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**Effect of various amendments on the fractions of potentially toxic
elements in a contaminated environmental sample**

Bachelor Thesis

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I declare that the Bachelor Thesis Effect of various amendments on the fractions of potentially toxic elements in a contaminated environmental sample is my own work and all the sources I cited in it are listed in Bibliography.

Prague, Czech Republic

Signature

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Abstract

It was hypothesised that soil acidification significantly decreases the bonding of PTE in soil to the easily exchangeable fractions as well as non-labile fraction. Addition of clay to soil samples will result in greatly decreased plant available content of PTE.

Clay amendments, kaolinite and vermiculite, were added at a 10% application rate to contaminated garden soil to reduce plant available metal(loid)s as well as other criteria that will reduce the impact of PTE on the environment. The garden soil samples were taken from Kutná Hora and after determining the soil properties, the samples were split into two groups; normal soil and acidified soil. The original soil was acidified by the addition of HNO₃, to observe if pH would influence the adsorption of the metal(loid)s. The normal and acidified soils were prepared into 50 ml beakers in triplicates and a control, and were placed in room temperature with occasional watering. Samples were analysed three times over an eight-week period to evaluate the effect of the clays on arsenic, cadmium, lead and zinc bioavailability, pseudo total concentrations and different fractions of a BCR sequential extraction. After eight weeks, the pseudo total values remained either slightly over or under the original values, with no significant changes in absolute values. Soils amended with vermiculite, vermiculite in acidified soil and kaolinite in acidified soil had values increase in week four, only to decrease in week eight. Kaolinite in the original soil managed to decrease plant available zinc by 0.6% over eight weeks. Vermiculite, vermiculite in acidified soil and kaolinite in acidified soil decreased plant available zinc by 0.3-1.1% in week four, but concentrations increased by 0.2% in week eight. Vermiculite and kaolinite proved to reduce plant available metal(loid)s in the soil, but not to the extent as it was expected. The pH influence from the acidification of the soil was not as significant as it was expected for our experiment. The studied elements had lower concentrations in the acidified soil but conclusively values remained higher than the controls.

Key words: Clay amendment, soil acidification, contamination, PTE

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1. Introduction

Mining and smelting are well known causes of environmental contamination, releasing copious amounts of potentially toxic elements (PTE) into soils, water sources and the atmosphere.

Deposits of waste tailings and slag have substantially elevated levels of toxic elements in the soil, consequently affecting surrounding agricultural areas. Contaminated agricultural areas therefore creates a pathway in which PTE are exposed to humans.

This paper focuses on contaminated soil from Kutná Hora, a Czech city located in the Central Bohemian Region. During the 13th to 16th century the mining and smelting industry blossomed and transformed Kutná Hora into one of the largest mining towns in Europe; producing 5-6 tons of silver and 50-100 tons of copper annually (Pauliš and Mikuš, 1998).

During the 20th century the mining and smelting processes slowly came to a halt. Due to the heavy mining and smelting, the land surrounding the area, including agricultural and garden areas have become subjected to heavy metal contamination through processes such as leeching, and aeolian or colluvial deposition.

Amendments have become a popular, cost effective method for immobilising heavy metal(loid)s. There is a vast array of amendments that can be used for immobilising heavy metal(loid)s. However, some amendments work better with certain toxic elements compared to others (e.g. tests have proven arsenic work best with Fe based amendments (Warren et al., 2003; Warren and Alloway, 2003). This paper focuses on using clay as an amendment to immobilise PTE in a contaminated garden soil sample from Kutná Hora. Two clays, kaolinite and vermiculite, have been chosen for this experiment due to their different structures, bonding and cation exchange capacities. The paper focuses on the effect of kaolinite and vermiculite on four of the highly-concentrated PTE present in the soil: arsenic, cadmium, lead and zinc.

2. Aims

- To describe the fraction distribution of potentially toxic elements (PTE) according to BCR sequential extraction, in a contaminated soil sample before and after pH adjustment and amendment with different clays.
- To study the effects of clay amendments on metal availability; specifically, arsenic, cadmium, zinc and lead in contaminated soil.

3. Objectives

- To review the literature on the use of clay amendments to contaminated soils in order to choose the most appropriate clay types for study, with regard to decreasing availability of PTE in soil samples.
- To test the effects of clay amendments toward the fraction distribution of PTE in a contaminated solid sample by means of their application in a series of reaction vessels.
- To determine the effect of pH on the availability of PTE in the contaminated sample.

