



Quality and suitability of fecal biochar in structurally stable urban tree substrates

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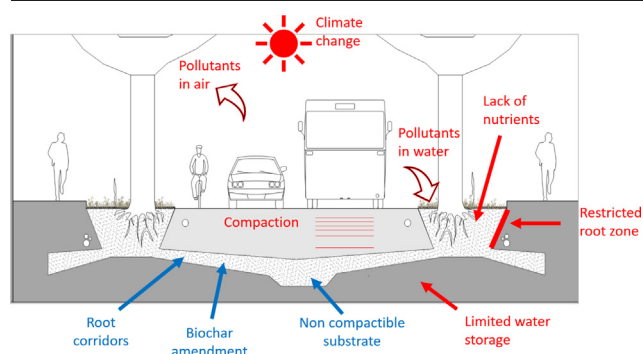
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HIGHLIGHTS

- Biochar-amended and structurally stable tree substrates improve the conditions of city trees
- The main factor is the increased water retention capacity provided by the biochar
- Fresh biochar amended structurally stable tree substrates, leach easily soluble ions in the first 3–4 months
- Biochar from fecal matter may be a valuable option as nutrient source in structurally stable tree substrates

GRAPHICAL ABSTRACT



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ABSTRACT

This study examines the effects of pyrolyzed materials (biochar) from horse manure, plant residues and human feces on the growth of 1-year old birch seedlings cultivated in a novel, structurally stable tree substrate. For this purpose, the composition of the produced biochar, the leachate from the substrates and the health and growth rate of the birch seedlings were observed over a period of 322 days. The results show that each of these biochars complies with the European Biochar Certificate (EBC) guidelines. No toxic heavy metals were detected. Furthermore, the presence of the biochar in the substrates enhanced the survival rate of the birch trees.

1. Introduction

In view of climate change and global urbanization, trees are increasingly becoming a central element of urban greenery (Zölch et al., 2016). However, due to extreme site conditions in street spaces, many trees can no longer meet the demands placed on them. Urban trees increasingly suffer from stressors such as tree pits that are too small and restrict root growth, soil compaction in the root zone, pollutants in seepage water or the air, or a lack of nutrient salts such as potassium or phosphorus (Roloff, 2013; Zuber, 2013). Roloff et al. (2008) have confirmed that tree vitality is declining in urban areas due to a significant deterioration in growing conditions

and predict that expected increases in droughts and heat waves will lead to the partial failure of urban trees over the next 50 years. Roloff (2013) points out that today's urban trees only achieve 50% of their life expectancy, and trees on street only achieve 25% due to site specific problems that affect their vitality. Thus, on average, a newly planted urban tree rarely reaches an age of more than 30 years (Roman and Scatena, 2011).

However, ecological, social, and aesthetic services, such as shading and cooling in cities, can only be provided by flourishing trees that have optimal growing conditions (Duthweiler et al., 2017). In the debate on sustainable urban development, the role of urban trees in sustainable water management is now increasingly discussed under the term "sponge city" because plants and their surrounding substrate layers filter and remove pollutants and reduce runoff into sewer systems through direct water uptake, storage, and long-term evapotranspiration (Zevenbergen et al., 2018; Zölch et al.,

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2016). Trees that die too early cannot provide these ecosystem services over the long term, leading to increased maintenance and costly new plantings (Roloff and Rust, 2019). A tree-friendly solution to providing adequate root space for trees in limited street spaces is to use substrates with a high proportion of large-grain sizes that have a stable open pore volume, are less compactible and maintain a high air capacity. If these so-called structurally stable tree (SST) substrates are introduced into the tree pit and the adjacent substructure of the road space, they improve root activity and can serve as a subterranean root corridor to adjacent green spaces thus optimizing site conditions (Balder, 2002; Grabowsky and Bassuk, 2002). To counteract the other stressors of urban trees, the substrate should have a high capacity to hold water and store nutrients (Schönfeld, 2017) and should not contain any heavy metals. SST substrates can potentially retain water in the root zone due to their large pore sizes, allowing for continued evapotranspiration during dry periods. This process can also be an important component of sustainable stormwater management measures for resilient cities (Zevenbergen et al., 2018; Embrén et al., 2008).

In Switzerland the city of Basel is already successfully using an SST substrate based on granite gravel. The city's focus has primarily been on stability and water permeability, and long-term nutrient supply has so far only played a minor role (Saluz, 2017). Early results from Stockholm, however, show that the first SST substrates which meet civil engineering requirements and contain a proportion of organic compost and biochar have been having positive effects on urban trees (Embrén, 2016).

Various authors have demonstrated the positive effect of biochar – a product obtained through the pyrolysis of biomass – on soil and plant growth (Dai et al., 2020; Purakayastha et al., 2019; Frenkel et al., 2017; Glaser et al., 2002). For example, biochar can increase the nutrient storage capacity and reduce the leaching of important plant nutrients compared to mineral fertilizers (Dai et al., 2020). At the same time, a slow release of nutrients such as phosphorus can improve the nutrient balance in the soil over the long term (Glaser and Lehr, 2019; Gwenzi et al., 2018; Chan, 2009; Steinbeiss et al., 2009). Moreover, biochar also has the potential to improve soil enzymatic activity, to immobilize heavy metals, such as nickel (Ni) and to influence plant phenological traits at the same time (Turan, 2022).

Since biochar has a highly porous structure, its bulk density is often low. This property positively influences its capacity to hold water and soil aeration. Since the biochar combines with the organic soil matter to form stable aggregates, it demonstrably improves the structural stability of the soil. These positive properties mean that plant biochar can be used as a supplement to organic matter in urban tree substrates (Gul et al. 2015; Abel et al., 2013; Verheijen et al., 2010).

Due to its abundant macronutrients, pyrolyzed fecal biochar also has great potential as a tree substrate component that replaces mineral fertilizers and closes nutrient cycles using natural recycled products, as shown in Fig. 1 (Bleuler et al., 2020; Ilango and Lefebvre, 2016). However, while much research is available on the effect of biochar from plant biomass, there are few studies on the use of fecal biochar from animal or human excreta.

The use of fecal biochar has not been extensively researched and it is rarely used in practice, probably due to the fear of diseases and heavy metals that may be present in the fecal matter (Bleuler et al., 2020; Ilango and Lefebvre, 2016). However, studies by Ebert et al. (2021), Bleuler et al. (2020) and Gold et al. (2017) indicated that the heavy metal content of fecal biochar is even lower than in previously studied sewage sludge-based biochar. Furthermore, the high temperatures of pyrolysis treatment eliminate pathogens (Bleuler et al., 2020). Nevertheless, little is known about the behavior of fecal biochar in SST substrates and there is a lack of knowledge of the optimal composition for tree growth.

This study aims to evaluate the technical feasibility of using SST substrates containing biochar from fecal matter as a nutrient carrier. It also examines its suitability as a tree substrate. It will be compared to a reference substrate (control) containing crushed stones and sand with little amounts of organic matter, as is the case in commercially available tree substrates. Fertilizer will be added to the positive control substrate, but not to the negative control. The small amounts of organic matter in commercially available tree substrates is one of the reasons why leaching of nutrients has often been observed (Saluz, 2017; Schönfeld, 2017).

The following hypotheses were formulated:

1. Substrates containing fecal biochar leach lower levels of nutrients than control substrates.
2. The addition of fecal biochar has no negative effects on the growth and health of birch seedlings.
3. Substrates with fecal biochar improve tree growth in comparison to substrates with plant-based biochar, the positive control substrate and the negative control substrate.
4. Substrates containing biochar allow roots to grow more strongly than the positive and negative controls.

