

EFFECTS OF HYDROPEAKING ON BENTHIC ALGAE IN ALPINE RIVERS

Master thesis

**In partial fulfilment of the requirements
for the degree of
Diplomingenieur**

submitted by:

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Abstract

In Austrian rivers there are about 5200 hydropower plants in operation. Alongside the impacts by the construction of the stations, the operation, like hydropeaking, affect different components of riverine ecosystems causing severe ecological problems. As benthic algae are the primary energy resource in rivers, the aim of this paper is to investigate the effects of hydropeaking on the biomass, productivity and composition of benthic algae in two different river habitats. Therefore, three different locations alongside the river –Salzach-, which is influenced by hydropeaking, and two alternate reference locations along a non-affected tributary, the –Lammer-, have been investigated in summer and late autumn 2018. The results for the shore line habitat showed a significant lower chlorophyll-A content in the –Salzach- river during autumn. Also for the photosynthesis activity parameters the –Salzach- had significantly differences between the seasons. For the permanent immersed habitat, the periphytic biomass content was significantly higher in the –Lammer-. The photosynthesis parameters showed a significant higher value for the summer in the -Salzach-. In case of the elemental composition, both habitats showed a significantly higher value for the C/N distribution in the –Salzach-, whereas the phosphor was not significantly different between rivers. The pigment composition of the algae showed no significant differences in algae divisions between the two rivers, but a shift between the two seasons in both habitats.

Zusammenfassung

In den Flüssen Österreichs sind ca. 5200 Wasserkraftwerke installiert. Neben den Auswirkungen durch die Konstruktion der Bauwerke, ist es vornehmlich die Betriebsart, wie zum Beispiel der Schwallbetrieb, der die Gewässerorganismen beeinflusst und schwerwiegende ökologische Probleme hervorrufen kann. Da benthische Algen die Primärproduzenten in Flüssen darstellen, ist das Ziel dieser Masterarbeit die Effekte von Schwallbetrieb auf die Biomasse, Produktivität und Zusammensetzung der benthischen Algen näher für zwei Lebensraumbereiche zu erforschen. Dafür wurden drei verschiedene Standorte entlang der Salzach, die

geschwallt wird, mit zwei unterschiedlichen Standorten an der ungeschwallten Lammer (Referenz), ein Zufluss der Salzach, im Sommer und Herbst 2018 miteinander verglichen. Die Ergebnisse für die Uferzone zeigten einen signifikant geringeren Chlorophyll-A Gehalt der Salzach während des Herbstes. Auch der Photosynthese Parameter (IK-Wert) zeigte einen signifikant geringeren Wert für die Salzach über die Jahreszeiten. In der permanent untergetauchten war die Biomasse in der Lammer signifikant höher. Die Photosynthese Parameter wiesen im Sommer einen signifikant höheren Wert in der Salzach auf. Im Falle der Elementzusammensetzung zeigten beide Zonen einen signifikant höheren Wert bei der C/N-Verteilung in der Salzach, wohingegen Phosphor keinen Unterschied zeigte. Bei der Pigmentzusammensetzung der Algen wiesen beide Zonen keine signifikanten Unterschiede zwischen den zwei Flüssen auf. Allerdings kam es zu einer Änderung der Zusammensetzung bei der Betrachtung der Jahreszeiten, Sommer und Herbst.

1. Introduction

The limited availability of natural resources and fossil fuels, combined with an expanding world population, is leading to a massive rise in energy demand. This requires innovations and new solutions in order to gain renewable resources for energy consumption. One of the most important renewable resources used is hydropower (Bejarano et al. 2018). Besides the positive aspects of this energy resource, to reduce greenhouse gas emissions and be independent of fossil fuels, major and severe ecological problems of aquatic ecosystems have to be faced, especially due to hydropower plants (Wagner et al. 2015).

In Austria, about 55% of electricity is generated through hydropower plants (E-Control 2018). 2882 hydropower facilities are linked to the electricity grid. If the small scale plants for the own consumption are taken into account too, Austria has about 5200 hydropower plants generating energy. To cover the basic load of daily energy demand, two third of the hydropower plants are operating as run-of-river plants (Wagner et al. 2015). Run-of-river plants operate continuously, using small gradients, throughout the day. In most cases it is not possible to change the discharge of a run-of-river plant in dependency of energy demand. But in some hydropower stations the opportunity to store water during a time of little energy demand as a reserve for peak demand is possible (Wasserkraftverband Mitteldeutschland e.V. 2019). To meet these requirements in the grid system, the other one third is run as storage or pumped-storage facilities (Wagner et al. 2015). Storage plants use the great drop in height between the impounding reservoir, which is fed by the water of a dammed up natural river, and the hydropower station at lower altitude. In case of a pumped-storage facility the impounding reservoir is filled by the water pumped upwards from a lower leveled basin (Wasserkraftverband Mitteldeutschland e.V. 2019).

Major ecological problems exist due to the involved regulations of the river itself. Through the implementation of hydropower plants in the river channels physical disturbances are likely to occur (Wagner et al. 2015). On the one side by constructional interventions, for example, the straightening of the river bed and the implementation of weirs and dams (Schmutz and Moog 2018). And on the other side

by the operation system of hydropower stations impacting flow velocity, water temperature and water level up and downstream the dam and the hydropower station facilities (Schmutz and Moog 2018).

By the implementation of a hydropower plant, the movement of organisms are disrupted and flow regimes, water temperature and biochemical cycles are modified (Wagner et al. 2015). Critical components like magnitude, frequency, timing of the flow regime and the duration of high flows, impairs for example distribution of species, their reproduction and survival in the river (Bejarano et al. 2018). Here reduced floodplain inundations and prolonged low flows can result in decreased growth rates or mortality as well as declined or eliminated areas of riparian plant cover (Table 1). Furthermore, the sudden flood recession can lead to a failure in seedling establishment (Zeiringer et al. 2018)

1.1 Benthic algae

Algae, which are attached or associated with the river substrate, are referred to as benthic algae (Figure 1). Those organisms include different size classes ranging from microphytobenthos to metaphyton. Therefore the sizes range from less than five micrometer to up to meters in length (Likens and Stevenson 2009).



Figure 1: Appearance of benthic algae (Vidyasagar 2016)

Benthic algae dominate the shallow sections of clear-water rivers, where they prefer to live in shaded areas (Horne and Goldman 1994).

The most common benthic algae found in freshwaters are cyanobacteria, green and red algae and diatoms (Stevenson et al. 1996). Pennate diatoms and cyanobacteria gliding over the substrat, filamentous green algae cannot move. The classification into various algal divisions is related to the pigment composition. Carotenoids and chlorophyll are the main groups of algal pigments (Horne and Goldman 1994).

Benthic algae are the primary energy resource for food webs in river systems. Due to the fact that benthic algae sticks to substrata and thus overgrow sediments and sand, which are than less vulnerable to be mobilized if current rises. Benthic algae can be seen as stabilising agents of these ecosystem. Due to overgrowing of substrate, benthic algae create a vital habitat for other organisms, like for example small invertebrates (Stevenson et al. 1996).

1.2 Environmental and biotic factors affecting benthic algae

Benthic algae are controlled by abiotic (discharge, water depth, grain size of sediments and shear stress) (Bruder et al. 2016) and biotic factors (competition, diseases, bioturbation and predation) (Likens and Stevenson 2009). But, if these sensitive balances of exterior influences get disturbed, the performance of the species are limited (Likens and Stevenson 2009).

The main changes in the natural environment of benthic algae are driven by alterations in hydrology (Polst et al. 2018). Algae are affected by physical disturbance, which include for example desiccation events, freezing, substrate movement and sudden increases in hydraulic forces, light and heat (Larned 2010). Greater differences between high and low flow conditions can lead to a loss in sensitive species leading to a reduced species richness itself (Table 1) (Zeiringer et al. 2018). In addition chemical parameters, like nutrients and pollutants, can be affected by flush events (Meile et al. 2005).

Table 1: Ecological responses to alterations in flow regime (according to Zeiringer et al. 2018)

Flow component	Alteration	Ecological response
Magnitude	Greater magnitude of extreme high and/or low flows	Life cycle disruption Reduced species richness Altered assemblages and relative abundance of taxa Loss of sensitive species
Duration	Decreased duration of floodplain inundation	Reduced growth rate or mortality Altered assemblages Reduced species richness
	Prolonged low flows	Reduction or elimination of plant cover Reduced species richness
Rate of change	Rapid changes in river stage	Drift (washout) and stranding
	Accelerated flood recession	Failure of seedling establishment

1.3 Hydropeaking

Hydropeaking is defined as an abrupt change in hydraulic conditions, like flow velocity, bottom shear stress and water depth (Schülting et al. 2016). Therefore, hydropeaking events are subdivided into three categories. Depending on duration, frequency and magnitude of the discharge, it is distinguished between a low base discharge on the first hand. In that case no electricity is generated. The second phase is characterized by a sudden increase or decrease of discharge, due to electricity generation. A high peak discharge is the third possibility, in which the energy generation is at its maximum (Bejarano et al. 2018). The consequences in all three cases are variations in the wetted river morphology, water quality, temperature and hydraulic parameters (Bejarano et al. 2018).

During hydropeaking events, different parts (morphology, runoff regime, water quality) of the river are affected (Figure 2). The main impact happens in the runoff regime. Due to the sudden change in discharge as well as in velocity, changes in the water quality are possible. For example rapid alterations in temperature or an increase in suspended load could lead to such a variation (Meile et al. 2005). With an increased amount of suspended sediments, a higher rate of abrasion of benthic algae could be expected, especially diatoms which are most likely to be damaged by suspended solids (Cashman et al. 2017). Even the morphology of a river can be influenced, if the shear stress is so great, the river bed surface itself starts to move. Another option could be bank erosion. Water depth, wetted cross section and flow velocity create the appearance of each individual river, which is characterized by its different habitats. Due to the fact that these three parts are increased by hydropeaking, also other habitats, which were not affected previously, are then influenced and thus, be altered substantially (Meile et al. 2005).

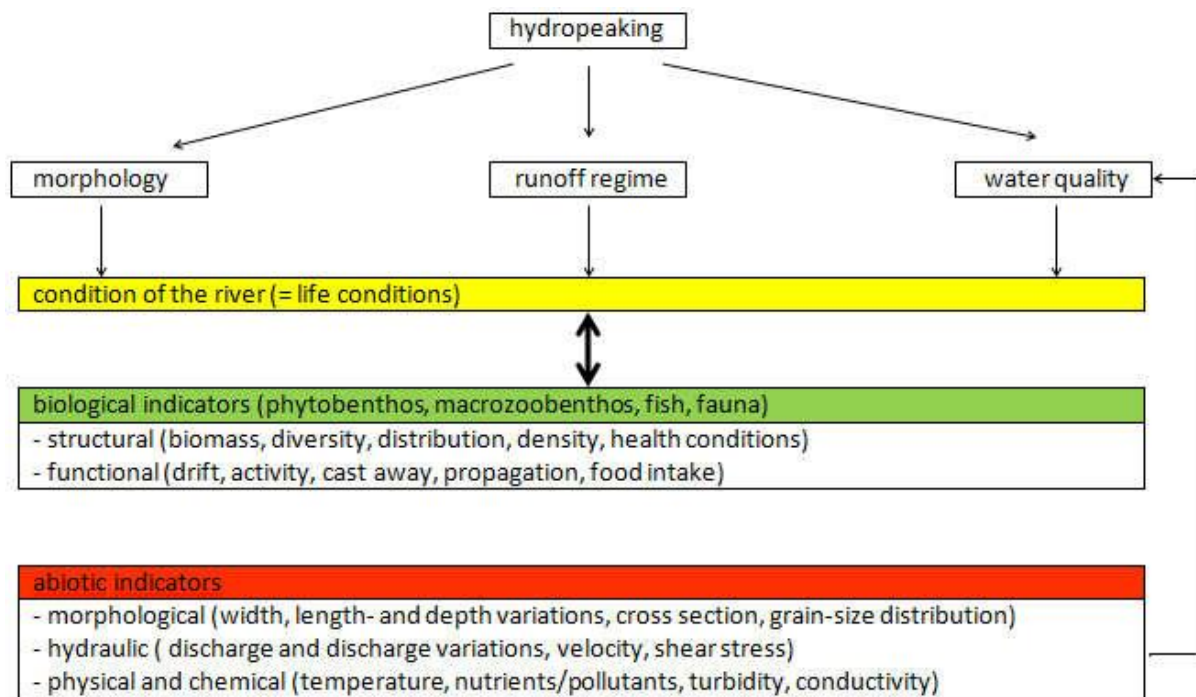


Figure 2: Influences on river conditions due to hydropoaking (according to Meile et al. 2005)

1.4 Hydropoaking effects on benthic algae

Despite natural flood events, hydropoaking lead to different pressures on river biota. Natural floodings are stochastic events leading to far-reaching effects on river ecosystems. But usually their frequency is lower (not every day or week, more seasonal). That is the reason why river organism hardly adapted to the frequent disruption by hydropoaking events (Meile et al. 2005). The occurrence of hydropoaking events can occur daily or more frequent, depending on the actual energy request (Cashman et al. 2017). Frequent, sudden and short-term fluctuations in water discharge and water level downstream of hydropower stations are results of hydropoaking (Bejarano et al. 2018). Consequently there is not enough time for benthic algae to recover. The actual recovery time of periphyton after such hydropoaking events would might take up to a couple of weeks (Cashman et al. 2017).

