counting and turbidity measurements. Obtain the data by initializing worst case scenarios at the sampling sites; and
- evaluate the collection efficiency of different separation techniques by different separation methods.

3 Fundamentals

3.1 Definition of microplastics

3.1.1 Definition of plastics and microplastics

Plastics is a common term for a broad family of organic materials of typically high molecular weight suitable for the manufacture of industrial products. This term is often interchangeably used with the term polymers. This is however only partially true; a common phrase says “All plastics are polymers but not all polymers are plastics”. Other polymeric compounds are of biological and inorganic origin, including starch, proteins and DNA. Their polymeric nature is just one of many chemical properties of plastics (Gorycka, 2009). Plastics are synthetic, non-metallic and moldable. They consist of repeating macromolecules derived from the polymerization of monomers extracted from gases or oils (e.g. naphtha) (GESAMP, 2010). The type of plastic produced depends on the different alteration of monomers during the polymerization process. The polymerization process can lead to the creation of weak or strong carbon-hydrogen bonds. The weaker of the two characterizes the amorphous structure (e.g. polyethylene), appearing like a loose pile and entangled strings. The stronger bonds are typical for the crystalline structure (e.g. polystyrene) in which the molecules are packed tightly together. The final products are processed, for example by molding, to various shapes and forms.

Two groups of plastics can be distinguished, thermoplastics and thermosetting plastics. Thermoplastics can be repeatedly thermally softened and hardened by cooling, which means that these kinds of plastics can be remolded and reused practically indefinitely. For this reason, most thermoplastic products are suited to a wide range of purposes, like packaging, food containers and other household products. More durable and heat resistant thermoplastics are used in automobiles, machinery and electrical products. Thermosetting ones on the other hand, harden permanently after being heated. Due to their high melting points they are used mainly for high-heat resistant products (Gorycka, 2009).

The term plastic covers a huge spectrum of materials for instance plastics, rubber, elastomers, textile fibers and technical fibers (GESAMP, 2010). Though they have only existed for just over a century, the adaptability of plastics has led to a great increase in their use over the past three decades, and they have rapidly found their way into everyday life (Derraik, 2002). The plastics’ favorable characteristics, being lightweight, strong, durable and low cost, as well as their excellent oxygen/moisture barrier properties and bio inertness, make them attractive for the manufacture of a very wide range of products. Plastic materials have been replacing conventional materials such as glass, metal and paper as packaging material of equivalent or superior design.

These mentioned properties are the reasons why plastics are a serious hazard to the environment. Since they are very buoyant, a huge load of plastic debris is being dispersed over long distances, and when they finally settle, they may persist for centuries (Goldberg, 1997).

The threat of plastic pollution has been ignored over centuries, and its serious harm to the lacustrine ecosystem has been only recently recognized. The first reports of plastic litter in the marine system in the early 1970s drew minimal attention from the scientific community. In the following decades, with accumulating data on ecological consequences of such debris, the topic received increasing, sustained research interest. A particular concern is the occurrence of smaller pieces of plastic debris including those not visible to the naked eye, called microplastics.

The annual global production for plastics has consistently increased over the last 50 years and rose in 2013 to 299 million tonnes, meaning a 3.9 % increase compared to 2012 (Fig. 1). However, the plastics production in Europe has been stable since 2002 (~ 57 million tonnes/year) (Plastic Europe, 2015).

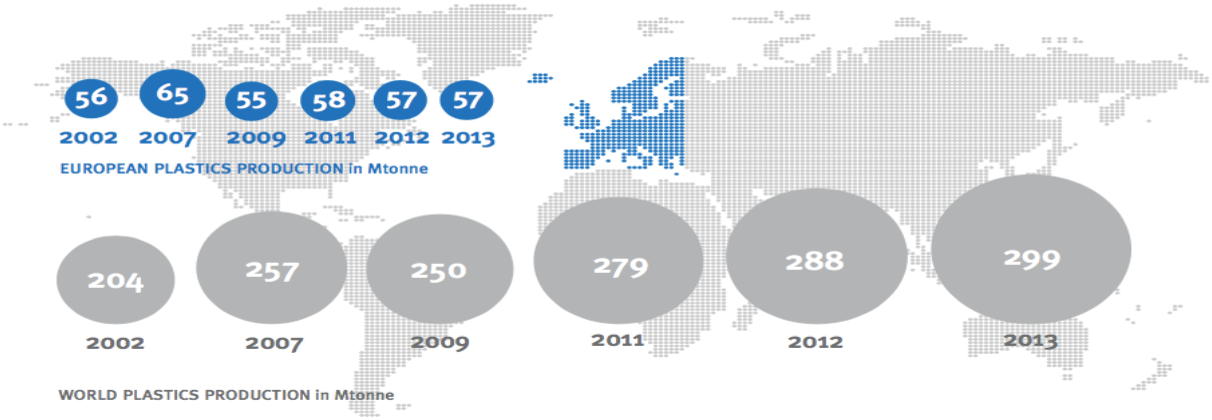

Figure 1: Global and European plastic production in million tonnes (Source: Plastice europe, 2015)

Worldwide, a special focus has been placed on plastic debris at the microscale, as it is widespread in the environment and has accumulated in marine as well as lacustrine systems and sediments rapidly in recent years, with maximum concentrations reaching 100.000 particles per m^3 (Wright et al., 2015).

The term, microplastics, has been defined differently by various researchers and a clear, uniform definition as well as a precise delimitation for nanoscale plastics has not yet been determined. The following designation seems to be enforced by Burkhard (2014).

- Macroplastics: > 5 mm
- Microplastics: 50 µm - 5 mm
- Nanoplastics: < 5 µm

The term microplastics refers more or less to a particle size < 5 mm (5.000 µm) (Browne, 2015). According to Storck et al. (2015) the term “micro” implies 1 µm. However, most studies investigated particles > 300 µm. Currently, the categories “large” (1 mm to 5 mm) and “small” (< 1 mm) have been introduced (Imhof et al., 2012). The lower limit is mostly determined by the mesh size of the sieve or net used for sample filtration and by the application of spectral and optical analysis for identification (Hidalgo-Ruz et al., 2012).

The standardization of sizes and materials of microplastics at an international level is a pre-requisite to make study results comparable.

Figures 2 and 3 show microplastic size categories in mm of two different studies, i.e. Browne (2015) and Hidalgo-Ruz et al. (2012).

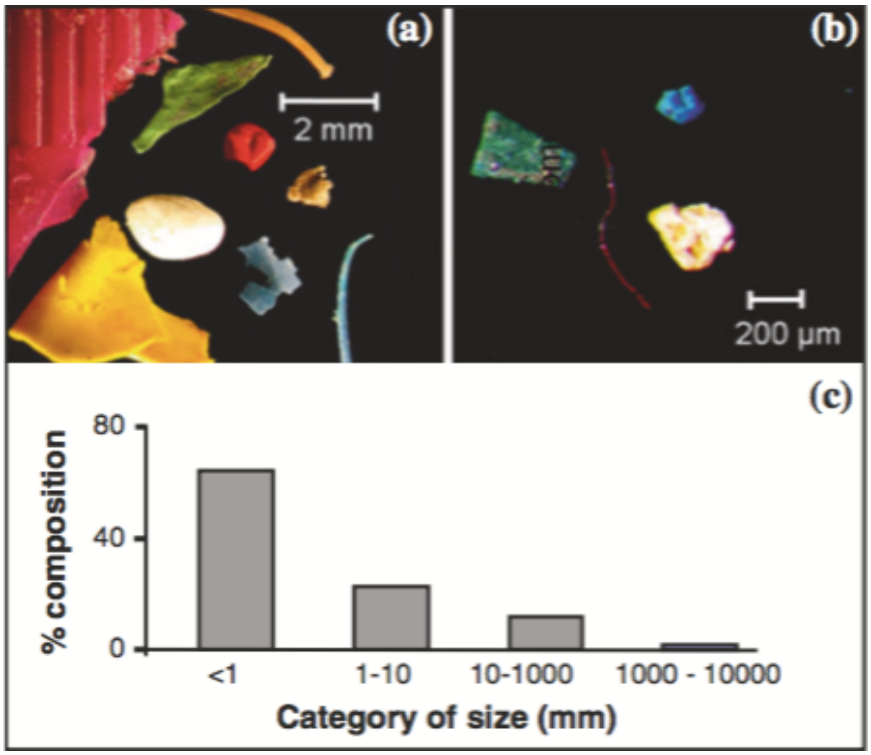

Figure 2: Size categories of microplastics in mm (Source: Browne, 2015)

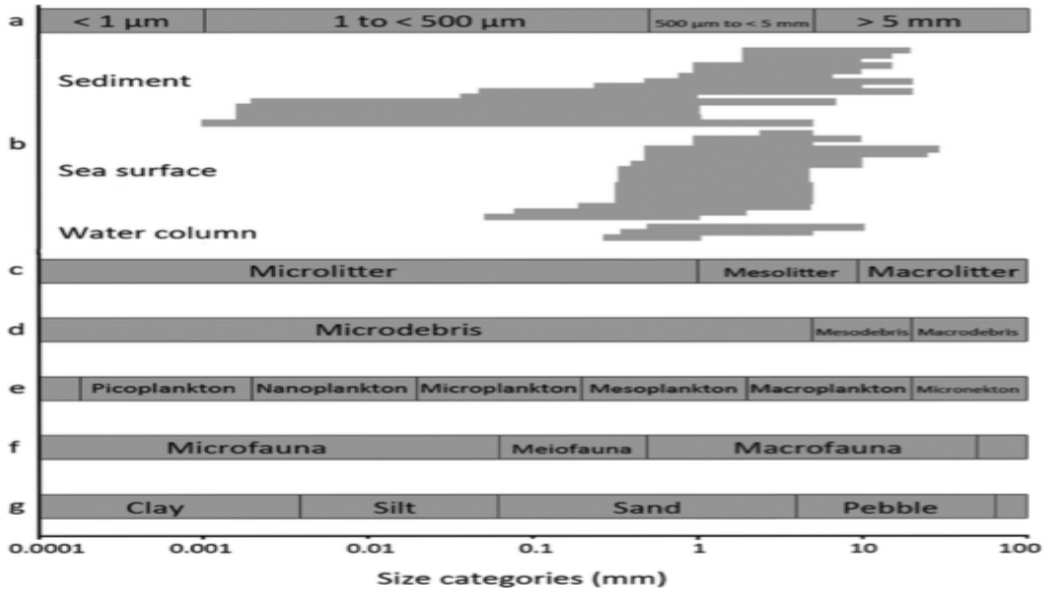

Figure 3: Size categories in mm (Source: Hidalgo-Ruz et al., 2012)

3.1.2 Primary and secondary microplastics

The particles' history and the planned use of the plastic particles determine whether they should be classified as primary or secondary microplastics.

The primary microplastics include plastic pellets which are directly used or used as product additives. Granules, which are intended for further industrial processing, are also part of the primary microplastics. They are typically used in facial cleansers and cosmetics, or as air blasting media, whilst their use in medicine as vectors for drugs is increasingly reported.

Secondary microplastics describe tiny plastic fragments derived from the breakdown of larger plastic debris. Over time, a culmination of physical, biological and chemical processes can reduce the structural integrity of plastic debris, resulting in fragmentation. Secondary microplastics serve no purpose but they are the consequence of the degradation of larger plastic parts by sunlight radiation, microorganisms, oxidation, or by mechanical abrasion. The fragments become smaller and smaller over time.

It is thought that microplastics might further degrade to be nanoplastics in size, although the smallest micro particles reportedly detected in the oceans at present are 1.6 µm in diameter (Cole et al., 2011).

The range of materials found corresponds with the plastic types of use. The most commonly used plastics are Polyethylene (PE) and Polypropylene (PP).

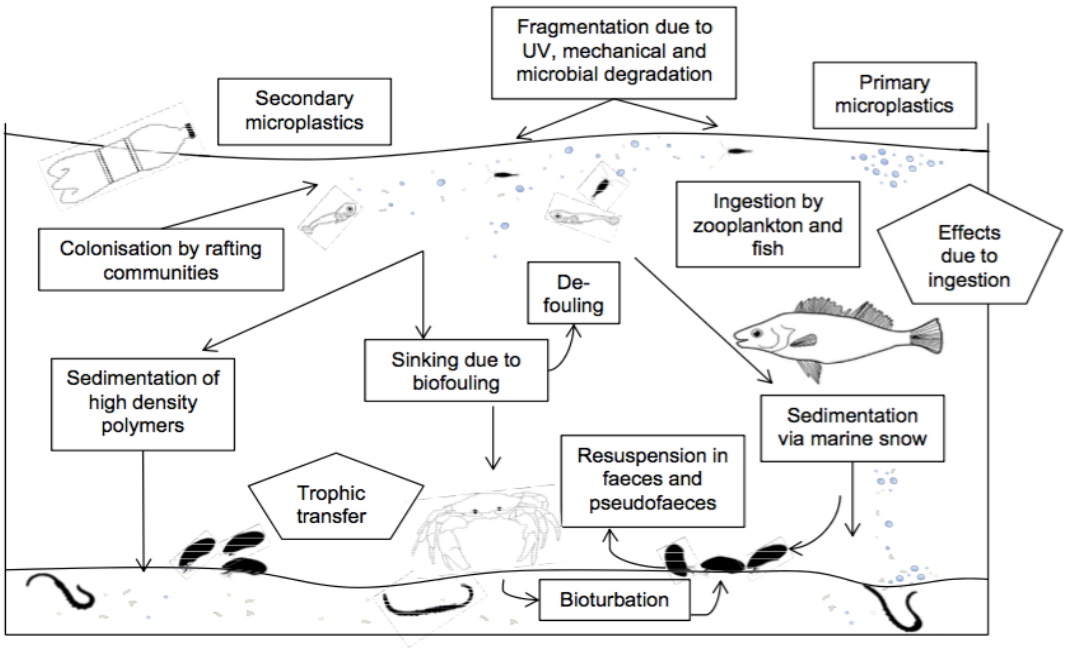

Figure 4: Origin of primary and secondary microplastics and potential pathways for their transport (Source: Wright et al., 2013)

Regarding the effects on the environment, it is important to know whether they are primary or secondary microplastics. As the latter becomes more persistent over time due to exposure to the environment, they are commonly associated with the aging of plastic surfaces. An aged, roughened, porous surface offers more adhesive surfaces for environmental pollutants (Liebmann et al, 2015).

3.2 Types of microplastics

Forensic techniques that compare the size, shape, and type of polymers found that the major types of microplastics in the studied habitats are polyethylene, polystyrene and polypropylene (Browne et al., 2011). Burkard (2014) described the most common types of microplastics and their structures:

3.2.1 Polyethylene (PE)

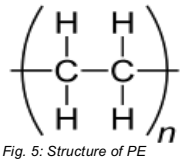

Fig. 5: Structure of PE

C_2H_4 is the chemical formula of polyethylene. Polyethylene is, with a share of about 29 %, the world's most widely produced plastic. In 2001, 52 tonnes were produced. Approximately 67 % of this amount goes to packaging and about 68.000 tonnes worldwide were used for plastic bags. This corresponds with a share of 0.71 % or 0.83 kg per inhabitant.

3.2.2 Polypropylene (PP)

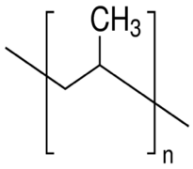

Fig. 6: Structure of PP

Worldwide more than 45 million tonnes of polypropylene are manufactured annually (2001) and it is chemically defined as C_3H_6. It is odorless and hypoallergenic and has a higher rigidity, hardness and strength than PE. It is suitable for applications in the food and pharmaceutical industry and is physiologically harmless.

3.2.3 Polyamide (PA)

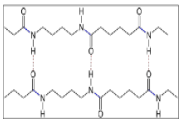

Fig. 7: Structure of PA

Polyamide is a linear polymer with regular repeating amides along a complex chain. This material obtains its importance through the production of nylons or aramids. Due to their chemical resistance, high tear resistance, as well as the uniform, smooth surface with high lubricity, the fibers are suitable for textiles and even sewing material in surgery. Polyamides are very sensitive to the moisture content of the surrounding environment and react with reversible water uptake or release, and thus also take up contaminations.

3.2.4 Polystyrene (PS)

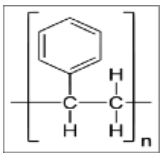

Fig. 8: Structure of PS

Polystyrene is chemically defined as C_8H_8. Polystyrene is a kind of white foam and used as a lightweight packaging material and effective insulation. Approximately 20 % of the polystyrene production is used in electronic applications, 39 % in packing materials and about 15 % are used in construction processes as insulation material. In general, the material is physiologically safe and approved without restrictions for food packaging, however the addition of flame retardants in insulation and foam panels make it very toxic for the aquatic flora with unknown long term effects. Furthermore, the addition of pesticides and algae protection products increases the hazard to the environment.

3.2.5 Polyvinylchloride (PVC)

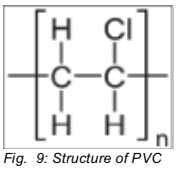

Fig. 9: Structure of PVC

PVC, a thermoplastic product with brittle and hard properties is chemically defined as C_2H_3Cl. Through additives, primarily stabilizers and plasticizers, it is adjusted to various application areas and its physical properties are improved, especially its sturdiness and moldability. The plasticizer phthalate is considered highly hazardous to our health, because it interferes with the hormonal balance of humans and damages the reproduction or development of cells. Through the incineration of PVC, hydrochloric acid and various carcinogenic organic substances are released.

3.3 Impacts of microplastics

Under normal environmental conditions, plastic particles that eventually end up in the surface water are not easily biodegradable. The degradation rate of synthetic polymers is extremely low, of course depending on the type of plastics and environmental conditions. It probably ranges from a few decades to several centuries, resulting in an accumulation of microplastics in the aquatic environment. The risk of microplastics is primarily caused by the combination of persistency of these materials and their potential accumulation in the food chain (Roex et al., 2011).

Microplastics occupy the same size fraction as sediments so they are potentially bioavailable to a wide range of organisms. They can be ingested by low trophic suspension, filter and deposit feeders, detritivores and planktivores. The factors influencing the availability of microplastics mentioned below have been described by Wright et al. (2013).

3.3.1 Size

A key factor contributing to the availability of microplastics is their small size, making them available to lower trophic organisms. Many of these organisms exert limited selectivity between particles and capture anything of appropriate size, directly or indirectly. Alternatively, higher trophic planktivores could passively ingest microplastics during normal feeding behavior or mistake particles for natural prey.

3.3.2 Density

The density of the plastic particles determines bioavailability in the water column, so the type of plastic ingested may vary between organisms and their preferred habitat. This cyclic pattern may make microplastics available to organisms occupying different depths of the water column at different times. The buoyancy of plastic is influenced by biofouling, for example, PE food bags displayed a well-developed biofilm within one week, which continued to increase throughout a three-week exposure period. By the third week, the PE food bags had started to sink below the sea surface, indicating neutral buoyancy. Fouling in the water column by foraging organisms is a potential pathway for microplastic particles to return to the sea air interface. Alternatively, fouled microplastics could continue to sink, as would high density plastics such as PVC, and would eventually reach the benthos. The rate of biofouling depends on parameters such as surface energy and hardness of the polymer, as well as water conditions.

3.3.3 Abundance

An increase in the abundance of microplastics in the environment will also affect their availability, as well as the enhanced chance that an organism will encounter microplastic particles. Therefore, the progressive fragmentation of microplastic items is likely to increase the amount of particles available for ingestion to a wider range of organisms.

3.3.4 Color

The color of the microplastics may interfere with the likelihood of consumption due to the similarity to prey. Studies reported that plastic particles sampled from the North Pacific exhibited size variation related to color; white plastic particles consistently decreased in abundance with decreasing size classes. Some commercially important fish and their larvae are visual predators, preying on small zooplankton, and may feed on microplastics which most resemble their prey, for instance white, tan and yellow plastic. Microplastics uptake by food similarity can also be applied to pelagic invertebrates, since they are also visual raptorial predators.

Apparently, microplastics have become widespread and ubiquitous, however information on the biological impact of this pollutant on organisms in the environment is only just emerging (Cole et al., 2011).

3.3.5 Toxicity

Gorycka (2009) mentioned that the toxicity of microplastics is connected with the intrinsic toxicity of microplastics or more specifically of their additives and monomers and their large surface area. Released, desorbed monomers of microplastics (additive-derived pollutants and sorbed pollutants e.g. POPs) can be highly carcinogenic, creating abnormities in humans, invertebrates and rodents.

Plastics are usually already enriched with different additives during the manufacturing process. Furthermore, plastics are able to act like sponges by absorbing and concentrating hydrophobic contaminants. This includes the majority of persistent organic pollutants (POPs), like polychlorinated biphenyls (PCBs), at concentrations with magnitudes several orders higher than of the surrounding environment. These contaminated plastic particles may serve as a toxin source to organisms ingesting them. Furthermore, toxic effects are suggested by compound damage as a result of the impacts related to the plastic ingestion by organisms.

3.3.6 Accumulation

Another potential threat to the environment comes from the accumulation of plastic debris. According to the UNEP report (2006) more than 70 % of the total marine litter input sinks to the sea bed in shallow as well as deep parts of the oceans. The majority of consumer plastics are neutrally buoyant, hence grains of sand trapped in their fouling matter or seams make many plastics sink to the sea floor. The vertical organic and inorganic particulate sinking fluxes may be compromised by microplastics settling in the water column. This process can slow down the gas exchange and sequestration between the overlying waters and the pore waters of the sediments. By doing so it disrupts or smothers inhabitants of the benthos by causing hypoxia or anoxia and generally altering the normal functioning of the ecosystem. It is also most likely that benthic debris is interfering with carbon cycling in waters (Mucha Torre, 2015).

Additionally, as plastic continues to fragment, the potential for it to accumulate within the circulatory fluid and phagocytic cells of an organism is likely to increase, as the smaller the microplastics are, the greater the abundance available for translocation becomes (Wright et al., 2013).

3.3.7 Physical damages

Another concern is the physical damage. The harmful effects concern the ingestion of plastics followed by the possible mechanical blockage of the intestinal tract, gastric enzyme secretion, diminished feeding stimulus, lowered steroid hormone levels, delayed ovulation and reproductive failure. This can eventually cause a serious internal injury or even death. Several studies, however, indicate that some organisms may eventually regurgitate plastic particles together with other indigestible matter, hence reducing the harmful effects. In all cases, more research needs to be done (Wright et al., 2013).

3.3.8 Effects on the food chain

Some additives and POPs are hazardous and if adsorbed to microplastics the toxicological effects on the organisms which have ingested those particles would largely depend on the concentrations of the contaminants. However, it is still not known to what extent the microplastics can transport those pollutants and whether such transport would increase the threat to the environment (Gorycka, 2009). The exposure to a wide variety of organisms is facilitated by the microplastics' distribution combined with their small size. The transmission of microplastics up the food chain is also documented. Fish and mussels are very popular as a food source for humans, so the suggestion can be made, that microplastic pollutions may also be ingested by human organisms. Researchers have already begun to speculate on the impact that such a pollutant might have on the food web.

It is possible that both humans and animals are capable of taking up microplastics in their body tissues and/or fluids, causing adverse health effects. In marine organisms like lugworms, barnacles, mussels, lobsters, petrels and seals microplastics have already been discovered (Roex et al., 2011).

3.4 Sources of microplastics

To understand the emerging environmental problems caused by microplastics it is important to identify the key pathways into habitats.

3.4.1 Cosmetics

Fig. 10: Facial gel with microplastics

Microplastics in cosmetics contribute to a silky texture or affect the fluidity and stability of products. Hollow microplastic balls are also used to encapsulate active ingredients (Leslie, 2014).

The peeling effect, with its abrasive function, is the most prominent benefit (Fig. 10). In order to achieve this effect, rather sharp-edged particles are used, not spherical ones. The most efficient peeling effect can be achieved with particles of about 420 µm in size. Particles < 60 µm are less suitable (Gouin et al., 2015). Natural materials such as pumice, oatmeal, apricot pits or walnut kernels can be replaced by plastics (Liebmann et al., 2015).

The range of relevant plastics in the cosmetic industry is very broad and includes polyethylene (PE), polypropylene (PP), polyethylene terephthalate (PET), polyamide (PA), polytetrafluoroethylene (PTFE, "teflon"), polymethylmethacrylate (PMMA), polystyrene (PS), polyurethane (PUR), and various co-polymers (Leslie, 2014).

According to a publication in 2012 of Gouin et al., 4.360 tonnes of microplastics in applications of shower gels and scrubs were used across the EU (including Norway and Switzerland). No numbers are published regarding the amount used in other cosmetic products.

The microplastic additives in personal care products enter the drainage system directly after use (shower gel, toothpaste), makeup products like facial creams, powder, mascara and many more are washed away after a temporal delay. It is hard to quantify the potential absorption into the body (lipstick color), inhalation (loose powder) or the penetration of microplastics through the mucous membranes (mascara).

Currently, there is no empirical evidence about the destination of the microplastics from cosmetics in the environment. The most likely sink is the waste water treatment plant. The distribution of microplastic particles in the air or directly into the ground is rather unlikely.

In relation to the total amount of plastic, the contribution of the microplastics in cosmetics is assessed as very low (Liebmann et al., 2015).

3.4.2 Plastics industry

For industrial processing, the intermediate plastics are typically provided as granules (pellets) or powder solutions with relatively uniform dimensions. In this form it is easy to transport and it makes the feeding of injection molding machines or extruders easier. Plastic granules may originate from material recycling. The used plastics are sorted according to species, crushed, cleaned, melted and then granulated again.

For plastics manufacturing and processing plants a loss of pellets may occur during transport or waste and transfer operations. When there are rinsing processes involved, but no provision for removing these particles installed, for instance screens or sieves, it is possible that the microplastics are directly discharged into the aquatic environment or arrives as indirect discharge in municipal sewage treatment plants.

Pellets or processing residues, which are stored on open premises, are exposed to wind and rain and can therefore be scattered to surrounding soil and water surfaces or are flushed away from impermeable surfaces into drains.

According to the study of the Environmental Agency Austria (EAA) in 2015, 10 % of the microplastics in the Danube originate from industrial sources.

Guideline values for microplastics are absent. Only indirect limitation exists in the sum of filterable solids, 30 mg/l, written in the Austrian Ordinance on Waste Water Emission in the part “emission limitations § 4”.

3.4.3 Microplastics special application

Microplastics are used as an abrasive in sandblasting for paint stripping, cleaning, roughing or the finishing of surfaces. For this treatment, hard, glasslike thermosetting plastics as well as gentler thermoplastics are used. The standard particle sizes are 2 to 0.2 mm.

Since plastic based blasting agents can be reused many times, the prevention of loss of particulates is in the interest of the user. The advantages of plastic granulate are their abrasion resistance and low dust content. Nevertheless, spreading through the air is possible, as well as there are pathways due to waste water and losses into the environment through leaky enclosures. After use, the particles can be burdened with heavy metals or other pollutants.

There is no data available on the amount and the whereabouts of the microplastics from the mentioned special applications in the environment (Liebmann et al., 2015).

3.4.4 Littering

Littering is the contamination of surfaces and space by throwing away waste. A lack of awareness among the population leads to the pollution of the environment with microplastics. However, when compared internationally, littering is a minor problem in Austria, thanks to the very well-functioning waste collection system.

In many cases it is plastic packaging, carrier bags or disposable tableware, which are discarded in public places either carelessly or carried away from crowded trash reservoirs by the wind.

Under the influence of weathering, UV radiation and mechanical stress the fragmentation of larger plastic parts into smaller ones, secondary microplastics, occurs.

The amount of plastics from littering is only a rough estimate. Initial estimates indicate a littering content of less than 0.5 % of the total amount of plastic waste.

The disintegration mechanisms of larger plastic parts to microplastics are scientifically unexplored, there is only a variety of probable factors. The specific allocation of a microplastic particles to the source of littering becomes more difficult the smaller the particles are (Liebmann et al., 2015).

3.4.5 Traffic

Tires are made of plastics, which are attributed to the category elastomers. The most important elastomers, which are used up to 70 % for the manufacturing of tires, are styrene butadiene rubber and butadiene rubber.

Tire wear is considered an important source of dust emissions in road traffic.

In three different river basins (e.g. Seine, France) tire wear was found. Influencing factors of the concentration volume are: the distance to the road, the traffic volume and the soil properties. In 2010 the total abrasion amount was estimated to be 111.420 tonnes a year in Germany. Approximately 39 % are attributed to plastics, corresponding to 43.454 t/y. In the oceans entire tires are found; the project "Save the North Sea" reported that 9 - 25 % of the total amount of waste in the North Sea is plastic.