2. Material and methods

To test the hypotheses, biochar was produced and analyzed for nutrients and toxins. Subsequently, an SST gravel-based substrate containing a proportion of pyrolysis biochar was formulated. Different variations of

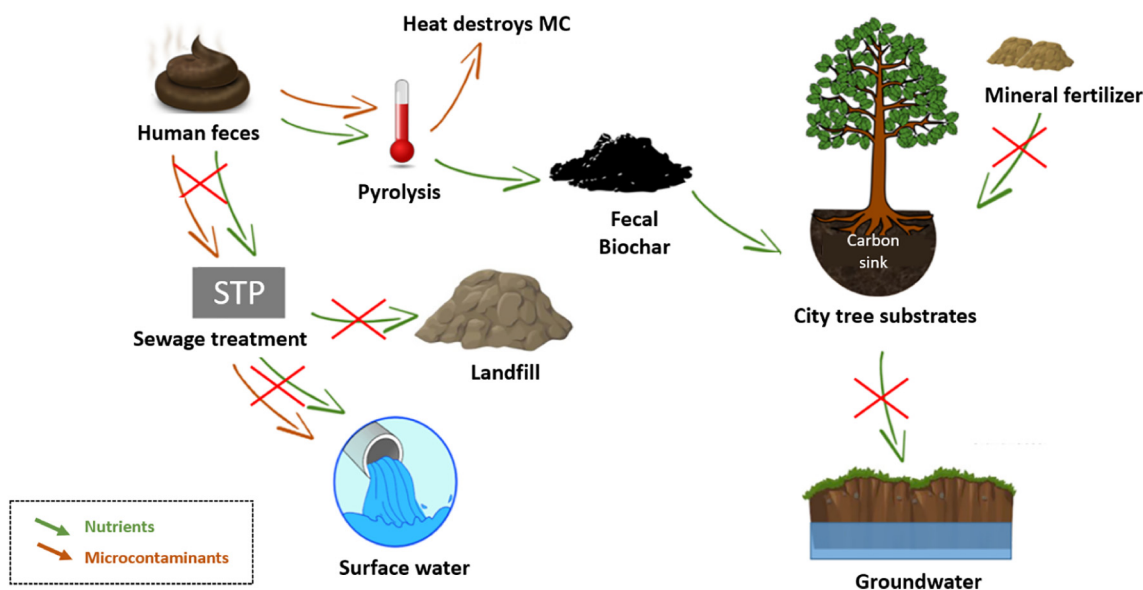


Fig. 1. Potential environmental benefits of biochar from animal and human feces for urban tree plantations (Bleuler et al., 2020).

this substrate were then tested by growing birch tree seedlings in them in an 11-month trial. The trial took place from June 6, 2018, to April 24, 2019 (322 days) in a foil tunnel on the Zurich University of Applied Science campus in Waedenswil, Switzerland.

The SST substrates consisted of 90% crushed granite gravel (<2 mm–16 mm), 5% expanded shale (2–50 μ m), 5% sand (2–50 μ m) and 5 or 10 vol% biochar, respectively. The substrate composition variants were based on research carried out at the ZHAW (Saluz, 2017), tree experiments in Stockholm (Embrén, 2016), experience gained in Basel with “structured soil” (S. Ramin, M. Sonderegger, City of Basel, 5.1.2017, personal communication) and experience from Cornell University’s experiments with “CU structured soil” (Grabowsky and Bassuk, 2002).

Six SST substrate variants, a positive control and a negative control were set up. This total of 8 substrate variants, each replicated 5 times, resulted in a total of 40 pots (see Fig. 2). For each substrate variant, five 20-liter planting pots were filled with the substrate mixtures. One-year old birch seedling (*Betula pendula*) were planted in each of the pots and randomly placed in a foil tunnel on the ZHAW campus in Waedenswil. No further organic material was added to the substrate, except for minimal amounts still attached to the bare roots. To characterize the biochar variants, chemical analyses of the biochars were performed according to the requirements of the European Biochar Certificate (EBC).

2.1. Source materials and pyrolysis

The horse manure used in this study was obtained from a horse farm at Schoenberg, Switzerland. Approximately 200 l of horse manure, consisting of fecal matter, bedding and straw served as feedstock for the pyrolysis. The sample was spread out and air-dried by turning it over several times over two days, in preparation for pyrolysis. For analysis purposes, a representative sample (2 l) was taken from the 200-l and further dried at 110 °C in a laboratory oven to achieve 100% dryness.

The dry toilet matter used in this study was collected from rental composting toilets (Kompotoi AG, Zurich, Switzerland). The substrate consisted of a mixture of human feces and urine, toilet paper, wood chips and leaves. This human waste was collected in 100-liter barrels underneath the toilets and transferred to a container, where 1 l of a microorganism culture (EM soil FIT) and 5 kg of Bokashi compost were added to each batch. Subsequently, the waste mixture was stored under pressure for 1 week and then stored (J. Linder, Kompotoi AG, Zurich, Switzerland, pers. communication). A representative sample was then taken and dried at 110 °C in a laboratory oven to achieve 100% dryness.

Entrance of greenhouse					Window
P11	V11	Cn1	K21	V21	
K22	Cn2	K11	Cp1	V12	
V13	Cp2	P12	V14	K12	
Cn3	P21	V22	Cn4	Cp3	
V23	P13	K23	V24	P22	
Cp4	K13	P23	P14	Cn5	
P24	K24	Cp5	K14	P15	
K15	V25	V15	P25	K25	

Fig. 2. Random Block-Design of the plant pot test.

Horse manure and human waste were pyrolyzed in a lab-scale pyrolyzer (Pyreka, Pyreg AG, Dörth, Germany) at a temperature of 550 °C with a reactor residence time of 20 min. During pyrolysis, the reactor was purged with 4 l of nitrogen every hour to ensure an oxygen-free environment and a steady flow of pyrolysis gases. After pyrolysis, the sample was cooled to room temperature. A representative subsample of the produced biochar was collected and stored in a freezer until analysis (–21 °C). The biochar intended for the plant experiments was stored in plastic bags for approximately one week prior to the start of the experiment.

2.2. Biochar analytics

The methods used to analyze the biochar were selected according to the guidelines of the European Biochar Certificate (EBC) (2012) to ensure that the requirements for a possible certification were met (Table 1).

2.3. Experimental design

The substrates and control setups were prepared and named as listed in Table 2. Five replicates of the SST substrate variants were produced for each basic setup and arranged in the greenhouse in a random-block design (Fig. 2). The pot labels included information on the setup and a replicate number (e.g., P15 stands for the setup with “5% biochar from horse manure, replicate #5”).

For the positive control, a liquid fertilizer (Wuxal, N:P:K = 8/3.6/5; <https://www.hauert.com>) was selected. The nitrogen content in the positive control Cp was adjusted to the nitrogen content of the composting toilet biochar (KT). The pots were irrigated with rainwater that was collected and stored in a cistern on campus. During the hot summer of 2018 refilling of the cistern was necessary. The refilling events were not recorded, and no data were collected on the quality of the irrigation water. For orientation purposes, the average EC and the average pH-value of the Wädenswil drinking water are used in some figures (Figs. 8, 10).

To mimic natural conditions, average quantities for watering the pots were calculated based on Waedenswil precipitation data (Meteosuisse, 2021). In the first two weeks after the start of the experiment on June 6, 2018, each pot was watered once a week with 3.6 l of rainwater from the cistern. From then on, the pots were watered twice a week (less in winter) with 1.8 l of water per pot and day. Water samples were obtained from the saucers of the plant pots.

2.4. Climate data

Temperature, relative humidity and the dew point in the greenhouse were measured hourly from June 29, 2018, until the end of the experiment using iButton sensors (iButtonLink, Whitewater, Wisconsin, USA). One sensor per substrate was placed on the pot surface.

Table 1
Methods of biochar analysis.

Parameter	Method
Yield	Measured in the continuous production process. Mass determination before and after pyrolysis
Bulk density	VDLUF A 13.2.1 (VDLUF A 1991)
pH	pH-probe (Hach-Lange PHC301)
Electrical conductivity (EC)	EC probe (Hach-Lange CDC401), ISO 10390
Ash content	Residue analyzed gravimetrically after heating in a muffle furnace (Nabertherm L3) at 550 °C according to EBC (2012)
Carbon, nitrogen, and hydrogen	CHN-Analyzer (Leco Truespec Micro) after DIN 51732:2014-07 (n.d.)
Chemical and other element compositions	ICP-OES analyzed after microwave digestion (ultraCLAVE4) at 250 °C and 120 bar for 10 min of 0.2 g sample with 5 ml HNO ₃ , 1 ml H ₂ O ₂ and 0.3 ml HF

Table 2
Substrate types in random block design.