It is already known, that a great magnitude of high and low flow conditions leads to a loss or at least a reduced species variation. Furthermore, the life cycle in the surrounding ecosystem is disrupted and causes alterations in the assemblage of organisms as well. Drift and stranding occur, due to rapid changes in the river stages and prolonged low flow situations, which favour the elimination of plant cover itself

(Zeiringer et al. 2018). However, compared to a natural flood event benthic algae, as well as other aquatic organisms, cannot react that fast to the new conditions. Additionally, the water level during the winter months is in general, at the lowest point. That means under natural circumstances the winter period is identified with a steady and low discharge. But especially throughout winter, the hydropower is used quite intensively, which is even more challenging for the biota in a river (Meile et al. 2005). Hydropeaking, as an operation rule, can lead to a change in the sediment transport patterns as well as the composition of algae (Hauer et al. 2017). Furthermore, the thermal regime and the chemical composition of the river water can be affected (Hauer et al. 2017). A variation in habitat quality and distribution can also affect the vegetation in the riparian areas of the river system, which is an additional impact of hydropeaking events (Hauer et al. 2017).

With the onset of these physical disturbance, as mentioned in the previous part (chapter 1.2 and 1.3), algae start to respond to it, relative to resistance or susceptibility. For example, changes in metabolism, algae biomass loss and taxonomic composition (Larned 2010). It is expected that diatoms and cyanobacteria are the more common algae group within a community exposed to pulsed flow conditions, because hydropeaking events favour the colonization for smaller and clinging algae like for example diatoms, which are less sensitive to shear velocity (Bondar-Kunze et al. 2016). An additional rapid rise in water discharge can cause drift or detachment of benthic algae (Bruder et al. 2016). That is one possible reason why the quantity of filamentous algae and epilithon are reduced downstream a hydropower station (Kokavec et al. 2017). Also the increased amount of suspended sediments, due to hydropeaking, tends to lead to the abrasion of benthic algae. Higher amounts of suspended solids, caused by increased shear stress due to a hydropeaking event, leads to higher turbidity in the river. This reduces the growth of benthic algae (Bruder et al. 2016). Furthermore, it can cause limitations in the production of biomass, because of less light availability (Hall et al. 2015).

Depending on the location of benthic algae assemblages, it can be differentiated between two habitats. The shore line, this is located right at the edge of the water line and the permanent submersed zone, which includes the deeper zones of the river. In these two habitats, various impacts, due to hydropeaking events, could affect benthic

algae. In the shore line the benthic algae are faced with desiccation stress (Eixler et al. 2006) by the dry out after a hydropeaking event. In contrast, the algae in the permanent submersed zone have a higher possibility of abrasion due to higher flow velocities and turbidity (Meile et al. 2005).

1.4.1 Photosynthesis and light

The light availability is fundamental for benthic algae to gain energy to convert inorganic compounds into biomass via photosynthesis (Stevenson et al. 1996). Phototrophic organism like benthic algae photosynthesize at a wavelength of 400 to 700nm (Stevenson et al. 1996). Photosynthesis is divided into two different parts.

On the one hand, there is the light reaction, which via the use of light energy by photosynthetic pigments produces Adenosin triphosphate (ATP) and Nicotinamide adenine dinucleotide phosphate (NADPH). These responses are not sensitive to temperature, because it is photochemical driven. During the light reaction it is distinguished between photosystem I (PS I), which is related with the reduction of NADP and photosystem II (PS II), which splits water molecules. Both photosystems have special molecules of chlorophyll-A, which are responsible for the electron transfer. Additional light-absorbing molecules, which absorb and conduct light energy towards the reactions centers. The PS I reaction center gets its energy mostly from chlorophyll-a molecules.

On the other hand there is the dark reaction which can also occur under the absence of light. In this process the reducing power of ATP and NADP and the chemical energy is used to reduce CO₂ to hexose.

The light variation depends on seasonal changes in solar insulation and cloud cover (Hall et al. 2015). Additionally, the riparian forest alongside the river also influences the actual light availability (Décamps et al. 2009). Together with the water depth, it regulates the light environment underwater (Hall et al. 2015). As light availability is crucial to keep up the life cycle and perform photosynthesis, periphyton is at its minimum during the winter months (Horne and Goldman 1994).

1.4.2 Nutrients and benthic algae

With the variation of light intensity and temperature, the nutrient uptake and growth is also affected. Phosphor and nitrogen are nutrients most likely to restrict growth (Stevenson et al. 1996). In general, phosphate and nitrogen are the macronutrients factors controlling biomass development.

Benthic algae, which are attached to the river bottom, are influenced by the water motions in streams and rivers. Therefore, the physicochemical environment surrounding the algal cell is under permanent change. Thus, the availability of dissolved nutrients and gases varies in the setting. In case of nutrient limitation, it is important to differentiate between the growth rate and the yield. By the term growth rate, the rate of biomass production is meant. Whereas yield quote the amount of produced biomass. The latter one is influenced by the total quantity of available nutrients, despite the first one, which is determined by the supply of the limiting nutrients (Stevenson et al. 1996). The influence of nutrients on benthic algae are probably not that strong, because the effects of varying light conditions, physical disturbance, due to fast flowing water or grazing, could be more important than nutrient limitation (Stevenson et al. 1996).

1.5 Research question and hypothesis

There are many scientific papers, which have analyzed the effects of hydropeaking on aquatic environments. But they mainly focus on fish or macroinvertebrates, which are affected by higher drift rates, reduction of macroinvertebrate biomass, change in fish behaviour or stranding of fish larvae and juvenile, which was detected for example in Holzapfel et al. (2017), Schülting et al. (2016) and Hauer et al. (2016). Kokavec et al. (2017) found out that, macroinvertebrates respond with a shift in community composition and species density due to changing hydraulic regimes.

Due to the lack of knowledge concerning the effects of hydropeaking on benthic algae, these need to be analyzed in more detail.

As mentioned in the previous sections, induced variations in the hydraulic conditions, for example discharge variations, increased velocity and shear stress (Meile et al.

2005) leads to additional stress for benthic algae, due to increased turbidity, chemical composition and habitat quality by alterations in hydrology (Hauer et al. 2017).

To address the knowledge gap about the possible links between hydropeaking events and benthic algal development, the thesis compares river sections with a distinct hydropeaking stressor and one without. Additionally, it is differentiated between two habitats within the river channel. The potentially impacted habitats are the shore line area on the one side, and the permanent immersed zone on the other side, which were compared seasonally between summer and autumn as well. It was hypothesized that:

1.

1.1 The biomass production in the shore line area will be reduced as benthic algae are affected by drought stress, due to frequent drying – wetting events related to hydropeaking. During dry conditions algal cells are not active, thus reduced biomass development is expected.

1.2 For the benthic algal composition, a reduction in algal divisions is expected due to the onset of desiccation in the shore line habitat, once the area falls dry. These frequent exposed zones should be dominated by sheathed cyanobacteria and non-mucilaginous diatoms (Larned 2010). As a result of that, green algae should be reduced due to prolonged drying (Ledger et al. 2008).

1.3 The photosynthesis activity is presumed to be decreased in the shore line habitat. The small changes in water depth, caused by hydropeaking changes the light availability and lead to a reduced photosynthesis activity (Bruder et al. 2016).

1.4 It is expected that in the shore line habitat the nutrient content will be reduced due to limited access to dissolved nutrients if the water level drops and desiccation starts.

2.

2.1 In the permanent immersed habitat, the algae are confronted with increased flow velocity, during a hydropeaking event. Based on Bondar-Kunze et al. (2016) a very high flow velocity has a negative influence on benthic algal biomass. Depending on the substrate size, which the benthic algae are attached to, the possibility rises that smaller grain sizes start to move, due to a higher flow velocity and shear stress

(Meile et al. 2005). If turnovers happen, the benthic algae maybe have less recovery time to establish again after a hydropeaking event. Both impacts are controlled by peak-discharge and can be the reason for a reduced biomass production in this habitat.

2.2 The abrupt rise in discharge, with the resulting changes in water depth, flow velocity and turbulences affect the algae in the permanent immersed habitat. Therefore, a higher percentage of cyanobacteria and diatoms are expected. Based on Bondar-Kunze et al. (2016) diatoms and cyanobacteria dominated the experimental flume with hydropeaking treatment. In contrast green algae were higher in the treatment without the hydropeaking effect and should dominate the river, which is not affected by hydropeaking.

2.3 Turbidity, caused by hydropeaking, will reduce the photosynthesis activity in the permanent submersed habitat (Bruder et al. 2016).

2.4 For the permanent immersed habitat it is presumed that due to the higher flow velocity, the dissolved nutrients are flushed away and the algae are not able to store them fast enough.

3.

3.1 Furthermore, it is supposed that in summer the biomass production is higher in both river habitats compared to autumn, because the metabolism of the benthic algae slows down (Horne and Goldman 1994).

3.2 In addition, it is supposed that in autumn the river, influenced by hydropeaking shows an even higher amount of diatoms and cyanobacteria in both habitats (shore line and permanent immersed) due to a higher intensity of hydropeaking events (Meile et al. 2005).

3.3 In summer the photosynthesis activity should be higher in both river habitats in contrast to the autumn, because during the winter month hydropeaking events are increased due to a higher demand of energy (Meile et al. 2005).

2. Site description

2.1 The river “Salzach”

The river “Salzach” has its source in mountains of the south-west part of Austria, the so called „Kitzbühler Alpen“. The alpine river rises at a sea height of about 2300 m. After a total distance of around 226 km the “Salzach” flows into the river “Inn” close to the village “Braunau”. The catchment size amounts to 6727 km². Above all, a water quality class of two was assessed (Bundesministerium für Land- und Forstwirtschaft 2009).

As a typical mountain river, the “Salzach” has its highest waterlevel during the summer month, due to glacier melting in the mountains. Due to the early regulation measures, caused by enlarged settlements, traffic- and infrastructural institutions, the river lost almost all typical characteristics, like a meandering watercourse. With the aim of flood protection the river bed of the “Salzach” was straightend and its banks were fixed by dams. The mean discharge ranges from 26 m³/s around the area of “Mittersill” in the upper reaches of the river to 240 m³/s at the town “Oberndorf” at the lower reaches of the river system (Bundesministerium für Land- und Forstwirtschaft 2009).

Like many other alpine rivers, the “Salzach” is used for electricity generation. Due to the use of hydro-storage stations to gain electricity, the water level can fluctuate up to 50 cm (Bundesministerium für Land- und Forstwirtschaft 2009). One of the five hydropower stations positioned in the middle courses, is located between “Werfen” and “Rauwerfen”. This hydropower station generates 76.5 million kWh a year, which provides energy for 22000 households on an average basis. The run-of-river plant produces about 50% of the total energy during the summer months (Mai until August). From October until March around 23 GWh of energy is generated (VERBUND Hydro Power GmbH 2018). In this site, a general water current of about 114 m³/s existed (Bundesministerium für Land- und Forstwirtschaft 2009).

2.2 The river “Lammer”

The origin of the river “Lammer” is located at the southern edge of the “Tennengebirge”, which the river surrounds in a wide curve (Fröschl 2019). The “Lammer” flows through the valley of “Abtenau” before it enters the canyon of the “Lammeröfen”, a hotspot for watersports like kayaking (Austria Forum, das Wissensnetz 2016 and Fröschl 2019). After about 40 kilometers the “Lammer” flows in the river “Salzach” southwards of the village “Golling an der Salzach”. In this area gravel bars dominate the morphology of the river (Fröschl 2019). The river “Lammer” is partly regulated (BMLFUW 2015). A weir is implemented in the watercourse about 30 kilometers before “Golling an der Salzach” (Dieckmann 2001). The weir itself is 3 m high with a resulting water height of 2 m (Hudelst 2016).

The water quality classes ranges from good to very good. The mean discharge at the location of Obergäu measures 18.1 m³/s (Austria Forum, das Wissensnetz 2016).

2.3 Sampling Design

The sampling in summer took place on May 30th 2018 and was done by a team of students. The site S15 at “Lammerspitz”, close to the village “Golling an der Salzach” were sampled in the morning, followed by the site S 10 at the “Fahrsicherheitszentrum”, which was sampled in the afternoon. In each site five transects (T1 to T5) by four columns (A to D) were defined. By multiplying five transects with the four existing rows, resulting in 20 individual samples, at each sampling site. The transection lines T1 and T2 were always close to the shore line, which frequently falls dry (shore line). T3 indicates the transition between the first two transects and the last two. T4 and T5 are located in the habitat, which are permanently submersed, and experience a change in flow velocities (Figure 4). For each individual sampling point, one stone has been chosen randomly. This leads to 20 samples for each river site at “Salzach” and “Lammer” and therefore in total 40 samples (Willner et al. 2018).