In Austria, the value corresponds to 6.766 t/year according to the calculations of the Federal Environmental Office. Regarding the lung disposal rate, particles > 10 µm, in Austria reach up to 677 tonnes/year. 1.128 tonnes/year are obtained as suspended particulates.

About the remaining approximately 4.961 yearly tonnes of tire wear, little can be stated in detail, it is probably dust and larger tire pieces. Based on these data it can be assumed that microplastics from tire wear enter the human body through the respiratory tract. Still questionable is, whether street banquets and drainage facilities satisfy a sufficient cleaning function, how far the plastics penetrate the ground, or if the groundwater is at risk (Liebmann et al., 2015).

3.5 Ways of distribution

A sharp differentiation between source and sink is hard to define.

3.5.1 Microplastics in waste water and waste water treatment plants

Sources of microplastics in waste water are cosmetics, industrial waste water as well as littered plastics. Furthermore, microplastics occur in the waste water of toilets and washing machines, for instance nylon, polyester and acryl fibers (Andrady & Neal, 2009). During one single washing cycle a piece of clothing looses 1.900 fibers (Brown et a., 2011). A large proportion of particle and fiber carriers are restrained by the treatment plant, which means that a high, but not a 100 % cleaning performance is achieved by the waste water treatment plant (Brandsma et al., 2013).

3.5.2 Microplastics distribution via sewage sludge

Since sewage sludge is a relevant sink for microplastics, its recycling and disposal paths must be considered. In Austria in 2010, the combustion predominated with 43 %, 17 % of the sludge was used in agriculture, about 32 % were attributable to the areas of composting, landscaping, warehousing and construction, while 8 % were used for landfill. The distribution in Europe looks slightly different, more than 1/3 of the sludge is used as fertilizer, about 40 % are used as landfill and 11 % is incinerated. It is possible that microplastics leach out from landfilling sludge. The combustion process is executed far beyond 300°C, therefore it can be assumed that even high-temperature resistant specialty polymers such as polyimides or PTFE (teflon) are thermally decomposed completely and no microplastics are present in ash or flue gas. Regarding the sludge for fertilization, microplastic particles are applied to agricultural land and are exposed to wind and rain, which can lead to further dissemination. How deep microplastic items can penetrate into the soil, is highly dependent on the soil's characteristics (Liebmann et a., 2015).

3.5.3 Microplastics distribution by waste landfill

It is estimated that plastics are present in whole or in fragments in the environment, unless they have been burned. In the EU 50 % of the plastic waste is still landfilled, which amounts to about 9.6 million tonnes/year (Barnes et al., 2009). Exceptions alongside Austria are, for instance, Germany, Switzerland and Belgium, which have a landfill ban. As alternative to landfill, energy recovery and recycling have been established. The average recycling rate is approximately 28 % in Europe. It is possible that microplastics are discharged with landfill leachate, however there has not been any scientific proof to this point (Liebmann et al., 2015).

3.5.4 Distribution by air, soil and groundwater

Microplastics in the air, can be transported by wind. Particulate matter can pass through the airways to the lungs and cause numerous diseases.Microplastics in soil are conceivable due to path sludge, littered secondary microplastics or compost. The heavier the plastic particles are, the more likely their deposition in the environment becomes. Generally, soils and sediments are an effective natural barrier for the propagation of particles and pollutants into groundwater. The risk of further displacement of microplastic particles into groundwater depends on the particle properties (e.g. size, shape, surface charge density) and on factors that affect the filtering effect of soils and sediments (e.g. particle size distribution, water saturation, flow rate, etc.). Existing preferential flow paths tend to increase the risk of further transport of microplastic particles from the surface to the underground (Liebmann et a., 2015).

3.6 Separation techniques and identification of microplastics

One of the fundamental challenges prior to the identification of microplastics is their separation from the initial matrix (Tagg et al., 2015). The separation of microplastics is still under development and is afflicted with many problems.

3.6.1 Density separation

The specific density of plastic particles can vary considerably depending on the type of polymer and the manufacturing process. Density values for plastics range from 0.80 to 1.40 g/cm^3, specifically for polypropylene from 0.85 to 0.94 g/cm^3, polyethylene from 0.92 to 0.97 g/cm^3, and for polystyrene from < 0.05 to 1.00 g/cm^3 and vary from study to study. Typical densities for sand or other sediments are 2.65 g/cm^3. This difference is exploited to separate the lighter plastic particles from the heavier sediment grains by mixing a sediment sample with a saturated solution and shaking it for a certain amount of time. After mixing, the sediment is expected to rapidly settle to the bottom, while the low density particles remain in suspension or float to the surface of the solution. Subsequently, the remaining supernatant with the plastic particles is extracted for further processing (Hidalgo-Ruz et al., 2012). For this method sodium chloride (NaCl), sodium iodide (NaI), or sodium polytungstate (SPT) solutions are used as separation agents (Tagg et al., 2015).

Fig. 11: MPSS (Source: Imhof et al., 2012)

Imhof et al. (2012) improved the density separation technique with the construction of the Munich Plastic Sediment Separator (MPSS). It allowed a reliable separation of different size classes of plastic particles from sediment samples. This separator enabled a successful separation of mesoplastic particles (20 - 5 mm), large microplastic particles (5 - 1 mm), as well as small microplastic particles (< 1 mm). As separation solution, a zinc chloride solution was used.

The MPSS is divided into three major components, the sediment container, the standpipe and the dividing chamber with the integrated filter holder and the sample chamber, entirely made of stainless steel and equipped with an electromotor (Fig. 11). Indeed, a maximum fill height of 4 - 5 cm is suggested with a diameter of 30 cm, which enables an analysis of a sample volume up to 6 liters in a single run, therefore, it cannot be used for separating small sample quantities.

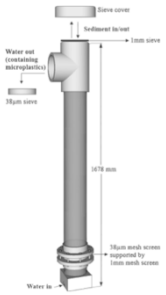

Fig. 12: Elutriation column (Source: Claessens et al., 2013)

Another method uses the principle of elutriation. Elutriation is a process that separates lighter particles from heavier ones using an upward stream of gas or liquid (Claessens et al., 2013).

According to Claessens et al. (2013), it is not possible to detect high density plastics such as polyvinylchloride (density 1.14 - 1.56 g/cm^3) or polyethylene terephthalate (density 1.32 - 1.41 g/cm^3) using a saturated salt solution, however, using high density chemicals like sodium iodide (NaI) could solve this problem, contradicting Hidalgo-Ruz et al. (2012) and Thompson et al. (2004). A device was developed to extract microplastics from sediment based on the principle of elutriation. (Fig. 12).

No universally recognized, satisfactory method for separating particles by density has been found so far.

3.6.2 Sieving

Microplastics can be separated from samples using sieves of variable mesh sizes. Materials retained in the sieve are collected and sorted, while those that pass through are usually discarded. The use of sieves with different mesh sizes allows distinguishing size categories of microplastics.

3.6.3 Filtration

The plastic particles are separated from the supernatant obtained through density separation by passing the solution through a filter with different pore sizes, usually aided by a vacuum or pressure. (Hidalgo-Ruz et al., 2012).

3.6.4 Flotation

Flotation relies on physical properties such as bulk density, particle size, shape, surface energy, and surface roughness. The most important feature is the wettability of the plastic. To facilitate the segregation between various plastic types, it is possible to modulate the wettability by altering the surface tension of the medium and/or chemical conditioning. Flotation of microplastics from sediments only enables a fragment recovery rate of 61 % (Harrison et al., 2012).

3.6.5 Visual sorting

Careful visual sorting of residues is necessary to separate the plastics from other materials or items. This is done by direct examination of the sample with the naked eye or with a microscope. Previously isolated plastic fragments can also be washed to remove other substances that adhere to their surface like sand and soil, for instance by ultrasonic cleaning in a liquid medium or with deionized water (Hidalgo-Ruz et al., 2012).

3.6.6 Identification of the chemical composition of microplastics

Infrared (IR) spectroscopy is the established technique for identifying regular shaped polymer materials and has been used extensively for identifying polymer materials larger than 100 µm (Robertson et al., 2015). This method compares the IR spectrum of an unknown plastic sample with spectra of known polymers. Micro-Fourier-transform infrared (micro-FT-IR) spectroscopy represents an ideal method for detecting microplastics in sediments, however this technique lacks a standardized operating protocol (Harrison et al., 2012). According to Hidalgo-Ruz et al. (2012) this is the most reliable method to identify the chemical composition of microplastics. Alternative spectroscopic techniques, like attenuated total reflectance (ATR) FT-IR spectroscopy, could also facilitate the identification of irregularly shaped microplastics that cannot be identified by FT-IR spectroscopy, however its main disadvantage is the high cost of the instrument used.

Another chemical analysis tool is the Raman spectroscopy, described in more detail below, which also gives information about the crystalline structure of the polymer.

Also, a differential scanning calorimeter could be used where temperature is applied simultaneously to an unknown sample and a reference material (Hidalgo-Ruz et al., 2012).

3.6.7 Separation problems and precautions

Samples can be preserved in their original form without initial sorting or they can be immediately sorted to store only the plastics from the original sample. Plastics separated from the sample should be dried and kept in a dark and temperature controlled environment at stable room temperature to reduce degradation during storage.

To avoid misidentification and underestimation of microplastics, it is necessary to standardize the plastic particle selection, following certain criteria to guarantee proper identification. This is particularly important when it is not possible to use more accurate methods, such as Fourier transform infrared spectroscopy (FT-IR). Pieces of microplastics toward the larger end of the size range (> 1 mm) can, to some extent, be visually distinguished according to the following criteria: no cellular or organic structures are visible, fibers should be equally thick throughout their entire length and particles must present clear and homogeneous colors.

New methods to separate microplastics from samples or from samples with large amounts of organic debris need to be developed to improve the efficiency of sampling programs. Molecular mapping made by focal plane array (FPA)-based imaging has recently been examined to detect microplastics by scanning the surface of filters obtained from density separation and filtration of samples. Enzymatic digestion of organic debris and other approaches could also be explored to facilitate the visual sorting of microplastics from large sample volumes.

Particles of unknown origin might also be erroneously characterized as microplastics, a problem that increases considerably with decreasing particle size.

3.7 The Danube River

The Danube is, with an average water flow of around 6.855 m³/s and a total length of 2.857 km, the second largest and second longest river in Europe after the Volga (Fig. 13). The entire river basin has a size of 817.000 km^2. It rises in the Black Forest (Germany) by the headwaters Brigach and Breg, opens into a wide delta and ends in the Black Sea (Romania). The river supplies much of central and southeastern Europe with water. It combines, as a waterway, very heterogeneous cultural and economic regions and flows through ten countries (Germany, Austria, Slovakia, Hungary, Croatia, Serbia, Bulgaria, Romania, Moldova and Ukraine), more than any other river on earth. Major tributaries of the Danube are, for instance, the Inn, Enns, Lech, Raab and Drau. It crosses the cities of Regensburg, Linz, Budapest, Bratislava, Belgrade and, of course, Vienna. The Danube is a transport and energy source and a popular attraction for tourists (Schmoller, 2014). For around 80 million people it serves as a source of drinking water. Various drinking water sources are regularly used. In the upper regions of the Danube basin as they are in Austria, springwater from the Alpine regions and/or groundwater are the main resources. Additionally, bank filtrate contributes at a low percentage to the drinking water supply in some major cities like Linz (Austria) or Ulm and Regensburg (Germany) (Storck et al., 2014).

Figure 13: Overview map of the Danube river basin (Source: ICPDR, 2005)

3.7.1 Water quality monitoring programs

For two decades, the International Association of Water Supply Companies in the Danube River Catchment Area (IAWD) has been conducting a yearly monitoring program with the general goal to determine physical, chemical, and microbiological parameters that are important for drinking water production. This monitoring program comprises most of the parameters which are relevant for drinking water surveillance under the EU Drinking Water Directive. Basic parameters like temperature, conductivity, pH value, oxygen as well as turbidity, nitrogen and phosphorus species, plus toxic elements, and heavy metals and organic surrogate parameters like TOC (total organic carbon), DOC (dissolved organic carbon), and AOX (adsorbable organic halogen compounds) are measured on a regular basis. Furthermore, the microbiological parameters *E. coli* / coliform bacteria, *Enterococci*, and *Clostridium perfringens* are examined.

The International Commission for the Protection of the Danube River (ICPDR) also implements a routine and comprehensive monitoring program with up to 116 sampling sites across the entire Danube basin every year. This approach takes less account of the requirements of drinking water production and mainly reflects the requirements of the EU Water Framework Directive.

The data and results of these monitoring programs provide a good overview on the water quality of the Danube River and its major tributaries (Storck et al., 2014).

3.8 Drinking water resources and supply

3.8.1 Supply structure in Austria and Vienna

With precipitation and the inflow of water from neighboring countries, a usable freshwater supply of 78.5 billion m^3 (large, dark blue cube) arises for Austria. Figure 14 shows that the total water demand is currently 2.2 billion m³ (medium, blue cube), with two-thirds used for agriculture and industry. From the amount of water theoretically available for usage, Austrian households use less than 1 % (small, light blue cube) (Ministerium FG, 2016).

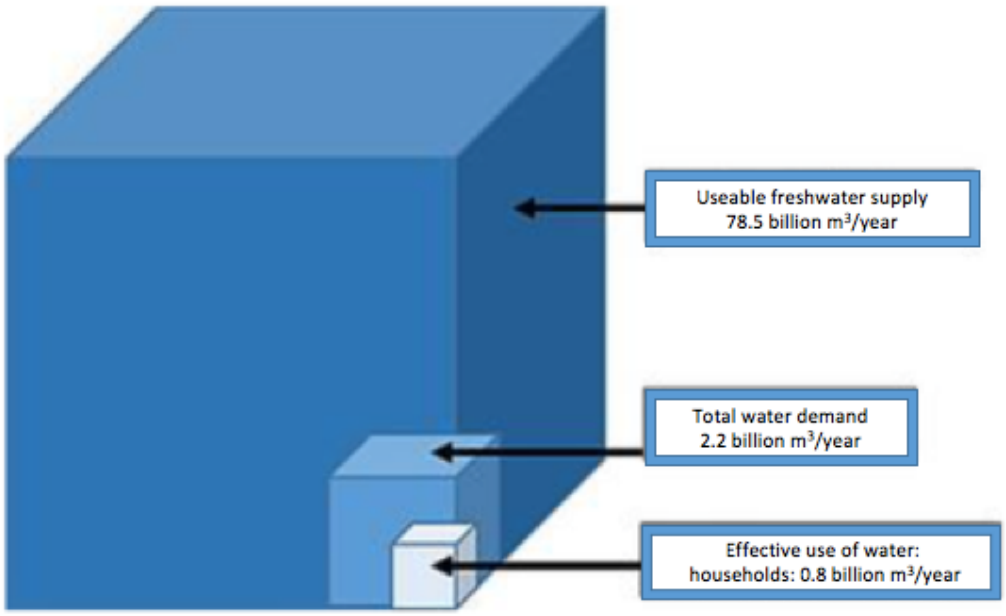

Figure 14: Usable freshwater supply for Austria (Ministerium FG, 2016)

The majority of the Austrian population lives in the provincial capitals (e.g. Vienna, Graz and Linz), where large utilities ensure the supply of drinking water from a central water supply network. The rest of the population is supplied by small water utilities or draws its drinking water from private wells or springs.

Austria owns a pipeline network of 77.000 km, 2.900 water containers, over a million household connections, 2.600 spring catchments and 1000 wells.

The average consumption per capita per day in Austria is about 135 liters.

99 % of the drinking water supply in Austria comes from groundwater, spring water and pore groundwater making up relatively equal amounts. 13 - 15 % of the Austrian households obtain their drinking water through their own private wells. Therefore, groundwater quality is of very high importance (Obersteiner et al.,2016).

The Viennese water supply is controlled by two mountain spring pipelines and various groundwater donors, which are integrated into the pipeline system in exceptional cases. About 589.000 m³ of drinking water can be routed to Vienna a day.

3.9 Bank filtration

3.9.1 Definition

The term bank filtered or artificially enriched groundwater describes groundwater which was formed as a result of anthropogenically induced infiltration of surface water (BMI, 1985). By contrast, groundwater from rainfall or from natural infiltration are formed by surface water. From a hydrological point of view, there is no difference between groundwater and bank filtrate. For practical applications, such as choosing the treatment process, the origin of the groundwater usually plays a minor role.

Bank filtration and artificial groundwater recharge are a commonly used method for drinking water extraction. In the underground passage a variety of substances are reduced or eliminated requiring a minimal amount of maintenance, energy and raw materials.

Surface water is infiltrated into the aquifer through the wells' potential gradient from surface water to groundwater (Ziegler, 2001). The positive gradient between the water level of the surface water and groundwater is produced naturally by floods or through artificial lowering of the water table by pumping water.

The possible rate of bank filtration is limited by the entrance resistance at the riverbank and sole, the permeability of the soil and the potential gradient. While in unsaturated groundwater the pores of the riverbank are kept free due to the high water pressure, clogging can occur, as fine materials may deposit in the pores or penetrate them. This is dependent upon the nature of the surface water, as well as microbial and chemical processes. Clogging can be reduced by floods, due to their exerted force. The water quality is changed while moving under ground, where suspended solids and some solutes are retained or degraded while the concentrations of persistent substances, such as chloride, mostly remain. The water recovered is a mix of natural groundwater and bank filtrate, whose ratio of either is dependent on the fluctuations of the water table and the amount of withdrawal quantity.

In Austria, to ensure microbiological quality, a residence time of at least 60 days underground or in the groundwater is relevant, since groundwater-foreign germs are normally inactive for this amount of time. This is based on the interaction of numerous limitation mechanisms in the soil. The distance of the extraction wells for surface water should be selected accordingly (Schmoller, 2014).

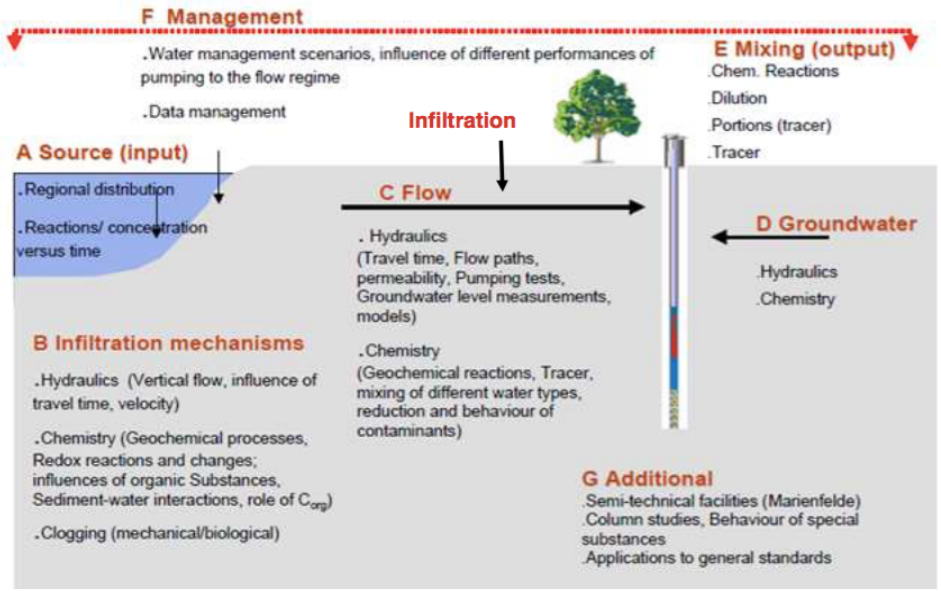

Figure 15: Bank filtration cleaning processes (Fritz, 2003; cit. Schmoller, 2014)

In quantitative terms, the use of surface water or bank filtrated water, depending on the size of the river basin and the climatic conditions, offers a uniform water supply option for many people. While during low water periods only groundwater is pumped into the filtration wells, the ratio of bank filtrated water may rapidly increase during shock loads. This improves the drinking water's quality through dilution or concentration equalization processes.

Despite the extremely complex effects of cleaning mechanisms and the relevant physical, chemical and biological conditions, this method can also be used in countries where technically sophisticated care and maintenance of water collection and treatment plants is not possible. Therefore, bank filtration is a cheap way to produce drinking water for all major cities along rivers in many developing countries. Water from well protected and well filtered aquifers can be used from a sanitary, microbiological point of view even without elaborate preparation steps for the drinking water supply (Schmoller, 2014).

3.9.2 Cleaning processes in the soil passages

The cleaning of the surface water occurs during the infiltration of the water through the different soil layers and materials. Four mechanisms are in action:

- Mechanical filtration
- Sedimentation
- Adsorption
- Microbiological degradation

While the two processes, mechanical filtration and sedimentation, are important cleaning mechanisms for particle removal, adsorptive forces play an important role in the deposition of particulate pollutants and microorganisms. Dissolved organic substances and small particulate matter are reduced in dependence of the consumption of electron acceptors (oxygen, manganese, iron, nitrate and sulfate) and the possible change in the redox conditions, as well as the pH value, especially through the activity of microorganisms. It has been proven, that biological activity decreases with increasing depth. The soil passage works as a filter for particles, colloids and pathogenic germs, removes ionic and hydrophobic organic substances by precipitation and sorption, removes ammonium and degradable organic substances in water by biological oxidation and inactivates germs and viruses through biological processes. The passage does not remove polar, poorly biodegradable organics and conservative ions. Mainly redox conditions, quality of the infiltrating water and the natural groundwater on site, the length of the flow path, the duration of the flow and soil and climatic conditions have an impact on the removal rates or on the cleaning performance (Ziegler, 2001).

Surface waters practically always contain particulate matter to which inorganic and organic contaminants adhere, as well as particulate microbiological contaminants (bacteria, viruses, protozoa and parasites). These contaminations should be removed by appropriate deposition. Significant influences here are the electrochemical properties of the particles and the pore water velocity. The transport distances of particles in the substrate can be significantly increased by the absorption of humic substances.

Particulate raw water contents are transported up to the orders of 50 - 500 nm with groundwater, where the particle retention in the soil passage occurs as surface as well as deep bed filtration (Ziegler, 2001). The underground passage readjusted to the bank filtration has a high deposition potential in comparison to suspended or colloidal stable particles. Regardless of solids exposure the measured turbidity levels in bank filtrate are mostly less than 0.1 FNU.

- **Microorganism**

Microorganisms (bacteria, protozoa, algae, etc.) and viruses are removed through the ground by filtration and sorption and biological inactivation. For viruses and bacteria especially, the length of the route through the ground is crucial. Bacteria are retained most effectively in the top few centimeters by adsorption, in which microflora and microfauna are extremely active. Also in deeper layers and in the underground passage, the sorption of substances to organic and inorganic surfaces as well as mechanical filtration plays an important role. With increasing filter grain size, the transport range of bacteria increases. Viruses are mainly removed via sorption through hydrophobic surfaces, whereupon, for the coated polysaccharide biofilms, the top infiltration zone has a significant impact (Ziegler, 2001).

- **Nutrients**

The nutrients ammonium, nitrate and phosphate primarily occur in waters with high levels of municipal waste water in intensively agriculturally used regions, but are usually not a problem for drinking water extraction through bank filtration (Ziegler, 2001).

- **Heavy metals and metal cations**

Iron, manganese and various heavy metals pass through the ground and are removed as a function of pH primarily by sorption. The elimination of heavy metals should not happen solely by natural or semi-natural processes (slow sand filters, soil passage), but in combination with ion exchange, precipitation, filtration and possibly complexation, which makes temporary, strong support possible (Schmoller, 2014).

- **Organic water contents**

Dissolved organic substances in water are composed of natural organic substances (humic substances, low molecular weight acids, proteins and carbohydrates) and various individual substances (such as pesticides, industrial chemicals, surfactants, drugs and disinfection by-products). They can be reduced through bank filtration in addition to mixing with groundwater mainly through biological and weak sorptive processes. The carbon content of all dissolved organic substances in water is described by the DOC parameters and captures the majority of the natural substances in water that can pass through a 0.45 µm filter. Depending on the composition of the infiltrating water, different proportions of DOC are implemented through the activity of soil microorganisms (Ziegler, 2001).

The concentration of degradable organic substances in the infiltrate is usually low and therefore a flow distance of less than 1 m is sufficient for their removal.

The organic carbon content only moderately decreases via anaerobic infiltration during bank filtration and underground passages, while, when using aerobic slow sand filtration and underground passage elimination, rates of about 70 % are possible due to higher microbial activity. First the metabolization and then the mineralization of organic compounds occur through the microbial metabolism.

Organic water pollutants can also be removed by sorption, depending on the characteristics of the materials and the surfaces of the base material. Aquifers and groundwater recharge basins consist of mineral oxides, clays and small amounts of organic matter, mostly quartz (sand, gravel). Nonpolar organic substances in the water, such as polycyclic aromatic hydrocarbons, which are used in the production of plastic, usually exist in low concentrations in the infiltrate and are eliminated in the infiltration zone where the organic fraction is highest. At the solid surface they are then microbially degraded (Ziegler, 2001).

Depending on the initial concentration, the prevailing redox conditions and other environmental factors may eliminate anthropogenic organic individual substances (pharmaceuticals, household and industrial chemicals, pesticides). Especially highly chlorinated and highly substituted aromatic hydrocarbons are difficult to degrade. Synthetic organic complex agents, aromatic sulfonates, aliphatic amines, pharmaceutics, alkylphenols/bisphenol A and pesticides are problematic organic substance groups summarized together that cannot, or only insufficiently, be removed during bank filtration (Schmoller, 2014).

3.9.3 Drinking water treatment processes according to the bank filtration

Due to the increase in anthropogenic water pollution, tightening of legal requirements, as well as the higher quality requirements of consumers, more treatment steps have been introduced in the past.

Combined processing operations are generally required, if the desired reduction or elimination rates of certain substances can only be achieved through several stages, each with limited effectiveness and if the combined phases appear more economical. By suitably arranging the steps, the adverse effects of treatment stages can be reduced or even prevented. Despite different raw water qualities, the same typical procedures are used.

The treatment of the bank filtrate is often carried out according to the "Düsseldorf Process" which is divided into the following steps:

- Bank filtration
- Ozonation
- Rapid filtration with demanganisation
- Activated carbon filter
- Deacidification
- Disinfection with chloride dioxide (ClO_2)

With favorable hydrogeological conditions, the underground passage with long residence times may even constitute an efficient, multiple stages process to eliminate microbial hazards and may also replace multiple technical stages.

Considering the existing water quality, system size, cost situation and the desired composition of drinking water, appropriate mechanisms are to be applied (Schmoller, 2014).

In this thesis, the quality of the bank filtrate is so high that either a UV or an additional chloride dioxide (ClO_2) disinfection is sufficient.

The disinfecting effect of UV radiation is based on the damage on the genes of microorganisms and viruses, therefore, their cell numbers cannot increase. A high disinfection efficiency is also recognizable towards parasites. To prevent a reactivation of the cells, a reduction equivalent dose of at least 400 J/m^2, based on a wavelength of 253.7 nm (100 mm thickness), is applied in Austria. When passing through the radiation zone of the radiation chamber, each particle is irradiated differently depending on location and duration. The disinfection efficacy can only be measured by the mortality rate of added microorganisms with known UV sensitivity.

In the case of disinfection with chloride dioxide (ClO_2), the addition is at least 0.2 mg/l ClO_2 and at most 0.4 mg/l ClO_2 in Austria. After a minimum reaction time of 15 minutes and sufficient mixing, a residual concentration of at least 0.05 mg/l ClO_2 must be detectable. For the undesired by-product, chlorite, formed in this process, the maximum permissible concentration when delivered to the consumer is 0.2 mg/l. Chloride dioxide is used to prevent the formation of trihalomethane, to reduce the development of odor and taste and due to better efficiency at higher pH values. Freely suspended microorganisms including bacterial and viral pathogens can be killed or inactivated. Parasites cannot be adequately eliminated, even at the maximum permissible dose of chloride dioxide (Schmoller, 2014).