Variant	Dosage of biochar	Basic setup	Number of pots
Negative control	–	Cn	5
Positive control (fertilized)	–	Cp	5
VE: plant-based biochar (charged with nutrients)	5 vol%	V1	5
	10 vol%	V2	5
PM: biochar of horse manure	5 vol%	P1	5
	10 vol%	P2	5
KT: biochar of fecal matter	5 vol%	K1	5
	10 vol%	K2	5
Total			40

2.5. Leachate analysis

Every 1–2 weeks after watering, the pH value and the electrical conductivity in the leachate were measured. Further analysis parameters (nutrients, heavy metals) were determined 1 to 5 times throughout the experiment since leaching of nutrients and heavy metals might impact the groundwater quality underneath a city tree planted in SST substrate. Analyses were conducted as listed in Table 3.

2.6. Plant observation

To assess the influence of the addition of biochar on tree and root growth different parameters were measured (Table 4). Furthermore, pictures were taken of all of the plants at the end of the experiment for further documentation.

2.7. Statistical analysis

Statistical comparisons of the experimental variants were performed with R software (<https://www.r-project.org/>) using Kruskal-Wallis and Wilcoxon tests, since most parameters did not show a normal distribution.

3. Results

3.1. Climate data

In summer 2018 and in spring 2019, the temperatures in the greenhouse fluctuated between 10 and 15 °C at night and up to 40–50 °C on hot days (Fig. 3). From mid-October 2018, temperatures decreased rapidly. They remained between 0 °C and about 10 °C throughout the mild winter of 2018/19, falling below 0 °C for only a few hours.

Relative humidity varied between from a minimum of about 20% during the day to about 80–90% at night from the start of the experiment to the end of October 2018 and from the beginning of February 19 to the end of the experiment in April 19 (Fig. 4). During the winter months,

Table 3
Methods of leachate analysis.

Parameter	Method	Frequency
pH	Hach-Lange PHC301, ISO 10390 (I2005)	Every 2nd week
Electrical conductivity	Hach-Lange CDC401, ISO 10390 (2015)	Every 2nd week
Na, NO ₃ ⁻ , NH ₄ ⁺ , PO ₄ ³⁻ , K, Cl, SO ₄ , Mg	Ion chromatography (IC)	4 × during trial
	Ion chromatography (IC)	5 × during trial
Non purgeable organic carbon (NPOC)	TOC-L (Shimadzu)	Beginning of trial and 2 months after beginning of trial
Total nitrogen (TN)	CHN Analyzer Leco TrueSpec Macro	Beginning of trial and 2 months after beginning of trial
Heavy metals	Inductively coupled plasma mass spectrometry (ICP-MS)	Beginning of trial

Table 4
Methods of plant observation.

Parameter	Method	Frequency
Height	Maximum length with scale	Beginning of trial, after 6 months and at the end of the trial
Chlorophyll	Dualex Force A	At the end of the trial
Root length	Maximum length with scale	At the end of the trial
Root volume	Average of two width measurements times maximum root length	At the end of the trial
Stem shoots	Visual assessment	At the end of the trial

relative humidity was higher, >80% to 100%. The reason for these dynamic changes was the temperature fluctuation in the foil tunnel.

3.2. Biochar characterization

3.2.1. Elemental composition

Fig. 5 (left) shows the total elemental composition (C, H, N, S, O) of the three biochars and the ash content (mineral residue). The composition of the KT biochar is similar like that of the VE biochar in terms of the main elements. The high ash content of the PM biochar can be explained by the high proportion of sand in the raw material “horse manure” (Fig. 5, right).

3.2.2. Nutrient content of the biochars

The concentration of the three main nutrients nitrogen (N), phosphorus (P) and potassium (K) in all three biochars are shown in Fig. 6. The VE biochar (commercial plant-based biochar, charged with nutrients) and the PM biochar (horse manure biochar) had very similar macronutrient contents, while the KT biochar (composting toilet biochar) contained slightly less potassium and statistically significantly more nitrogen and phosphorus.

The increased N and P content in the KT-biochar can be explained by the human feces in the source material. In addition to solid feces, the source material also contained sawdust soaked in urine, which contains a high concentration of these nutrients (Rose et al., 2015). The KT-biochar had the highest concentration of elements such as calcium (Ca), magnesium (Mg), iron (Fe) and sulfur (S). However, with the main exception of silicon, the VE- and PM-biochars had similar element concentrations. It can be assumed that the nutrients in the VE-biochar result from nutrient “charging” during the production process. The nutrients in the fecal biochar are most likely derived from the digestive systems of humans (KT) and horses (PM).

3.2.3. Heavy metal content of the biochars

For all three biochars, the content of the heavy metals arsenic (As), cadmium (Cd), copper (Cu), chromium (Cr), lead (Pb), mercury (Hg) and nickel (Ni) (Fig. 7) were lower than the limits specified for the “EBC-AgroBio” quality level of the European Biochar Certificate (EBC) (2012) (Fig. 7, left). The concentrations of Cu and Zn found in the KT biochar were significantly higher than in the other two biochars. In the case of zinc (Zn), the content in the KT-biochar was higher than permitted for “EBC-AgroBio” level but still met the requirement for “EBC-Agro”. The two-fold higher zinc and copper content of the KT biochar compared to the PM and VE biochars indicates its provenance from human feces because copper and zinc are essential micronutrients for human nutrition that are also excreted.

3.3. Characterization of leachates

3.3.1. Electrical conductivity and pH value of the leachates

At the beginning of the experiment, the electrical conductivity (EC) of the leachates (Fig. 8) showed large differences between the controls (Cn and Cp, app. 450 μS/cm) and the biochar amended substrates, ranging from 1000 μS/cm (K1, 5 v/v biochar) to >2500 μS/cm (P2, 10 v/v biochar). Between June 6, 2018, and October 6, 2018, the EC of the leachates decreased exponentially for all of the biochar-amended substrates. This is