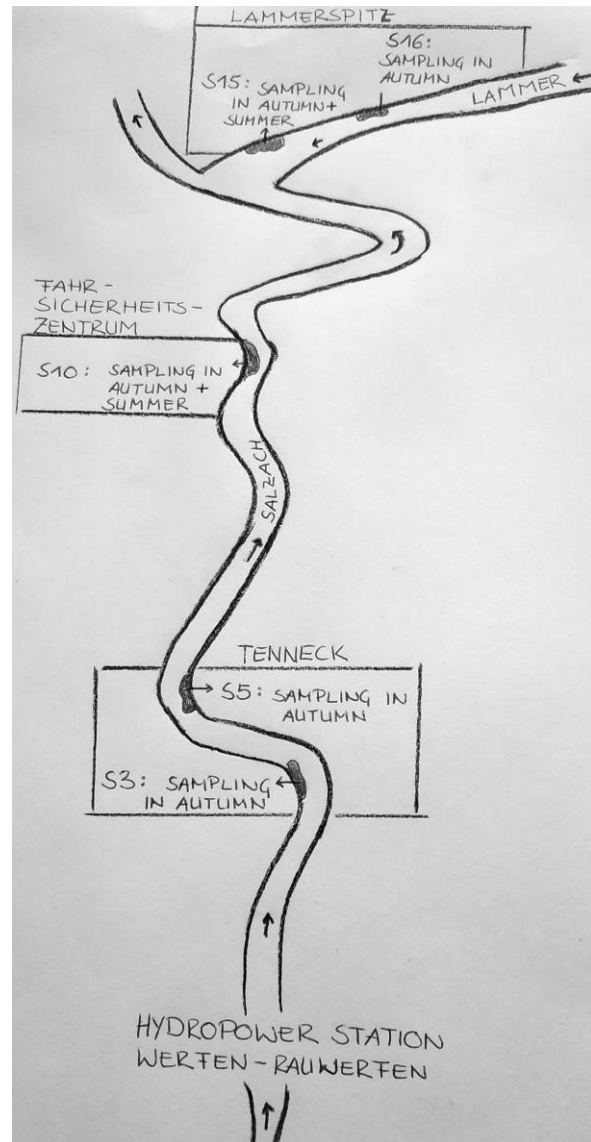


Figure 3: Orthophoto SAGIS (scale 1: 71086) and scheme of the sampling area

The sampling in autumn, under low tide conditions, took place from November 13th to November 15th 2018. Three sites along the river “Salzach” and two at its tributary “Lammer” were chosen. The “Lammer” is used as a reference, because it is not influenced by hydropeaking whereas the “Salzach” stands for the impacted river environment, due to the impacts of the hydropower stations within its river channel. The three sites within the “Salzach” were chosen, because they are close to the hydropower station “Werfen - Rauwerfen” and therefore the influence due to hydropeaking is strongest.

The first site, named “Tenneck”, closely downstream from the hydropower station “Werfen – Rauwerfen”. It is characterized by high flow intensity. Nevertheless, in this area the stream is meandering in a natural way. At “Tenneck” the samples were taken at the two gravel bars at the left (S3) and right turn (S5) of the river.

The second site, named “Fahrsicherheitszentrum” a bit further downstream from the first spot, is still characterized by high flow intensity and the river is still meandering in a natural way. At “Fahrsicherheitszentrum” the samples were taken at the gravel bar opposite to the river side of the Fahrsicherheitszentrum Stegenwald (S10).

The third site, named “Lammerspitz”, is just before the village “Golling an der Salzach”. It is characterized by moderate flow intensity. In this area the river is meandering in a natural way as well. Additionally there are the confluences of the “Lammer” and the “Torrener Bach”. At “Lammerspitz” the samples were taken at the gravel bar shortly before the “Lammer” opens out into the “Salzach” (S15) and further upstream of the “Lammer” (S16). For all five sampling sites (S3, S5, S10, S15, S16) the spatial design was used as illustrated in Figure 4 and described in the summer sampling set up (Willner et al. 2018).

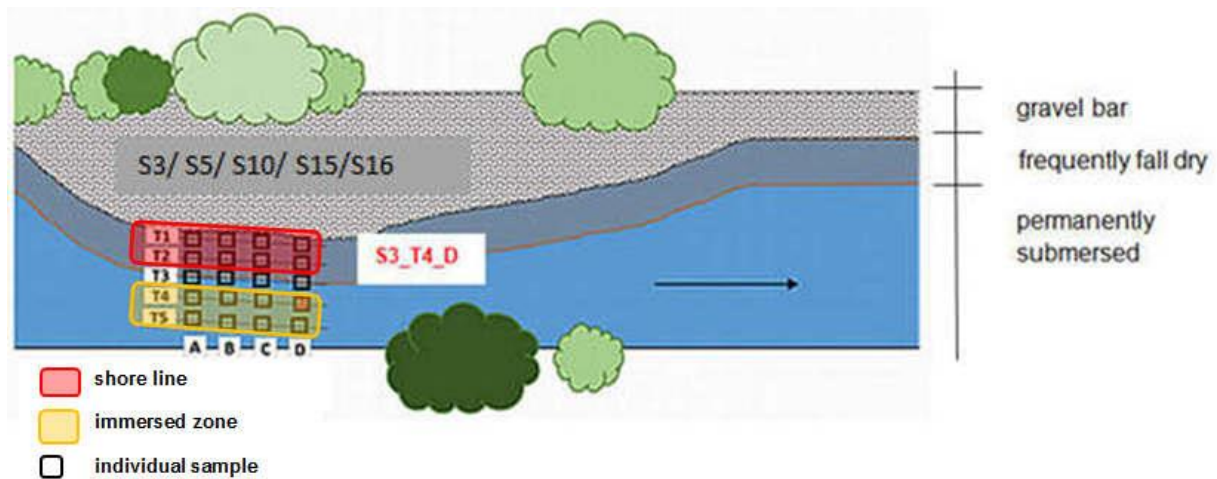


Figure 4: Illustration of the sampling design

3. Methods

For the summer sampling the flow velocity was determined for each individual sampling point and on each site the water temperature was measured. On site, the photosynthesis efficiency via the fluorometer (Phyto-PAM-II, WALZ) was measured. The fiber optic device was positioned over the algae surface of the stones to determine the effective quantum yield. As result, the electron transport rate (rETR) was captured (Willner et al. 2018). After the algae were cleared from the stones surface, the stones were washed with 50 ml of water and transferred into a tube. For

each stone one tube was used and stored in the freezer until further analyses were made. To receive the surface of the stones, the image processing program “ImageJ” was used. Therefore, a photo of the stone with a known scale like a ruler was taken. The uploaded picture were scaled by the function “set scale”, followed by contouring the stone as well as its surface. With the function “measure” the surface area could be estimated (Willner et al. 2018).

During the autumn sampling for each individual sample one to three stones with a diameter of around two to eight cm were picked. The selected stones were stored under a box for dark adaptation. Via the fluorometer (Phyto-PAM-II, WALZ) the first analysis started by gaining information about the PI-Curve and the biomass yield. Afterwards the benthic algae layer at the stones surface was carefully removed by using a known volume of water and brushing the stone surface with a toothbrush. After this process the biological sample was transferred to 50ml-tubes. A picture of each stone was taken onto a scaled paper. The sealed tubes of each individual sample, in total 100 samples, by adding up five sampling sites with 20 samples each, were carefully transported and stored in a shaded and air-conditioned box, set at approximately 7 °C, until they get analyzed in the laboratory.

The water temperature at each of the five sample sites was measured. Additionally, via the field probe (HDQ 40 device, Hach Lange) the electrical conductivity, the pH-value and the oxygen saturation were measured once at every sampling site. Furthermore, the water depth with a yardstick and the flow velocity with the HDQ device (Hach Lange) at each individual sampling point were taken. All the chemical analyses in the laboratory took place in the facilities of the “Wasser Cluster Lunz” at Lunz am See (Lower Austria).

3.1 Hydraulic conditions

The sampling points on which the hydraulic data of the Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement (2019) are based on for this research were post - located after the original sampling points in summer and autumn. Therefore the accuracy between the original sampling points at the gravel bar to the post – located points differ at a maximum 3 m.

The “Lammer” with its hydraulic conditions is represented by the sampling site S 15 (Figure 9 and 10, Figure 13 and 14). The “Salzach” is represented by the sampling site S 10 (Figure 11 and 12, Figure 15 to 17), because the sampling for summer and autumn was made at this site. In all figures the shore line (blue coloured line) and the permanent immersed habitat (orange coloured line) are represented. Due to simplifications in the used model for the hydraulic data, the hydrographs for the flow velocity and the shear stress have to be used and analyzed carefully.

The figures for the summer show one day before the actual sampling day (May 30th 2018). For the autumn the figures show the whole sampling week (November 13th to November 15th 2018) with an additional day before the sampling.

Table 2: sampling stone diameters

Location and season	Shore line habitat – stone diameter [cm]	Permanent immersed habitat – stone diameter [cm]
“Salzach” summer	4.0 – 6.0	3.2 – 11.6
	mean: 5.0	mean: 7.6
“Salzach” autumn	2.4 – 8.1	3.0 – 8.9
	mean: 4.1	mean: 5.5
“Lammer” summer	3.8 -7.7	5.6 – 12.4
	mean:6.0	mean:9.7
“Lammer” autumn	1.9 – 8.0	2.9 - 8.3
	mean: 4.5	mean: 4.9

Table 3: critical shear stress thresholds (according to Ingenieurbüro Kokai GmbH 2017)

Substrate	Diameter [mm]	Critical shear stress [N/m²]
Gravel sand (stable stocked, long-lasting overflowed)	0.63 – 6.3	9
Gravel sand (stable stocked, temporary overflowed)	0.63 – 6.3	12
Gravel	6.3 – 20	15
Coarse gravel	20 - 63	45

3.2 Chlorophyll-A analysis

Via a fluorometer (Phyto-PAM-II, WALZ), which emits light of certain wavelengths, the benthic algae get induced to become photosynthetically active. However, the photosynthetic active chlorophyll-A used a part of the energy for photosynthesis. The other part is reflected as fluorescence. The sensor captured this fluorescence and calculated the amount of existing chlorophyll-A, due to the intensity of the reflection. As result the chlorophyll-A content of the stone surface is determined (Gutowski et al. 2005). As pheophytin-A is a degradation product of chlorophyll-A, it has the same fluorescence and absorption as chlorophyll-A. Therefore, it is possible that it interfered with chlorophyll-A. Especially for phytobenthos, it is quite likely that pheophytin-A is included in the sample, too. Due to the acidification in the analysis, where by chlorophyll-A changed to pheophytin-A a correction for pheophytin-A could be made (Pitzl 2014a).

For the chlorophyll-A analyses via a photometer (Spektralphotometer DR 3900, Hach Lange), each individual sample got filtrated onto a GF/C-filter, cut into small pieces and together with 90 % acetone solution transferred into a glass vial. Before the samples get stored in the fridge overnight, the solutions got homogenized by an ultrasonic probe (Sonifier W-250 D from the company Branson). The following day the glass vials were treated in a centrifuge for 10 minutes by 2500 rounds per minute. 6 ml of the supernatant liquor got decanted into a 5 cm cuvette which is placed into the photometer (Spektralphotometer DR 3900, Hach Lange), where the wavelength 750 nm, 664 nm and 665 nm getting measured. Afterwards 200 µl of 0,1 N HCL was added and after 90 seconds the solution got measured again (Pitzl 2014a).

3.3 High performance liquid chromatography (HPLC)

The high performance liquid chromatography (HPLC) was invented in the 1960's as further development of the liquid chromatography. It is distinguished between two HPLC versions, the isocratic HPLC and the gradient HPLC. For these analyses the gradient HPLC was used. That indicated a permanent change between the interaction of eluent and stationary phase over the whole treatment. During the

analysis, the composition of the eluent changed, thus needing of an equilibration at the beginning of a substance separation. As a result of the analysis the distribution of the pigments are given (AlphaCrom AG 2019).

The first working steps (filtration, homogenization) are identical to the preparation steps of the chlorophyll-A analysis (according to 3.2). After the 24 hour storage, the glass vials were treated in a centrifuge for about 10 minutes by 2500 rounds per minute before a small amount of the supernatant liquor got transferred into the HPLC vials. Until the samples got measured in the HPLC device (Hitachi LaChrom Elite, VWR), they were stored in a dark and cold place (Pitzl 2014c).