3.10 Raman spectroscopy

Spectroscopy is the study of the interaction between matter and radiated energy, especially light. In particular, the electric component of the electromagnetic light interacts with electrons in a crystal and, depending upon the structure of the crystal, responds with a new signal that can be measured and interpreted. Raman spectroscopy, named after Sir C. V. Raman, is a spectroscopic technique based on inelastic scattering or so called Raman scattering of monochromatic light, usually from a laser in the visible, near infrared, or near ultraviolet range. Inelastic scattering occurs during an interaction, in which the incident light has a different wavelength than the dispersed light. That means that the frequency of photons in monochromatic light changes upon interaction with a sample. Photons of the laser's light are absorbed by the sample and then reemitted. The frequency of the reemitted photons is shifted up- or downwards in comparison to the original monochromatic frequency, which is called the Raman effect. This shift provides information about vibrational, rotational and other low frequency transitions in molecules. The Raman effect is based on molecular deformations in electric fields determined by molecular polarizability. The laser beam can be considered an oscillating electromagnetic wave with an electrical vector. Upon interaction with the sample it induces the electric dipole moment which deforms the molecules. Due to this periodical deformation, molecules start vibrating with characteristic frequencies. The amplitude of this vibration is called nuclear displacement. In other words, monochromatic laser light with a typical frequency excites molecules and transforms them into oscillating dipoles. Such oscillating dipoles emit light of three different frequencies when (Fig. 16):

- A molecule which is not in a Raman-active state absorbs a photon with a specific wavelength. The excited molecule returns back to the same basic vibrational state and emits light with the same frequency as the excitation source. This type of interaction is called an elastic Rayleigh scattering.
- A photon is absorbed by a Raman-active molecule which at the time of interaction is in the basic vibrational state. Part of the photon's energy is transferred to the Raman-active mode and the resulting frequency of scattered light is reduced. This Raman frequency is called Stokes Raman scattering.
- The third variation, the anti-Stokes Raman scattering, happens when a photon is absorbed by a Raman-active molecule, which, at the time of interaction, is already in the excited vibrational state. The excessive energy of the excited Raman-active mode is released, the molecule returns to the basic vibrational state and the resulting frequency of scattered light increases (B& W, 2016).

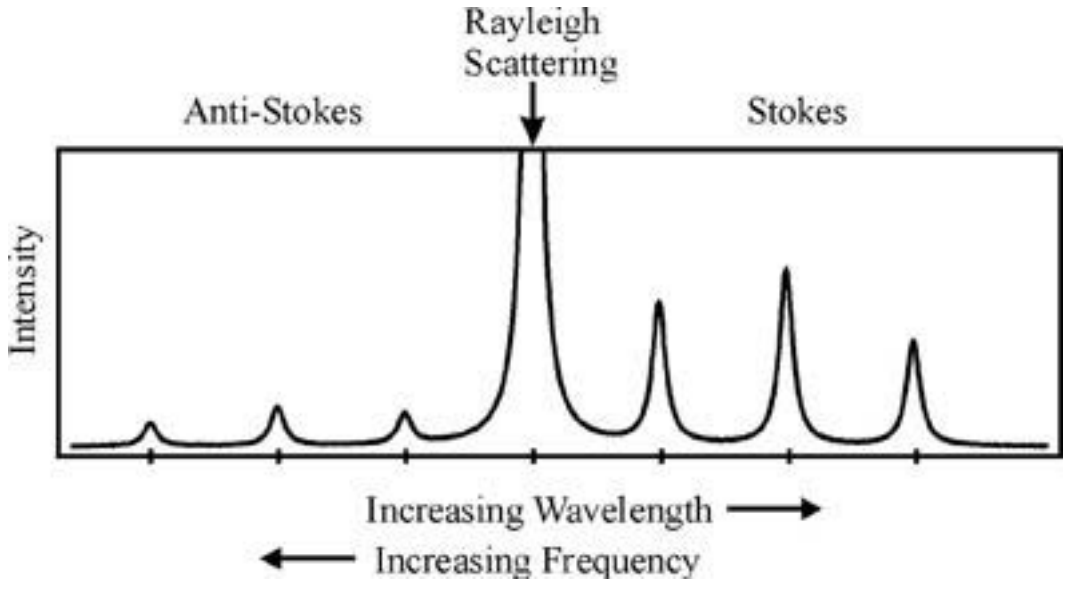

Figure 16: Rayleigh, Stokes, anti-Stokes Raman scattering (Source: UA Geoscience, 2016)

Unfortunately, more than 99 % of all photons undergo elastic Rayleigh scattering, however this type of signal is useless for the practical purposes of molecular characterization. Only about 0.001 % of the incident light produces an inelastic Raman signal. Spontaneous Raman scattering is very weak and special measures should be taken to distinguish it from the predominant Rayleigh scattering. Instruments such as notch filters, tunable filters, laser stop apertures and double and triple spectrometric systems are used to reduce Rayleigh scattering and obtain high-quality Raman spectra.

This a non-destructive technique and needs little sample preparation. In fact, Raman analysis can be conducted directly via glass, plastic bags, cuvettes, and other transparent containers. It is a highly selective, quantitatively as well as qualitatively usable technique, which can easily differentiate very similar molecules in known and unknown materials (B&W, 2016).

This is the currently used analysis method for the characterization of polymers and plastics. Earlier, these materials were examined using burn tests, chromatographic separation and wet chemical techniques, which tend to damage the sample (B&W, 2016).

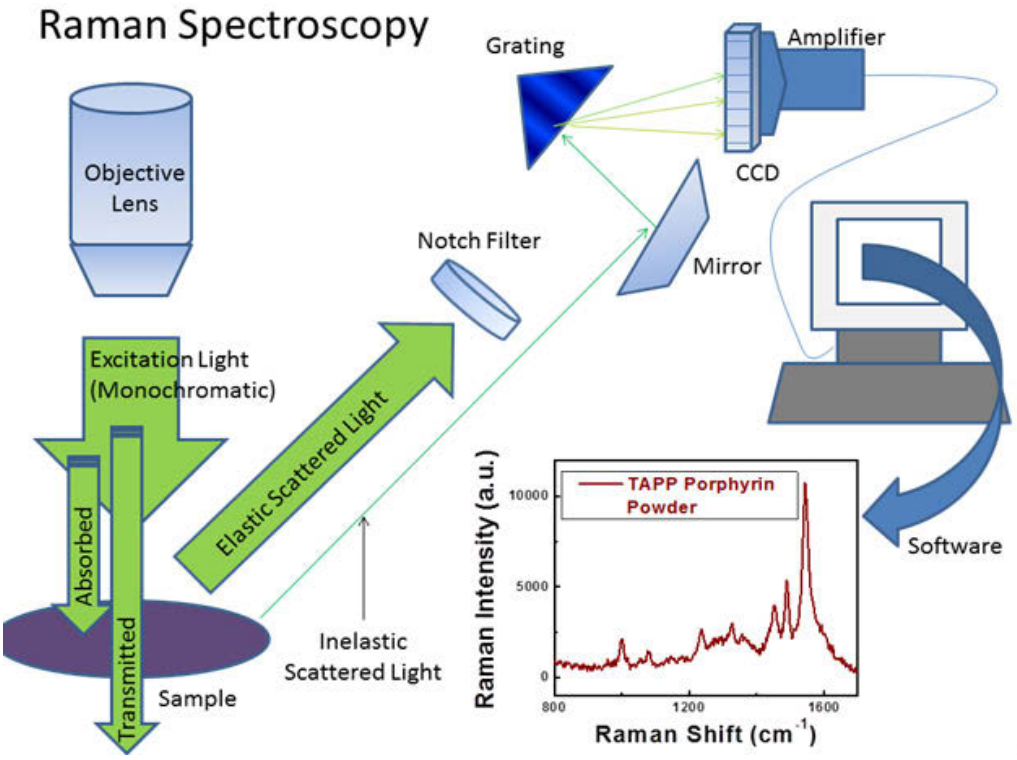

Figure 17: Scheme of the Raman spectroscopy (Source: American Chemical Society, 2011)

3.10.1 Raman spectrum

The result of the inelastic scattered light is the Raman spectrum that is used for further analysis. The horizontal axis of the Raman spectrum indicates the difference between the input laser light and scattering light energy, while the vertical axis indicates the intensity of the Raman scattering light (Fig. 18).

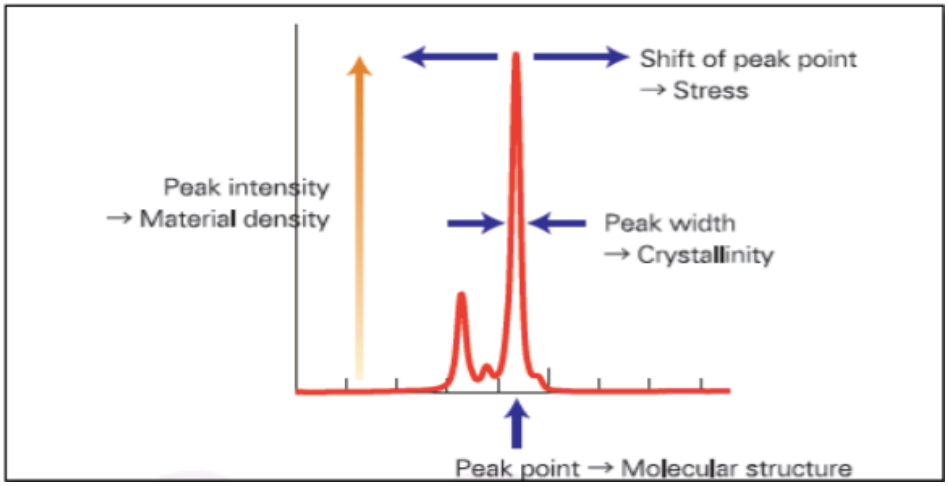

Figure 18: Scheme of a Raman spectrum (Source: Left coast instruments, 2011)

Different types of information can be obtained from a Raman spectrum. Every molecule has its own unique Raman spectrum, which can be correlated to a molecular fingerprint. Through this technique, slight differences in the matrices can be utilized for instance for analysis which otherwise would not be possible using conventional univariate techniques. Figure 19 illustrates the basic principles of a Raman analyzer, along with Raman spectra of four similar molecules. These Raman spectra can be clearly distinguished (B&W, 2016).

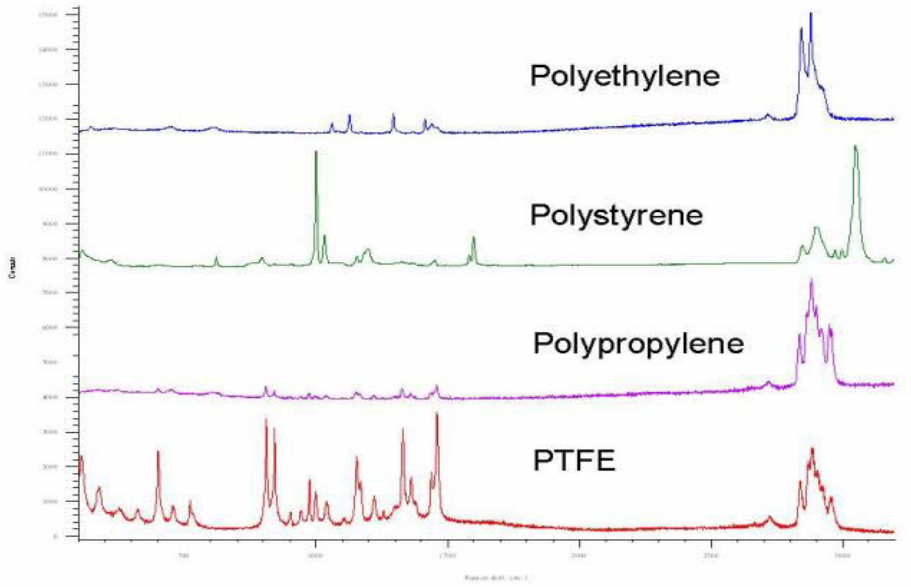

Figure 19: Raman spectra of different plastic types (Source: Ceriumlabs, 2008)

3.10.2 Raman application

Raman is used in a huge variety of applications. It is commonly used in chemistry, since vibrational information is specific to the chemical bonds and symmetry of molecules. Therefore, it provides a fingerprint by which the molecule can be identified. The fingerprint region of organic molecules is in the range of 500 - 2000 cm^{-1}. In the pharmaceutical industry, one major application for Raman is the detection and substantiation of incoming raw materials. The technique is employed for the analysis of liquids, tablets, and gel caps. Raman is also an essential tool in counterfeit-drug analysis. It can be used to identify explosives such as RDX, PETN, TNT and the binding agents inside explosive materials. The technique is also effective for identifying toxic solvents in biological warfare agents and for studying other potentially harmful agents (B& W, 2016).

4 Material and methods

The locations, materials and methods used in the experimental part of the thesis are described below and, for this research, the pictures and graphics were produced by the author.

All experiments were executed from October 20, 2015 to October 14, 2016.

4.1 Locations

The experiments and the sample taking were carried out in the laboratory of the Institute of Sanitary Engineering and Water Pollution Control (SIG), the Imaging Center of the University of Natural Resources and Life Sciences as well as the facilities of the water supply system, Vienna:

The five sampling sites are as follows:

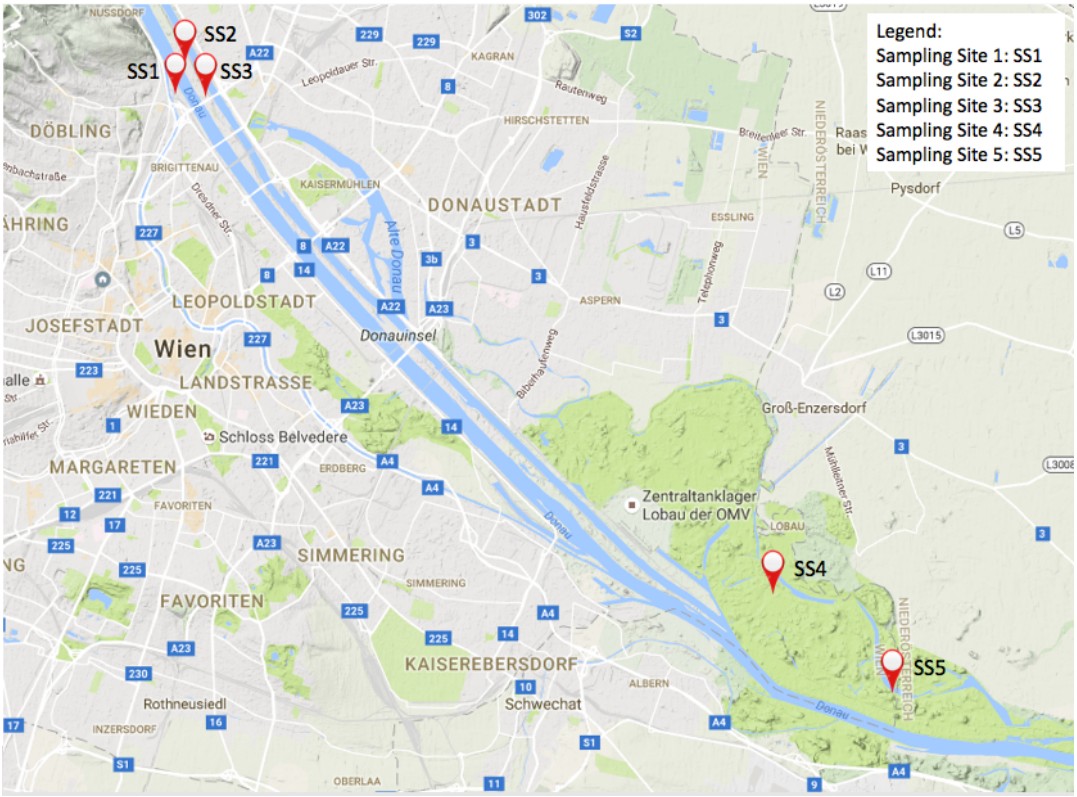

Figure 20: A cartographic representation of the sampling sites visited in this study

Sampling site 1 for instance is used to extract water from the banks of the Danube. The recovered water is fed into network operation 1 as raw water and/or into network operation 2 after disinfection by UV irradiation, or into the transport network as emergency water supply in emergency operations or is discharged into the Danube in operational condition.

The site is composed of: seven dug wells with a diameter of 2 m and a depth of approximately 14 m and two tube wells with a filter diameter of 780 mm and a depth of around 15 m, furthermore two collection wells exist. The collection well one is a concrete well with cast iron parts and inlet openings, a diameter of 5 m and a depth of circa 14.5 m. The collection well two is a concrete well with windows with a diameter of 5.5 and a depth of approximately 15.5 m.

All wells are equipped with pumps with a capacity of 70 l/s.

4.2 Methodology of the set-up and pretests of the Stainless Steel Cascade (SSC)

These tests were carried out to find the proper mesh sizes for the in situ sampling as well as to identify the problems of this method. The materials used were:

- Two stainless steel holders with a diameter of 10 cm and one with 14 cm by Satorius
- Nylon filters with pore sizes of 500 µm, 300 µm, 150 µm, 100 µm, 50 µm, 30 µm, 15 µm, 5 µm from Fa. Lactan
- Cellulose nitrate filters with a mesh size of 1.2 µm
- Four silicone tubes with Ø 12mm
- Classical PVC tube ¾”
- One water meter

4.2.1 Set-up of the Stainless Steel Cascade

This cascade is the basic referential method for the in situ sampling. The three stainless steel filter holders form the main units. The holders are connected by four silicon tubes; the inflow as well as the outflow tubes are conventional ¾” PVC tubes and the outflow hose features a water meter (Fig. 21). When the holders are equipped with filters of descending pore sizes (e.g. 15 µm, 5 µm and 1.2 µm) the water can flow in cascades through the units and a separation of particles according to size is achieved.

Figure 21: Set-up of the stainless steel cascade

4.2.2 Pretests of the Stainless Steel Cascade

The pretesting of the cascade as well as finding the suitable pore sizes is very important in order to work in the field without any problems.

The different materials got tested using Danube water, sewage sludge and tap water. The tested materials of varying pore sizes were: cellulose acetate, cellulose nitrate, teflon, high quality filter papers, microfiber filters of 100 % borosilicate and nylon filters.

4.3 Methodology of the set-up and pretests of the Blue Filter Cascade

This method has been developed to find a comparison to the SSC. The system got its name due to the blue closures.

The pretests were conducted to verify the design and the fitness for use of the cascade. For this, the following materials were utilized:

- A hot glue gun
- Cable ties
- Nylon filters with different mesh sizes
- Five tecnoplastic cartridge filter 5”
- Five tecnoplastic stainless steel mesh cartridge with a pore size of 100 µm
- A water meter
- Classical PVC tube ¾
- Sealing tape
- Connecting threads

4.3.1 Set-up of the Blue Filter Cascade (BFC)

The basis of the cascade are five filter units 5” with a maximum working pressure of 6 bar and a maximum operating temperature of 35°C and five stainless steel mesh cartridges with a mesh size of 100 µm (Fig. 22). The five filter units were connected with four connecting threads and, to achieve optimal sealing, the connecting threads were equipped with sealing tape. To create a cascade, the nylon filters, with different descending pore sizes, were fixed with a hot glue gun and thin cable ties around the cartridges. This method made it easy to replace the nylon filters after each sampling. Same as the reference method, the stainless steel cascade, the in- and outflow tubes were classical ¾” PVC tubes and the outflow tube was equipped with a water meter.

Figure 22: Set up of the Blue Filter Cascade (BFC)

4.3.2 Pretests of the Blue Filter Cascade

To examine the fitness for use of the cascade, several varying numbers of units were put together and differently polluted waters, Danube water, sewage sludge mixed with water and tap water were filtered. So as to find the optimum cascade to analyze water pollution, descending pore size combinations were varied. Only nylon was used as a filter material for the Blue Filter Cascade.

4.4 Methodology of the in situ sampling

The sampling was executed at five different sampling sites close to the Danube, which were selected strategically in order to be able to estimate the entry of the microplastics in the bank filtration passage. The sampling was carried out in October 2015 and September 2016 with the Stainless Steel Cascade (SSC) and the Blue Filter Cascade (BFC) to have approximately the same weather as well as hydrological conditions in the Danube. A further sampling in 2016 was necessary to confirm and enhance the results of 2015.

For the first measurements in 2015 only the SSC was used. Due to its easy handling and the simple exchangeability of the filters, samples could be drawn with the greatest possible accuracy. The first test measurements were carried out in the laboratory of the Institute of Sanitary Engineering and Water Pollution Control, therefore the application of the method in the field came with almost no surprises. The second cascade, the Blue Filter Cascade, was developed in 2016 to allow a comparison with the SSC.

Figure 23: On-site installation of the SSC and BFC

Sampled were bank filtrate at sampling site 3 and groundwater at sites 4 and 5. At site 1 bank filtrate and Danube water and at site 2 only Danube water were tested during operation (Tab. 1).

In order to simulate a worst case scenario for bank filtrate contamination, the water was gradually raised after a three-week standstill at sampling site one (SS 1) on September 06, 2016. After commissioning a discharge of 125 l/s at 12:30 the discharge was increased twice. At 13:20 the discharge was increased to 250 l/s followed by another at 14:15 at 380 l/s. However, the system shut down after the last increase and there was a restart at 14:26.

Table 1: Sampling sites, sampling dates, water types, and filter cascade types used

Sampling Site	Date	Water Type	Filter Cascade Type
SS 1	20.10.15	Bank filtrate	SSC
SS 2	28.10.15	Danube water	SSC
SS 3	28.10.15	Bank filtrate	SSC
SS 4	30.10.15	Groundwater	SSC
SS 5	30.10.15	Groundwater	SSC
SS 3	01.09.16	Bank filtrate	SSC & BFC
SS 1	06.09.16	Bank filtrate	SSC & BFC
SS 1	14.09.16	Danube water	SSC & BFC
SS 2	14.09.16	Danube water	SSC & BFC

At all sampling sites, the pore sizes of the filters were adapted to the water pollution. Filters with a pore size of 15 µm, 5 µm, 1.2 µm were selected at sampling sites where the contamination was low, and filters > 15 µm were chosen for Danube water sampling. Both cascades were set up as shown in Figure 23 and simply connected to the water connections of the sampling sites, then the filtration could be started. After each sampling process the filters were changed, stored in clean glass petri dishes, and new ones were put in for further sampling. The goal was to separate the bank filtrate, groundwater and Danube water contaminants by size fractions and to analyze them by Raman spectrometry in order to find microplastics.

Not only sampling with the cascades was executed, also several liters of the sampled water were bottled for the particle counting with the Abakus as well as for turbidity measurements with the turbidimeter, Turbiquant.

All samples were taken to the laboratory and stored in the fridge at 4°C.

Table 2 shows all applications used in this thesis.

Table 2: Summary of the analysis tools used at each sampling site

Sampling Site	Date	Applications						
		S::can	Sigrist WTM 500	Turbiquant 1100 IR	Klotz	Abakus	Zeiss Microscope	Raman
SS 1	20.10.15	x				x	x	x
SS 2	28.10.15	x				x	x	x
SS 3	28.10.15	x				x	x	x
SS 4	30.10.15	x				x	x	x
SS 5	30.10.15	x				x	x	x
SS 3	01.09.16	x	x	x	x	x	x	x
SS 1	06.09.16	x		x	x	x	x	x
SS 1	14.09.16	x		x		x	x	x
SS 2	14.09.16	x		x		x	x	x

4.5 S::can measurements

The turbidity, conductivity, temperature, permeability, NO_3-N (Nitrate), TOC, and DOC were measured continuously by the S::can micro::station at all sampling sites.

Both national laws and an increasing number of supranational recommendations are prescribing emission limits for reasons of public health. These limits may not be exceeded, and must be controlled, in order to not place consumers of drinking water in danger. The quality of raw and drinking water must constantly be monitored. The S::can uses the absorption of light as measuring principle. Yet, not only one or two wavelengths are measured, a continuous optical spectrum reaching from low ultraviolet to visible light is measured. The result of the measurement is the so called fingerprint (Fig. 24) (S::can, 2013).

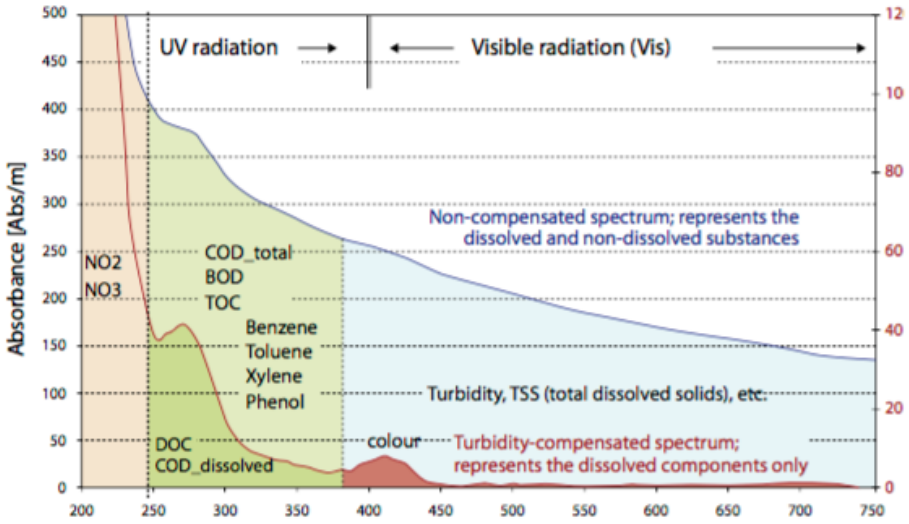

Figure 24: Absorption spectra of for instance COD, DOC and BOD (S::can, 2013)

The fully modular micro::station combines S::can instruments to a compact and versatile system (Fig. 25). It is a complete solution, it only has to be connected to a water supply and discharge in order to receive a variety of immediately available information and parameters. The components spectro::lyser, S::can probes and controller are assembled on a compact panel. It is designed for online monitoring of water quality parameters in clean media, such as drinking water. The spectro::lyser measuring principle is based on a UV-Vis spectrometry over its entire range (190 - 720 nm or 190 - 390 nm) (S::can, 2016).

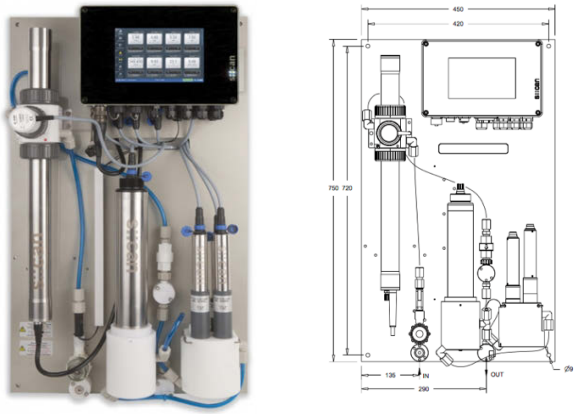

Figure 25: S::can micro::station (Source: S::can, 2016)

4.6 Particle counting and turbidity measurements

In addition to the sampling, particle counting and turbidity measurements were carried out on-site as well as in the laboratory.

At two sites, SS 1 and SS 3, the particle count was performed by a mobile online two-column particle counter from Fa. Klotz (Fig. 26).