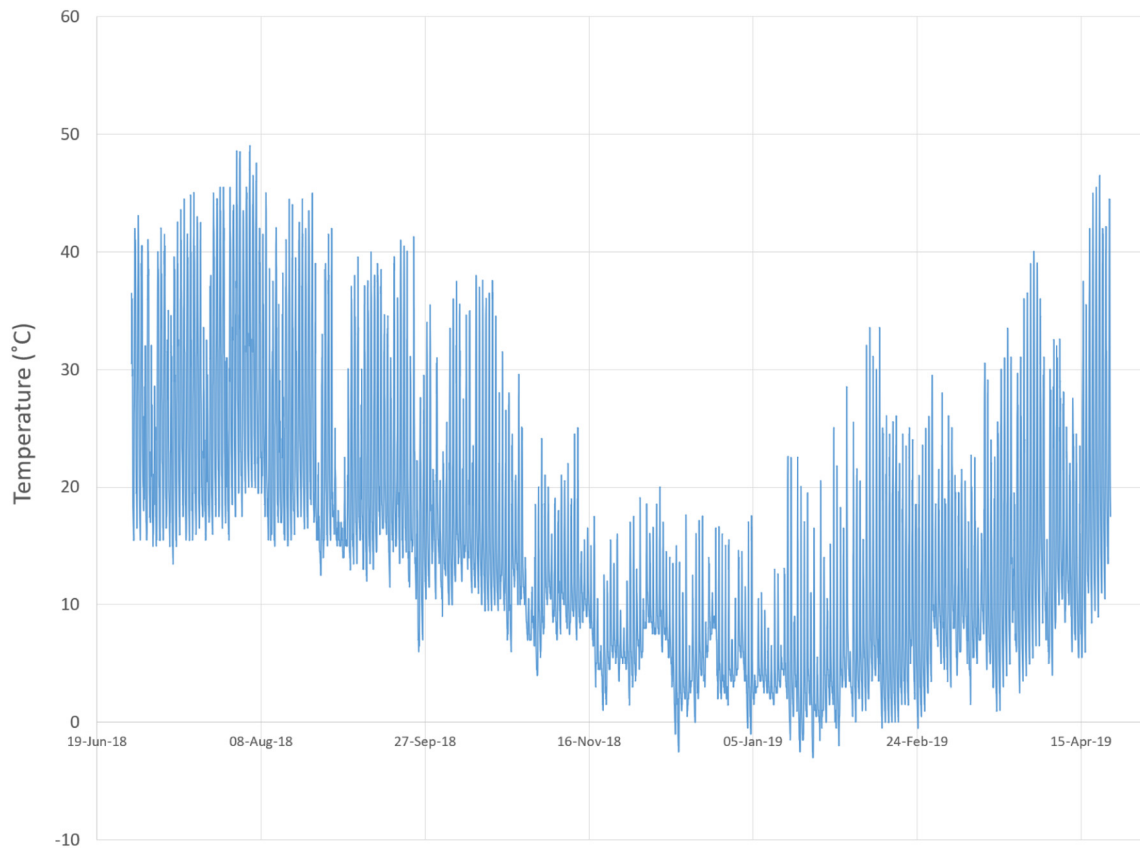


Fig. 3. Temperature profile during test period (June 6, 2018–April 24, 2019).

attributed to the leaching of ions from the substrates when watering the birch seedlings. After October 2018, the EC values in all of the leachates leveled out at an EC value slightly lower than the average of Waedenswil

drinking water. Therefore, it can be concluded that after approximately 4 months, the leaching process came to end because the leachable ions had been washed out.

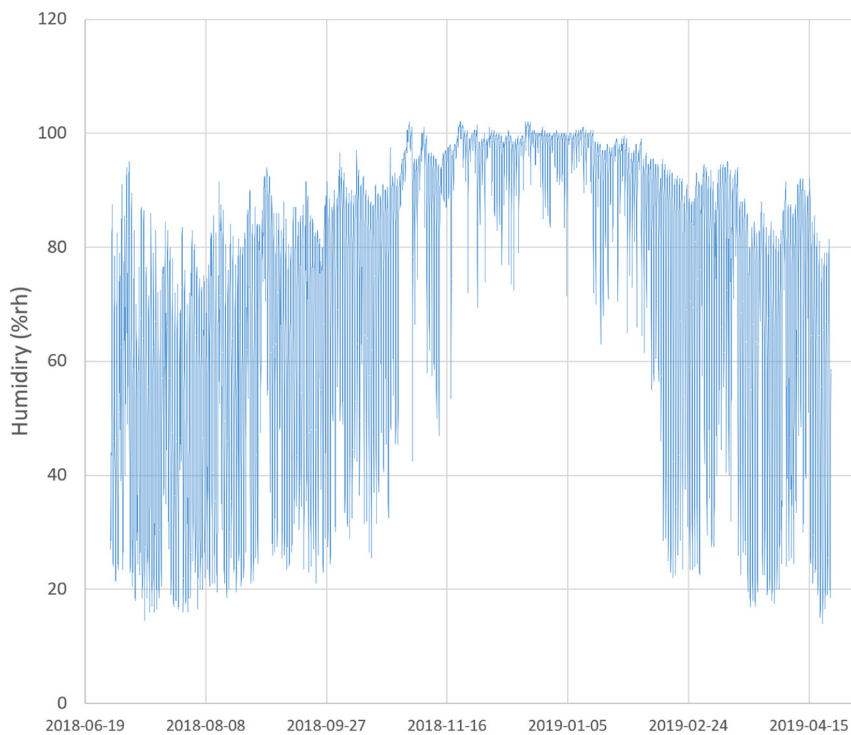


Fig. 4. Relative humidity during the test period (Sensor Pot V24, June 6, 2018–April 24, 2019).

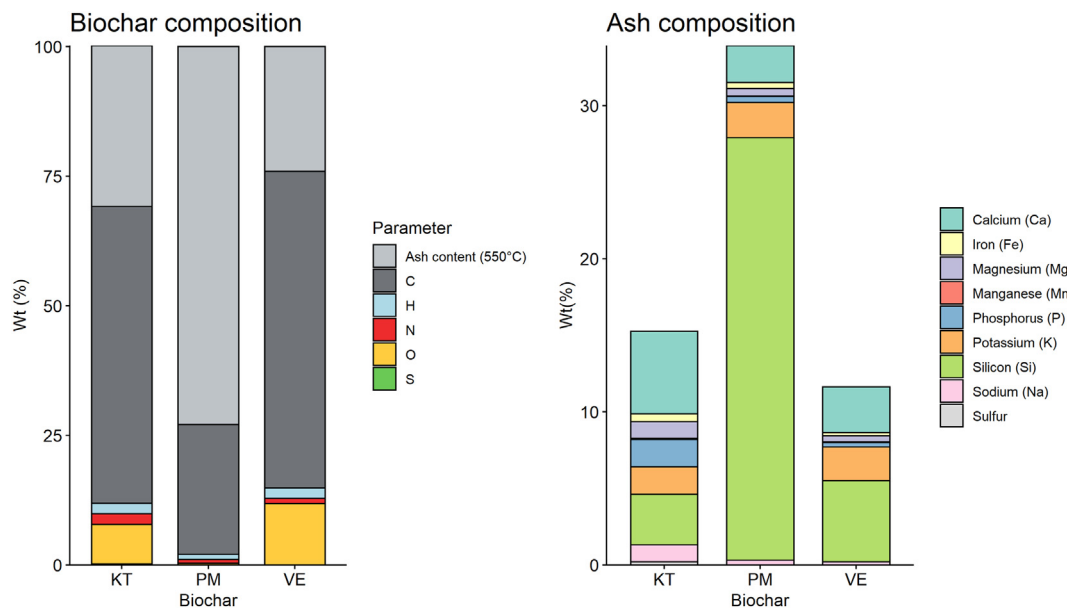


Fig. 5. Main components (wt.%) of the three biochars (left) and elemental composition of the biochar ashes (right).

At the beginning of the experiment, the leachates from the substrates with 10% carbon v/v (K2, P2) had an EC that was about 2 times higher than those with 5% carbon (v/v). This difference between the substrates also decreased during the first 4 months due to leaching. At the end of the experiment, the substrates with 10% carbon content still had 14–23% higher EC in the leachate compared to those with 5% carbon content (based on mean values between February 14, 2019, and April 24, 2019; $n = 6$ per setup).

In contrast, the pH-values (Figs. 9, 10) fluctuated throughout the summer with the highest values occurring at the end of August 2018 when there was a maximum of pH 9.6 that differed by a Δ pH of 1.6. From mid-October 2018 to mid-February 2019, all of the leachates had a pH of between 7.8 and 8.1. With the onset of spring 2019, the pH values started to fluctuate again, but did not differ from each other as much as in summer 2018. The substrates containing biochar VE (V1, V2) had the highest pH

values of all the setups. Fig. 10 also shows that all variants with 10 vol% biochar had a slightly higher pH than the variants with 5 vol%, especially in the first 2 1/2 months.

The magnitude of the pH differences between the controls and the biochar-amended substrates after Dec. 3, 2018 (Fig. 10) indicates that the type of biochar does not have a long-term effect on the pH of the leachate from the SST substrate. The pH-drop from as high as pH 9.5 to below pH 8 during the first 3.5 months coincides with the washout of the easily soluble ions and can probably be explained by alkaline ions in the leachate (see Section 3.3.2).