The solvents used for the analyses were the following. Solvent A consisted of methanol (HPLC grade) and 0.5 mol ammoniumacetat-solution (A.R. grade) in the ratio 8:2. Solvent B is made up of acetonitil (HPLC grade) and Milli-Q (pure) in the ratio 9:1. Solvent C consisted of pure ethylacetate (HPLC grade) and solvent D, which was just used to rinse the pillar, was made up of 30 % methanol (HPLC grade) (Wright et al. 1991). The areas of the peaks from the samples were than compared with the calibrated peaks, which resulted in the concentration of the pigments themselves (Wright et al. 1991). The resulting pigments fucoxanthin, diadinoxanthin and chlorophyll C2 indicate diatoms (Przytulska et al. 2016). Cyanobacteria were represented by the pigments echinenone, myxoxanthophyll and zeaxanthin (Bonilla et al. 2005) and the pigments for green algae were violaxanthin, chlorophyll B, lutein and neoxanthin (Bonilla et al. 2005).

3.4 PAM Measurement

To measure different types of benthic algae attached to the stones, as well as the yield, a fluorometer (Phyto-PAM-II, WALZ) was used. Due to the multiple wavelengths of 440 nm, 480 nm, 540 nm, 590 nm and 625 nm, it is possible to distinguish between four different pigment types, which can be indicative for green algae, cyanobacteria, diatoms and phycoerytherin. Thus, the software calculated different classes of algae. With the generation of a light curve it was possible to compare the effective quantum yield of photosynthesis II, called yield and the relative electron transport rate, abbreviated by ETR with the incident photosynthetic active radiation (PAR) (Heinz Walz GmbH 2016). Furthermore the values of the light

compensation point I_K , which symbolizes the point at which respiration and oxygen production were leveled out and the $rETR_{max}$, the saturation, which represents the highest value in the curve, were used to differentiate between the sites and seasons (Stevenson et al. 1996). Before every measurement, the stones were put underneath a box to enable them to adapt to the dark (Heinz Walz GmbH 2016).

3.5 C/N- Analyses

For the C/N analyses, the GF/F-filters got dried at 450 °C for four hours. Before and after the filtration the dried filters were weighted. The punches of the individual sample filters got transferred into a pre-weighted tin capsule (IVA Analysetechnik). Right after the transfer the capsule got weighted again. Afterwards they got sealed and were stored into a desiccator till the C/N analyses (Pitzl 2014b). The C/N samples were analyzed by an EA device (Flash 2000 – HT Plus, Thermo Fisher Scientific). For the analysis, the samples got transferred into a reactor which is filled with a layer of tungsten in the upper part for the oxidation and underneath copper rods for the reduction. The reactor had a temperature of 980 °C and is filled with helium. After this process the water got removed and the sample ran through a GC column in which nitrate and carbon got separated. With the thermal conductivity detector both peaks for nitrate and carbon could be analyzed. In the following (Conflow IV, Thermo Fisher Scientific) the reference vapours been added. The “amount% - calculation” to receive the values for nitrate and carbon was done by a comparison of the area of the standard with the area of the samples for nitrate and carbon separately (Thermo Fisher Scientific GmbH 2010).

3.6 P- Analyses

The frozen and filtered GF/F- filter got cut into small pieces and transferred into glass vials. Those ones got dried for four hours by 450 °C. When they were cooled down 5 ml of 90 mmol/l sulfuric acid were added and they were kept in a water bath at 96 °C for one hour. After the samples cooled down again, 2 ml eluate got pipetted into a new glass vial. Then 40 µl ascorbic acid solution and 40 µl mix reagent was added and everything was mixed up together. After 10 to 30 minutes the absorption by a

wavelength of 880 nm was measured with a 1 cm cuvette in the photometer (Spektralphotometer DR 3900, Hach Lange) (Pitzl 2016).

3.7 Statistical analysis

The statistical analysis was performed with “IBM SPSS Statistics Version 24” software. To test the significant differences between the habitats (shore line and permanent submersed) in the two rivers, as well as the variation in the habitats within the “Salzach” over the seasons, the non-parametric test Kruskal-Wallis one-way ANOVA was used. All analysis were considered as significant at $p < 0.05$. For the further statistical evaluation the values for pheophytin had been excluded, because the calculated values were too low (not measureable to $0.614 \mu\text{g}/\text{cm}^2$). The results for the summer sampling are based on the data set analyses made by Willner et al. (2018).

4. Results

4.1 Hydraulic conditions

For the results of the hydrology in the “Salzach” only the hydrograph for the spot S 10 was used, because the hydrographs for the other two spots (S 3 and S 5) are similar in their appearance. The used ranges for the hydraulic parameters (water depth, flow velocity, shear stress) included all spots, as well as both considered habitats (shore line, permanent immersed habitat) in the “Salzach”. For the “Lammer” the same approach was made. Here just the hydrograph for the spot S 15 was used. To get a better impression about the hydrology in the two rivers a bigger time period was considered for the following characterizations. Exemplary for this bigger time period Figure 5 to Figure 8 were put in front of the hydrology description. The mentioned ranges of the parameters include the shore line (smaller value) and the permanent immersed habitat (higher value).

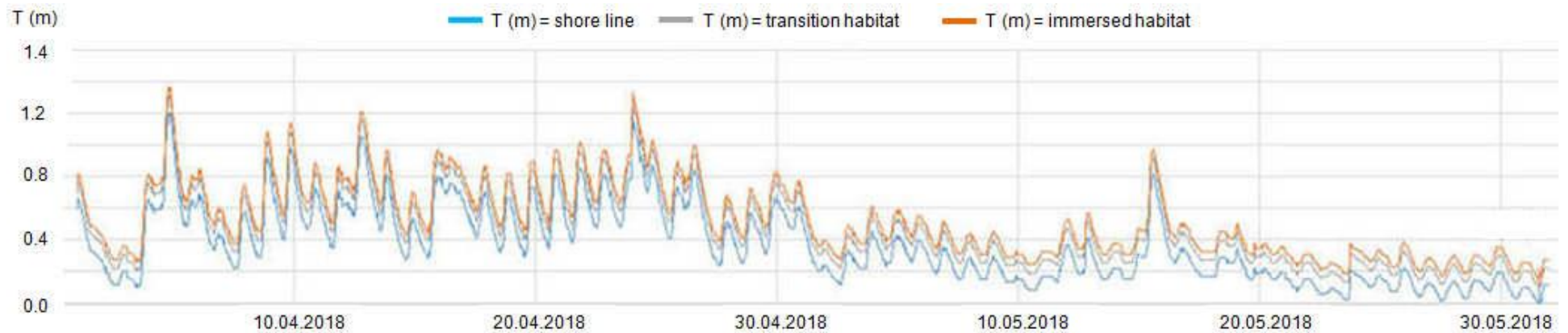


Figure 5: water depth of "Lammer" (April - Mai 2018) (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

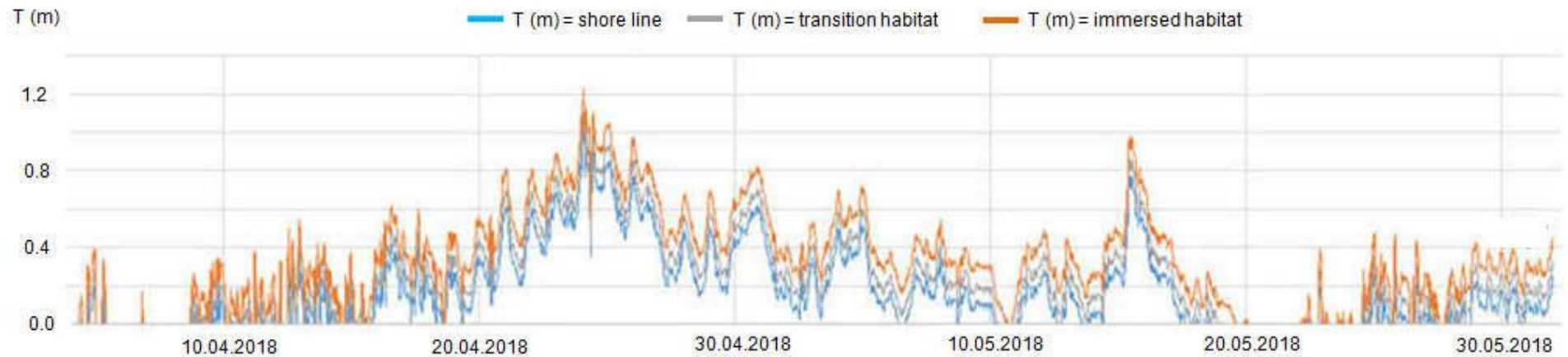


Figure 6: water depth of "Salzach" (April - Mai 2018) (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

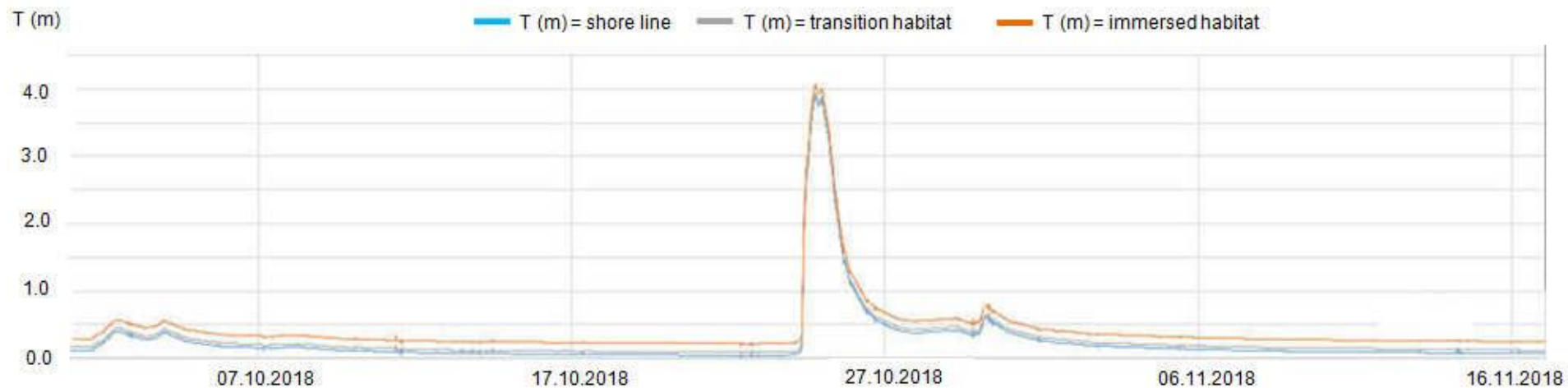


Figure 7: water depth of "Lammer" (October - November 2018) (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

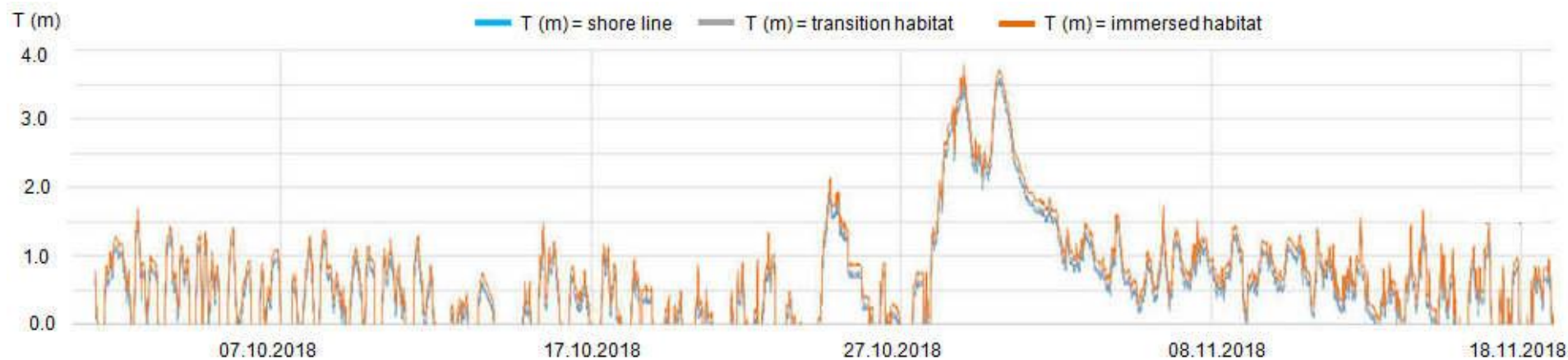


Figure 8: water depth of "Salzach" (October - November 2018) (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

The “Lammer” showed more fluctuations over the summer compared to the autumn. Even though the mean value for the water depth with 0.4 m to 0.5 m were two times higher than in the considered time in autumn, the means for flow velocity (0.3 m/s to 0.5 m/s) and shear stress (2.7 N/m² to 6.7 N/m²) stayed almost the same compared to the values in autumn.