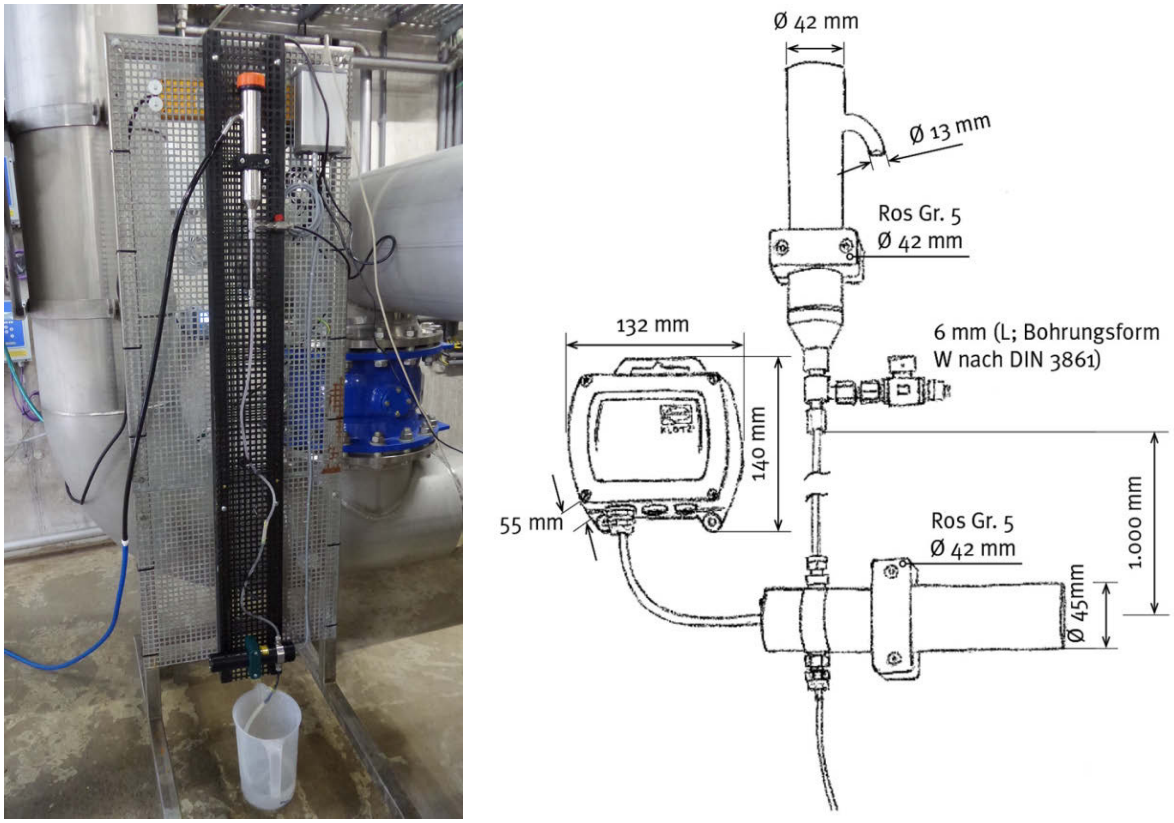

Figure 26: On-site installation of the mobile particle counter from Fa. Klotz and a drawn scheme (Source: Klotz)

The two-column counter CSS-Fluid, with the sensor 255, is housed in a splash-proof die-cast housing. The liquid is supplied by means of the riser pipe or a pressure holding valve. Alternatively, a liquid pump can be used in pressureless systems. All parameters, particle size, time and limit values can be parameterized using a laptop. The particle number can be transferred to an evaluation system via the 4 - 20 mA current interface.

The software allows single and multiple measurements. The evaluation can be carried out cumulatively or distributively, however in this case cumulative evaluation was chosen due to the particle numbers. The measured particle sizes were between 1 - 100 µm and the amount of particles per liter were evaluated.

After the on-site sampling, further particle counting was performed in the laboratory with the Abakus particle counter from Fa. Klotz (Fig. 27). The particle counting system “Abakus mobile fluid” was used to count particles of a magnitude of 1 - 100 µm. The unit is equipped with the LDS 30/30 sensor. The samples can be conveyed from the beaker, equipped with a magnetic stirring bar and placed on a magnetic stirrer, with the self-priming pump DPS-1. With the evaluation software "Log and Show", the measured results can be exported and further processed via the built-in interface on the PC (Klotz). It is possible to execute a single measurement or continuous measurements with different cycle times. A continuous measurement was chosen to identify trends. For each sample 10 measurements of 10 ml were performed twice to calculate the mean value, the standard deviation and relative standard deviation later on. The presentation of the results is either cumulative or distributive; the cumulative presentation was chosen.

The Abakus is a laser based particle counter. The method of static laser diffraction analysis for the measurement of particle size distributions is not a method in which individual particles are counted. The distribution of particle sizes is performed by size classes (e.g. 1 - 2 µm followed by 2 - 5 µm and up to > 100 µm). In the case of the static laser diffraction analysis, the diffraction pattern of a particle collective is measured. Through the application of mathematical methods, the particle size distribution can be calculated from the detected diffraction pattern. It is an optical method in which the scattered light emerging from the particle collective at a specific spatial angle is detected. Large particles bend the laser light with high, small particles with lower intensity. The recorded diffraction and scattered light patterns were evaluated.

Bank filtrate and Danube water samples were processed. It was not necessary for the bank filtrate samples to prepare dilution series however for the Danube water samples it was, due to the fact that the Abakus can only operate a maximum of 120000 particles per ml and the particle numbers in the Danube water samples were substantially higher. For the dilution a 1000 ml glass flask and an automatic 100 ml pipette were used. A dilution of 1:10 was made. The sample bottles were shaken beforehand to ensure a uniform distribution of the particles and then 100 ml of the sample were filled into 900 ml of ultra-pure water.

The end result of the particle count is the distribution of the particles in size classes.

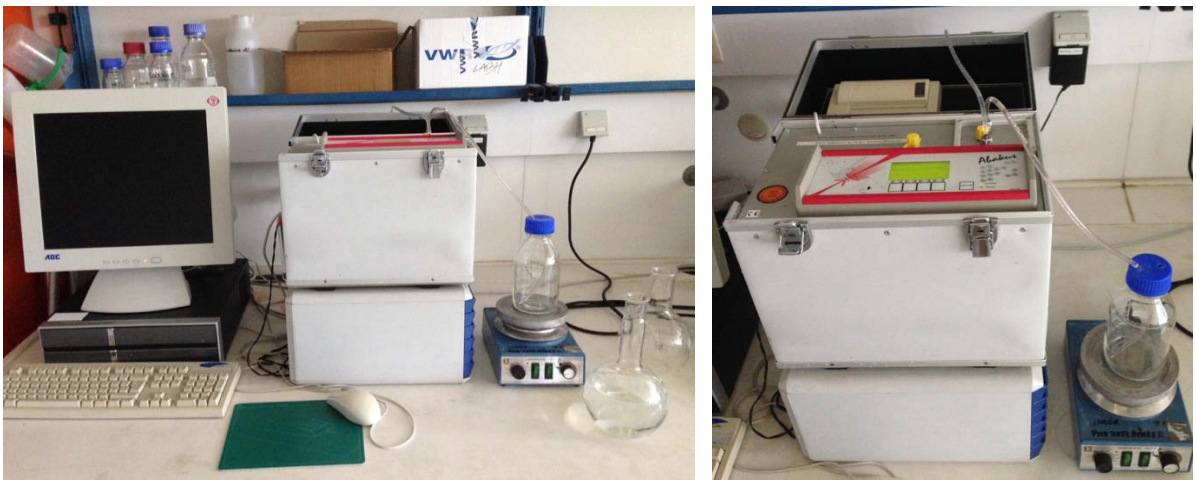

Figure 27: Abakus mobile fluid from Fa. Klotz

4.7 Turbidity measurements

Fig. 28: Sigrist WTM 500

A turbidity measurement works as follows: Particles scatter light in many directions. The light scattered at 90° is measured. The clearer a liquid is, the lower the turbidity. The units of turbidity are NTU (nephelometric turbidity unit) or FNU (formazin nephelometric unit).

At one site (SS 1) a mobile turbidimeter, Sigrist WTM 500, was installed (Fig. 28). The Sigrist WTM 500 is an online turbidimeter that provides non-contact measurement of the 90° scattered light in a free-falling water stream. Automatic adjustment using a fixed internal reference standard enhances measurement reliability and minimizes the need for cleaning and calibration. The WTM 500 has a nominal range of 0 to 500 formazin nephelometric units (FNUs) in eight selectable scale ranges, with a maximum resolution of 0.001 FNU (Sigrist).

Fig. 29: Turbidimeter- Turbiquant

Furthermore, a portable turbidimeter, Turbiquant 1100 IR from Merck, was used for all taken samples (Fig. 29). For each sample, 3 measurements were performed and the mean was calculated. The liquid sample was transferred into the glass cuvette, placed in the turbidimeter and slowly rotated clockwise. The cuvette was turned back to the position of the smallest measured value, and the device determined the turbid value.

The measurement of the turbidity was executed to compare it to the particle counter's results. The degree of the turbidity of water is a measure of water contamination. A liquid is turbid when it contains undissolved suspended particles. Therefore, the turbidity correlates with the number of particles.

4.8 Microscopic sorting of the particle fractions

The filters were examined with an optical microscope from Zeiss and a ten-fold magnification (Fig. 30). The microscope and the axiocam were connected directly to the computer on which the images were processed with the AxioVision imaging software. AxioVision is a high-performance, flexible image processing and analysis system for modern microscopy. The recording, archiving and processing of the images can be done according to personal requirements (Zeiss, 2009).

The filters were placed under the microscope and live images of the filters were transferred directly to the computer. The particles were measured and classified into size fractions.

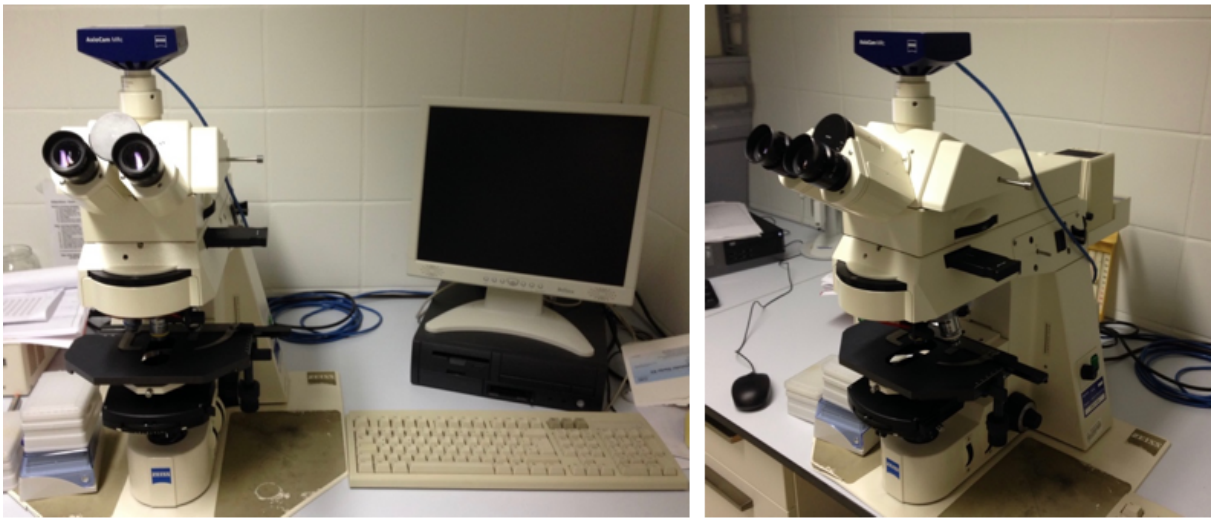

Figure 30: Zeiss microscope with an axiocam

4.9 Methodology of the density separation technique

Fig. 31: Saturated NaCl solution

The main product used for this separation technique was a saturated solution of sodium chloride (Fig. 31). 86 g of sodium chloride were dissolved in 1000 ml of ultra-pure water at a temperature of about 20°C. Thompson et al. (2004), Hidalgo-Ruz et al. (2012) and Hohenblum et al. (2015) successfully used this method in their studies. However, the values of the saturated solution density vary between studies. The density of 1.1640 g/cm^3 was suggested by Lide (2004) and a density of 1.1998 g/cm^3 was published by Thurmond et al. (1984).

The change in density allowed the light microplastics to float in the supernatant or on the surface, therefore collection was possible but complicated.

A solution made of the filter residues, from sampling at SS 2 with the BFC on September 14, 2016, and the saturated sodium chloride solution was prepared. This technique is based on two separation steps.

Due to the small sample quantities the 100 ml separatory funnels were chosen. The weight of the first 100 µm filter material was 2166.1 mg the weight of the second was 1804.7 mg and the weight of the third was 665.6 mg (Fig. 32). A total of 4636.4 mg/46 liters were filtered using the Blue Filter Cascade. The cascade was exclusively equipped with three 100 µm filters.

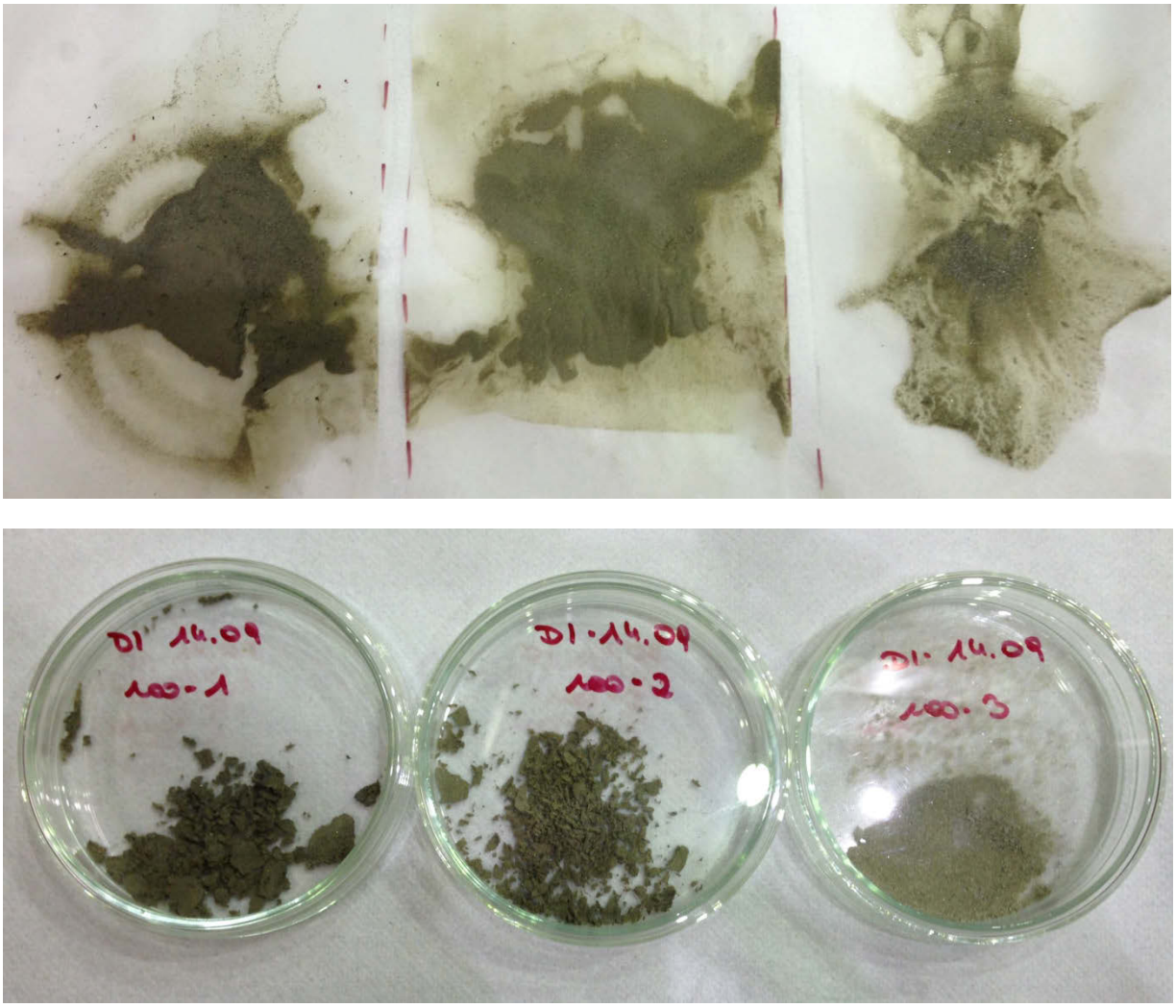

Figure 32: Sample material for the density separation technique from SS 2, in wet, freshly sampled and dry condition

The sample material was filled into a separating funnel and about 100 ml of the saturated sodium chloride solution were added (Fig. 33) and carefully shaken for two minutes. After the homogenization, and a sedimentation time of 45 minutes the sediments at the bottom of the funnel were gravitationally extracted. The supernatant was collected in a glass beaker.

Figure 33: Density technique by 100 ml separating funnels

The supernatant was isolated using a 0.45µm teflon filter and a vacuum pump to extract the microplastic particles.

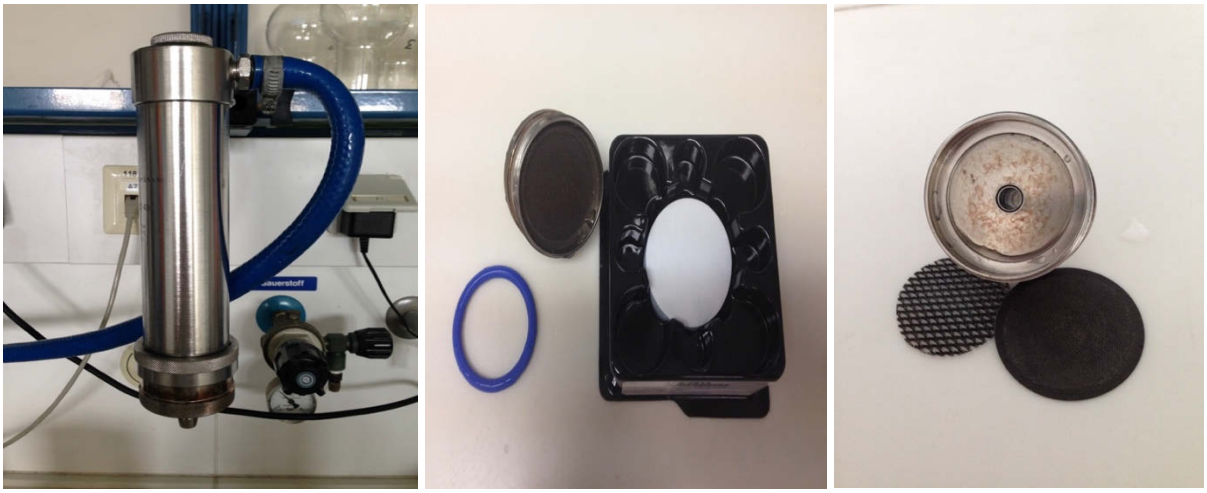

Figure 34: Pressure filtration system with the filter unit and teflon filters (0.45µm)

For the identification and further determination of the particles the Raman spectrometer was applied.

4.10 Methodology of the separation by chemical technique

In order to perform the separation by chemicals the Blue Filter Cascade was used, equipped with nylon filters with pore sizes of 100 µm, 50 µm, 30 µm, 15 µm and 5 µm. For this experiment 290 mg of fluorescent microplastic particles and 50 mg of commercial microplastic particles were used. After weighing, the dry particles were filled directly into the PVC tube at the inlet of the filter cascade and after connecting the cascade to the water supply line, the water was turned on. After 10 minutes, 150 liters of water had flown through the cascade and a distribution by particle size was ensured (Fig. 35).

Figure 35: BFC with fluorescent microplastics

After the mechanical separation, 500 mg common detergents, 10 ml hexane and 200 µl Triton X-100 were added to the filter units and the separation efficiency was observed. In the preliminary evaluation for the recovery of fluorescent microplastics following organic solvents were used: toluene, benzene, cyclohexane and hexane. Of these solvents, hexane was best suited for separation (Mucha Torre, 2015). The use of a solvent like hexane, which is non-polar, had the task of attracting the microplastic particles to move from the liquid part to the solvent part by means of london disperse forces (Ashenhurst, 2012). It is the weakest intermolecular force and leads to the attraction between two non-polar molecules. Parts of the mixture of hexane and microplastic particles as well as the mix of Triton X-100 and microplastics were put on a glass slide for further investigation with the Raman spectrometer.

The separation test was observed and followed by the UV light chamber.

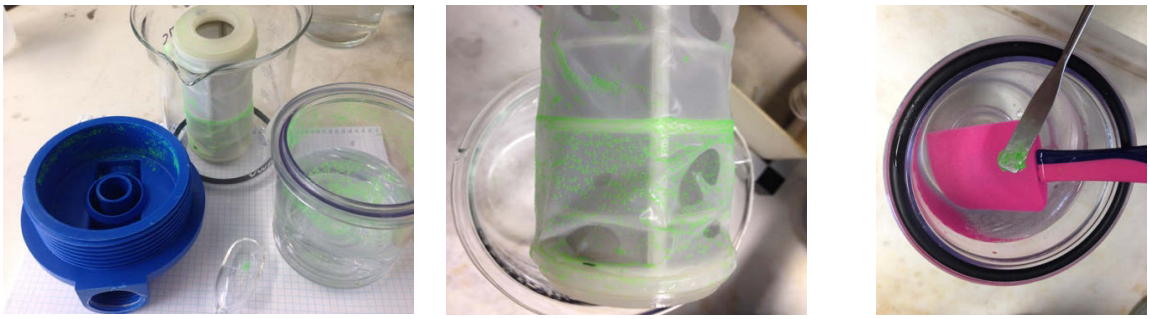

Figure 36: Separation by chemical technique

Since not all particles resuspended, the contaminated parts were placed in the vacuum chamber oven at 60°C overnight, as dry particles are easier to remove.

Lastly, the remaining particles were washed off the beakers and filter units with ultra-pure water and filtered in a water jet pump and a vacuum pump through a 0.45 µm teflon filter each and dried in an oven at 60°C overnight (Fig. 37).

Figure 37: Separation by water jet pump and pressure system

4.11 Methodology of the separation by ultrasonic

Figure 38: Branson 3510 sonicator

First, the fluorescent microplastic particles with a diameter of 75 - 90 µm were attached to filters of different mesh sizes and rubbed carefully into the pores. Two beakers, one containing ultra-pure water and one with a saturated sodium chloride solution, were prepared. The filters were added into the beakers for 10 minutes. The beakers were then put in the Branson 3510 sonicator (Fig. 38) for another 10 minutes and the separation was observed.

The separation by means of acoustic forces is a promising alternative to conventional methodologies like filtration or sedimentation (Gröschl, 1998). For this method ultrasound, usually ranging from 20 - 400 kHz, and an appropriate solvent like water or a saturated sodium chloride solution are used to free the filters from the fluorescent microplastic particles. Ultrasonic cleaning uses cavitation bubbles induced by high frequency pressure waves to stir a liquid. The agitation generates high forces on the microplastics. The intention is to remove almost all particles adhering to or embedded on the filter. The interaction of standing ultrasonic waves, with particles dispersed in a fluid or attached to a solid, produces forces on the particles, which can be utilized for the separation of the dispersed particles from the fluid (Benes et al., 2001).

4.12 Methodology for the microplastics recovery test

Before the test was started it was relevant to do some preliminary work. The filters as well as the fluorescent and commercial microplastics were weighed.

Fig. 39: Filter unit with microplastics

Approximately 245 mg of microplastics were used for each of the three experiments.

For these experiments, the stainless steel cascade was equipped with nylon filters with pore sizes of 300 µm, 100 µm and 30 µm. The dry microplastic particles were transferred directly from the weighing boats into the influent silicon tube of the first unit, which was then connected to the water supply line of the laboratory (Fig. 39). The water was turned on for 10 minutes.

Figure 40: SSC with the microplastic particles

The microplastic particles were distributed in the cascade as a function of the size. The water was turned off and the filters were placed in glass petri dishes to prevent contamination and ensure safe transport and storage.

The filters were weighed again in order to calculate the recovery rate and examined in the UV light chamber.

4.13 Methodology of the microplastics analysis in bottled water

Figure 41: Plastic bottles

This analysis was carried out to verify the presence of microplastics in bottled water. In order to avoid contamination of the filters, clean laboratory cotton cloths were worn during the entire experiment.

Three bottles of 1.5 liters each were purchased from ten different brands.

After placing an unpolluted teflon filter (0.45 µm) into the pressure filtration system, the water was poured in small steps of 250 ml into the apparatus. At the end of the filtration the filter was taken out and placed in a clean glass petri dish.

To attain statistical significance, the process was repeated three times for each brand.

The filters were evaluated with an optical microscope with a ten-fold magnification. The particles and fibers were counted and measured. Particles resembling microplastics were assessed with the Raman spectrometer.

4.14 Methodology of the particle characterization

The XploRA INV from Horiba, which was the selected Raman spectrometer is an inverted, integrated confocal system. The confocal microscope is coupled with a full Raman module mounted above which includes a 200 mm focal length spectrograph equipped with four-grating turrets, laser sources and detectors. The different excitation wavelengths are supplied by two internal lasers, with a wavelength of 532 nm with a cut off < 60 cm $^{-1}$, and a 785 nm- cut off < 50 cm $^{-1}$. The optical path is split into two parts: the illumination (laser) path and the collection (Raman signal) path. On the incoming laser path, the laser beam is reflected towards the microscope by the use of a special filter (dielectric edge rejection filter). It uses an injection/rejection mode which firstly directs the laser into the microscope and then filters out the Raleigh scattered light as it returns to the spectrograph, allowing only the Raman scattered light to be transmitted to the confocal hole and the entrance of the spectrograph. The spectrograph itself is used to disperse the various multichromatic Raman spectral lines onto the CCD detector for detection and analysis (Fig. 42).

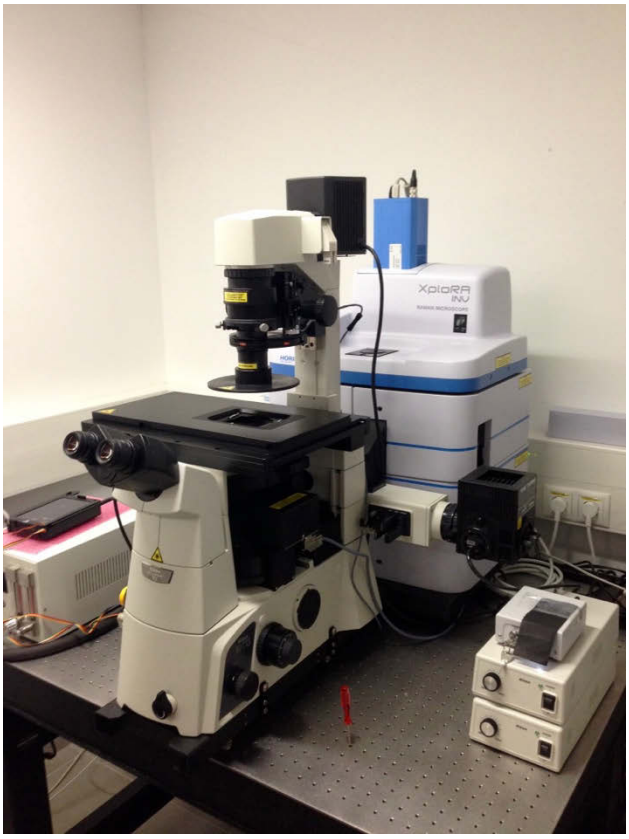

Figure 42: The Raman spectrometer, XploRA INV, of the Imaging Center at University of Natural Resources and Life Sciences, Vienna

Before the actual application of the spectrometer, some preparations were made.

As a first step reference spectra were created as the taken samples consisted of an unknown composition and thus differentiation was easier. Leaves, peppers and tomatoes were used to obtain spectra of cellulose and lignin. Organic and mineral materials, quartz, vermiculite, apatite, graphite and sand were evaluated as it was assumed that these materials provide the basis of the unknown samples and various plastics, polyethylene, polypropylene and polystyrene were investigated as they were target to answer the thesis' question.

The beforehand investigation of different filter materials was also necessary, since it was not clear which filters would have background spectra and would influence the result. Tested filter materials were cellulose acetate, cellulose nitrate, teflon, high quality filter papers, microfiber filters of 100 % borosilicate and nylon filters.

After the on-site sampling the filters were weighed with the filter material and then without the sampled material to determine the amount of filter residues.

The spectrometer was used to examine only the filter residues without the filters, making a separation of filter and filter residues necessary. After the particles had already parted from the filters by simple air drying, they were dried in the oven and the filter residues were tapped off. Due to the unsatisfactory results of the chemical and ultrasonic separations, only the density separation in combination with the pressure system was applied. Using the other separation methods too much filter material would have been lost during the separation process.

Prior to each use of the XploRA INV a calibration of the lasers, 532 nm and 785 nm, had to be carried out (Fig. 43).