3.3.2. Concentration of NO_3 , PO_4 and K in the leachates

Figs. 11–13 show the leachate concentrations of NO_3 , PO_4 and K at the beginning of the experiment (June 6, 2018), after 68 days (August 13, 2018) and after 322 days (April 24, 2019). All of the figures show the

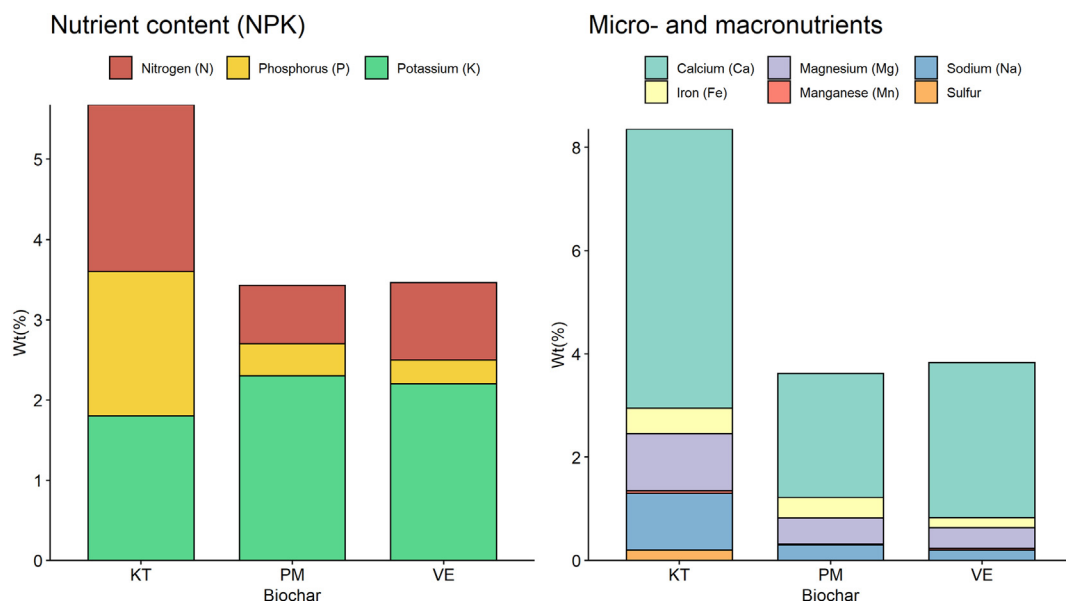


Fig. 6. Concentration of macro- and micronutrients for each biochar (wt.%)

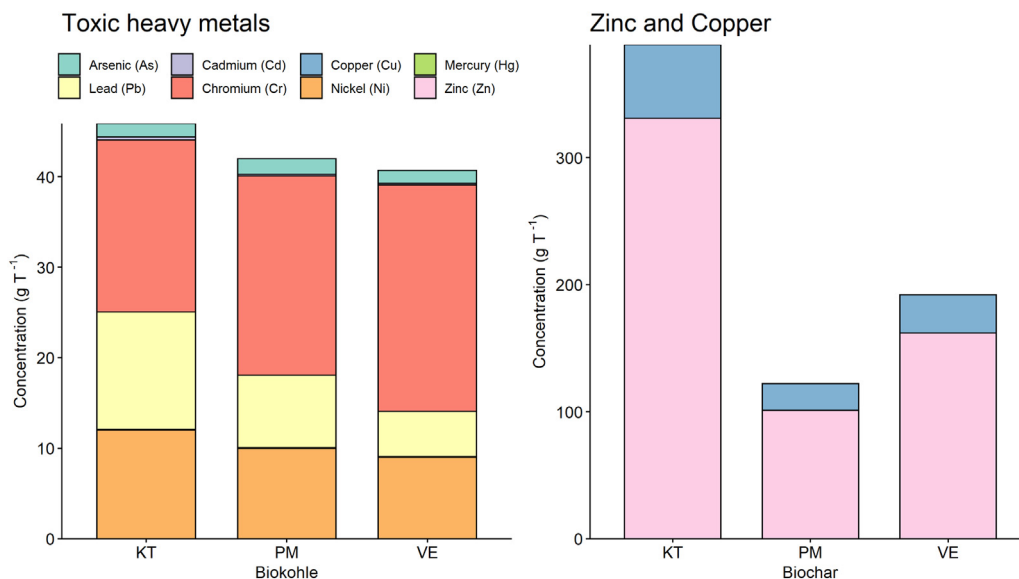


Fig. 7. Heavy metals (g/1000 kg) in the three biochars.

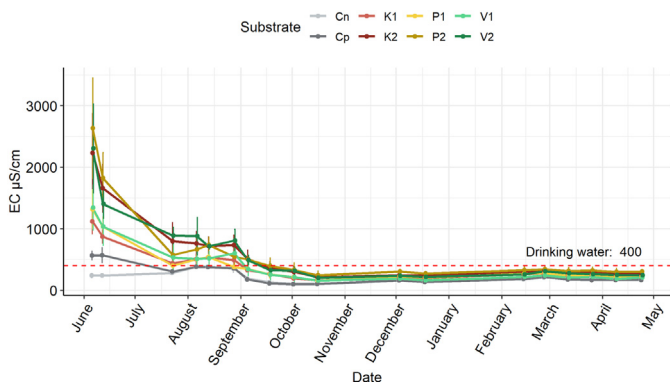


Fig. 8. Electrical conductivity (EC) of the leachates in $\mu\text{S}/\text{cm}$.

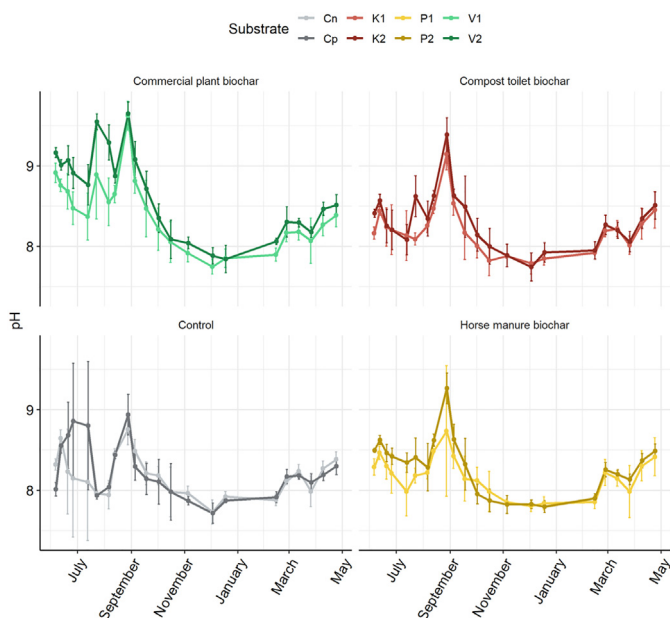


Fig. 9. pH of the leachates in different substrates.

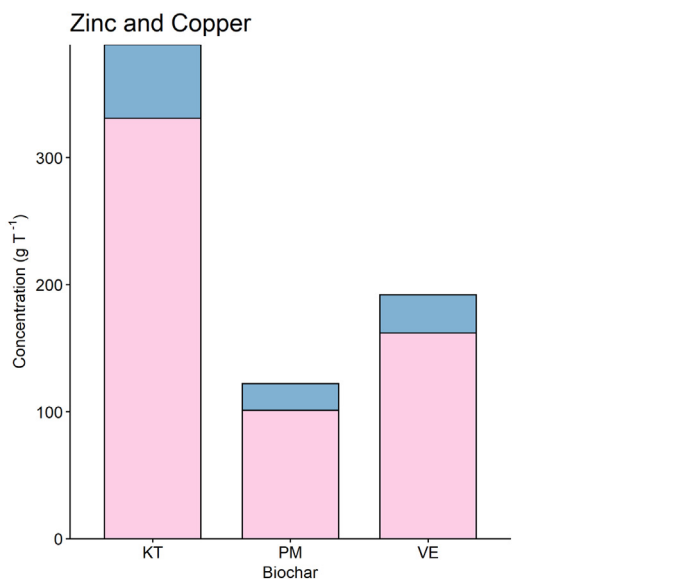


Fig. 10. pH values in leachates over time, for each substrate type.

concentrations in the leachates (A), the concentration as a percentage of the first leachate sample (B) and the proportion of the respective elements in the three biochars (C).