4. Literature review

There has been a growing interest in remediation and usage of amendments to decrease the availability of risk elements in the soil. Plenty of research in this field has generated a wide range of literature focused on remediation and stabilisation of contaminated soil and amendments (e.g. Alloway, 2013; Basta and McGowen, 2004; Arcon et al., 2005; Kumpiene et al., 2008). The methods used for amending contaminated soil depends on the elements present in the soil as well as the binding capacity the amendment has. The areas of study found in the literature greatly vary according to climate, soil and the type of mining; therefore, these factors must be taken into consideration when reviewing the methods presented in the literature.

4.1. Soil contamination

4.1.1. Sources

Heavy metal(loid)s derive mostly from lithogenic and/or anthropogenic sources. The lithogenic sources refer to the heavy metal(loid)s, which derived from the geological parent material of which the soil has developed from. The lithogenic sources are the dominating sources of concentrations of heavy metal(loid)s in world soils. While major elements such as oxygen, iron, aluminium, sodium and another six other elements make up 99% of the earth's crust, only 1% of the earth's crust accounts for all the risk elements (Alloway, 2013).

Anthropogenic sources are considered sources that contaminate or pollute the soil, including acid rain, particulates emitted into the atmosphere through combustion processes and mining deposition. There are multiple mechanisms by which contaminants have entered the soil from their original source (Alloway, 2013). Air currents cause the deposition of dust and is the perfect carrier of aerosol sized particles and gaseous forms of metal(loid)s. Runoff and flooding aid the movement of elements from its source to rivers, streams, gardens or agricultural fields. Disposal of contaminated soils into heaps by machinery on non-contaminated land can cause leaching of contaminants (Alloway, 2013).

Atmospheric deposition tends to be the most extensive form of contamination as transportation allows for long distance deposition. Most of the metal(loid)s emitted into the atmosphere come from coal, electricity generating stations, smelting and transport emissions (Alloway, 2013; Gul et al., 2015).

Flooding and sediment deposition play a key role in the transportation of contaminants. Metal(loid)s are carried in suspension in water sources, thus during flooding the suspended metal(loid)s get deposited on alluvial soils. The traces of elements that have been deposited onto the soil are fine particles called 'tailings' that are produced from mining and are often discharged into streams as waste bi-products (Alloway, 2013)

Localised contamination occurs through agriculture and horticulture when animals are given feed that has been influenced with risk elements, which in turn is released into the environment in the form of manure. Biosolids from waste water treatments are key sources of nitrogen, phosphorus and organic matter, giving them a significant fertiliser value (Alloway, 2013). The primary macronutrients in inorganic fertilisers are nitrogen, phosphorus and potassium, however, secondary macronutrients include calcium, magnesium and sulphur. Thus, inorganic fertilisers along with

essential risk elements that are applied to the soil, are prevalent sources of metal(loid)s in agricultural soils worldwide.

Non-ferrous metalliferous mining and smelting activities are directly related to metal(loid) contamination. Mining and smelting activities contribute to environmental contamination through their extraction and processing of gangue minerals and metal ores that naturally contain high concentrations of metal(loid)s. The activities that take place in close proximity to agricultural land are responsible for the excessive contamination of agricultural soil and the crops that are produced. The effect of human consumption of produce grown on contaminated land is very serious and hazardous (Zhuang et al., 2009).

Smelting involves converting sulphide ore minerals into oxides in a furnace and then separating them by the different molten metal(loid)s. In the past, inefficient processing and converting of the sulphide ore minerals were responsible for air, water and soil pollution, which are still present to this day.

4.1.2. Concentrations

Soils worldwide accommodate a majority, if not all, of the metal(loid)s present on the periodic table. However, their concentrations vary and certain metal(loid)s could be under the limit of detection. The concentrations of these elements present in the soil are categorised as total content and plant available. The availability of the element depends on multiple factors including pH, temperature, redox status and water content (Alloway, 2013). Total concentrations of elements in the soil can be in any form, ranging from ions bound in the crystal structure of minerals, those that are adsorbed to clays and oxides and free ions. The total concentration does not however predict the potentially plant-available fractions of heavy metal(loid)s.

The total concentrations can be determined through total digestion involving strong inorganic acids, followed by spectrometry measurement. In order to properly investigate an area for its concentration of metal(loid)s it is necessary to collect sufficient samples from apparently uncontaminated sites to provide a proper comparison (Alloway, 2013).