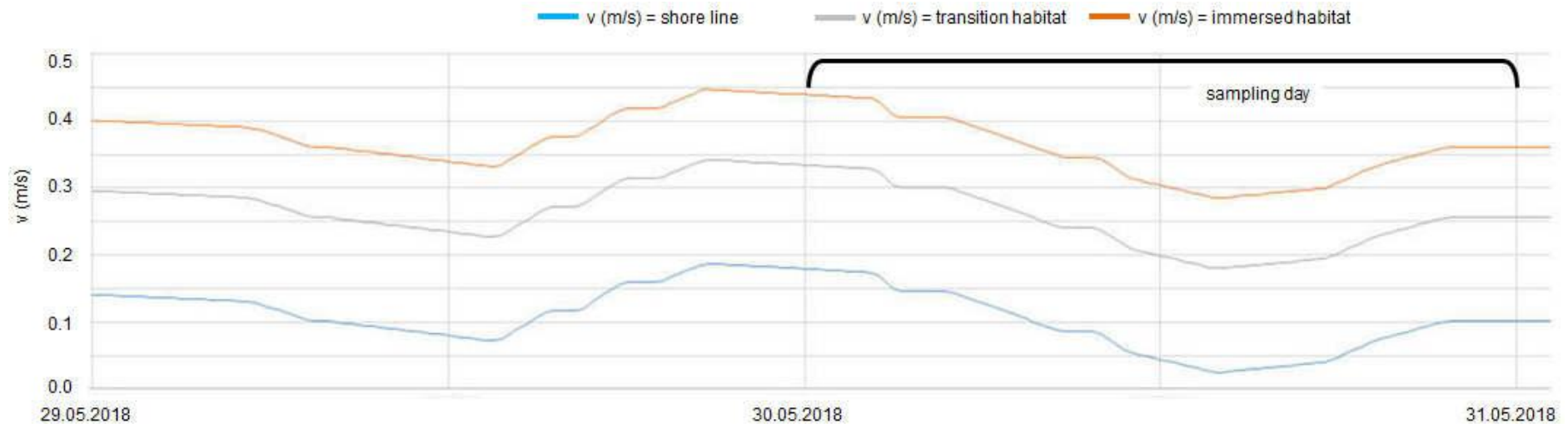


Figure 9: flow velocity graph of "Lammer" for late Mai 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

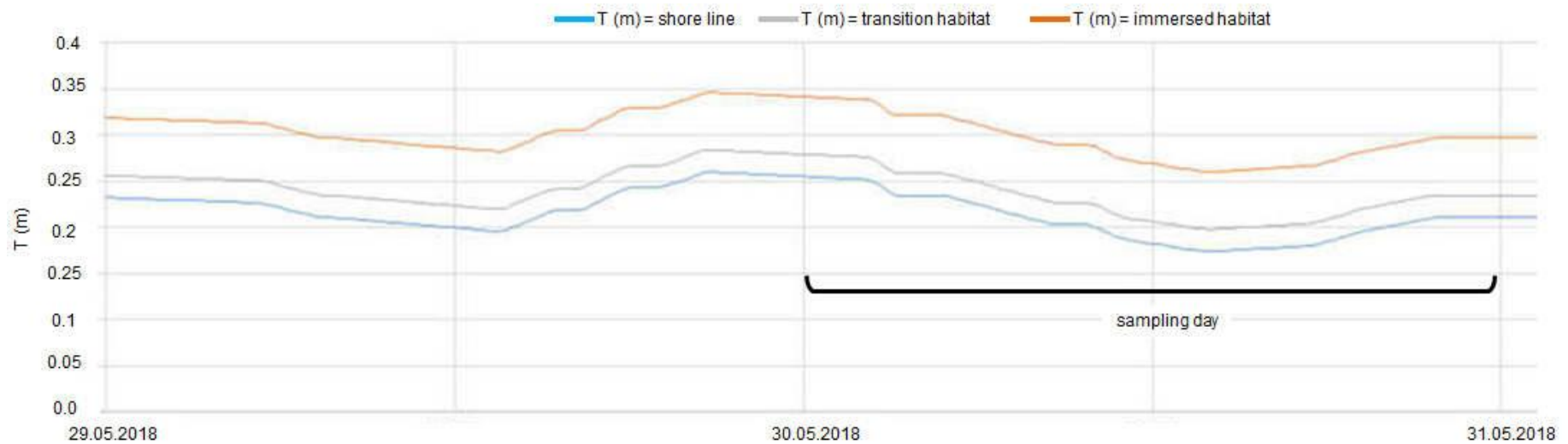


Figure 10: water depth graph of "Lammer" for late Mai 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

During the considered summer months the "Salzach" showed fluctuations (Figure 11 and Figure 12). But compared to the autumn, the peaks were smaller and in general the amplitudes were more even. The mean values for the water depth reached from 0.03 m to 0.2 m. The mean flow velocity ranged from 0 m/s to 0.7 m/s and the shear stress showed mean values of 0 N/m² to 3.0 N/m². In contrast to the autumn, the summer mean values were almost three times lower in case of the water depth and up to four times lower for the shear stress.



Figure 11: shear stress graph of "Salzach" for late Mai 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

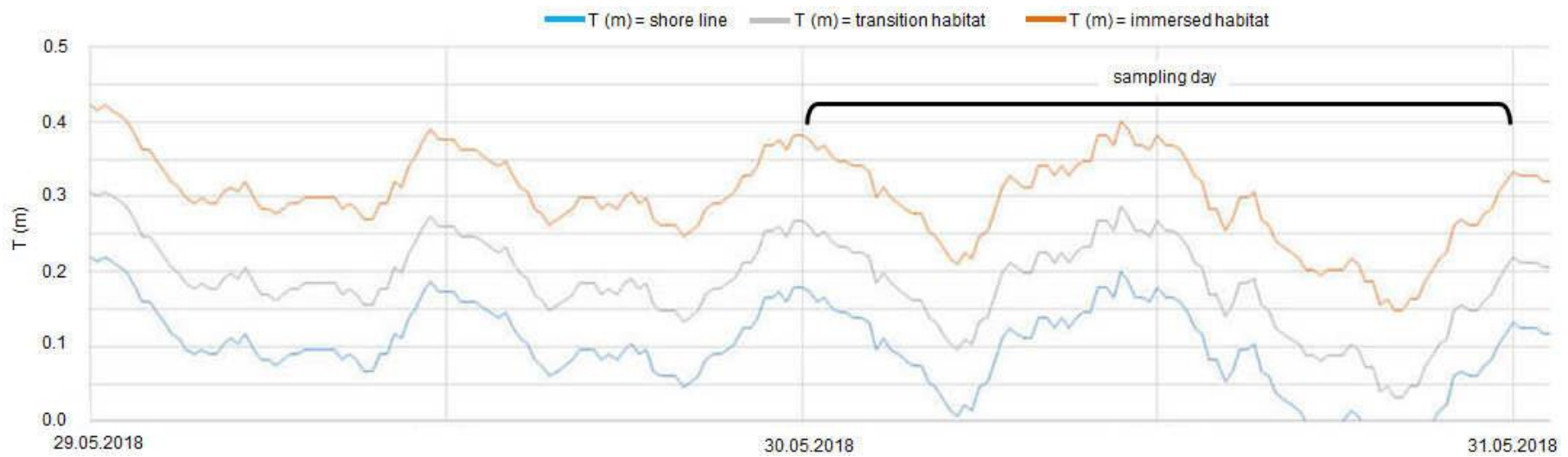


Figure 12: water depth graph of "Salzach" for late Mai 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

In contrast to the summer, the “Lammer” had hardly any fluctuations in November 2018 (Figure 13 and Figure 14). It also had a peak event building up from 24.10.2018 to 25.10.2018. For these days maximum values of 3.4 m to 4.1 m for the water depth, 1.6 m/s to 2.2 m/s for the flow velocity and 18.7 N/m² to 28.1 N/m² for the shear stress were reached. The mean values for the remaining time ranged from 0.1 m to 0.3 m water depth, 0.2 m/s to 0.8 m/s flow velocity and a shear stress of 0 N/m² to 7.8 N/m².

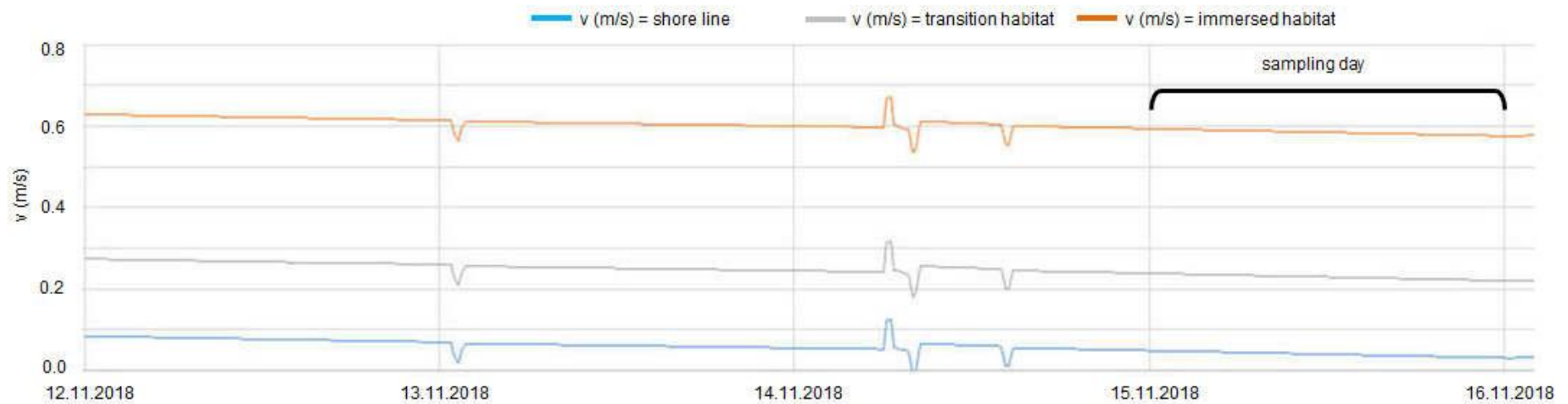


Figure 13: flow velocity graph of "Lammer" of the sampling week in 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

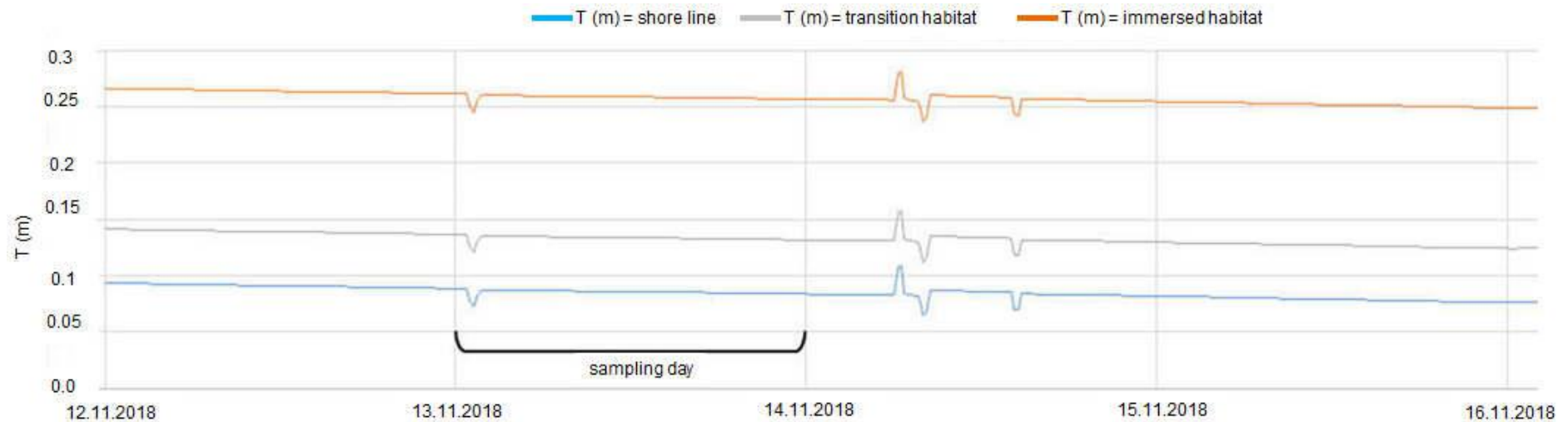


Figure 14: water depth graph of "Lammer" for the sampling day 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

The "Salzach" showed massive fluctuations over the considered time period in late autumn. The graphs for flow velocity, shear stress and water depth showed a peak building up from the 28.10.2018 until the 30.10.2018. During these times the water depth maximum values ranged from 2.7 m in the shore line area till 3.5 m in the permanent immersed habitat. The flow velocity reached maximum values of 2.2 m/s to 3.6 m/s and the shear stress showed highest values of 49 N/m² to 113 N/m². Over the remaining time the mean values for the water depth were 0 m to 0.4 m, the flow velocity ranged from 0 m/s to 1.0 m/s and the shear stress reached mean values of 0 N/m² to 12,2 N/m².