3.3.2.1. Nitrate (Fig. 11). The highest nitrate leachate concentration – approximately 20 times higher than in all other SST substrates – was found in the leachate from Cp (positive control) at the start of the experiment. The 95% decline on day 68 showed that most of the nitrate added to Cp as a fertilizer had been leached out. The other 7 setups exhibited the same pattern from a lower starting level. Interestingly, the nitrate concentration in the leachates from Cn (negative control) was significantly higher than those in all the setups with biochar. This suggests that the three biochars did not contribute to nitrate leaching and might even have slowed down nitrate leaching.

Finally, compared to the leachates from the KT and PM, the SST substrates based on VE (V1 and V2) had a significantly higher nitrate concentration in the leachate than P1 and P2. At the end of the experiment, on day 322, the nitrate leachate concentrations were between 0.5 and 0.68 mg/l, with Cp still being significantly higher (0.92 ± 0.06 mg/l).

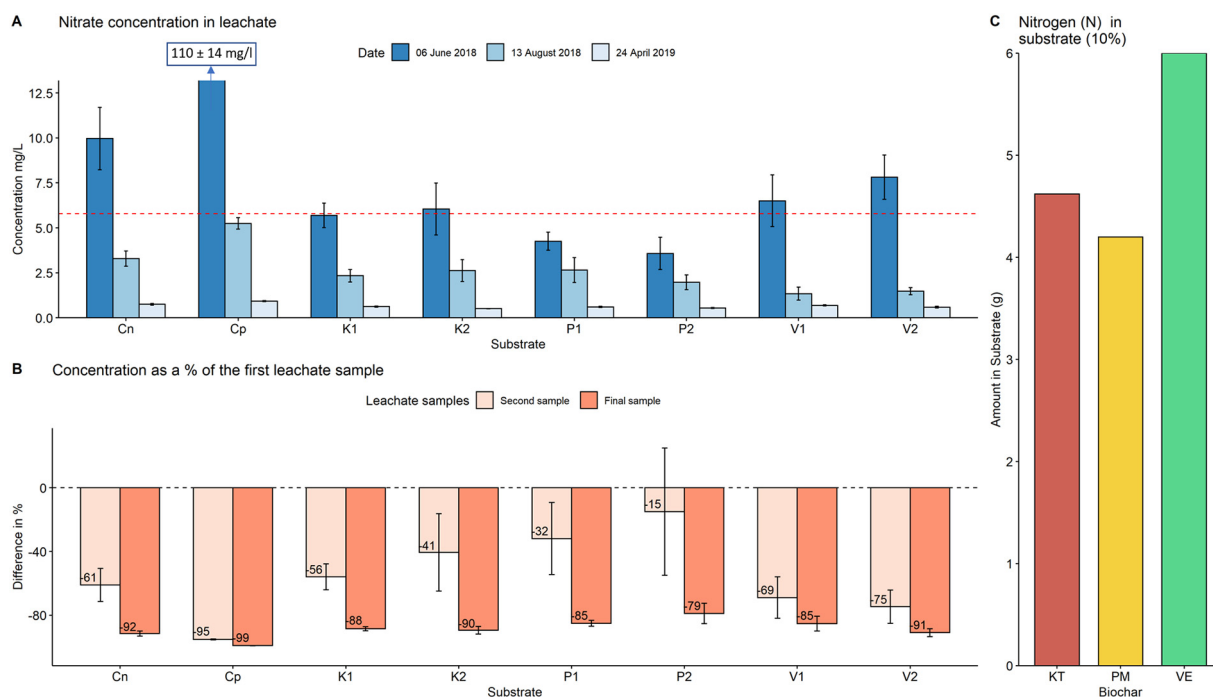


Fig. 11. A) Nitrate concentration at three time points. Red line = drinking water in Waedenswil. B) Decrease of nitrate in leachate in % of the first measurement. C) Nitrogen content in wt.% of the biochar.

The nitrogen content of the three biochar setups did not seem to influence nitrate leaching. A comparison of the nitrate content with Waedenswil drinking water indicated that the cistern water used for watering was mainly rainwater on the three sampling days.

3.3.2.2. Phosphorous (Fig. 12). The highest initial leaching concentration of phosphate was 7–14 times higher in V2 and 3.5–6.9 times higher in V1 than in all of the KT- and PM-based setups. The 87–93% decline in V1 and V2 on

day 68 shows that most of the mobile phosphate ions had been leached out by then. One reason for this is the nutrient loading of biochars V1 and V2 with compost extract (Verora GmbH, pers. communication), as well as the addition of liquid mineral fertilizer in the positive control (Cp). Interestingly, the phosphate in the leachates for all of the setups increased slightly (and on a very low level) between day 68 and day 322, except for K2 and P2. The phosphate content of the three biochar setups did not seem to influence the nitrate leaching.

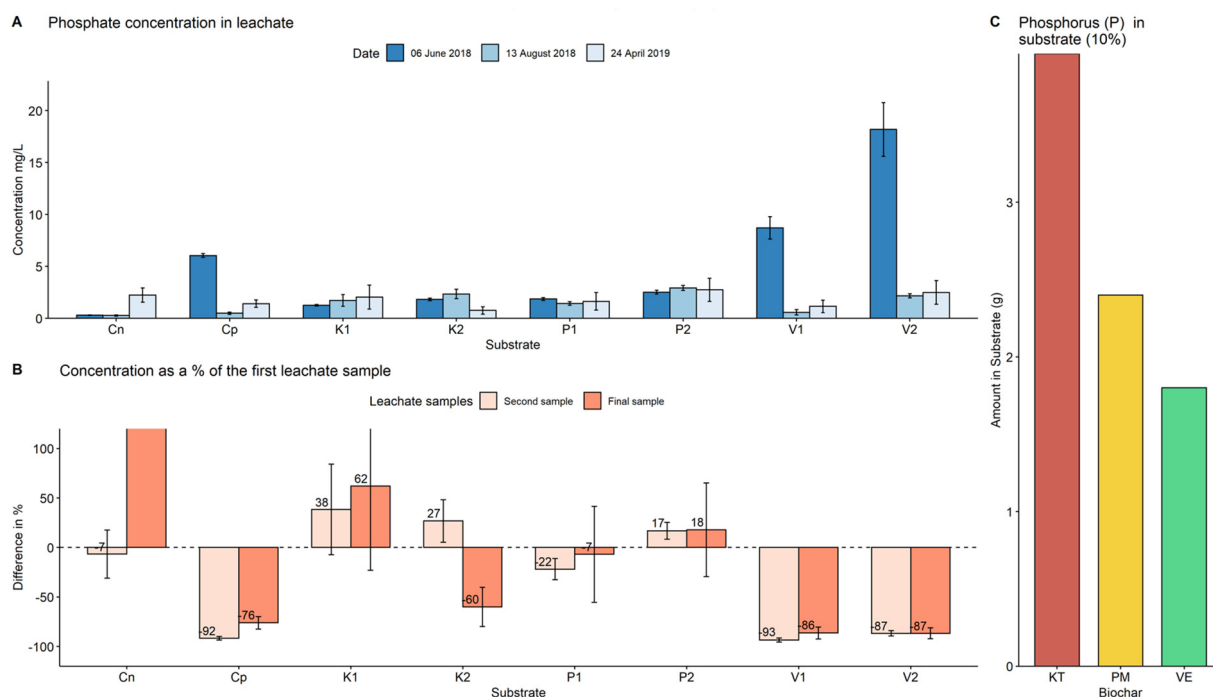


Fig. 12. A) Phosphate concentration at three time points. B) Decrease of phosphate in leachate in % of the first measurement. C) Phosphorus content in wt.% of the biochar.