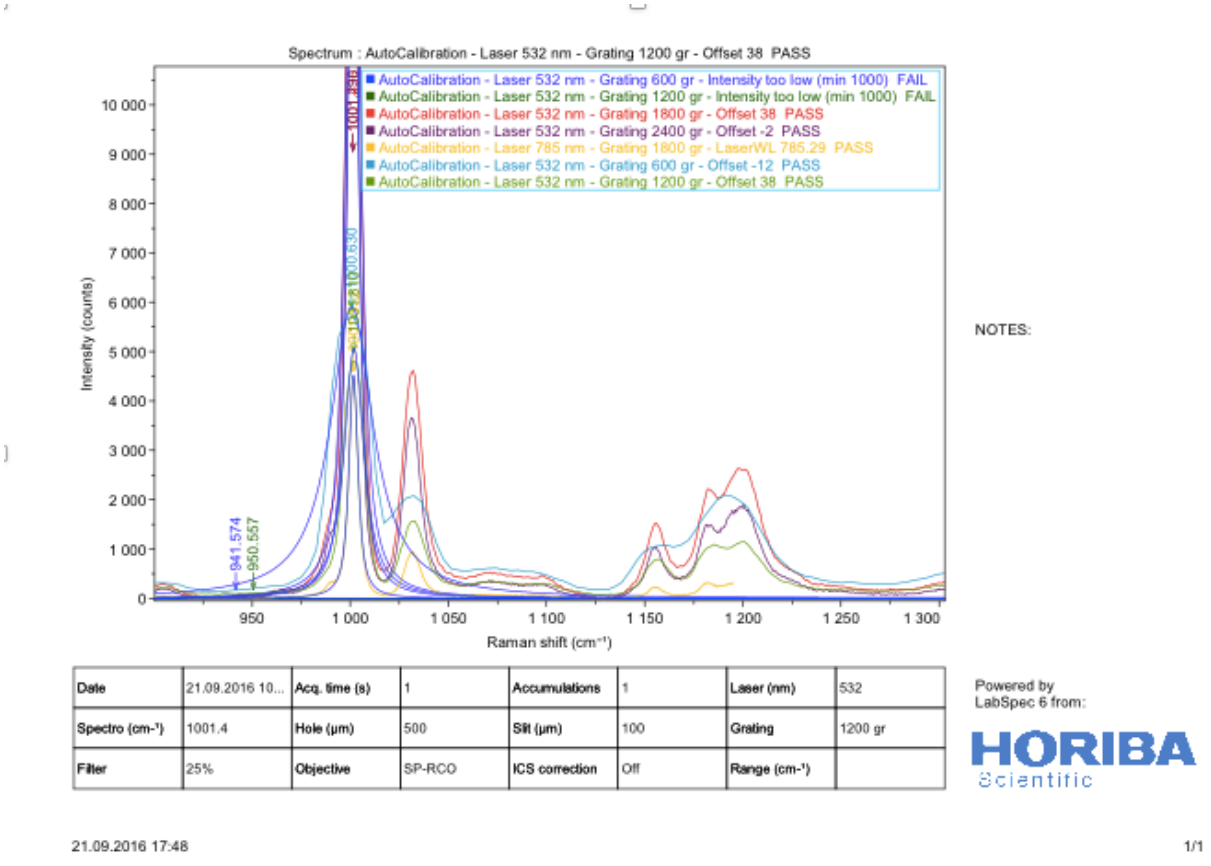

Figure 43: Autocalibration spectra from laser 532 nm with all gratings

A ten-fold magnification of the microscope Nikone Eclipse Ti-U was used for all investigations. The wavelength interval was chosen between 200 and 3700 cm^{-1}. The choice of the laser, the intensity of the laser, the acquisition time, the accumulation and the grating had to be adapted in every single experiment. Furthermore, each time the slit and hole parameters had to be adjusted and played with the Z axis. Raman imaging can be carried out in all three dimensions (X, Y and Z) in any desired combination. 2D surface imaging (XY plane), 1D depth profiling (Z axis), 2D optical cross-sectioning (XZ and YZ) and full 3D volume imaging (XYZ) are possible (Horiba, 2014b).

The sample material was placed on an extremely thin IBIDI glass dish and positioned under the Raman inverted, confocal microscope. After finding one or more suitable particles, a single spectrum or a hyperspectral map were created, which appeared immediately on the PC. All data, whether a single spectrum or a hyperspectral map comprising hundreds of thousands of spectra, can be processed with standard spectroscopic functions, including baseline subtraction, smoothing, solvent subtraction, and full mathematical manipulation.

The whole principle is based on the interaction of laser light with a sample resulting in a Raman spectrum, a very unique fingerprint.

The XploRA INV combines the standard XploRA Raman microscope with the capabilities of an inverted microscope (Horiba, 2014a). An inverted microscope is a microscope with its light source and condenser on the top, above the stage, pointing down, while the objectives and turret are below the stage, pointing up. The stage of the microscope is fixed, and the focus is adjusted by moving the objective lens, in this case with a ten-fold magnification, along a vertical axis to bring it closer to or further away from the sample. The focus mechanism has a dual concentric button for coarse and fine adjustment. Furthermore, the sample can be focused with the eye or via the built-in camera (Fig. 44).

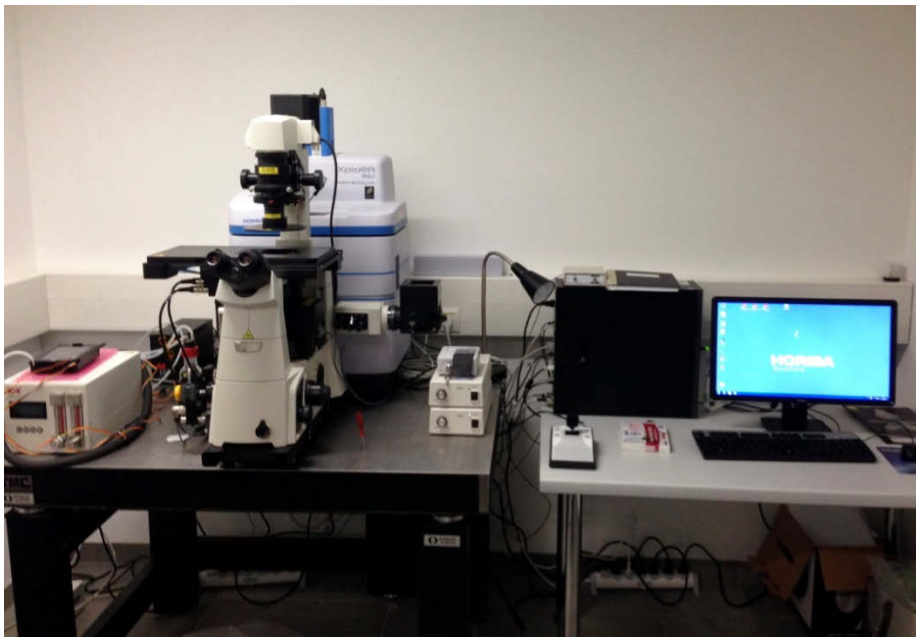

Figure 44: Installation of the Raman spectroscope (XploRA INV)

The purpose of this application was to find and identify microplastics in bank filtrate, Danube water and groundwater. A Raman spectrum contains information on material identity (e.g. characteristic Raman bands), material composition (e.g. peak intensity or multivariate analysis scores), molecular structure or strain (e.g. peak position) and crystallinity or phase (e.g. peak width) (Horiba, 2014b).

5 Results and discussion

5.1 Set-up and pretests of the Stainless Steel Cascade (SSC)

The chosen material was nylon, due to the large availability of different pore sizes, and cellulose nitrate. The nylon filters had mesh sizes between 500 - 5 µm and the cellulose nitrate filters a pore size of 1.2 µm. Filters with pore sizes between 15 µm, 5 µm and 1.2 µm were selected for sampling sites with low contamination and filters with pore sizes > 15 µm for Danube water samples.

The filters were tested in various combinations with different water qualities.

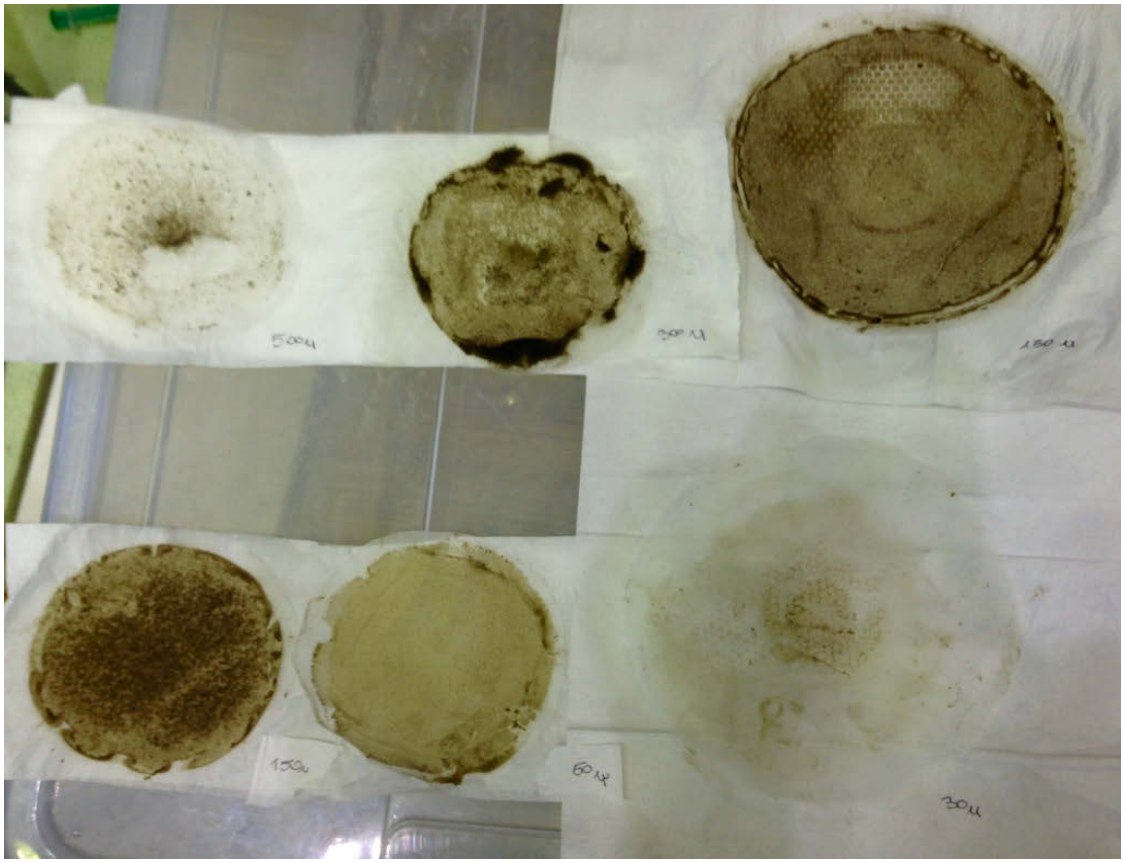

Figure 45: Pretest results with sewage sludge on a 500, 300, 150 / 150,50, and 30 µm filter

It can be seen that the different pore sizes lead to a separation of the size fractions. Thus, the desired effect was already generated before the in situ application without problems.

Figure 45 shows that sewage sludge mixed with water was filtered through a cascade with mesh sizes of 500, 300 and 150 µm and was collected and filtered again through a cascade with filters of smaller pore size (150, 50 and 30 µm) in order to be able to follow the purification process of the mixture. A clear reduction of the pollution be observed and thus a cleaning of the water with decreasing pore size is recognizable. If the water had been further filtered, it would become even cleaner with a further decrease in pore size.

The cleaner the experimental water was the smaller pore sizes could be selected and vice versa (Fig. 46).

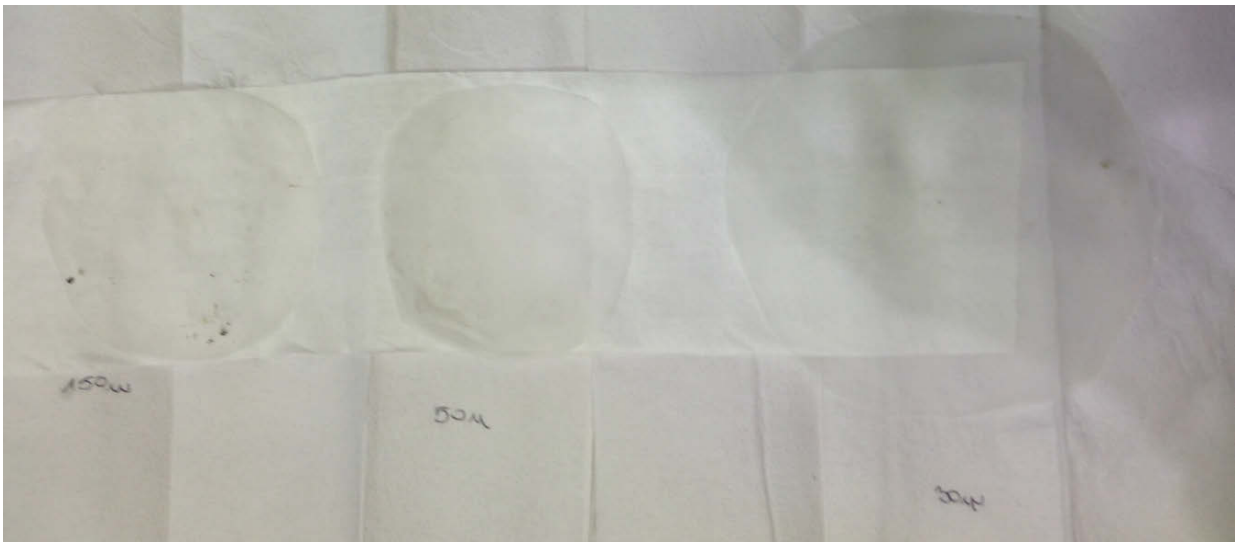

Figure 46: Tested bank filtrate on 150, 50 and 30 µm filters

5.2 Set-up and pretests of the Blue Filter Cascade (BFC)

It has been found that the optimal descending composition of the filters were 100 µm, 50 µm, 30 µm, 15 µm and 5 µm for bank filtrate, while for Danube water the filter units were only equipped with 100 µm filters. The individual particle fractions were optimally separated and the contamination of the water was of secondary importance due to the greater capacity of the filter units and the larger flow volume in comparison to the SSC.

Figure 47: Filter tests with Danube water and 3 filter units

5.3 Comparison of the two filter systems

According to the results of both cascade tests it was obvious that the pore sizes always have to be adapted to the contamination of the water and the desired discharge. During field application, the sampled water type and desired discharge must be known beforehand.

Problems with the SSC occurred during the sealing process but these could be remedied with simple aids. Silicon rings were cut and placed around the filters. It was necessary to seal the filter units since they were not tight otherwise.

The BFC had to be ventilated before each filtration, as otherwise the units would not fill up entirely with water.

The aim was to filter $1m^3$ with these methods, but the objective was only achieved with the BFC. Both methodologies have advantages as well as disadvantages. The great advantage of the SSC is that all particles are collected directly on the filters and the filters, no matter the mesh size, can be easily exchanged. However, it's not possible to have the same amount of water flow through the cascade in a certain amount of time compared to the BFC. Thus, this method is much more time consuming. The BFC works very efficiently in terms of water quantity and time, though admittedly it is very difficult to get all the filtered particles out of the filtration unit because the particles are not only on the filters but also on the cartridge and this means additional separation steps.

Nylon was not the originally desired filter material, since nylon is ultimately also plastics, but due to the lack of appropriate alternatives with suitable mesh sizes it was the only option. An advantage was that the material could be tailored, since the material was ordered in one big piece ($1\ m^2$), and processed to fit any type of request, whether, as in this case, fit on a stainless steel filter or on a cartridge.

In summary, a system that combines the advantages of both cascades should be developed and a filter material which is not plastics but still comprises a large range of mesh sizes should be found for the filtration.

5.4 In situ sampling

Table 3 shows the pore size as well as the sampling volume and time. Sampling site 2, 3. Filtration, showed a special case. The selected filter pore sizes were much too small for the contaminated Danube water, the pores were immediately blocked, so only half a liter of water could flow through the cascade and the sampling time is accordingly small. This result clearly points out how important the selection of the pore size, based on the location factors, is. The sampled volumes were in the range of 0.5 to about 420 liters and the sampling time was in the range of 0.5 - 150 minutes. The filter pore sizes used comprised 500 - 1.2 µm. The most frequently used pore sizes with the SSC were 15, 5 and 1.2 µm for bank filtrate and groundwater, and 500, 300 and 150 µm as well as 150, 100 and 50 µm for Danube water.

Table 3: Sampling sites, sampling dates, sampling volume and time as well as the filter pore sizes of the SSC

Sampling Site	Date	Sampling Volume [l]			Sampling Time [min]			Filter Pore Size [µm]		
		1. Filtration	2. Filtration	3. Filtration	1. Filtration	2. Filtration	3. Filtration	1. Filtration	2. Filtration	3. Filtration
SS 1	20.10.15	262	529		60	90		50/30/15	15/5/1.2	
SS 2	28.10.15	130	132	0.5	30	30	0.5	500/300/150	150/100/50	50/30/1.2
SS 3	28.10.15	189	-	-	90	-	-	15/5/1.2	-	-
SS 4	30.10.15	344	-	-	60	-	-	15/5/1.2	-	-
SS 5	30.10.15	406	-	-	60	-	-	15/5/1.2	-	-
SS 3	01.09.16	422	-	-	150	-	-	15/5/1.2	-	-
SS 1	06.09.16	232	-	-	140	-	-	15/5/1.2	-	-
SS 1	14.09.16	91	285	260	12	42	33	150/100/50	500/300/150	150/100/50
SS 2	14.09.16	4	127	78	2	30	30	150/100/50	500/300/150	150/100/50

Table 4 lists the sampling volume and time as well as the corresponding flow rate. The flow rates happened at a range of 1.0 - 6.8 l/min. The greater the sampling time was, the higher was the volume and thus the flow rate.

Table 4: Sampling volume, sampling time and the flow rate of the SSC

Sampling Site	Date	Sampling Volume [l]			Sampling Time [min]			Q [l/min]		
		1. Filtration	2. Filtration	3. Filtration	1. Filtration	2. Filtration	3. Filtration	1. Filtration	2. Filtration	3. Filtration
SS 1	20.10.15	262	529		60	90		4.4	5.9	-
SS 2	28.10.15	130	132	0.5	30	30	0.5	4.3	4.4	1
SS 3	28.10.15	189	-	-	90	-	-	2.1	-	-
SS 4	30.10.15	344	-	-	60	-	-	5.7	-	-
SS 5	30.10.15	406	-	-	60	-	-	6.8	-	-
SS 3	01.09.16	422	-	-	150	-	-	2.8	-	-
SS 1	06.09.16	232	-	-	140	-	-	1.7	-	-
SS 1	14.09.16	91	285	260	12	42	33	7.5	6.7	8.7
SS 2	14.09.16	4	127	78	2	30	30	2	4.2	2.6

With the Blue Filter Cascade, the flow rate depends on the volume and time as well. The larger collection capacity and therefore higher flow rates make it possible to trap much bigger quantities of particles without blocking the cascade (Tab. 5). The sampled volume was in a range of 46 - 2106 liters, the sampling time between 10 - 150 minutes and the flow rates in a range of 4.60 to 14.04 liters/minute. The filters used were of a size range of 5 - 100 µm. At SS 3 and SS 1 the same mesh sizes were used for the bank filtration. At SS 1 and SS 2, Danube water was sampled with 100 µm filters, at SS 1, however, one filter unit less was used because the water was cleaner than at SS 2.

Table 5: Sampling sites, sampling dates, filter pore sizes, sampling volume and the flow rate of the BFC

Sampling Site	Date	Filter Pore Size [µm]	Sampling Volume [l]	Sampling Time [min]	Q [l/min]
SS 3	01.09.16	100/50/30/15/5	2106	150	14.1
SS 1	06.09.16	100/50/30/15/5	2106	150	14.1
SS 1	14.09.16	2x100	312	23	13.6
SS 2	14.09.16	3x100	46	10	4.6

The much larger sampling volume of the BFC compared to the SSC immediately catches one's eye, as well as the accordingly larger discharges. The largest sampled volume was 529 liters for the SSC and 2106 liter for the BFC. This results in a difference of 8.14 liters per minute in favor of the BFC. Due to the five filter units the particles were separated better by size fraction during the processing of the samples, however, more separation steps were necessary afterwards than with the SSC. It is simply a great advantage and reduces the processing time that all particles were collected directly on the filter. In the case of the SSC, only the separation of the particles from the filters, in the case of BFC the separation of the particles from the filters and from the remaining filter units was necessary.

The weight of the filter reflects the pollution of the water.

At SS 2, Danube water was sampled and the most particles were filtered with the SSC and BFC in both years, 2015 and 2016. However, with the Blue Filter Cascade almost 4 times more material was caught, than in the first filtration on the October 28, 2015 and in the second filtration on the September 14, 2016 (500, 300, 150 µm) with the SSC.

During the filtration of bank filtrate with the SSC and BFC, between 1 and 20 g of particulates were filtered. Here, no distinction between the particle weights using the two systems is possible.

Tables 6 and 7 display the filter pore sizes and particle weights for the SSC and BFC are displayed.

Table 6: Sampling sites, sampling dates, filter pore sizes and dry particle weight for the SSC

Sampling Site	Date	Filter Pore Size [µm]			Particle Weight [mg]		
		1. Filtration	2. Filtration	3. Filtration	1. Filtration	2. Filtration	3. Filtration
SS 1	20.10.15	50/30/15	15/5/1.2		0.8	4.2	
SS 2	28.10.15	500/300/150	150/100/50	50/30/1.2	1295	43.4	710
SS 3	28.10.15	15/5/1.2			7.9		
SS 4	30.10.15	15/5/1.2			7.1		
SS 5	30.10.15	15/5/1.2			19.6		
SS 3	01.09.16	15/5/1.2			18.1		
SS 1	06.09.16	15/5/1.2			18.4		
SS 1	14.09.16	150/100/50	500/300/150	150/100/50	65.2	5.4	9.6
SS 2	14.09.16	150/100/50	500/300/150	150/100/50	60.2	943	59.7

Table 7: Sampling sites, sampling dates, filter pore sizes and dry particle weight for the BFC

Sampling Site	Date	Filter Pore Size [µm]	Particle Weight [mg]
SS 3	01.09.16	100/50/30/15/5	13.3
SS 1	06.09.16	100/50/30/15/5	11.4
SS 1	14.09.16	2x100	41.7
SS 2	14.09.16	3x100	4636

5.5 S::can measurements

S::can is installed at each sampling site.

The data from 2015 were obtained by a single measurement per site. In 2016, several readings were made per location and mean values were formed (Tab. 8).

Table 8: S::can sensor results

Sampling Site	Date	Turbidity [FTU]	Abs/m	NO3-N [mg/l]	TOC [mg/l]	DOC [mg/l]	Conductivity [uS/cm]	Temperatur [°C]
SS 1	20.10.15	0.28	1.25	1.18	0.81	0.5	409	16
SS 2	28.10.15	297	12.3	0.92	49.6	5.07	-	14
SS 3	28.10.15	0.06	1.33	1.35	0.81	0.51	396	16
SS 4	30.10.15	-	-	-	-	-	542	13
SS 5	30.10.15	-	-	-	-	-	509	12
SS 3	01.09.16	0.12	2.23	1.44	1.12	0.71	379	16
SS 1	06.09.16	0.19	1.76	1.09	0.89	0.61	400	15
SS 1	14.09.16	16.4	13.4	1.16	5.26	2	-	16
SS 2	14.09.16	114	14.2	1.14	5.94	2.81	-	20

On the basis of the turbidity, it is clearly recognizable that Danube water is being registered at SS 2. However, the value of 2015 is still far higher than the value of 2016. At SS 1, Danube water was sampled on September 14, 2016, however the turbidity data are barely comparable to the data of SS 2. These great differences were also the reason why, during the sampling with the BFC, at sampling site 1 only two filter units were used and at SS 2 three were applied.

Site 2 also stands out clearly in the data regarding other parameters. NO_3-N is the only parameter with values similar to the other sampling locations. According to drinking water regulation (2012), the maximum allowed guideline value for nitrate (NO_3^-) is 50 mg/l. 1 mg/l NO_3-N corresponds to 4.43 mg/l NO_3^-. This means that 11.3 mg/l NO_3-N are allowed. All results are < 1.5 mg/l so the values are far below this guideline value. The values of organic carbon for clean water range between 1 - 2 mg/l. The Danube water samples exceed these values enormously, while the values of the bank filtrate sampling points, however, lie in between these values.

Sampling site 1 stands out only in the turbidity values compared to sampling site 2, otherwise the values of SS 1 resemble the values of the other parameters of SS 2.

The conductivity was measured only for bank filtrate. The water temperature, however, was registered for all stations and is between 12°C and 20°C.

At sites 4 and 5, groundwater is conveyed and no turbidity, permeability, and TOC data are recorded.

5.6 Particle counting and turbidity measurements

The particle counts for all sites were performed with the Abakus particle counter in the laboratory (Tab. 9).

On the basis of the data, a large difference in the quantity of particles between the Danube water, bank filtrate/groundwater samples were recognizable, as well as a large variability between the size classes. If the bank filtrate, groundwater and Danube water values are examined separately, homogeneity within size classes can be seen. The highest particle counts were achieved at SS 2 in 2015. A comparison with the values of the turbidity measurement with the S::can probe confirms this result.

Particle sizes < 1 µm and between 2 - 30 µm were generally detectable during the entire measurement, whereas larger particles were not always present. Since the quantity of particles > 30 µm strongly decreases, said size was used as a cut-off point.

Table 9: Abakus particle counting results of all sites

Sampling Site	Date	Water Type	< 1 µm	1- 2 µm	2- 5 µm	5- 10 µm	10- 15 µm	15- 30 µm	30- 50 µm	> 100 µm
SS 1	20.10.15	Bank filtrate	1018	251	52	11	2	1	0	0
SS 2	28.10.15	Danube water	12757500	3740650	1099540	356900	64451	2356	981	95
SS 3	28.10.15	Bank filtrate	1511	386	55	14	4	2	2	0
SS 4	30.10.15	Groundwater	738	192	49	17	2	0	0	0
SS 5	30.10.15	Groundwater	1807	540	80	20	6	3	1	1
SS 3	01.09.16	Bank filtrate	26283	9834	2732	740	252	64	9	0
SS 1	06.09.16	Bank filtrate	15403	4116	823	129	39	8	1	0
SS 1	14.09.16	Danube water	1540408	4121460	37044	2484	461	50	1	0
SS 2	14.09.16	Danube water	5019285	2835160	1006543	235316	66196	7544	86	0

Figure 48 displays the particle distribution according to size classes of all sites where bank filtrate and groundwater were sampled. The lines of the graphs in Figures 48 and 49 are ordered as described in Table 9, from top to bottom.

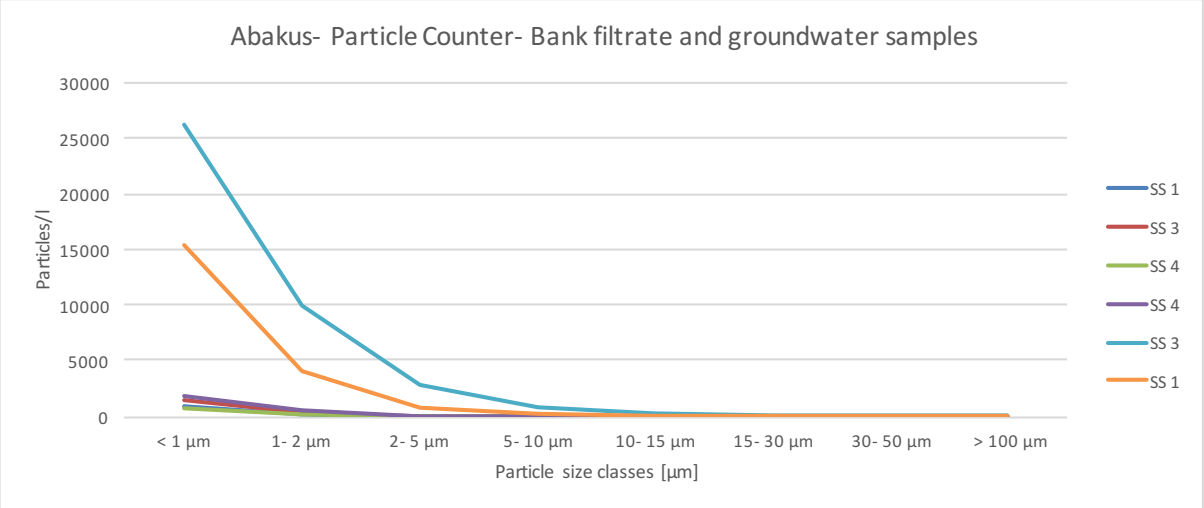

Figure 48: Distribution of particles by size classes of bank filtrate and groundwater samples

Figure 49 shows the particle counting results of the Danube water samples taken in 2015 and 2016.