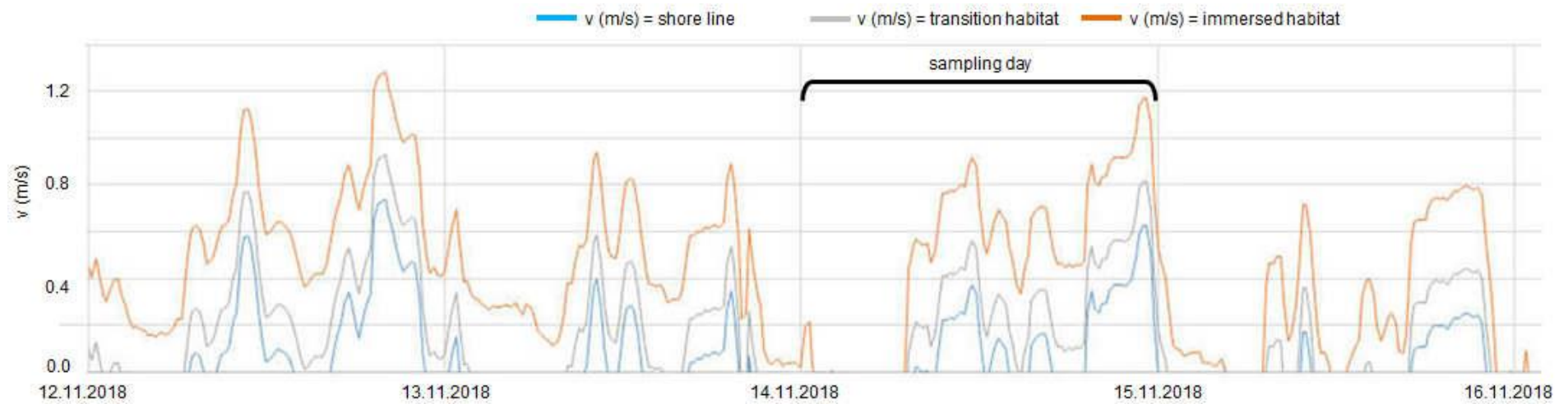


Figure 15: flow velocity graph of "Salzach" for the sampling week in 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

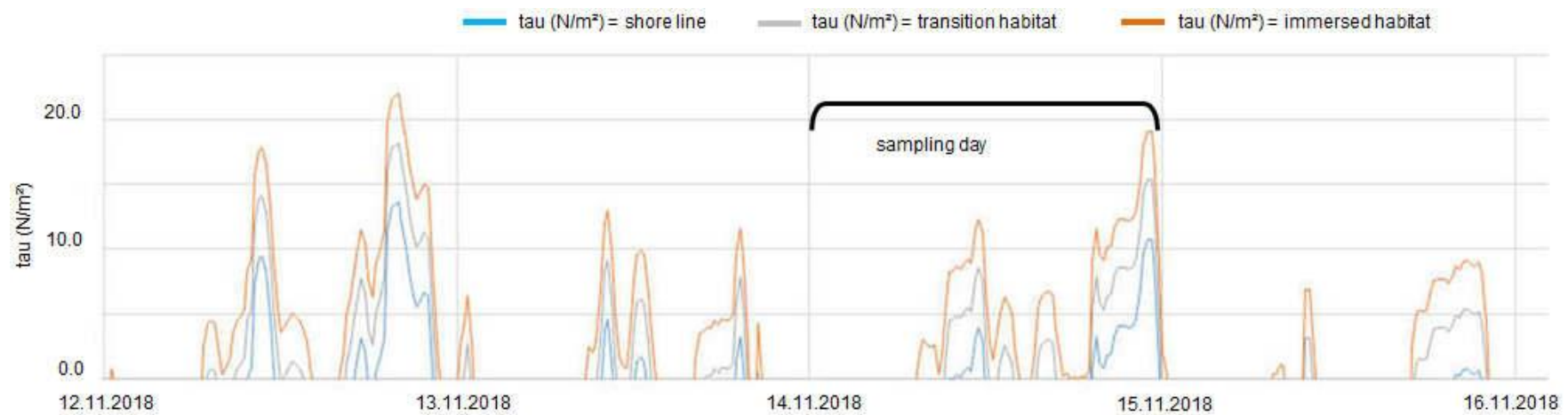


Figure 16: shear stress graph of "Salzach" for the sampling week in 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

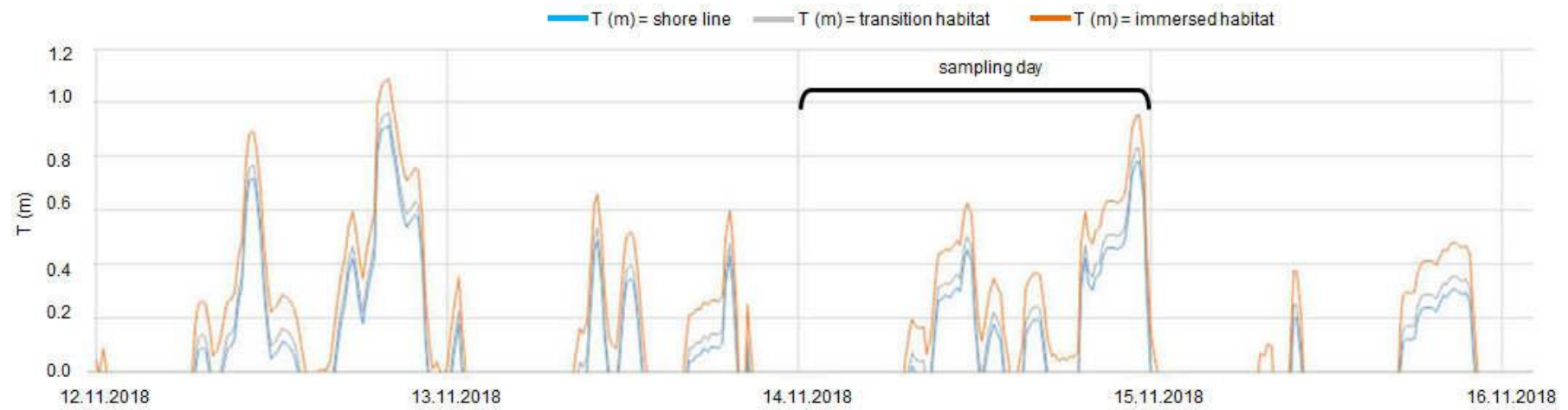


Figure 17: water depth graph of “Salzach“ for the sampling week in 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019)

4.1.1 Hydraulic analysis for the shore line habitat

In case of the flow velocity and the shear stress the Kruskal-Wallis test showed a significant higher value for the “Salzach” by the comparison between the two rivers. For the comparison of the “Salzach” over the seasons, none of the hydraulic parameters (water depth, flow velocity and shear stress) pointed out a significant result.

The stone diameters, which were sampled during the autumn in the “Salzach” reached from 2.4 cm to 8.1 cm. In the “Lammer” the stone diameter reached from 1.9 cm to 8.0 cm. Taking into account that the shear stress ranged from 0 N/m² to 12.2 N/m² in the “Salzach” and 0 N/m² to 7.8 N/m² in the “Lammer”, however the critical shear stress threshold value for coarse gravel (Table 3) is up to six times higher for these diameters. The same results were achieved for the summer sampling too.

4.1.2 Hydraulic analysis for the permanent immersed habitat

For the flow velocity the Kruskal-Wallis test showed a significant higher value in the “Salzach”. The “Salzach” pointed out no significant result, by the comparison of the hydraulic parameters (water depth, flow velocity and shear stress) over the two seasons.

The stone diameters, which were sampled during the autumn in the “Salzach” reached from 3.0 cm to 8.9 cm (Table 2). The shear stress ranged from 0 N/m² to 12.2 N/m². However the critical shear stress threshold value for coarse gravel (Table 3), at which the gravel would start to move, is four times higher for these diameters. The same results for the summer. In the “Lammer” the stone diameter reached from 2.9 cm to 8.3 cm and the shear stress were 0 N/m² to 7.8 N/m².

4.2 Results for the shore line habitat

The chlorophyll-A development showed a higher periphytic biomass production in the “Salzach”. That was unexpected due to the hydropeaking in the “Salzach” a lower biomass content compared to the “Lammer” was presumed. However, the Kruskal-Wallis test showed no significant value for the shore line habitat.

Comparing the distribution in the “Salzach” over the seasons the Kruskal-Wallis test showed a significant difference for the shore line habitat ($p < 0.05$). The “Salzach” showed the expected higher chlorophyll-A development for the summer sampling compared to the autumn sampling (Table 4).

Table 4: mean chlorophyll-A distribution over the seasons (no. of observations: summer = 8; autumn = 24)

	distribution within the river Salzach (shore line)	
	summer	autumn
Chlorophyll-A [$\mu\text{g}/\text{cm}^2$]	0.56 *	0.11

* = ($p < 0.05$)

In the following figures the different colored bars represent the specific pigments for the various algae groups. The grayish colored segments symbolizes the diatoms (Przytulska et al. 2016). The blue colored represents the cyanobacteria and the greenish parts show the pigments for the green algae (Bonilla et al. 2005).

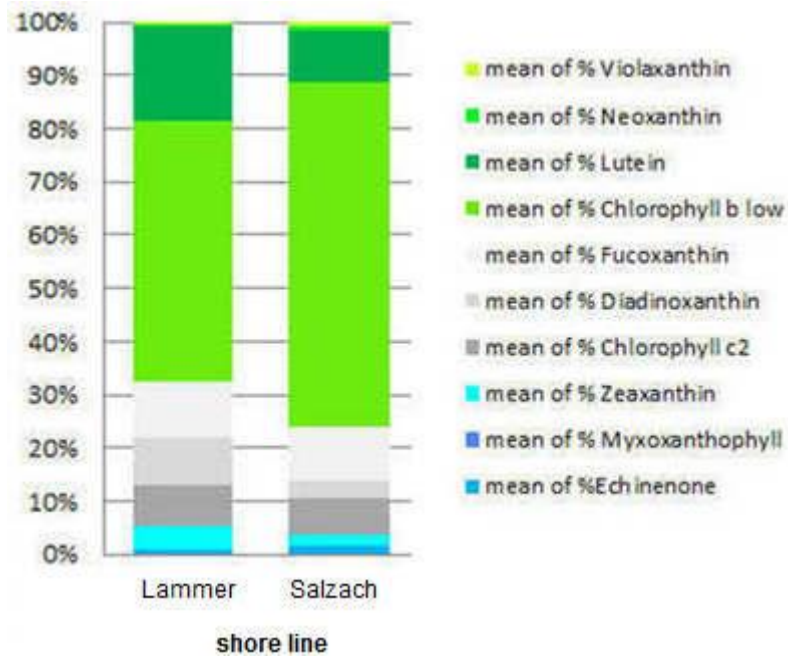


Figure 18: Algae pigment composition over the rivers

The difference between the algae pigment distribution among the two rivers is minor (Figure 18). But instead of the expected dominance of diatoms and cyanobacteria in the “Salzach”, which are affected by hydropeaking treatment, the majority was green algae.

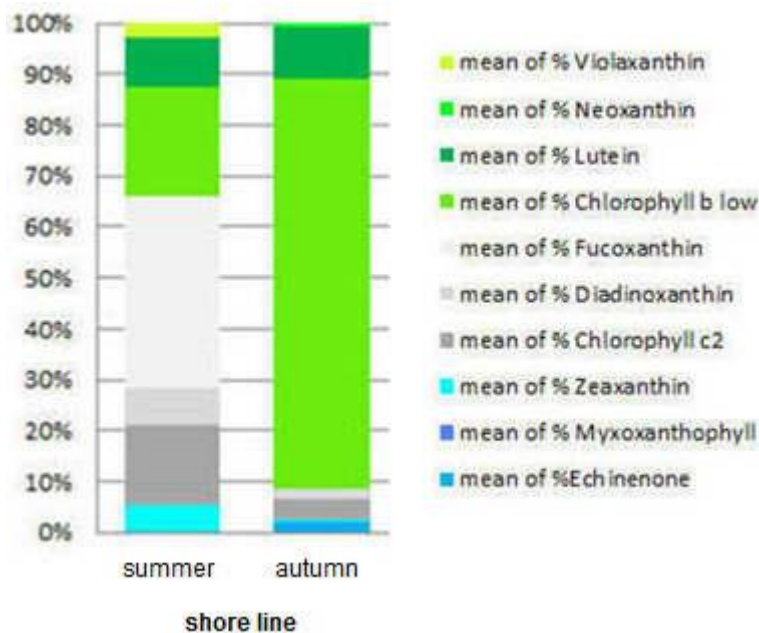


Figure 19: Algae pigment distribution in the Salzach for the two seasons

Figure 19 illustrates that from summer to autumn a shift from dominated by diatoms to dominated by green algae took place. Whereas in summer the percentage ratio

between diatoms and green algae were 59:33 in the “Salzach”, in autumn the ratio ended up to 6:88 in the “Salzach”. That is a huge decline of diatoms in the “Salzach” for the late autumn.