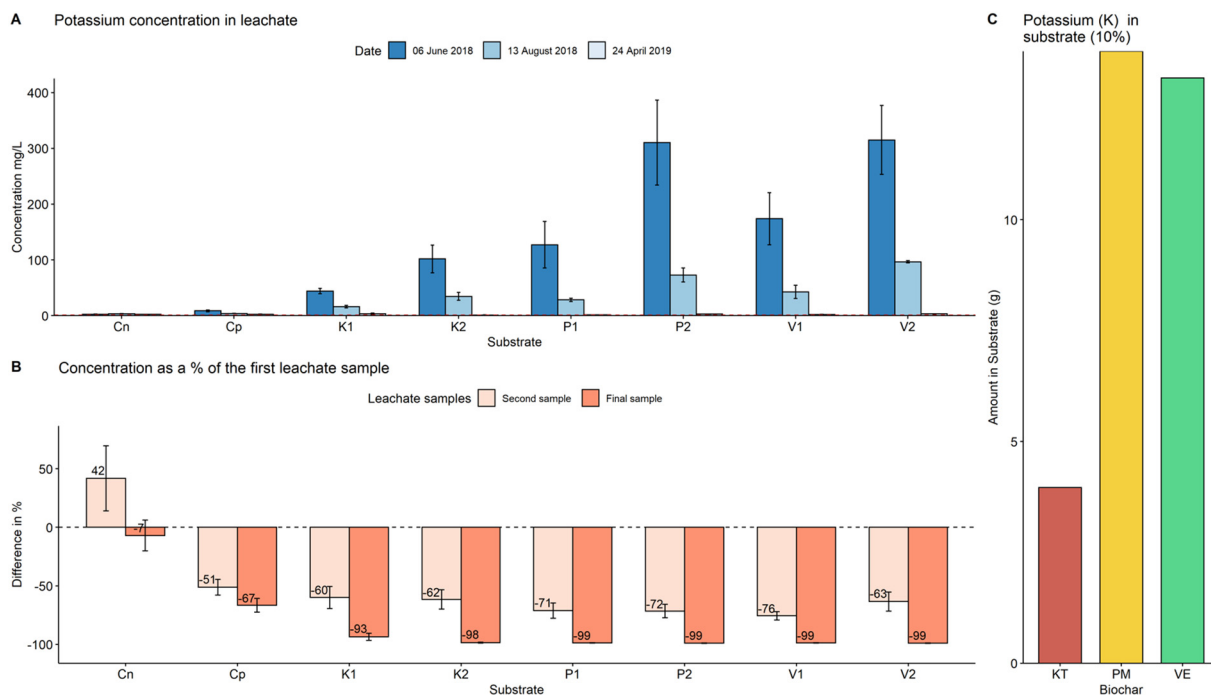


Fig. 13. A) Potassium concentration at three time points. B) Decrease of potassium in leachate in % of the first measurement. C) Potassium content in wt.% of the biochar.

3.3.2.3. *Potassium* (Fig. 13). Compared to nitrate and phosphate, the leachate concentrations of potassium on day 1 were higher in all of the biochar-based substrates than in Cn and Cp (Fig. 13). By the end of the experimental period, virtually no leaching occurred. All three biochar substrates leached significantly more potassium than the negative and positive controls.

Furthermore, the Ca²⁺, Cl⁻, Mg²⁺, Na⁺ and SO₄²⁻ ions were leached out significantly more from the KT and PM substrates than from the VE substrate and the two control substrates, Cn and Cp. The relatively high proportion of these two ions can be explained by the presence of human urine and horse urine in the source material for KT and PM.

For all substrates, the potassium concentration in the leachate seemed to correlate with the proportion of biochars in the substrate. The substrates with compost toileting biochar had the lowest concentrations, while the substrates with horse manure and the nutrient-charged plant biochar had similarly high values. All setups with 10% v/v of biochar had higher concentrations of potassium in the leachates than those with 5% v/v.

3.3.3. *Heavy metals*

The heavy metal concentrations in the leachate were only measured once at the beginning of the experiment (June 6, 2018). Very small amounts were found, with arsenic only present in three of the biochar

amended substrates (V1, V2, P2) at concentrations very low and below the predetermined thresholds (Table 5).

3.4. *Tree and root growth*

The growth of the birch tree seedlings during the 322 days was low (Fig. 14). Their relative size increase with respect to their initial size is a more interesting parameter than their absolute size. The stagnation of the growth in Cn (negative control) and the dieback in the positive controls Cp were evident. In all biochar-amended substrates the birch tree seedlings survived and showed some growth. This is an indication that the addition of plant-based biochar can have a positive effect on seedling development.

At the end of the experiment, after 322 days, the root volume of the birch tree seedlings in biochar-amended substrates was 270–310% higher than in the negative controls (Fig. 15). An amount of 5% biochar in the substrate was sufficient to significantly improve the conditions for the birch trees, most likely due to the improved capacity of the SST substrate to hold water. The appearance of the roots and shoots of the eight test setups is documented in Fig. S1.

4. *Discussion*

Birches are very fast and tall-growing woody plants that can reach growth heights of up to 7 m after only six years. When fully grown, they

Table 5

Heavy metal concentrations in the first leachate sample (6.6.2018). Blue: measured value above the lower detection limit. <LOD: measured values are below the detection limit.

Setup	Cu (mg/l)	Pb (mg/l)	Zn (mg/l)	As (mg/l)	Cd (mg/l)	Cr (mg/l)	Ni (mg/l)
Cn	0.012	<LOD	0.014	<LOD	<LOD	<LOD	<LOD
Cp	0.012	<LOD	0.033	<LOD	<LOD	<LOD	0.013
P1	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
P2	<LOD	0.011	<LOD	0.014	<LOD	<LOD	<LOD
K1	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
K2	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD	<LOD
V1	<LOD	<LOD	<LOD	0.014	<LOD	<LOD	<LOD
V2	<LOD	<LOD	<LOD	0.029	<LOD	<LOD	<LOD

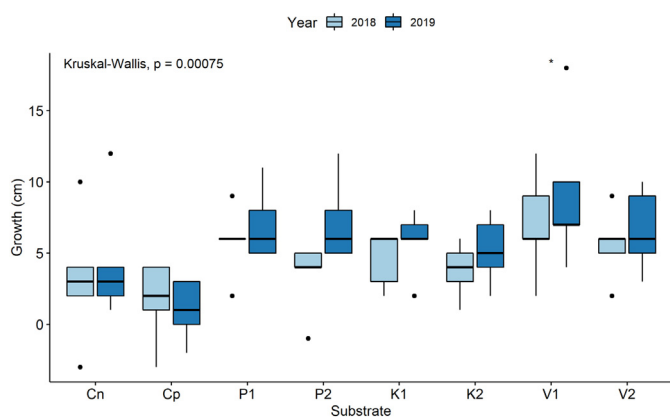


Fig. 14. Absolute tree growth (cm) by substrates after 4 months (October 2018) and at the end of the experiment (April 2019).

can reach up to 30 m, even higher in individual cases, with specimens reaching ages of up to 160 years (Roloff and Bärtels, 2018). Our experiment focused on the second year in the life of birch trees.

During the 322 days of the experiment, the birch trees grew on average less than 10 cm, which is low. The birch seedlings had to survive under poor conditions: The SST substrate itself did not have a high-water storage capacity and the temperature fluctuations during the growing season were high (with temperatures up to >50 °C on the pot surface), probably resulting in the substrate drying out at least temporarily.

In the case of severe drying of soils, it is known that water initially runs off superficially and can wash away fertilizers or pesticides until the soil is completely wetted (Gimmi, 2005). A similar effect could have occurred in this experiment (strong leaching from the dried substrate). The birch seedlings may also have been severely stressed by the drying out. From July to October 2018, the pH value in the leachate was always higher than pH 8, with a maximum pH > 9.5. The leachate from the V2 batch (biochar VE, 10%) had the highest pH value and the leachates with 10% biochar had consistently higher pH values than the leachates with 5% biochar. However, the negative and positive controls consistently had the lowest pH values. Beginning in mid-October, the pH of all of the leachates stabilized at approximately pH 7.7 to 8 before rising again to pH > 8 from mid-February. The pH fluctuations at the beginning of the measurement period were likely due to intermittent drying and rewetting. The experiment

demonstrated that leaching of easily soluble ions, such as K⁺, Na⁺ or Cl⁺, from the biochar amended SST substrates occurred initially, but ended after 3–4 months. This was probably because most of the easily soluble ions had leached out by then.