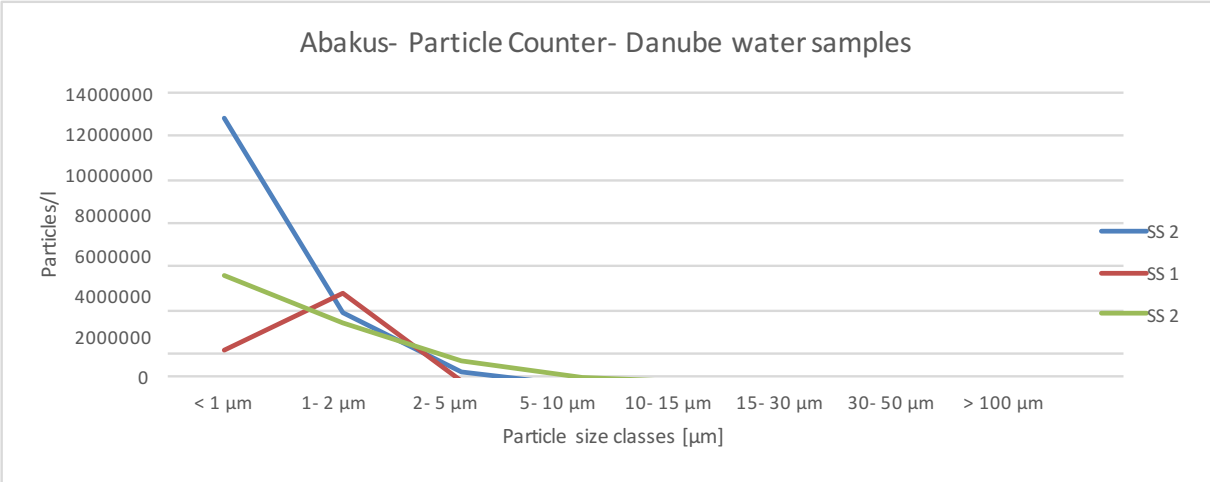

Figure 49: The quantity of particles of different size classes of Danube water samples

The results of the particle counting with the Abakus and Klotz, as well as the turbidity measurements of the worst case scenario, will be described next.

At 12:30 the sampling site was started, which can be clearly recognized due to the high number of particles. These figures were generated by the deposits during the three-week standstill. At 13:20, the discharge was increased but there was no recognizable increase in particle numbers while measuring with the Abakus. At 14:15, an increase of discharge took place and this time an increase in particle numbers, which was probably also caused by deposits in the system, could be seen (Tab. 10).

Tables 10 and 11 show the same results as Table 9. Particles < 30 µm, were present during the entire measurement. Particles between 30 and 100 µm with a low concentration of particles/liter were detected in the online system, as well as in the laboratory, however their number was small.

Table 10: Abakus, particle count, average results per liter (SS 1, 09/06/2016)

Sampling Time	Q [l/s]	< 1 µm	1- 2 µm	2- 5 µm	5- 10 µm	10- 15 µm	15- 30 µm	30- 50 µm	> 100 µm
12:30	125	44752100	15536980	2117530	156600	49190	9950	810	30
13:20	250	2458430	847220	117910	14450	3630	560	60	20
14:15	380	4336580	1433120	189870	20020	4470	686	90	20

Table 11: Klotz, average particle counting results per liter (SS 1, 09/06/2016)

Sampling Time	Q [l/s]	< 1 µm	1- 2 µm	2- 5 µm	5- 10 µm	10- 15 µm	15- 30 µm	30- 50 µm	> 100 µm
12:56	125	767728	373718	57531	6505	1838	177	6	0
13:20	250	1712316	709489	96142	9610	2257	158	6	0
14:15	380	2655338	1182496	186113	20244	5081	276	11	6

Figure 50 shows the extremely high increase in particles after the activation of the sampling site. Compared to the first rise, the growth in quantities after the two increments of the discharges are very small.

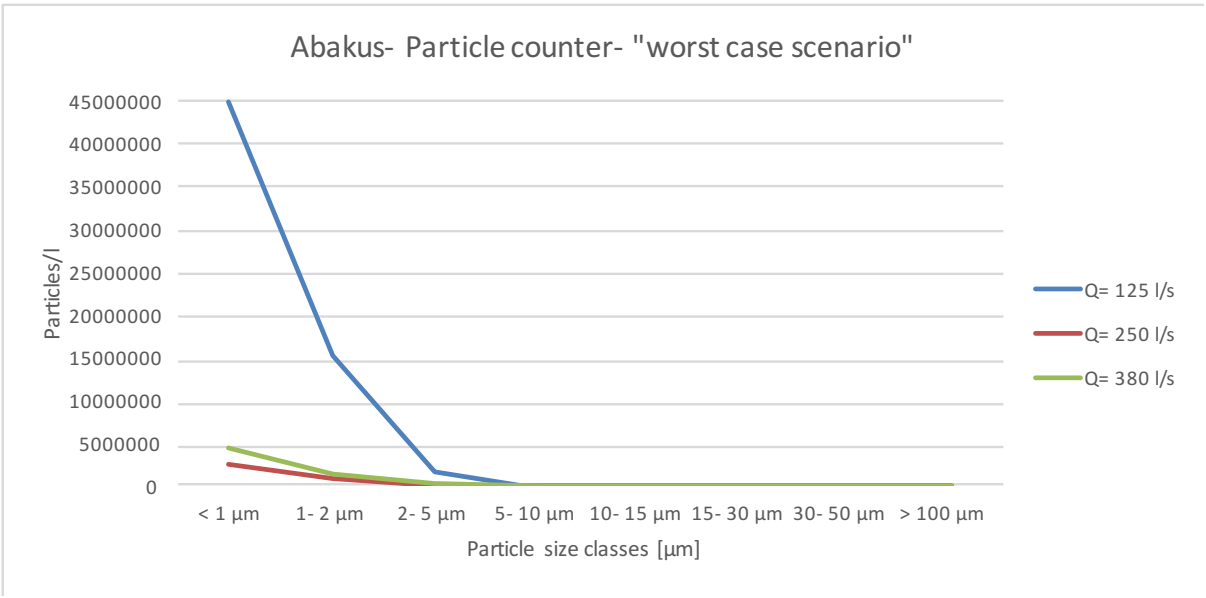

Figure 50: The quantity of particles of particular particle sizes classes at a discharge rate of 125, 250 and 380 l/s (SS 1, 09/06/2016)

To make a comparison of the data of the two particle counters possible, the discharge at 12:30 of the Abakus was deleted for Figure 51 as measuring with the Klotz did not start until 12:56 due to calibration needs. The highest particle numbers were measured at the highest discharge, by means of the above-mentioned effect. The discharge rate of 250 l/s indicated average particle numbers and the smallest particle numbers were measured at the smallest discharge. The higher the flow rate, the more deposits were carried away.

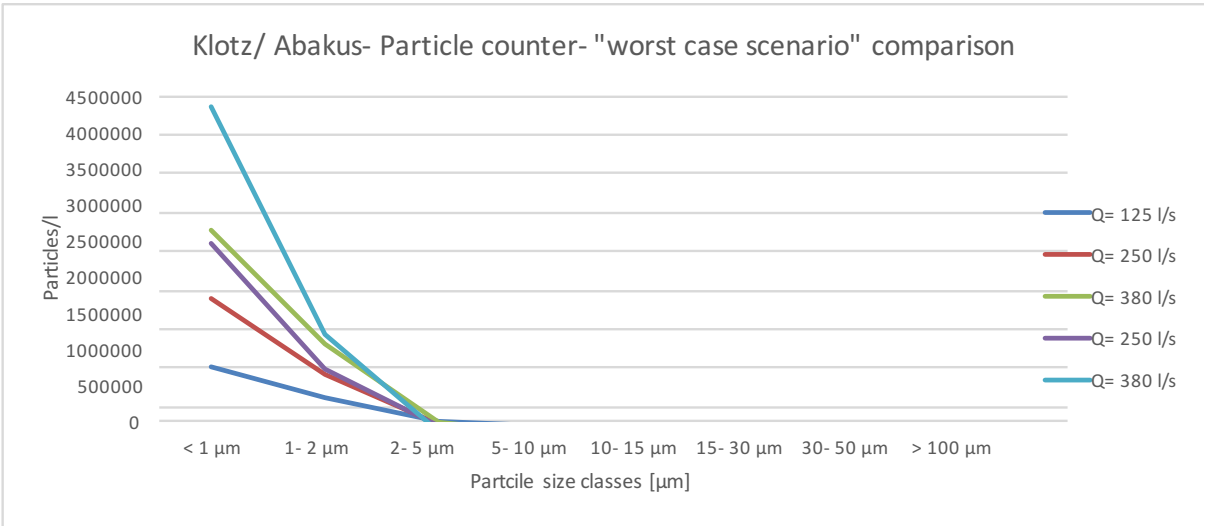

Figure 51: The quantity of particles of a particular particle size class at a discharge rate of 125, 250 and 380 l/s (Klotz-blue, red and green line) and 250 and 380 l/s (Abakus-purple and turquoise line), (SS 1, 09/06/2016)

The peak at 13:20 shows a significant increase but also drops faster than the peak at 14:15. Due to the short failure of the site, a longer and not so high rise appeared. At 14:15, after the start of operation of all pumps and wells, there was a breakdown of the system, therefore a minimal decrease can be seen, however the peak rose again after the restart of the sampling site at 14:26. The particle sizes 1, 2 and 5 µm are visible over the entire measurement period and confirm the values in Tables 9, 10 and 11.

Due to the continuous recording of the Klotz particle counter, air bubbles can be detected in the system as well, which are presumably responsible for the high peak in addition to the increase in particles. These air inclusions cannot be detected with the Abakus, since grab samples were taken only punctual and no continuous measurements, measured over a certain period of time, can be carried out.

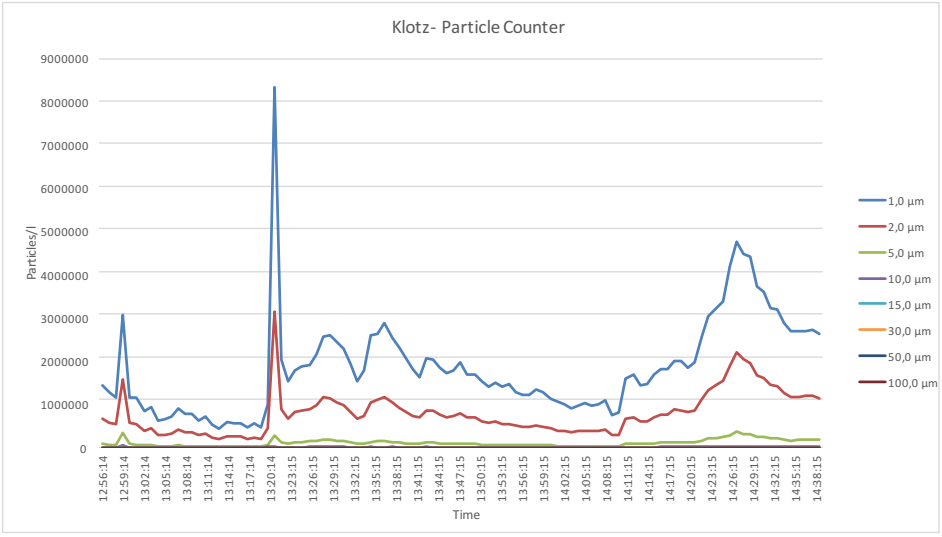

Figure 52: The change in particles/liter between different size fractions over a certain amount of time (SS 1, 09/06/2016)

An important indicator for the quantity of particles is the turbidity. The more turbid a liquid, the more suspended solids it contains.

The turbidity increased with the particle increase at 13:20 and slightly at 14:15, as well as 14:26. The turbidity clearly correlated with the increase in particles, as shown in Figures 52 and 53.

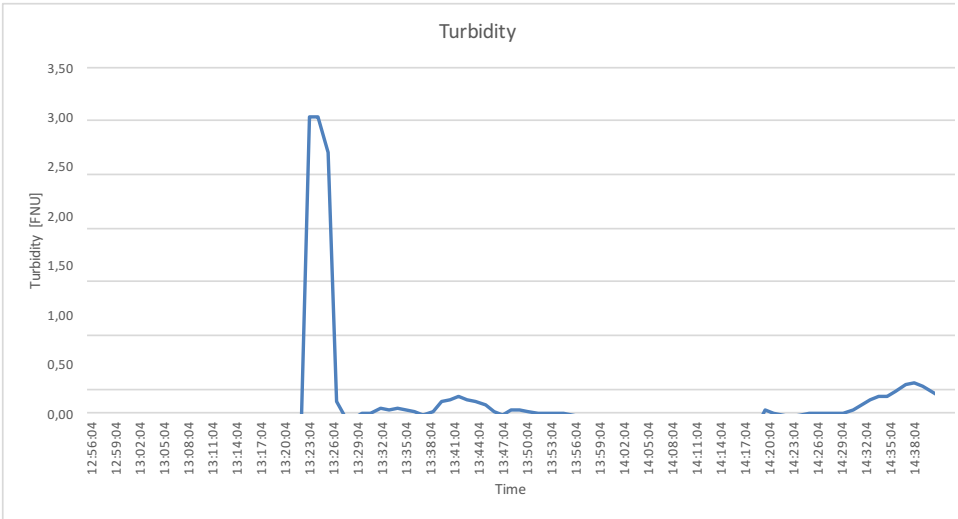

Figure 53: Measurement of the turbidity over a certain period of time with the Sigrist WTM 500 (SS 1, 09/06/2016)

The turbidity was measured manually with the turbidimeter, Turbiquant 1100 IR, using samples taken when the discharge was increased as well as between the increments. For each sample three measurements were made to obtain statistically significant results. A coincidence of the two turbidity measurements and the particle measurement, measured with the Klotz, can be seen.

Table 12 reveals the values of the turbidity measurements.

Table 12: Turbiquant measurement results (SS 1, 09/06/2016)

Time	1. Measurement	2. Measurement	3. Measurement
12:32	0.97	1.78	1.55
13:23	0.05	0.01	0.01
14:05	0.04	0.08	0.01
14:15	0.37	0.12	0.08
14:30	0.82	0.63	0.83
14:50	0.09	0.02	0.00

Figure 54 presents the results of Table 12.

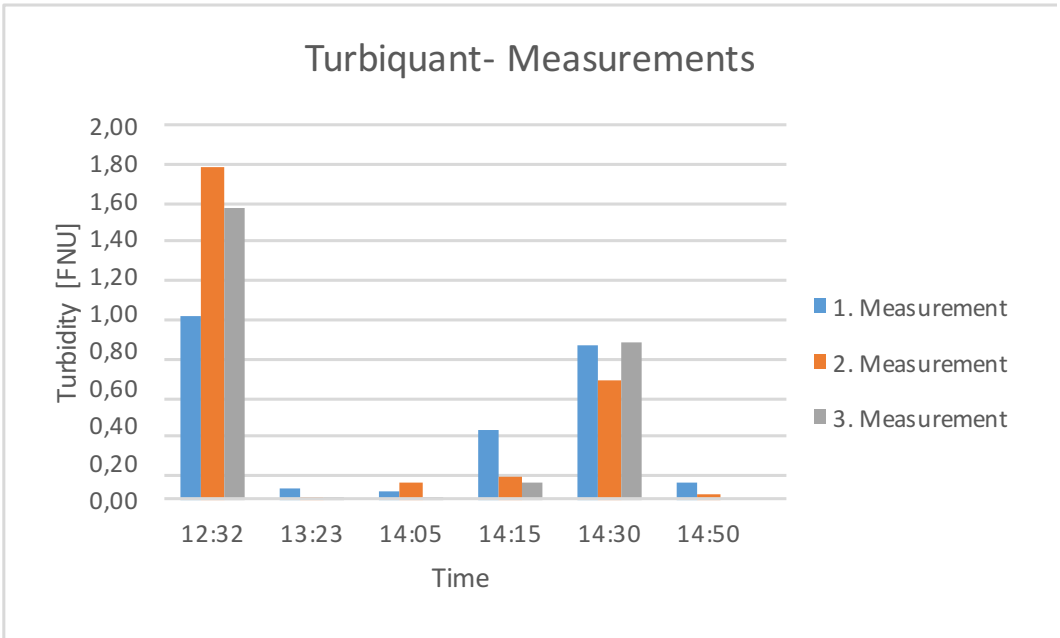

Figure 54: Changes in turbidity at different sampling times (SS 1, 09/06/2016)

Higher turbidity values at the increased discharges are also noticeable in this measurement since the values of the intermediate measurements are significantly lower (Fig. 54). Furthermore, these measured values indicate the actual turbidity values since the measurement with the Sigrist WTM 500 can detect air bubbles and these inclusions may increase turbid values.

In general, this means that, each time the sites are put into operation, a large amount of sediments is released into the system and can be measured indirectly by turbidity rates.

5.7 Microscopic sorting of the particle fractions

Apart from determining the size and appearance, the particles were also counted and quantification was attempted, however this goal was not possible to achieve in all cases.

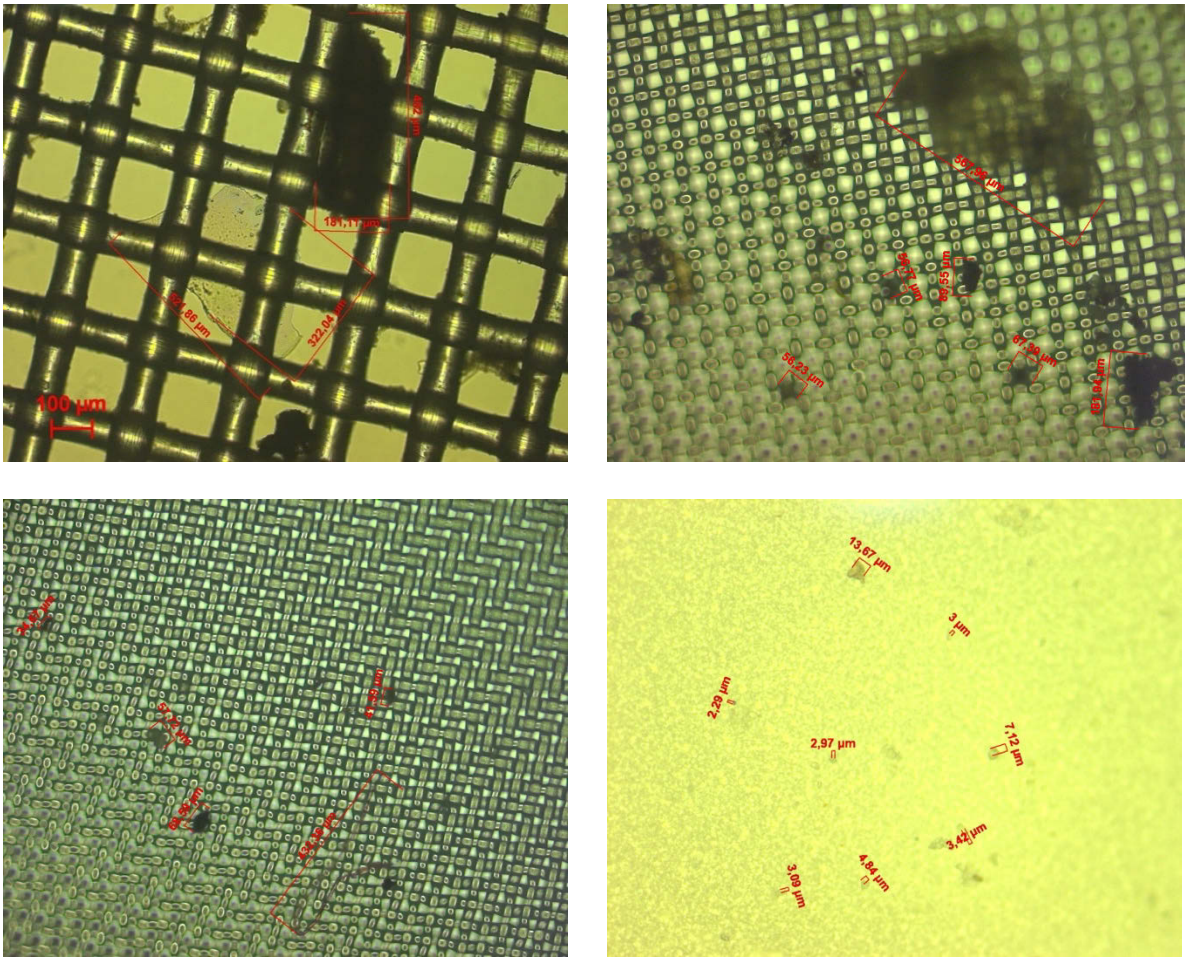

Figure 55: Filters used with the SSC at SS 3- 150 µm (Pic. 1), SS 1- 50 µm (Pic. 2), SS 2- 15 µm (Pic. 3), SS 5- 1.2 µm (Pic. 4)

Depending on the filter pore size the particles could be very well separated into size fractions, depending, of course, on the first filter's pore size. As can be seen in Figure 55 (Pic. 2), this filtration started with the 50 µm filter, since particles larger than 50 µm are also present thereon.

The particles measured ranged from > 500 to 2 µm in size.

Most of the particles or fragments were black and transparent. Some fibers were red, blue or green (Fig. 55, Pic. 3). This observation is similar to the results of the experiment about microplastics in bottled water. As displayed in Table 15, the granules and fragments were black while the fibers were blue, green and red as well.

5.8 Results of the density separation technique

In general, this technique is a normal solid liquid separation process, extended by the addition of a saturated sodium chloride solution.

During the sedimentation process in the separatory funnels, layers with particles of different sizes were already visible (Fig. 56).

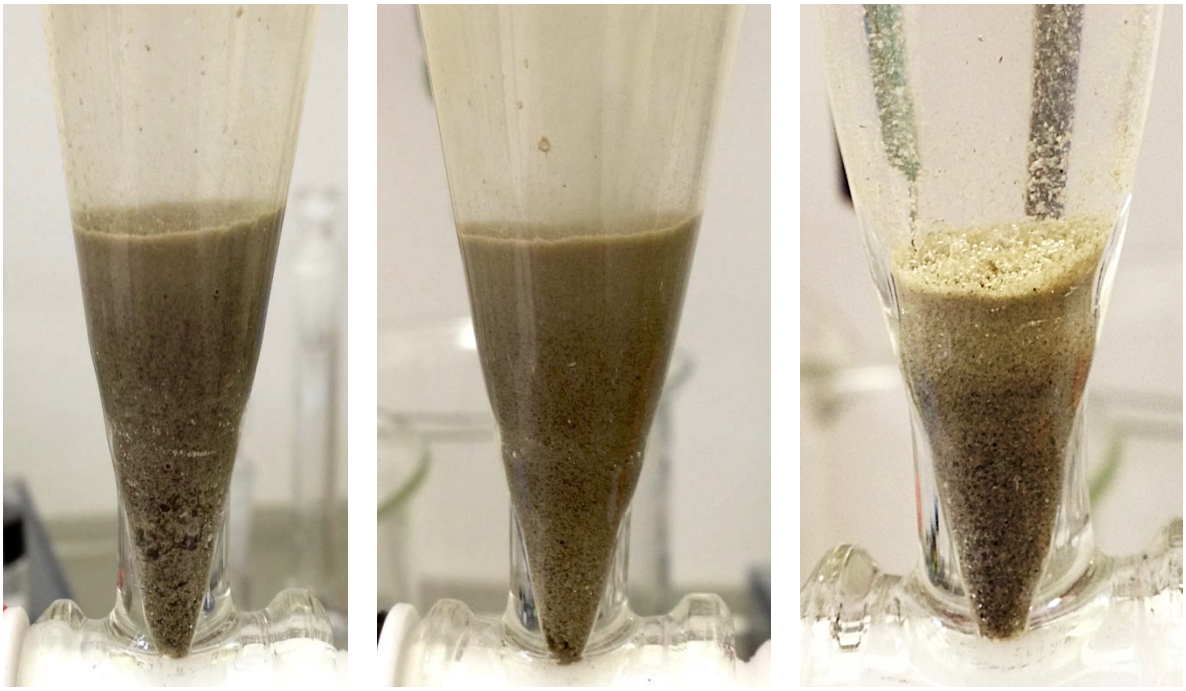

Figure 56: Settled layers of 3x 100 µm filters of the BFC, in 100 ml funnels with a sodium chloride solution

The deposited particles are layered according to their density and size due to their different sedimentation velocities. The low-density particles remain in the solution or float at the surface of the saturated sodium chloride solution. Since the rate of sedimentation is essentially determined by density, different substances may deposit at different velocities. Therefore, the layers are not solely dependent on particle size. The change of the density due to the added sodium chloride (NaCl) solution made it possible to extract the light microplastics.

The various extracted layers as well as the less dense particles, isolated with a pressure filter system, were examined with the Raman. It was also attempted to provide isolation using the water jet pump, this, however, failed.

The main component of the sediment was quartz, as displayed in Figure 57. The comparison with the reference spectrum confirmed this assumption, which was prepared in advance using known samples.

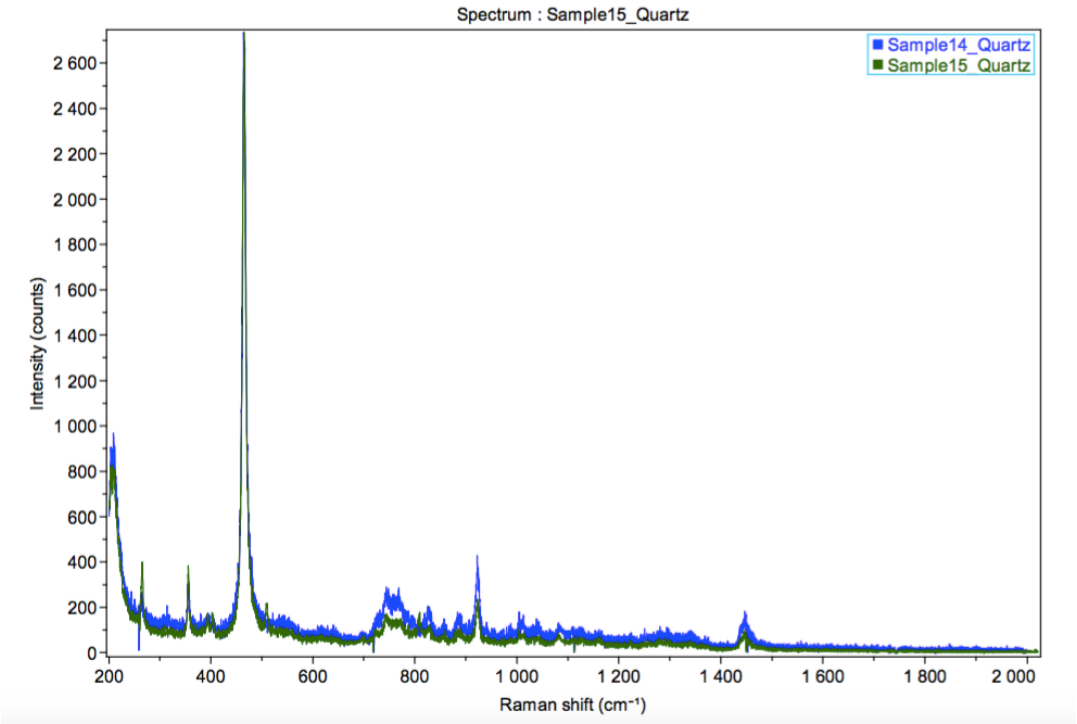

Figure 57: Two spectra of quartz sand

On the teflon filters polyethylene was found. These results are discussed in point 5.14- Particle characterization.

This technique is a very efficient way to examine samples of unknown composition and with particles of different densities.

The specific density of plastic particles can vary considerably depending on the type of polymer and the manufacturing process. Typical densities for sand or other sediments are 2.65 g/cm^3. Density values for plastics range from 0.8 to 1.4 g/cm^3, for polypropylene (PP) from 0.85 to 0.94 g/cm^3 and polyethylene from 0.92 to 0.97 g/cm^3 (Hidalgo-Ruz et al. 2012) to be more specific. These large differences in density were utilized while using this method. The lighter microplastic particles were separated from the heavier sediment by mixing the filtrate with the saturated sodium chloride solution. In other studies, however, other solutions like a sodium polytungstate solution with a density of 1.4 g/cm^3, tap water and seawater were applied. Imhof and his research team (2012) used a zinc chloride solution with a density of 1.6 - 1.7 g/cm^3 and Claessens et al. (2013) used a high density sodium iodide (NaI) solution for the separation. According to the investigation of Hidalgo-Ruz et al. (2012) the shaking time varies between the studies. It ranges from 30 seconds up to 2 hours. In this study, a time of 2 minutes was sufficient due to the small sample quantity.