The cyanobacteria played in both seasons a more subordinated role. For the pigment myxoxanthophyll, which is part of the cyanobacteria indicator pigments, showed no values for the summer and autumn sampling. Also for the pigment echinenone, which is part of the cyanobacteria indicator pigments too, no values could be detected in the summer samples.

Instead of the expected lower photosynthesis activity in the “Salzach”, the “Lammer” showed slightly lower values for the parameters. But for both parameters, the light compensation point I_K , as well as for the saturation $rETR_{max}$ the Kruskal-Wallis test did not receive significant values for the comparison between the two rivers.

Table 5: mean distribution of photosynthesis parameter over the seasons (no. of observations: summer = 8; autumn = 24)

	distribution within the river Salzach (shore line)	
	summer	autumn
I_K	112.5 *	147
$rETR_{max}$	16.01	15.73

* = ($p < 0.05$)

Comparing the distribution of the photosynthesis parameters in the “Salzach” over the seasons, the summer showed an unexpected lower value for the compensation point I_K . In the Kruskal-Wallis test the difference in the I_K – value for the shore line was significant ($p < 0.05$).

The median of the mean total phosphor content for both rivers are almost identical. Even though the Kruskal-Wallis test displayed no significant differences for total phosphor for the shore line habitat, nevertheless a trend of a higher total amount of phosphor in the “Salzach” compared to the “Lammer” can be seen. For the analysis of the nutrients carbon (C), nitrogen (N) and phosphor (P) only data out of the fall sampling existed.

The C/N ratios showed a significant difference for the shore line. The measured contents were higher in the “Salzach” with a maximum of 13. In the tributary “Lammer” the mean value was around 8.

4.3 Results for the permanent immersed habitat

Comparing the data between the two rivers, the result of the chlorophyll-A development showed the expected higher periphytic biomass production in the “Lammer” (Table 6). The Kruskal-Wallis test showed a significant value for the immersed habitat.

Table 6: mean chlorophyll-A distribution of the rivers (no. of observations: Lammer = 24; Salzach = 32)

	distribution between the two rivers (immersed habitat)	
	Lammer	Salzach
Chlorophyll-A [µg/cm ²]	0.25 *	0.04

* = (p < 0.05)

Comparing the distribution over the seasons the “Salzach” showed a higher chlorophyll-A development in the summer. This outcome was expected but the Kruskal-Wallis test showed no significant difference (p < 0.05) for the permanent immersed habitat.

The difference between the algae pigment distribution among the two rivers is minor (Figure 20). But instead of the expected dominance of diatoms and cyanobacteria in the “Salzach”, which are affected by hydropeaking treatment, the majority was green algae.

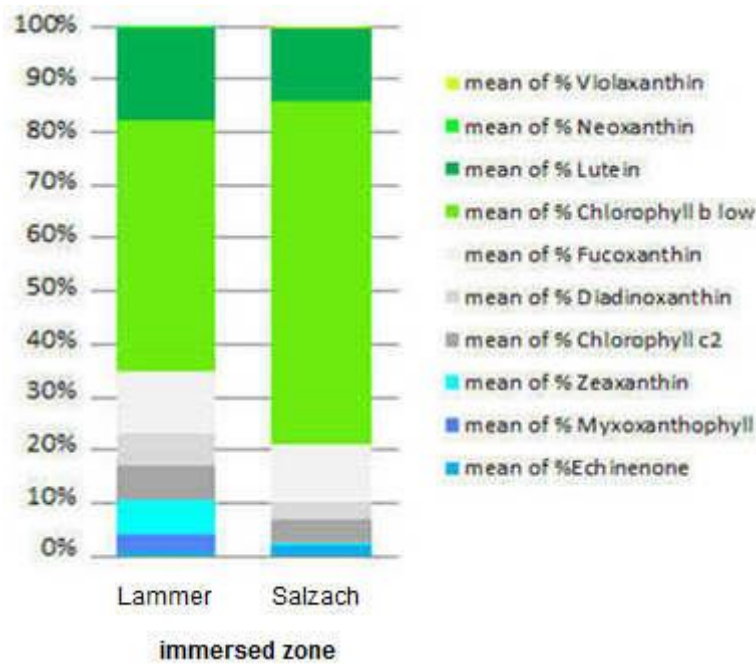


Figure 20: Algae pigment composition over the rivers

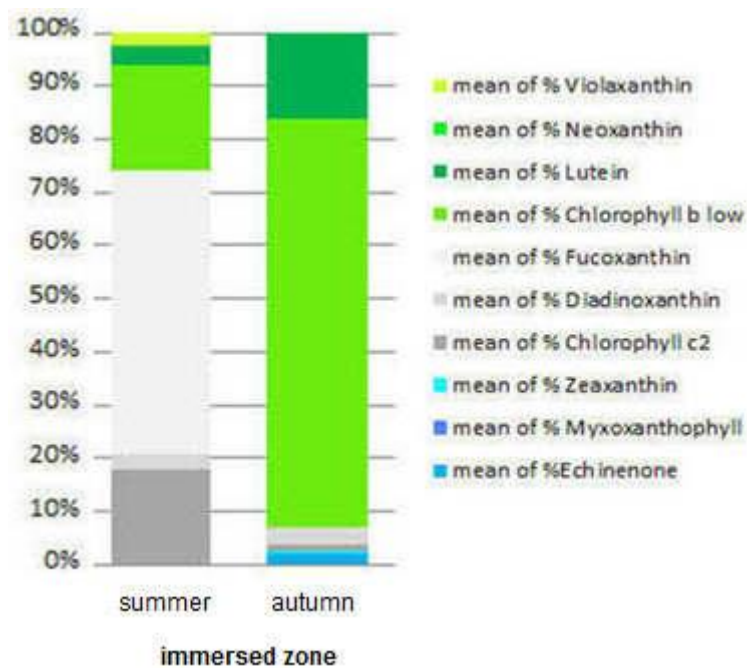


Figure 21: Algae pigment composition in the Salzach for the two seasons

Figure 21 illustrates that from summer to autumn, a shift from being dominated by diatoms to dominated by green algae took place. Whereas in summer the percentage ratio between diatoms and green algae were 72:25 in the “Salzach”, in fall the ratio ended up to 4:91 in the “Salzach”. That is a big decline of diatoms in the “Salzach” for the late autumn. It was expected that during the summer period the “Salzach” would

be dominated by green algae and in autumn the dominance of diatoms and cyanobacteria.

The cyanobacteria played in both seasons a more subordinated role. For the pigment myxoxanthophyll, which is part of the cyanobacteria indicator pigments, showed no values for the summer and autumn sampling. Also for the pigment echinenone, which is part of the cyanobacteria indicator pigments too, no values could be detected in the summer samples.

For the photosynthesis activity the “Salzach” showed the expected lower values compared to the “Lammer”. But for both parameters, the light compensation point IK as well as for the rETRmax, the Kruskal-Wallis test did not show significant values for the comparison between the two rivers.

Comparing the distribution of the photosynthesis parameters in the “Salzach” over the seasons (Table 7), the summer showed the expected higher value for the compensation point IK. In the Kruskal-Wallis test the difference in the IK – value for the immersed habitat were significant ($p < 0.05$).

Table 7: mean distribution of photosynthesis parameter over the seasons (no. of observations: summer = 8; autumn = 24)

	distribution within the river Salzach (immersed habitat)	
	summer	autumn
IK	131.59 *	70.35
rETRmax	18.44	15.51

* = ($p < 0.05$)

The Kruskal-Wallis test displayed no significant values of total phosphor for the permanent immersed habitat. Nevertheless the total amount of phosphor is lower in the “Salzach” compared to the “Lammer”. That was expected, because the “Salzach” is affected by hydropeaking events and therefore a lower content of phosphor was presumed.

The C/N ratio showed a significant value for the immersed zone. The content in the “Salzach” was measured with a maximum of 20 whereas in the tributary the mean value stayed around 8.

5. Discussion

5.1 Shore line habitat

The hypothesis concerning the higher biomass content in the “Lammer” compared to the “Salzach” was not verified. Surprisingly the amount of chlorophyll-A was higher in the shore line habitat of the “Salzach” instead of the river “Lammer”, pointing out the opposite outcome of the research done by Bondar-Kunze et al. (2016). The hydrographs for the water depth in the “Salzach” (Figure 12 and 17) illustrated, that the habitat fell dry at least every third day. In contrast, the shore line in the “Lammer” (Figure 10 and 14) did not fall dry. In combination with the additional data of macrozoobenthos sampled in April 2018 (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019) the grazer community within the “Lammer” consumed benthic algae in this habitat. Especially grazers scrape algae biomass off their habitat surfaces. Grazers could be snails, larvae from mayflies, stoneflies or caddisflies or water-scavenger beetles (Schönborn and Risse-Buhl 2013). The scientist group for the Surema Plus project found around 48 % grazers in the area of S 15 in the “Lammer”. Compared to 28 % grazers at the spot S 3, 29 % at the gravel bar S 5 and none at S 10 in the “Salzach” (Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement 2019). With the nearly two times higher amount of grazers in the “Lammer”, the outcome of the biomass research in the shore line habitat was related to the natural eating habits of macrozoobenthos.

The expectations concerning the algae pigment distribution in the shore line habitat were not fulfilled comparing the two rivers to each other. Instead of a dominance of diatoms and cyanobacteria in the “Salzach” the majority was green algae. That was unexpected, because based on the research of Bondar-Kunze et al. (2016) a higher content of diatoms as well as cyanobacteria due to hydropeaking in the river was presumed for the “Salzach”. The flow velocities in the “Salzach” were on average 0.5 m/s (Figure 15). This flow velocity intensity is above the critical value of 0.15 m/s (Bondar-Kunze et al. 2016). Therefore, the amount of diatoms should have been higher. Figure 9 and Figure 13 shows the steady flow of the “Lammer” with flow velocity hardly above the critical value of 0.15 m/s, which corresponded with the high green algae content for the pigment distribution in the “Lammer”.

The photosynthesis activity in the “Salzach” was not reduced compared to the “Lammer”, like it was expected. The hydrographs for the water depth in the “Salzach” (Figure 12 and 17) illustrated that the water level fluctuated permanently, whereas the shore line in the “Lammer” (Figure 10 and 14) had a constant water level. Therefore, the changes in the water level did not have the significant impact on the photosynthesis activity of the benthic algae. In contrast the permanent fluctuations in water depth in the “Salzach” (Figure 12 and 17) resulted in the higher storage of phosphorus in the benthic algae. This was not expected, but Eixler et al. (2006) detected in his research, that benthic algae store even more phosphorus after a period of starvation and desiccation stress.

5.2 Permanent immersed habitat

In the permanent immersed habitat, the algae are confronted with the increased flow velocity, during a hydropeaking event. After the research of Bondar-Kunze et al. (2016) flow velocities greater than 0.1 m/s has a quantitative (decrease in biomass) influence on benthic algae. This is represented in the chlorophyll-A content, which showed a significant reduction in the biomass production for the “Salzach” compared to the tributary without hydropeaking. Taking into account that about two weeks before the sampling, a peak event with occurred shear stresses of 57.73 N/m² to 113.14 N/m² in the “Salzach” took place the stones, of the later sampled diameter, started to move. During this peak event the critical shear stress for coarse gravel was exceeded (Table 3). Based on the research of Cashman et al. (2017) benthic algae need several weeks to recover from this flooding. Thus, the biomass content was reduced in the permanent immersed habitat in the “Salzach”.

Even though the values for the photosynthesis activity showed no significant values, they corresponded with the biomass content, because the parameters IK and rETRmax were reduced in the “Salzach”, which is impacted by hydropeaking. With mean shear stress up to 21.9 N/m² (Figure 11 and 16), the critical shear stress for gravel sand mixtures was exceeded (Table 3). Therefore, the turbidity in the river rose and was limiting the photosynthesis activity (Bruder et al. 2016).

The algae pigment distribution between green algae on the one side and diatoms and cyanobacteria on the other side were almost the same in the two rivers. Based

on the research by Bondar-Kunze et al. (2016) a greater difference between the rivers was presumed. The flow velocities in the “Salzach” were on average 0.8 m/s (Figure 15). This flow velocity intensity is above the critical value of 0.15 m/s (Bondar-Kunze et al. 2016) therefore, the amount of diatoms should have been higher. Figure 9 and Figure 13 shows the steady flow of the “Lammer” with flow velocity around 0.7 m/s on average for the immersed habitat. The total amount of phosphor was reduced in the “Salzach” compared to the content in the “Lammer”, which was expected. The flow velocities in the permanent immersed habitat reached up to 1.0 m/s (Figure 15) thus, the nutrient content was flushed away. This is represented in the lower phosphor content in the “Salzach” compared to the “Lammer”.

5.3 Distribution in the “Salzach” between the seasons

For the comparison over the seasons both habitats (shore line and permanent immersed habitat) in the “Salzach” showed the expected higher biomass content for the summer. The graphs of the hydraulic parameters illustrating a higher fluctuation pattern during the autumn (Figure 15 to 17) compared to the considered summer month (Figure 11 and Figure 12). Thus, the summer values were higher due to reduced impact of increased water depth, flow velocity and turbidity.