The KT and PM fecal biochars and the nutrient charged VE plant biochar used in this project contained significant amounts of the nutrients N (ammonium/nitrate), P and K. The fertilizer dosage of the Wuxal liquid fertilizer in the positive control (Cp) was selected so that the nitrogen content in the fertilized substrate corresponded to that in the variant with composting fecal biochar (KT). The concentrations in the leachate in the individual approaches can be classified in decreasing order as follows:

- Nitrate: Cp > (all other biochars in this project)
- Ammonium: Cp > VE > (all other biochars in this project)
- Phosphate: VE > Cp > PM > KT > Cn
- Potassium: VE = PM > KT > Cp = Cn

Leaching was highest in the positive control (Cp) fertilized with the Wuxal mineral fertilizer (nitrate, ammonium, phosphate) as well as the nutrient charged VE (phosphate). In terms of leaching potassium, the two fecal biochars (PM and KT) were almost on par with the loaded plant-based biochars. Although the addition of fertilizer to the positive control (Cp) was adjusted based on the nitrogen content of the initial KT biochar, the NO₃⁻ concentration in the leachate of Cp was over 20 × higher than in the leachate from the biochar in KT. This indicates that the nitrogen contained in the biochar is fixed and hardly washed out. Furthermore, the higher biochar dosage in the substrate experiment did not result in more nitrate in the leachate. Results from a previous leachate study (Bleuler, 2016) confirm this finding.

The sodium and chloride ions are leached significantly more, exhibiting a clear relationship to the content in the biochars. Thus, this should be adjusted for different plants and soil conditions. Common salt (NaCl) is excreted in urine, and the analysis of the biochars shows that sodium in the fecal biochar (KT) was about 4–5 times more concentrated than in the charged plant biochar (VE) and horse dung biochar (PM). This was also evident in the leachate samples. Substrates combined with little amounts of biochar can, however, provide a better nutrient holding capacity, as shown for phosphate and nitrate in this study. This is due to the absorption capacity of biochar, a fact that was also confirmed by the studies of Tauqeer et al. (2022) and Abel et al. (2013). Hypothesis 1 was thus confirmed for nitrate and phosphate, but not for the highly soluble potassium, chloride, and sodium ions (data not shown).

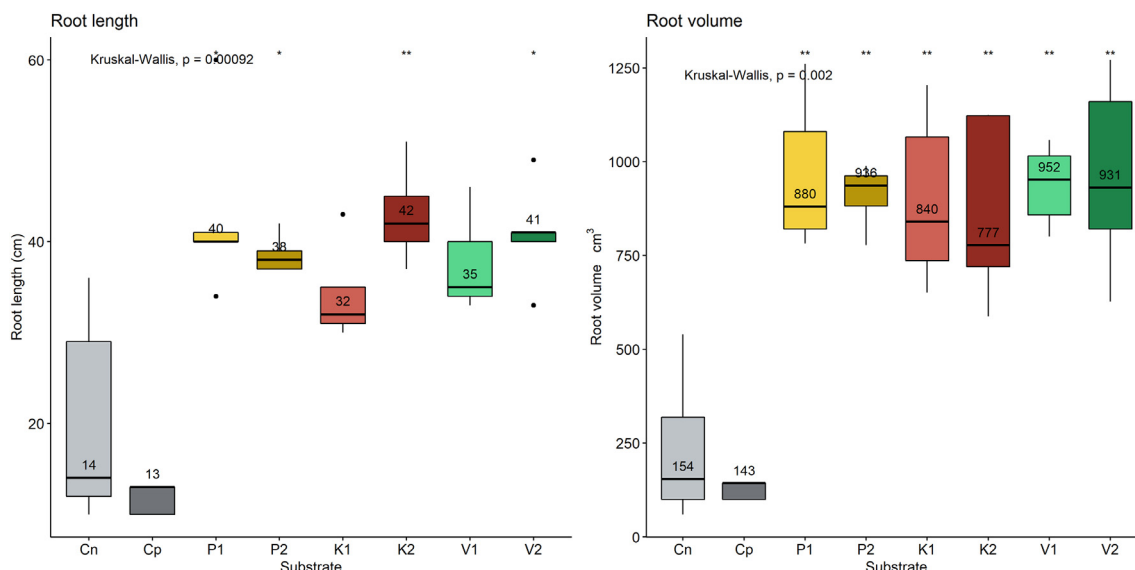


Fig. 15. Root length (cm) and root volume (cm³) by substrates over experimental period.

At the end of the trial (leaf sprouting had just occurred in April 2019), the birch tree seedlings in the K1 (5% human fecal matter biochar) and P1 (5% horse manure biochar) variants made the healthiest impression. Even though the substrates with fecal biochar did not grow significantly better than the seedlings in the other biochar-amended SST substrates, they did survive. Hypothesis 2 was therefore confirmed.

Birch seedlings in the SST substrates with fecal-based biochar (setups K1, K2, P1 and P2) did not grow better or worse than the V1 and V2 setups with plant-based biochar. Hypothesis 3 was therefore not confirmed in this study. Finally, all of the biochar-amended substrates had a positive effect on root length and -volume compared to the Cp and Cn controls, thus confirming hypothesis 4.

The slow growth of the birch tree seedlings cannot be explained by the presence of heavy metals. Cu, Pb, Zn, As, Cd, Cr and Ni were below the detection limit of 10 µg/l in the leachate (June 6, 2018) with only few exceptions. Arsenic (As) was detected just above the detection limit in the P2, V1 and V2 setups at the start of the experiment. However, the EBC quality standard was clearly met. A toxic effect caused by heavy metals can therefore be excluded.

SST substrates amended with biochar and organic material may enable multifunctional urban spaces to be made more tree friendly. This study supports the assumption that substrates containing biochar allow for a better root growth than the control substrates. A better capacity to develop roots in substrates leads to better tree health. Thus, by using biochar amended SST substrates, it seems possible to increase the ecosystem services of green spaces and to incorporate them into a strategy for the mitigation of the urban heat island effect. The substrates serve as a root space with nutrients, a foundational layer and water storage.

The use of fecal biochar in tree substrates may contribute to closing water and nutrient cycles in cities and put value on an otherwise unused resource. The use of fecal biochar in urban sponge city elements that incorporate a filter layer, and a revitalized topsoil layer could have a positive impact on flood control, groundwater protection and nature conservation, whilst also reducing wastewater treatment and disposal costs. Furthermore, the results demonstrate that the addition of biochar to an SST substrate can significantly improve conditions for tree seedlings in terms of tree survival. The main reason for this is likely to be its effect on increasing the substrate's water storage capacity and the provision of nutrients. Biochar can be used in addition to compost in SST substrates due to its structural stability and durability (Saluz, 2017).

The vitality of the birch seedlings in the setup with 5% fecal biochar suggests that the roots may also be able to tap the nutrients bound in the biochar structure. However, the extent to how much the biochar supports tree growth cannot be answered in this study. It shows, however, that both the nutrient and water storage properties of the 5 Vol% biochar may have an impact on the vitality of the trees.

The experiment also demonstrates the need for long-term studies to quantify the impact of biochar on aging in the soil and on plant productivity and vigor. The environmental risk of biochar can be minimized by controlling the source of the raw material and the temperature of the pyrolysis process, as shown by Lu et al. (2013). Further studies to confirm the nutrient and pollutant content results shown here may help to ensure that the increased use of fecal biochar in the future will not only reduce the volume of waste, but also provide a valuable product for closing material cycles.

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CRedit authorship contribution statement

Andrea Gion Saluz: Conceptualization, Methodology, Investigation, Writing – original draft. **Mira Bleuler:** Conceptualization, Methodology, Investigation. **Nikita Krähenbühl:** Data curation, Visualization. **Andreas Schönborn:** Conceptualization, Supervision, Data curation, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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