The literature review of Hidalgo-Ruz et al. (2012) found that out of 68 studies 44 focused on microplastics in sediments. However, analysis of the studies' results has a number of limitations. First, often different sampling methods are used, resulting in different units. When sampling beaches for instance, the reported units for microplastic concentrations are expressed per m^2 or m^3. Other studies report the observed abundances of microplastics per weight of sediment sampled (Claessens et al., 2013). This makes comparison between studies difficult. The method most often applied for processing the sediment samples is density separation. 65 % of all studies extracting microplastics from sediments use density separation (Hidalgo-Ruz et al., 2012).

5.9 Results of the separation by chemical technique

After the filtration with the BFC it was easy to see that the particles were mainly stuck in the pores of the 100 µm and 50 µm filter units and that the particles had aggregated there (Fig. 58). In the other filter units of the BFC no particles were found. The distribution exactly fit the size of the fluorescent microplastic particles of 70 - 90µm, which indicated that the mechanical separation of the cascade worked excellently.

Figure 58: Well distributed und aggregated fluorescent microplastics in the BFC

The results of the filtration were examined in the UV light chamber. The contamination of the whole unit, the beaker, the closure and the filter was clearly recognizable (Fig. 59). Because of this, no recovery test was carried out with the BFC, since this distribution was sufficient to establish that no exact quantitative and only qualitative tests could be performed. In the field, this method has nevertheless been successfully applied, since the particle quantity is significantly higher and the mineral and organic residues of the samples did not adhere to the filter unit compared to the fluorescent microplastic particles which adhere extremely well.

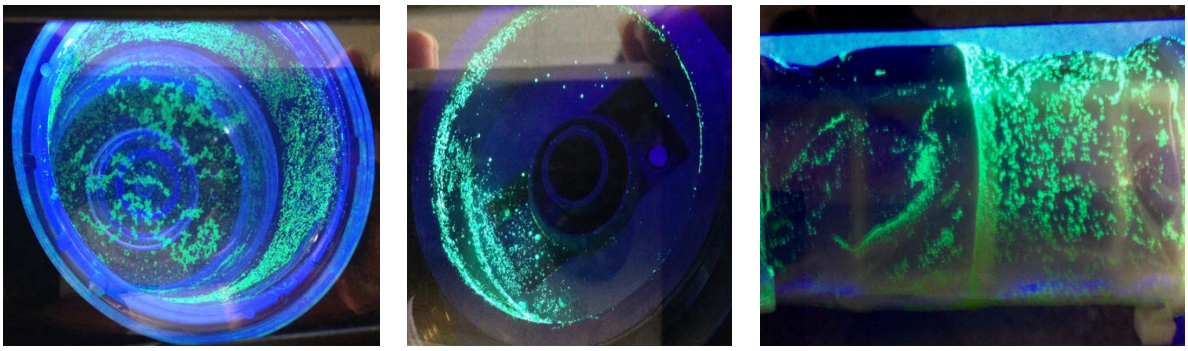

Figure 59: Contamination of one filter unit

By using 500 mg common detergents, 10 ml hexane and 200 µl Triton X-100, the chemical separation was carried out.

The collection efficiency of hexane was given, but too many particles remained in the unit itself, as well as in the filters, and further separation steps had to follow. The common detergents helped in changing the surface tension and the interaction with the water but had no separating effect and were only used as supporting agent for the hexane. The addition of hexane gave rise to another problem, since the mixture of hexane and particles was not identifiable in the Raman spectrometer. The Triton X-100 is a release agent, yet the same problems as with the addition of hexane occurred (Fig. 60). It partly enabled the separation but further separation steps were required.

The aggregation of the particles can be explained through the strong hydrophobic forces between the particles. The hydrophobic forces are stronger than the hydrophilic ones. Hydrophilicity refers only to the interaction with water and neither to the solubility nor to the ability to attract and bind water. Thus, the particles in this case were not hygroscopic. Common detergents are hydrophilic and lipophilic, referred to as amphiphilic, and have been used to weaken the hydrophobic forces between the particles. The detergent did not help separating the particles means, that the added concentration was too small for the hydrophobic attraction between the particles and had to be increased. Hexane is widely used as cheap, relatively safe, largely unreactive and easily evaporated nonpolar nonionic solvent. Triton X-100 is a nonionic surfactant consisting of a hydrophilic polyethylene oxide chain and is part of an aromatic hydrocarbon lipophilic or hydrophobic group. Both have a good separation ability couldn't be used for particle separation in this case and were unusable for further processing of the sample with the Raman. The separation with these two media is not sufficient and thus further separation steps are necessary and make this method very time consuming.

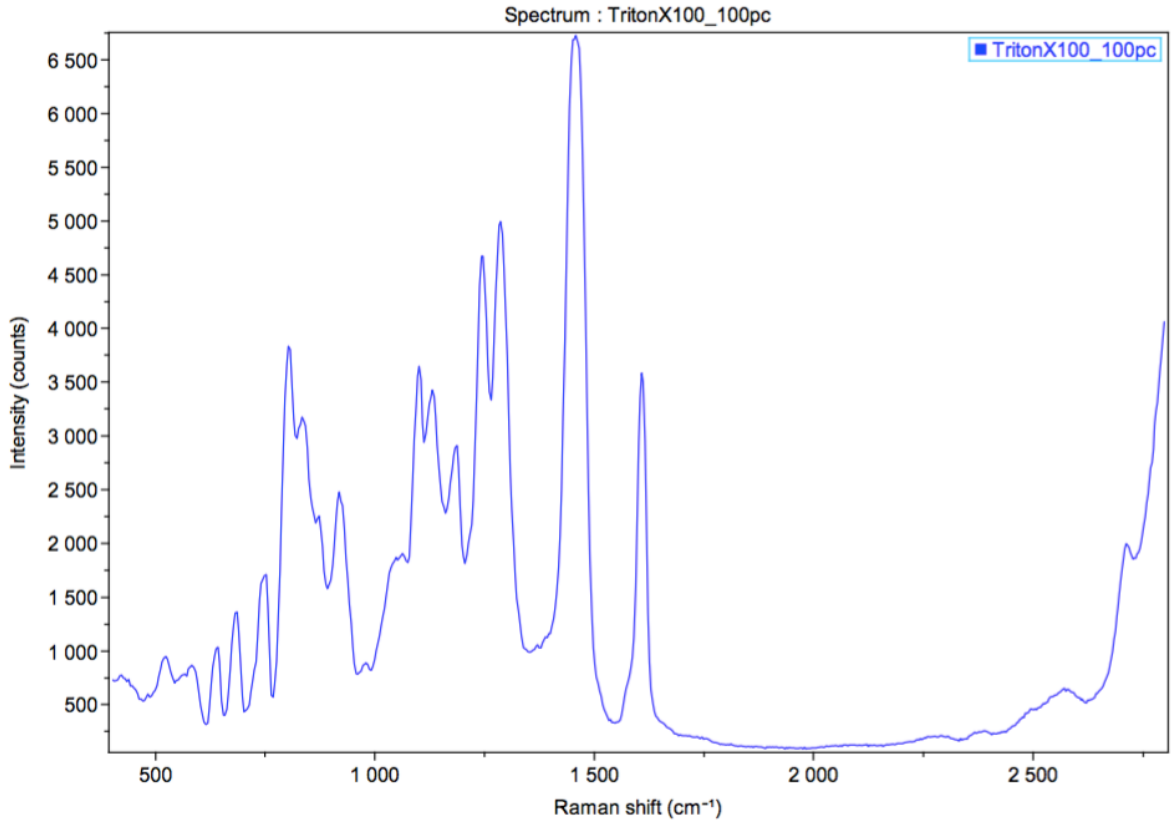

Figure 60: Raman spectrum of Triton X-100 without a visible microplastic spectrum

After the separation attempts with the agents, the separation was performed with a pressure filtration system equipped with 0.45 µm teflon filters and a water jet pump. The separation with the water jet pump failed, just like in the results of the density separation. The water was not sucked up quickly enough and the particles settled in the funnel. It can therefore be concluded that the vacuum pump is recommended for this separation as well as for the density separation.

The results were unsatisfactory; it was not possible to remove all particles from the filter units. During the separation steps a lot of particles were lost and it was not possible to make any valid quantitative statements.

5.10 Results of the separation by ultrasonic

This separation method did not work at all. The particles did not resuspend from the filter very well and, even when the items did, they attached themselves to the beaker, in which the filters were positioned. If they were not in the water, then they were still in the pores of the filters. During the attempt of removing the filters from the glass beakers to filter out the particles in the water, for instance with a vacuum pump, the particles were again attracted to the filter.

It also made no significant difference whether the filters were in ultra-pure water or in a saturated sodium chloride solution. In the salt solution more particles separated from the filter than in ultra-pure water but in both cases the removal of the filter from the beaker was the problem. Attempts to allow the beaker to overflow without removing the filter from the beaker and to catch the particles in another beaker also failed. During the overflow process particles were lost and then stuck to both beakers.

Drying in the oven was considered, but a complete evaporation of the water was not possible and the problem of the trapped particles in the filter and the distribution in the beaker would not be solved.

Moreover, this method did not make it possible to quantify the particles.

This method is a good solution in applications where resuspension of particles is not necessary. However, this method is not suitable for a separation of filter and particles. In addition, further separation steps would be necessary, which would again be time consuming.

5.11 Comparison of the separation techniques

Among the separation techniques of microplastics, the best performance was achieved using the density technique. The second best efficiency was obtained by the separation by chemical technique and the poorest collection efficiency was maintained by use of the ultrasonic separation technique.

5.12 Microplastics recovery test with the SSC

The characteristics of the examined microplastics:

Table 13: Characteristics of microplastics used in this experiment

Characteristics	Commercial Microplastics	Fluorescent Microplastics
Size range [µm]	500-20	75-90
Color	white, blue, pink	green
Shape	irregular	round

After weighing the filters and subtracting the final weight from the initial weight, the percentage of the recovered particles was calculated.

Table 14: Recovery test results (09/26/2016)

Experiment No.	Recovery Percentage [%]
1	92
2	84
3	90

The comparison of the results suggested that particles were lost on the way through the cascade or during the injection process. During the investigation of the cascade with the UV light chamber it could be established that the fluorescent microplastic particles were stuck in small amounts in the filter unit and washing the cascade with tap water over a paper filter showed that commercial microplastics also remained in the cascade.

However, the results also show that only a small portion of the particles, 2.6 mg, 3.1 mg and 1.4 mg, remained in the unit and most of it was retained on the filter.

Figure 61: First assessment of the filters after the test

The evaluation of the filters, visually and with UV light, also confirmed the numerical results.

In experiment No. 1 (Fig. 62), there were more commercial microplastics on the first filter with a pore size of 300 µm than on the second filter with a 100 µm pore size and almost all fluorescent microplastic particles where found on the 30 µm filter.

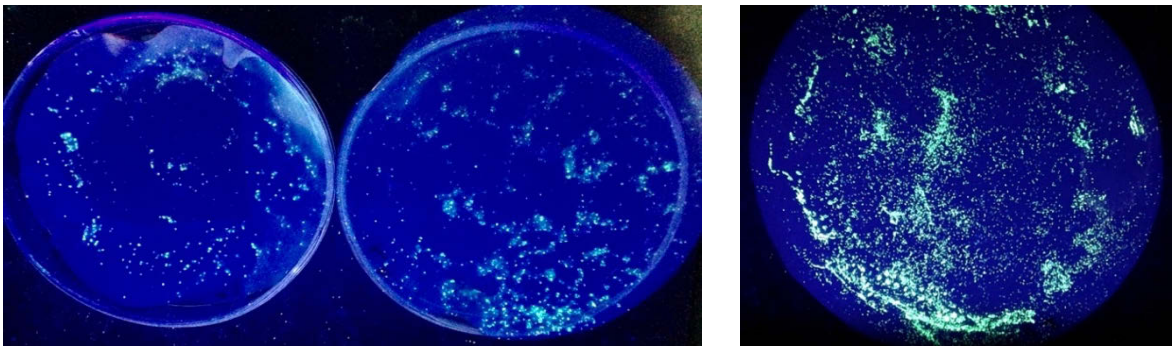

Figure 62: Results of experiment No. 1 (filter pore size 300 µm- left, 100 µm- middle and 30 µm- right picture)

Experiment No. 2 showed that more commercial microplastics were on the second filter than on the first and like in experiment No. 1, most of the fluorescent ones were on the 30 µm filter. However, it is also clear to see that in Experiment No. 2 most of the fluorescent particles have been lost (Fig. 63).

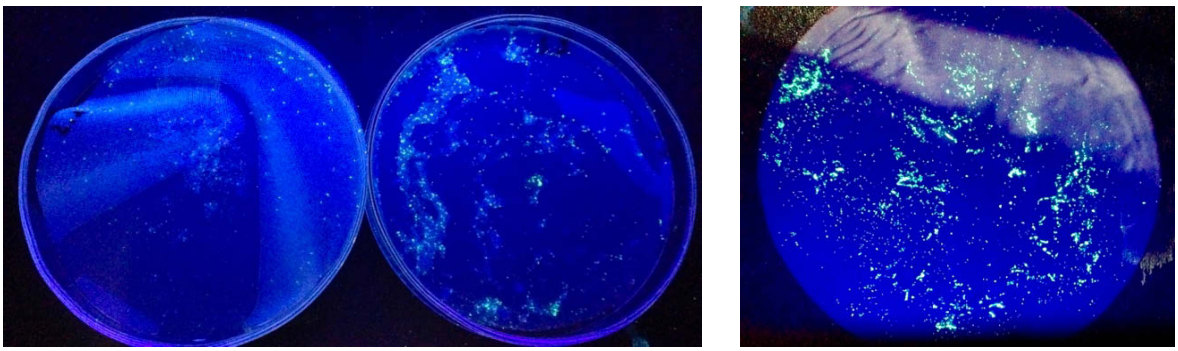

Figure 63: Results of experiment No. 2 (filter pore size 300 µm- left, 100 µm- middle and 30 µm- right picture)

Experiment No. 3 confirmed the results of experiment No. 2. Here, too, most of the commercial microplastic particles were found on the 150 µm filter and less on the 300 µm filter. The appearance of the third filter also confirmed the results, since it had the smallest loss of particles (Fig. 64).

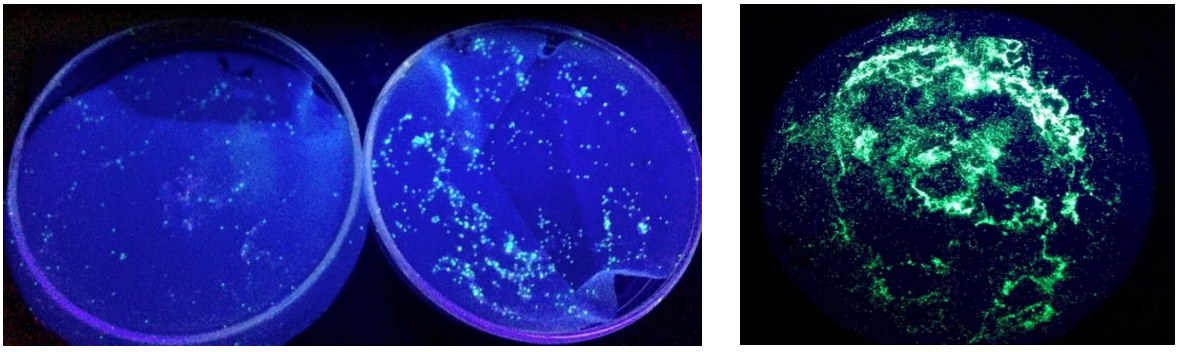

Figure 64: Results of experiment No. 3 (filter pore size 300 µm- left, 100 µm- middle and 30 µm- right picture)

To sum up the results, it seems that most of the commercial microplastic particles used were between 100 µm and 300 µm in size. The fluorescent microplastics with sizes between 75 µm and 90 µm were found on the filter with the pore size of 30 µm, since the particles were smaller than 100 µm.

5.13 Microplastics analysis in bottled water

Table 15 reveals the particle numbers as well as the size fractions of three experiments using water from ten different brands.

Table 15: Plastic bottle experiment results (09/13/2016)

Brand No.	Particles/ 1,5 Liters				Particle Size Fragments/ Granulates [µm]	No./ Particle Size Fibers [µm]	Color of the Particles (fragments, granulates,fibres)
	1. Experiment	2. Experiment	3. Experiment	Mean			
1	3	2	4	3	10- 30	1/ 750	black/red/blue/transparent
2	3	2	0	2	150- 350	-	black/transparent
3	5	0	4	3	10- 50	-	black/transparent
4	3	5	1	3	30- 50	1/ 200	black/transparent
5	3	2	6	4	10- 20	-	black/transparent
6	5	0	2	2	10- 30	-	black
7	5	4	7	5	30- 50	3/ 50-200	red/green
8	9	3	6	6	10-20	2/ 400-800	black/blue/transparent
9	10	8	9	9	20-30	1/ 300	black/red/transparent
10	0	2	1	1	50- 100	-	black

Table 15 clearly shows that not more than ten particles per 1.5 liters could be found per bottle in the experiment. The results range from 0 to 10 particles. The particle size of the fragments and granular materials vary between 10 µm to 350 µm. Fibers were not found in every bottle, but if they were present, they were of the order of 200 to 800 µm. Fragments and granulates were the most abundant items while fibers had comparatively low numbers. Most fibers were red, blue, black or green, while most fragments and granules were black or transparent.

When comparing the brands, only two brands, No.8 and No.9 had slightly higher particle numbers in their bottles, and, even when comparing the experiments within a brand, the particle numbers per bottle were relatively constant. The variability between the brands was greater than within the experiments.

The reason why teflon, (PTFE – polytetrafluorethylen), was chosen as a filter material is obvious. The material is hydrophobic, has a high natural porosity and a good strength, which was a great advantage when removing the particles for further investigation with the Raman.

For a comparison, the clean teflon filters (0.45 µm) were used as a reference (Fig. 65).

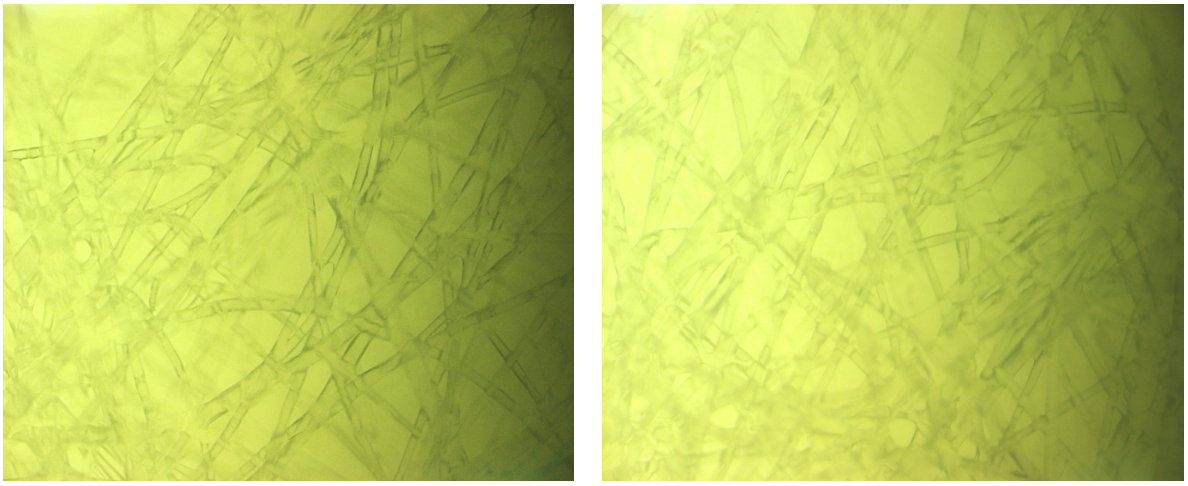

Figure 65: Clean teflon filters

Figure 66 presents the images of the investigated teflon filters (0.45 µm) received by the investigation with the Zeiss microscope and the Raman spectrometer XploRA INV.

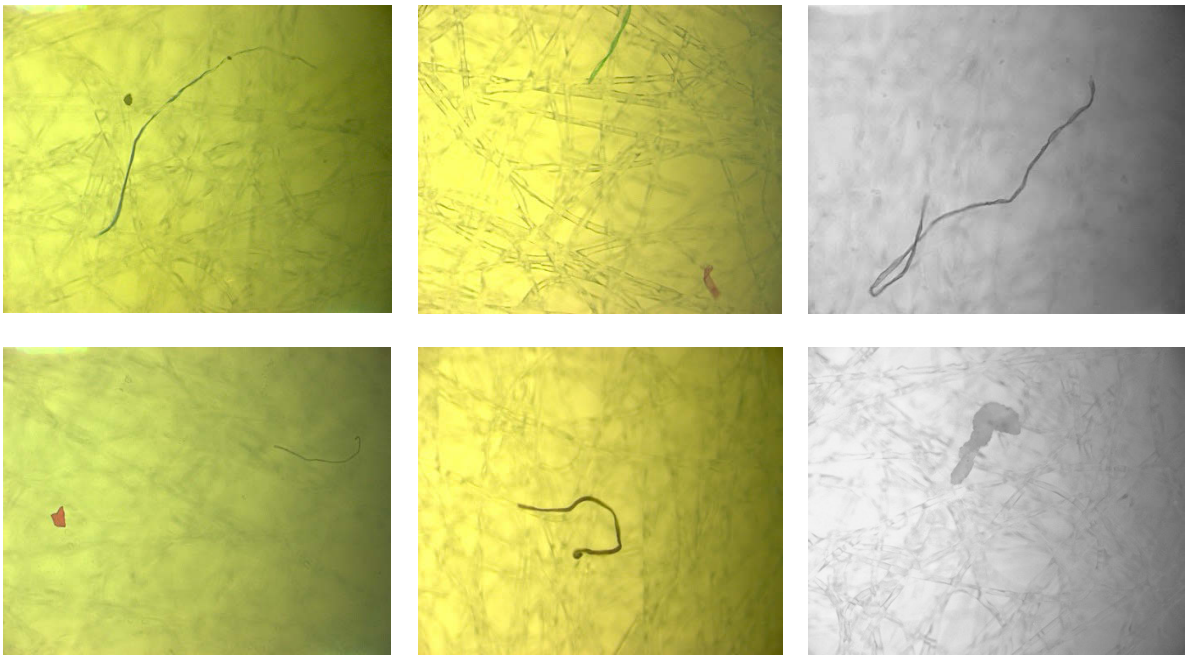

Figure 66: Investigation of the teflon filters with an optic microscope and the spectrometer with a 10x magnification

The investigation of the particles and fibers with the Raman confirmed the assumption that the materials found were microplastics. It was possible to identify polyester (Fig. 67). Polyethylene terephthalate (PET) is a thermoplastic made of polycondensation. PET is used for the production of plastic bottles, which are commonly known as PET bottles. Moreover, fleece materials are also fabricated from recycled PET bottles.

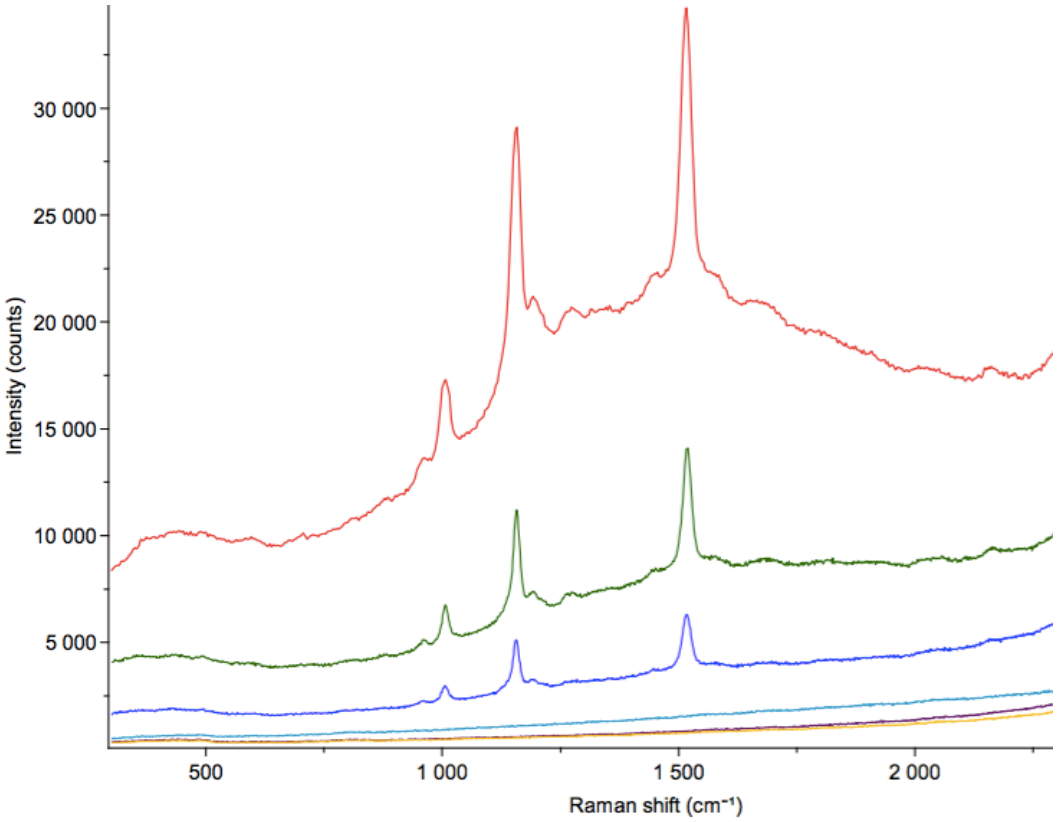

Figure 67: Raman spectra of polyester compared to a reference spectrum (red) of the KnowItAll Raman library

According to a study by Professor Gerd Liebezeit (2014), microplastics were discovered in potable water recovered from PET bottles and 24 different beer brands. A publication conducted by Markt (2014), which is based on the study of Professor Liebezeit, says that it is suspected, that these fibres can be derived from textiles, specifically from fleece materials, which are made of polyester, and reaffirm the results of this study. The researcher concentrated only on the number of particles and not on the size.

The German Farmers' Union engaged an Institute of the Technical University of Munich to investigate Liebezeit's results. No microplastics could be detected, neither in beer nor in water. The bottled water producers also conducted a research. No microplastics were found therein, however, the examination methods have not been published.

No other published studies and thus comparisons on this topic were available.

The small number of the items found in this research does not seem very alarming, but an eye should be kept on this as the use of plastics increases in all areas of life.

5.14 Particle characterization

During the preparation of this characterization, for instance, spectra of peppers were developed to get an idea how lignin or cellulose spectra would look.

Figures 68 and 69 show different species of peppers (yellow and green), but a clear relationship between the species is recognizable by the peaks. Both have peak characteristics at 1000, 1200 and 1500 cm^{-1}.

The spectra resulted from an acquisition time of 4 seconds, 1 accumulation, and a laser intensity of 532 nm. The other parameters were: hole- 500 µm, slit- 100 µm, grating- 600 gr/mm, filter- 10 % and a ten-fold objective.