Looking at the algae pigment distribution both habitats showed a higher contribution of diatoms and cyanobacteria in the summer, despite the expected dominance of diatoms and cyanobacteria for the autumn sampling. Due to the fact that the hydropower station in “Werfen – Rauwerfen” generates about 50% of the total energy during the summer months (VERBUND Hydro Power GmbH 2018) the pigment distribution is explainable. Furthermore, Larned (2010) and Ledger et al. (2008) showed that rivers, impacted by hydropowering show a dominance of diatoms and cyanobacteria which also contribute the outcome of the pigment distribution during the summer period.

In case of the photosynthesis activity the two habitats reacted differently. Unexpectedly the shore line showed a reduced activity for the summer. Whereas the permanent immersed habitat showed the higher photosynthesis activity for the summer compared to the autumn. The outcome of the permanent immersed habitat

correlates with the hydraulic parameter graphs, illustrating a higher fluctuation pattern during the autumn (Figure 15 to 17) compared to the considered summer months (Figure 11 and Figure 12). With shear stresses up to 21.9 N/m² in the autumn, the turbidity in the “Salzach” was higher compared to the summer with shear stresses of 3.0 N/m² on average. Therefore the photosynthesis activity was limited during the autumn (Bruder et al. 2016).

6. Conclusion

The shore line habitat showed no effect, on account of hydropeaking when comparing the affected and not affected river (see column “River” in Table 8). For the distribution between the seasons in the “Salzach” only the hypothesis concerning the biomass production was verified. The results in the permanent immersed habitat were different. Three out of four hypotheses concerning the distribution between the two different rivers (“Lammer” without hydropeaking operation and “Salzach” with hydropeaking) were verified for this habitat (see column “River” in Table 8). Looking at the outcome for the season, the hypotheses for biomass production and photosynthesis activity were verified. Therefore, the conclusion is that the permanent immersed habitat is more affected and more heavily influenced by hydropeaking events in different seasons, as well as in the river itself compared to the shore line habitat.

Table 8: Overview over the verified hypothesis

Hypothesis	Habitat	River	Season
Biomass	Shore line	No	Yes
	Immersed habitat	Yes	Yes
Photosynthesis	Shore line	No	No
	Immersed habitat	Yes	Yes
Element composition	Shore line	No	No data
	Immersed habitat	Yes	No data
Pigment composition	Shore line	No	No
	Immersed habitat	No	No

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References

- AlphaCrom AG. 2019. "Chromatographie Grundlagen." Retrieved January 16, 2019 (<https://www.alphacrom.com/de/hplc-grundlagen>).
- Austria Forum, das Wissensnetz, AEIOU. 2016. "Lammer." Retrieved February 9, 2019 (<https://austria-forum.org/af/AEIOU/Lammer>).
- Bejarano, María D., Roland Jansson, and Christer Nilsson. 2018. "The Effects of Hydropeaking on Riverine Plants: A Review." *Biological Reviews* 93(1):658–73.
- BMLFUW. 2015. "HOCHWASSERRISIKO- MANAGEMENTPLAN 2015 RISIKOGEBIET : Lammer, Oberscheffau 5012."
- Bondar-Kunze, Elisabeth, Stefanie Maier, Doris Schönaauer, Nicolas Bahl, and Thomas Hein. 2016. "Antagonistic and Synergistic Effects on a Stream Periphyton Community under the Influence of Pulsed Flow Velocity Increase and Nutrient Enrichment." *Science of the Total Environment* 573:594–602.
- Bonilla, Sylvia, Valérie Villeneuve, and Warwick F. Vincent. 2005. "Benthic and Planktonic Algal Communities in a High Arctic Lake: Pigment Structure and Contrasting Responses to Nutrient Enrichment." *Journal of Phycology* 41(6):1120–30.
- Bruder, Andreas, Diego Tonolla, Steffen P. Schweizer, Stefan Vollenweider, Simone D. Langhans, and Alfred Wüest. 2016. "A Conceptual Framework for Hydropeaking Mitigation." *Science of the Total Environment* 568:1204–12.
- Bundesministerium für Land- und Forstwirtschaft, Umwelt und Wasserwirtschaft. 2009. "Die Salzach: Ein Fluss Bewegt." Retrieved June 7, 2018 (<https://docplayer.org/64426562-Die-salzach-ein-fluss-bewegt.html>).
- Cashman, Matthew J., Gemma L. Harvey, Geraldene Wharton, and Maria Cristina Bruno. 2017. "Wood Mitigates the Effect of Hydropeaking Scour on Periphyton Biomass and Nutritional Quality in Semi-Natural Flume Simulations." *Aquatic Sciences* 79(3):459–71.
- Décamps, H., R. J. Naiman, and M. E. McClain. 2009. "Riparian Zones - Encyclopedia of Inland Waters." *Encyclopedia of Inland Waters*.
- Dieckmann, Mathias. 2001. "Lammer Salzburger Land / Österreich." Retrieved May 15, 2019 (<http://www.kajaktour.de/lammer.htm>).
- E-Control. 2018. "No Title." Retrieved October 18, 2018 (<https://www.e->

- control.at/konsumenten/oeko-energie/basiswissen/oekostrom-arten/wasserkraft).
- Eixler, Sebastian; Ulf; Karsten, and Uwe Selig. 2006. "Phosphorus Storage in *Chlorella Vulgaris* Trebouxiophyceae , ..."
- Fröschl, Christine. 2019. "Die Lammer: Vom Ursprung Bis Zur Mündung." *Salzburger Land Tourismus GmbH*. Retrieved February 9, 2019 (<https://www.salzburgerland.com/de/magazin/die-lammer-vom-ursprung-bis-zur-muendung/>).
- Gutowski, Antje, Julia Foerster, and G. Hofmann. 2005. "Untersuchung Der Benthischen Mikro- Und Makroalgen in Der Tide-Elbe Auf Eignung Zur Beurteilung Des Gewässers Gemäß EG-Wasserrahmenrichtlinie." *Transport* 87.
- Hall, Robert O., Charles B. Yackulic, Theodore A. Kennedy, Michael D. Yard, Emma J. Rosi-Marshall, Nicholas Voichick, and Kathrine E. Behn. 2015. "Turbidity, Light, Temperature, and Hydropeaking Control Primary Productivity in the Colorado River, Grand Canyon." *Limnology and Oceanography* 60(2):512–26.
- Hauer, C., P. Holzapfel, P. Leitner, and W. Graf. 2016. "Longitudinal Assessment of Hydropeaking Impacts on Various Scales for an Improved Process Understanding and the Design of Mitigation Measures." *Science of the Total Environment* 575:1503–14.
- Hauer, C., A. Siviglia, and G. Zolezzi. 2017. "Hydropeaking in Regulated Rivers – From Process Understanding to Design of Mitigation Measures." *Science of the Total Environment* 579:22–26.
- Heinz Walz GmbH. 2016. "Phytoplankton Analyzer Software V 3 Principles of Operation." (0).
- Holzapfel, P., P. Leitner, H. Habersack, W. Graf, and C. Hauer. 2017. "Evaluation of Hydropeaking Impacts on the Food Web in Alpine Streams Based on Modelling of Fish- and Macroinvertebrate Habitats." *Science of the Total Environment* 575:1489–1502.
- Horne, A. and R. Goldman. 1994. *Limnology*.
- Hudelist, Michael. 2016. "11-Jähriger Ertrinkt in Lammer." Retrieved May 15, 2019 (<https://infomediaworx.wordpress.com/2016/08/26/11-jaehriger-ertrinkt-in-lammer/>).
- Ingenieurbüro Kokai GmbH. 2017. "Kritische Schubspannung." Retrieved April 19, 2019 (<https://www.bauformeln.de/wasserbau/feststofftransport/kritische-schubspannung-tabelle/>).

- Kokavec, Igor, Tomáš Navara, Pavel Beracko, Tomáš Derka, Ivana Handanovičová, Andrea Rúfusová, Zuzana Vráblová, Tomáš Lánczos, Marta Illyová, and Ferdinand Šporka. 2017. "Downstream Effect of a Pumped-Storage Hydropower Plant on River Habitat Conditions and Benthic Life-a Case Study." *Biologia (Poland)* 72(6):652–70.
- Larned, Scott T. 2010. "A Prospectus for Periphyton : Recent and Future Ecological Research." *Journal of the North American Benthological Society* 29(1):182–206.
- Ledger, Mark E., Rebecca M. L. Harris, Patrick D. Armitage, and Alexander M. Milner. 2008. "Disturbance Frequency Influences Patch Dynamics in Stream Benthic Algal Communities." *Oecologia* 155(4):809–19.
- Likens, Gene E. and R. J. Stevenson. 2009. "Algae of River Ecosystems." *Encyclopedia of Inland Waters*.
- Meile, T., M. Fette, and P. Baumann. 2005. "Synthesebericht Schwall/Sunk Publikation Des Rhone-Thur Projektes." *Projekt Report* 48.
- Pitzl, Beate. 2014a. "26_SOP-Chlorophyll a Photometer - Methode Description." 4.
- Pitzl, Beate. 2014b. "28_SOP-CN Vorbereitung - Methode Description." 2.
- Pitzl, Beate. 2014c. "SOP_Pigmente Extrahieren - Methode Description." 1.
- Pitzl, Beate. 2016. "03_SOP-Wasser PP - Methode Description." 4.
- Polst, Bastian H., Christine Anlanger, Ute Risse-Buhl, Floriane Larras, Thomas Hein, Markus Weitere, and Mechthild Schmitt-Jansen. 2018. "Hydrodynamics Alter the Tolerance of Autotrophic Biofilm Communities Toward Herbicides." *Frontiers in Microbiology* 9(December):2884.
- Przytulska, A., J. Comte, S. Crevecoeur, C. Lovejoy, I. Laurion, and W. F. Vincent. 2016. "Phototrophic Pigment Diversity and Picophytoplankton in Permafrost Thaw Lakes." *Biogeosciences* 13(1):13–26.
- Schmutz, Stefan and Otto Moog. 2018. "Dams: Ecological Impacts and Management." in *Riverine Ecosystem Management*.
- Schönborn, Wilfried and Ute Risse-Buhl. 2013. *Lehrbuch Der Limnologie*. second edi. Stuttgart: Schweizerbart.
- Schülting, Lisa, Christian K. Feld, and Wolfram Graf. 2016. "Effects of Hydro- and Thermopeaking on Benthic Macroinvertebrate Drift." *Science of the Total Environment* 573:1472–80.
- Stevenson, R. Jan, M. L. Bothwell, and Rex L. Lowe. 1996. *Algal Ecology\rfreshwater Benthic Ecosystems*.

- Thermo Fisher Scientific GmbH. 2010. "Thermo Scientific FLASH HT Plus Fully Automated Multi-Element Isotope Analysis for C , N , S , O and H."
- Universität für Bodenkultur Wien - Institut für Hydrobiologie, Gewässermanagement. 2019. *Surema Plus Projekt - Hydrologische Daten*.
- VERBUND Hydro Power GmbH, Salzburg AG. 2018. "Kraftwerk Werfen-Pfarrwerfen." Retrieved September 13, 2018 (<https://www.verbund.com/de-at/ueber-verbund/kraftwerke/unsere-kraftwerke/werfen-pfarrwerfen>).
- Vidyasagar, Aparna. 2016. "No Title." *Livescience*. Retrieved September 19, 2018 (<https://www.livescience.com/54979-what-are-algae.html>).
- Wagner, Beatrice, Christoph Hauer, Angelika Schoder, and Helmut Habersack. 2015. "A Review of Hydropower in Austria: Past, Present and Future Development." *Renewable and Sustainable Energy Reviews* 50:304–14.
- Wasserkraftverband Mitteldeutschland e.V. 2019. "Verschiedene Arten von Wasserkraftwerken." Retrieved March 19, 2019 (<https://www.wasserkraftverband.de/pages/wissenswertes-zur-wasserkraft/verschiedene-arten-von-wasserkraftwerken.php>).
- Willner, Magdalena, Viktoria Kobel, and Eva Kunz. 2018. *Bericht: Konstruktives Projekt Unveröffentlichter Praktikumsbericht*.
- Wright, S. W., S. W. Jeffrey, R. F. C. Mantoura, C. A. Llewellyn, T. Bjornland, D. Repeta, and N. Welschmeyer. 1991. "Improved HPLC Method for the Analysis of Chlorophylls and Carotenoids from Marine Phytoplankton." *Marine Ecology Progress Series* 77(2–3):183–96.
- Zeiringer, Bernhard, Carina Seliger, Franz Greimel, and Stefan Schmutz. 2018. "River Hydrology, Flow Alteration, and Environmental Flow." *Riverine Ecosystem Management* 67–89.

7. Affirmation

I certify, that the master thesis was written by me, not using sources and tools other than 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.

Wien, 21.05.2019 Magdalena Pöhm