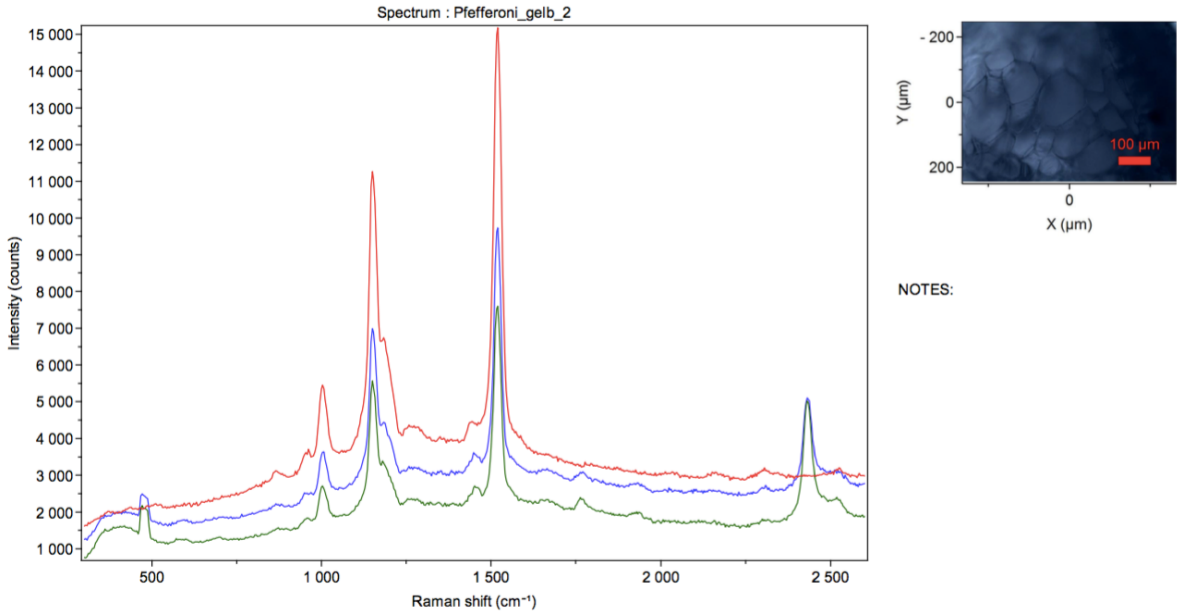

Figure 68: Spectra of a yellow pepper

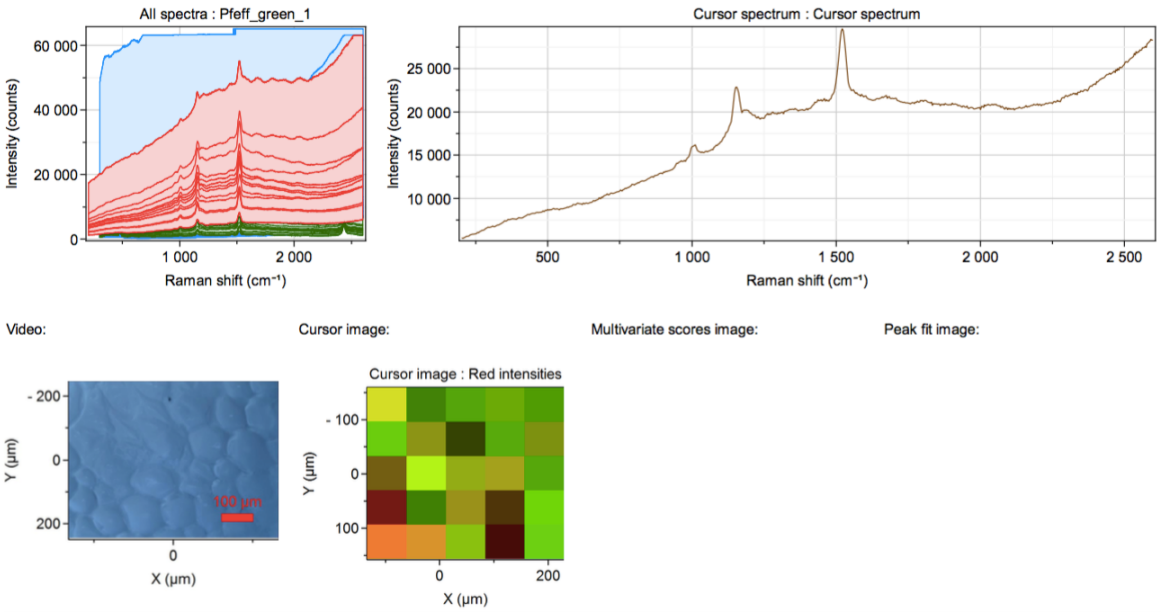

Figure 69: Map of a green pepper

To investigate the main question of this work, all samples were examined with the XploRA INV by Horiba.

Microplastic particles > 30 µm could only be detected and characterized in the Danube water samples of SS 2 in 2015 and 2016.

The concentrations of the particles in the Danube water samples were 2 microplastic particles/132 l and 4 microplastic particles/46 l.

The six particles were 100 - 350 µm in size and were composed of polyethylene. The fact that the particles are polyethylene is not surprising since this is the plastic type that is produced most and therefore the most widespread plastic type in the world.

In 2016, the particles were detected on the first 100 µm filter of the BFC. The particulates could only be discovered after density separation with the separatory funnels and supernatant filtration with the pressure filter system. Without these separation steps, characterization would probably not have been possible.

Microplastic particles are hard to observe, the particles are often hidden in complex meshes or aggregates composed of organic and mineral materials. When there are microplastics in an aggregate, it is possible to see them, providing the right focus level and that a 3D mapping is carried out for half a day.

Moreover, the size of microplastic particles of interest (> 30 µm), is much smaller than those of the aforementioned mineral or organic objects. This fact combined with the infinitesimal number of plastic particles made the Raman mapping extremely difficult. On a certain given focus plane (and in the corresponding confocal Z-volume) it is hardly possible to find microplastics. Time-consuming mapping of large areas and various focus planes did not seem feasible when it comes to establish a relatively fast workflow (protocol) in order to examine microplastics > 30 µm.

The rest of the samples consisted of sand, various minerals and organic materials.

Figure 70 pictures a spectrum of a microplastic particle sampled on September 14, 2016 at SS 2 and separated from a 100 µm by density separation.

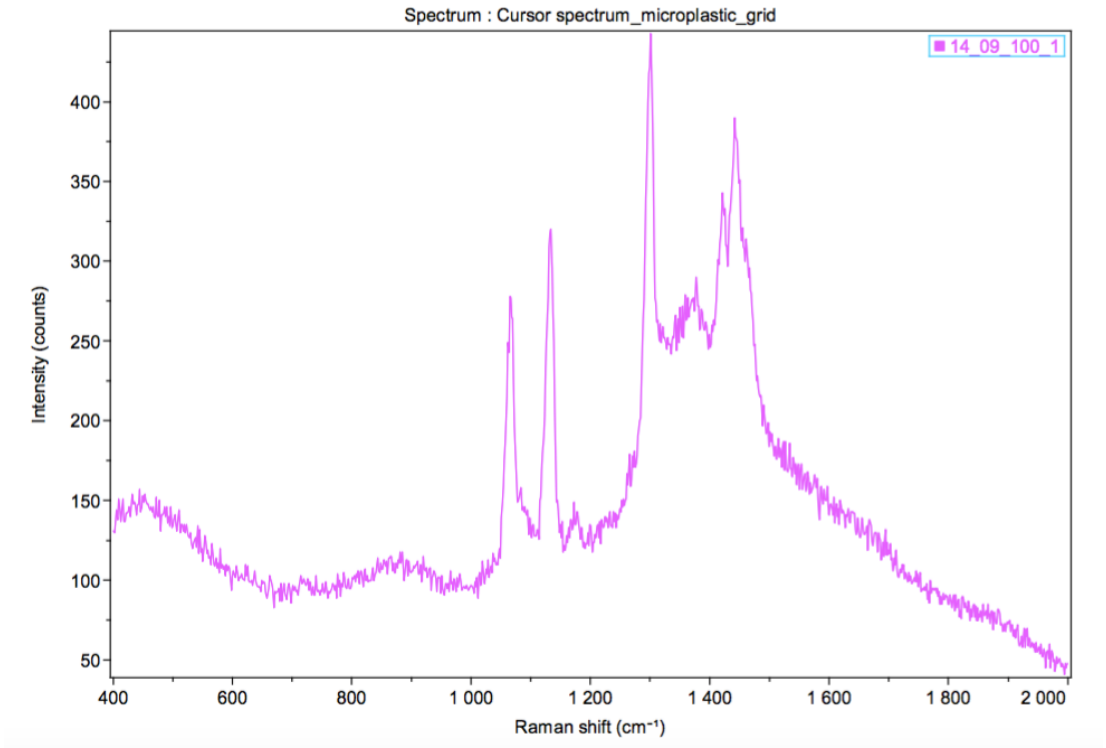

Figure 70: Polyethylene spectrum from the sampling with the Blue Filter Cascade at SS 2 on 09/14/2016

The results were confirmed using the reference spectra. These were derived by microplastics in shower gel. The pink spectrum is from the Danube water samples and is put into relation with the other two spectra. A clear confirmation of the material is possible (Fig. 71).

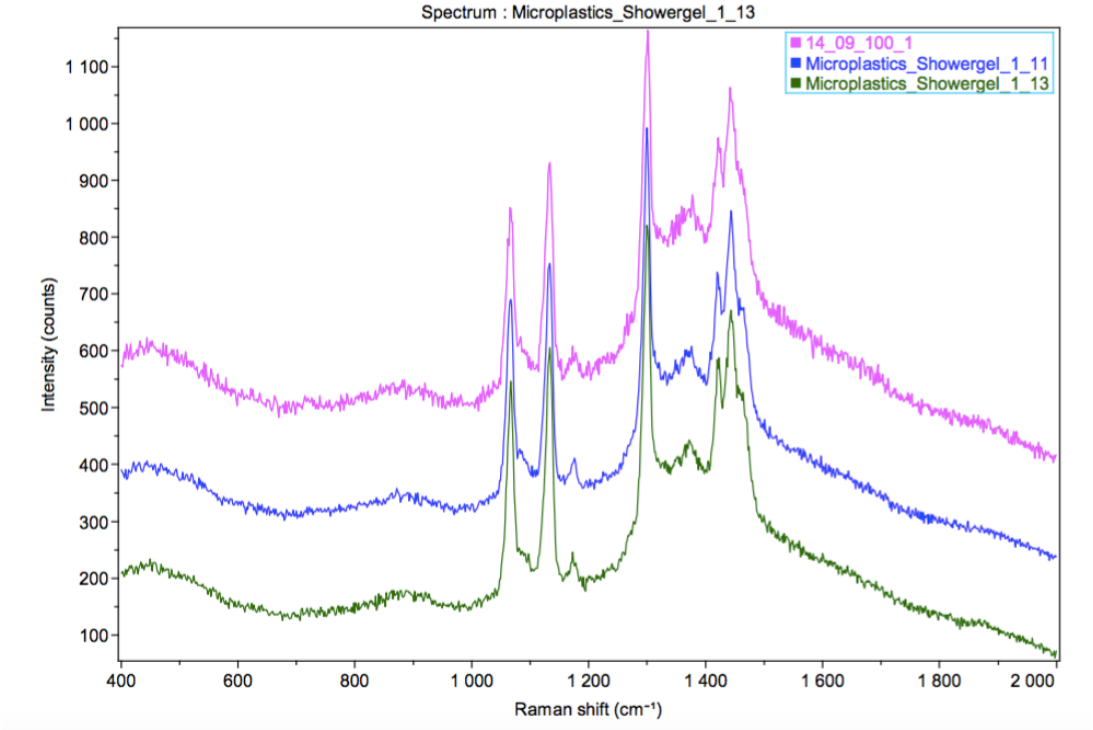

Figure 71: Comparison of the Danube water spectrum (pink) with polyethylene reference spectra

The same procedure was also carried out for the sample in 2015.

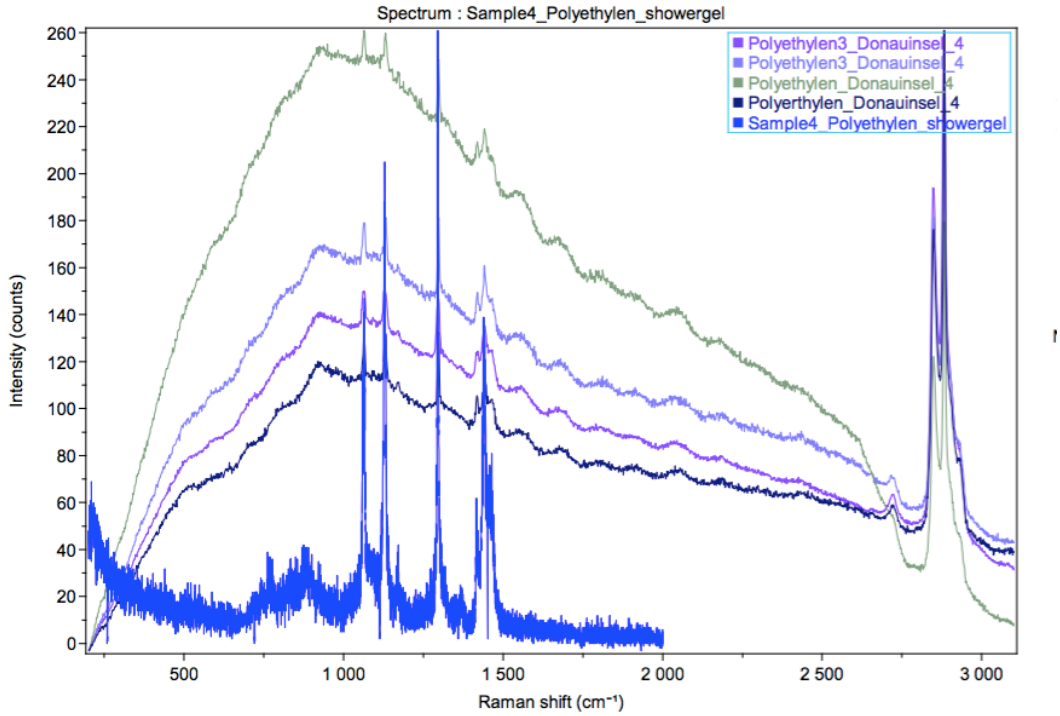

Figure 72: Comparison of the polyethylene spectra from sampling at SS 2 on 10/28/2015 with a reference spectrum (blue)

This map (Fig. 73) of a microplastic particle was created with an acquisition time of 3 seconds, 2 accumulations, and the 785 nm laser. The hole and slit parameters were 100 µm with a grating of 100 gr/mm, a 100 % filter and a ten-fold magnification.

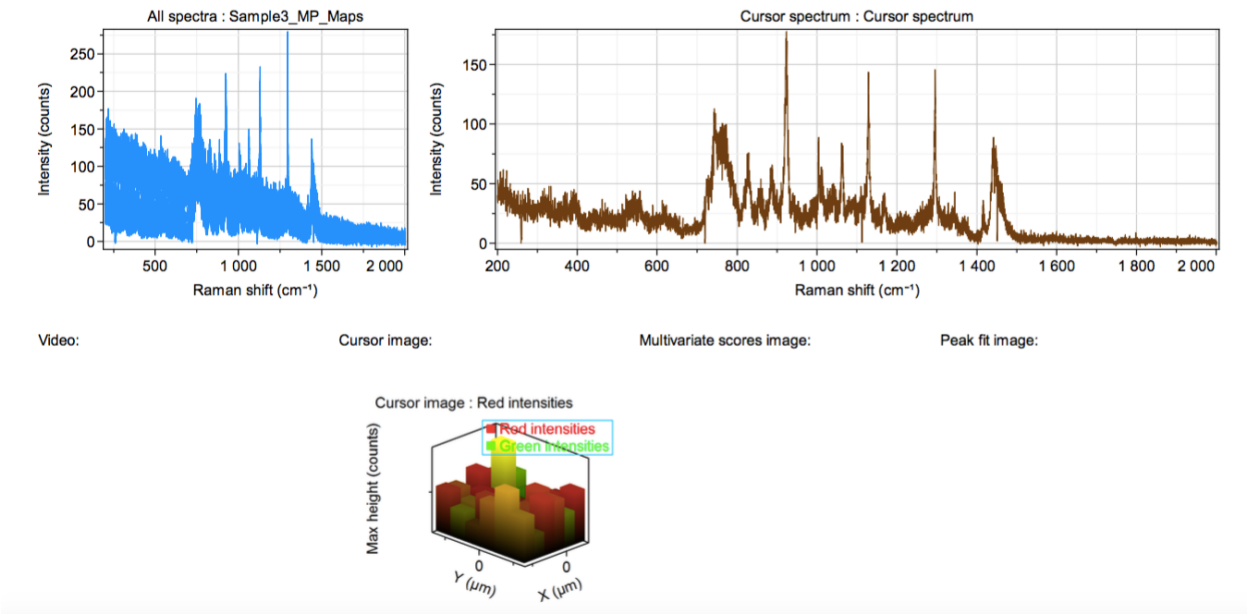

Figure 73: Map of a microplastic particle derived from the sampling with the BFC in 2016

The images of the microplastic particles were also recorded directly with the integrated spectrometer camera. The magnification is the same for all three images (10x magnification) the zoom, Z axis and depth profiling parameter, however, is different for each image.

As can be seen in Figure 74, the shape of the particles is irregular, sometimes sharp edged. In picture 1 a 150 µm, in picture 2 a 100 µm and in picture 3 a 200 µm microplastic particle is displayed.

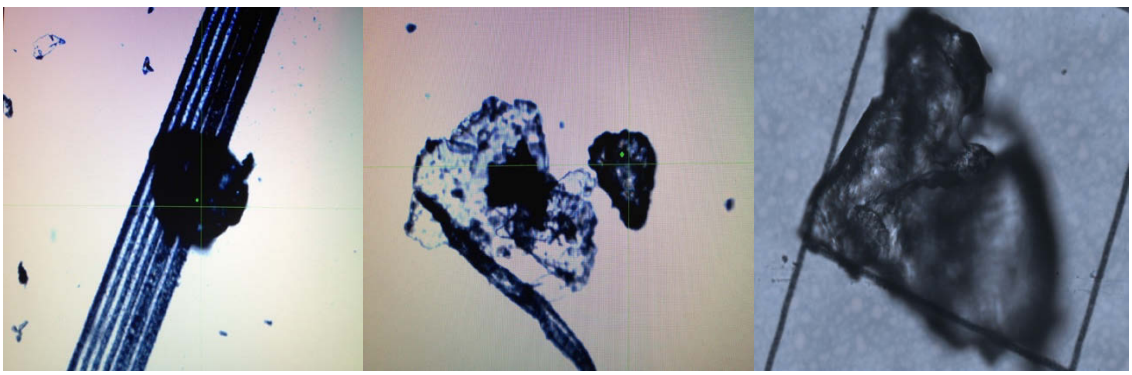

Figure 74: Raman pictures of the microplastic particles and their sizes: Pic.1: 150 µm Pic. 2: 100 µm Pic.3: 200 µm

The main problems in the investigation were the spectrometer itself and the unknown composition of the samples. It was almost impossible to get denoised spectra of any material or to detect microplastics, which might appear individually, aggregated, or covered. The right parameter selection and the highly variable composition of the samples made this evaluation method enormously time consuming.

In pure, highly concentrated samples, the Raman gave excellent results and is a great analytical method. Where very high spatial resolution is required, Raman spectroscopy can also be used excellently. In the case of unknown samples, as they were in this case, it was very difficult to obtain results, as different materials and thus spectra were superimposed.

The spectra were compared with the reference spectra prepared beforehand. An online spectra library, KnowItAll, was also available, but it was not very reliable.

For further investigations using this method, an upright spectrometer would be advantageous. With this Raman spectrometer, the particles always had to be focused through an object, in this case the IBIDI glass dish, an upright one would be able to focus directly on the particles. With an upright Raman, the separation steps would not be necessary since the filters would not be on the image and the spectra. With the XploRA it was always necessary to subtract the background spectrum of the IBIDI glass dish or the filter. However, even with an upright Raman the problem with the superposition of the particles would remain.

Tagg et al. (2015), Fries et al. (2013), Hildalgo-Ruz et al. (2012) and GESAMP (2015) mention the Raman spectroscopy as a reliable identification method for polymers. One thing, however, all these studies have in common. None of these researchers mentioned how the spectroscopy was applied, with which Raman they generated their results, or which plastic types were investigated. They always refer to each other only. But none have used Raman spectroscopy as a proper analytical method, except for samples of known composition (Mucha Torre, 2015). Therefore, no comparative literature for this application is available.

5.15 Conclusion

- The research question of this thesis is: "Are there microplastics in bank filtrated Danube water?" According to the results, this question can be answered with a clear no. There were no microplastics > 30 µm detected in bank filtrate and groundwater. However, microplastics were found in the Danube water samples of both 2015 and 2016. The bank filtrate passage efficiently removed the microplastic particles > 30 µm. The particles were filtered, settled or adsorbed by the changing properties of the soil passage. Nonpolar organic substances are normally eliminated in the infiltration, where the organic fraction is highest. At the solid surface a further microbial degradation takes place. The underground passage readjusted to the bank filtration has a high removal potential against contaminants.

  Due to the occurrence of microplastics in Danube water, this result is very gratifying. There is only a low risk of microplastics entering drinking water, but no particles < 30 µm or nanoparticles were examined and no particles < 100 µm were found.

- The two filtration methods applied in this thesis worked well, however they are still under development. A need for further method development exists, there is also a requirement for standardized sampling protocols. Regular data acquisition must also be carried out. A sampling of the bank filtrate passage and a study of the soil regarding the presence of microplastics would also be interesting to conduct.

- In order to achieve better separation efficiency, further studies dealing with samples of unknown composition would be advantageous.

- Further independent studies on microplastics in bottled water should be performed, since a single study is not sufficient as comparison and confirmation.

- The characterization of the particles with Raman spectrometer XploRA INV was not the best choice for this research. An upright Raman would have been better. This method is not suitable for an analysis of samples of unknown composition. The analysis is enormously time consuming, expensive and too much preparatory work is necessary before the actual application. Other methods of characterization should be considered (e.g. FT-IR).

5.16 Outlook

Microplastics are ubiquitous in marine systems as well as fresh water systems, where they interact with a variety of organisms. As research on microplastics in freshwaters still is in a very young phase, only starting out in the last five years, many questions remain and further research needs to be done. There is a lack of standardized sampling protocols and analytical methods to determine the occurrence of microplastics in fresh water resources including drinking water. Retention during drinking water treatment and potential exposure of consumers to microplastics via drinking water must be investigated. Information on particles < 100 µm is urgently needed. The following steps are seen for the near future:

- development of a methodology for monitoring microplastics in freshwater systems and definition of size fractions relevant for future monitoring programs;
- development of analytical methods for identification of polymers and quantification of particle sizes and particle concentrations;
- development of a sampling protocol for microplastics;
- investigation of microplastics retention during drinking water treatment;
- understanding the degradation behavior including particle lifetimes and ultimate fate in freshwater;
- assessing the potential of rivers to be a source of the microplastics reaching the oceans;
- assessing and understanding microplastic interactions with biota;
- assessing microplastics' impacts on ecosystem services; and
- evaluating the consequences of microplastics for humans.

Globally, freshwater is a decreasing natural resource and is in a fragile state. The process of identifying, monitoring, and dealing with water pollution will be essential.

Attention and research similar to that recommended for microplastics in marine systems is needed for freshwater systems. Progress on this issue requires support from a scientific knowledge base and would benefit from cooperative efforts by the relevant legal bodies and legislative frameworks at the international, national, and regional levels. An implementation of educational programs, the cooperation of urban and rural facilities and, above all, persuasion through practical examples of environments that easily and directly exhibit proper control of waste, will be required.

Finally, the toxicology of microplastics and overall relevance for drinking water production must be evaluated.

6 Summary

Worldwide plastic production (thermoplastics, polyurethanes and other plastics excluding PET, PA, PP and polyacrylic fibers) increased from 230 to 322 million tonnes/year between 2005 and 2015, however the EU share (EU 28 plus Norway and Switzerland) has remained the same (PLASTIC EUROPE, 2016) at approximately 60 million tonnes/year. Microplastics are tiny plastic particles up to 5 millimeters in diameter (UNEP, 2016) although the discussion of size is not yet complete. In the GESAMP study (GESAMP, 2015), particles between 1 nm and < 5 mm are referred to as microplastics, but sampling in surface water is usually carried out with planktonic nets with mesh widths of 330 µm. In the ISO working-draft "Plastics", microplastics (ISO, 2016) are divided into large microplastics between 1 mm and 5 mm and small microplastics < 1 mm (usually in the range 200 - 800 µm). The literature distinguishes between primary and secondary microplastics. Primary microplastics include plastic pellets such as granules intended for further industrial processing and plastic particles used directly or as product additives. Sources of microplastics include cosmetics and personal care products, textiles and clothing (synthetic fibers), traffic (dust from tires), cleaning agents or compressed air cleaning additives and the emissions from plastic producers and processors (plastic resin pellets). A variable proportion of microplastics is released by sewage treatment plants, whereby, depending on the purification procedure, the differences are significant (UNEP, 2016). The secondary microplastics designate minute plastic fragments, which are formed though physical, e.g. UV or mechanical abrasion, biological or microbiological and chemical (e.g. oxidation) processes and come from larger plastic waste fractions. Over time, the structural integrity of plastic waste can be lost and microplastics could become nanoplastics. The smallest microparticles detected in the oceans are 1.6 µm in diameter (Cole et al., 2011).

The risk of microplastics is caused by a combination of persistence of these materials and their possible accumulation in the food chain (Roex et al., 2011). Microplastics occupy the same size fraction as sediments and are potentially available to a wide range of organisms. It can be confused with food (plankton) by fish and is a source of chemicals such as softening agents and adsorbed traces of other chemicals.

Microplastics have attracted special attention in Austria because Lechner et al. (2014) found in a study, that the number of microplastic particles in the plankton nets exceeded those of fish larvae during sampling in the Danube. A study commissioned by BMLFUW showed that plastic particles (> 500 µm to 5 mm) are found in the flowing water of the Danube in Austria. For the entire fraction, the annual freight in Aschach is < 14 t per year, in Hainburg < 41 t per year. However, no plastic particles could be identified in the examined fish (Hohenblum et al., 2015).

Since plastic particles have been found in the Danube, the question arises whether these can also be passed through bank filtration and into drinking water.

In order to clarify this question, this diploma thesis on the subject was carried out at the Institute for Sanitary Engineering and Water Pollution Control.

This research is divided into three main components. The first part describes basic information regarding microplastics. The second part comprises the materials and methods used for the different tests as well as experiments. The third part is focused on the results obtained through the different undertaken analyses to answer the thesis's question, the best separation techniques to recover most of the particles from the samples and characterization with the Raman spectrometer applied for identification of size fractions and polymer types.

The experimental part of this research was carried out at five sampling sites close to the Danube and in the laboratories of the Institute of Sanitary Engineering and Water Pollution Control. The sampling and the experiments were carried out from October 2015 to October 2016.

The sampling was carried out twice using two different methods, the Stainless Steel Cascade (SSC) and the Blue Filter Cascade (BFC). Both were set up and applied only for this research. The SSC was made up of 3 stainless steel filter holders, which were connected by silicone tubes. The filter holders were equipped with nylon filters of decreasing mesh sizes (500 - 1.2 µm) in order to create a cascade and to separate the particles by size. The Blue Filter Cascade consisted of five connected cartridge filters equipped with nylon filters of 100 - 5 µm in size. Both systems could be connected directly to the water supply system, and both had a water meter at the outlet to measure the sampled volume. The methods were used at five sites to extract particles of different size fractions of Danube water, bank filtrate and groundwater.

In addition to sampling, particle counting and turbidity measurements were carried out, both on-site and in the laboratory. The particle counting was executed using an online two-column particle counter from Fa. Klotz and an Abakus mobile fluid. On-site the turbidity was measured with a Sigrist WTM 500 and a manual turbidity measurement was done with the turbidimeter, Turbiquant 1100 IR, by Merck. Microscopic particle size sorting on the filters were made with a common Zeiss microscope.

In the laboratory, various separation and collection methods such as density separation with a saturated sodium chloride solution, chemical separation and ultrasonic separation were performed. The density separation was conducted via100 ml separatory funnels and a saturated solution of sodium chloride (NaCl). The supernatant was collected in a glass beaker and afterwards the particles were isolated with a pressure filtration system. The chemical technique used a solution of common detergents, hexane and Triton X-100, to separate fluorescent microplastics, again a following separation with the pressure filtration system was necessary. During ultrasonic separation, the particles were separated from the filter by ultrasonic waves. The best results were obtained by density separation.

With both sampling methods, recovery experiments with commercial and fluorescent microplastic particles were carried out to investigate quantitative as well as qualitative questions. Moreover, microplastics analyses in bottled water were performed. In this analyses, fibers of polyester were identified using the spectrometer.

Further investigations regarding the separated particles were executed with the Raman spectrometer. With this laser-based characterization method it was possible to identify microplastic particles in two Danube water samples of sampling location 2. The concentrations of the particles in the Danube water samples were 2 microplastic particles/132 l (> 100 µm) and 4 microplastic particles/46 l (> 100 µm). The six polyethylene particles were 100 - 350 µm in size.

No microplastics could be detected in the bank filtrate or in groundwater. Thus, the research question of this thesis can be answered with, no, there are no microplastics > 30 µm detected in bank filtrate.

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8 List of figures

9 List of tables

10 Affirmation

I certify that the master thesis was written by me, not using sources and tools other than those quoted and without use of any other illegitimate support.

Furthermore, I confirm that I have not submitted this master thesis, either nationally or internationally, in any form.