Table 1. Median and Maximum total metal(loid) concentrations in top-soils in various countries as well as world average values (mg/kg)

	Europe [a]	England and Wales [b]	Netherlands [c]	Denmark [d]	USA [e]	World [f]
No. of Samples	852	5,692	100	393	1,903	-
Arsenic	6.0 (<27.3)	-	5.6 (<39)	3.3 (<48)	-	4.7
Barium	65.0 (<1,700)	121 (<2,973)	32.4 (<620)	-	-	362
Cadmium	0.145 (<14)	0.7 (<41)	0.14 (<2.5)	0.16 (<0.85)	0.16 (<41)	1.1
Cobolt	7.0 (<255)	9.8 (<322)	2.4 (<22)	-	7 (<347)	6.9
Chromium	22.0 (<234)	39.3 (<838)	15.7 (<106)	9.9 (<64)	24 (<4,107)	42
Copper	12 (<239)	18.1 (<1,508)	10.2 (<141)	7 (<93)	17.3 (<1.836)	14
Manganese	382 (<6,480)	577 (<42,603)	-	-	372 (<8,071)	418
Nickel	14 (<2,560)	22.6 (<440)	4.94 (<66)	5 (<76)	18.3 (<4,497)	18
Lead	15 (<886)	40 (<16,338)	15.6 (<451)	11.3 (<102)	10.1 (<1,650)	25
Zinc	48 (<2,270)	82 (<3,648)	40.3 (<316)	26.8 (<135)	56.5 (<6,173)	62

Salminen et al. 2005 [a]; McGrath and Loveland (1992) [b]; Brus et al. (2009) [c]; Bak et al. (1997) [d]; Burt et al. (2003) [e]; Kabata-Pendias and Mukherjee (2007) [f]

4.1.3. Arsenic

Arsenic naturally occurs in several oxidation states between $-III$ and $+V$ and is known for its toxicity to the environment. Uncontaminated soils can contain concentrations of arsenic between 0.1 and 55 mg/kg, nevertheless soils contaminated by mining, smelter and other industrial activities give rise to much higher concentrations (Alloway, 2013).

In aerobic conditions arsenic is primarily associated with iron (oxy)hydroxides whereas arsenic sulphide minerals tend to precipitate in anaerobic conditions. Since arsenic has multiple oxidation states it forms bonds with sulphur and carbon more quickly and undergoes biological transformations which result in volatilisation from soils (O'Neill, 1990; Sadiq, 1997).

Despite its toxicity arsenic in low concentrations has been discovered to be essential in animals (Uthus, 1992) and beneficial in plants (Evans et al., 2005).

Arsenic originating from soil parent materials mainly come from weathered parent materials with shales, clay and phosphorites (phosphate) contain higher levels of arsenic compared to sandstone and limestone. Atmospheric deposition plays a role in arsenic contamination in vicinities that surround mining operations. Fifty to eighty percent of arsenic deposited into soils by atmospheric deposition is in soluble form (Haygarth and Jones, 1992). In agriculture, the application of fertilisers, lime, pesticides etc, raise the inputs of arsenic into the soils.

Arsenic in soils is commonly associated with its primary minerals derived from parent material, secondary minerals formed through mineral weathering and arsenic that has adsorbed to mineral surfaces (Alloway, 2013).

Arsenic sorption by different compounds has been shown to be dependent on the pH of the system. In a study of arsenate adsorption to 49 surface and subsurface soil horizons belonging to six different soil orders in the US Soil Taxonomy, arsenate adsorption tended to increase with increasing pH solution, showing maximum at around pH 6-7, and decreased past the maximum (Goldberg et al., 2005).

Soils polluted with arsenic are abundant world-wide, especially in areas that are within the vicinity of mining, smelter and metal processing activities. The total arsenic concentration at an industrial site in Belgium was found to be as high as 360,000 mg/kg (Cappuyins et al., 2002). Arsenic depositions that have accumulated on forest floors from the past continue to leach into mineral soils for prolonged periods after being released from their source (Erbanova et al., 2008). Highly polluted soils have been observed to act like net sinks of arsenic (Erbanova et al., 2008) except for

two sites in the Czech Republic where the retreat of soil acidification through atmospheric deposition has been identified as a possible explanation of enhanced leaching (Erbanova et al., 2008). Surface soil contamination is thought to be associated with enhanced uptake of arsenic by plants despite most plants being arsenic excluders (Brandstetter et al., 2000).

4.1.4. Cadmium

Cadmium minerals can be found as greenockite and otavite, but more commonly occurs in zinc minerals. Cadmium concentrations in rocks rise with increasing phosphorus, sulphur and carbon concentrations. Cadmium concentrations in non-contaminated soils generally increase with decreasing percentage of sand as cadmium is associated with finer particles (Baize, 1997).

Cadmium is a non-essential metal that naturally occurs in all soils as a divalent cation at concentrations varying between 0.1-1.0 mg/kg. The toxicity of cadmium in the soil is persistent since its residence time exceeds decades and because its bioavailability does not decrease in the long run (Smolders et al., 1999). Cadmium contamination of agricultural soils indirectly affects humans, through crops containing levels of cadmium that pose a risk to human health.

Soil cadmium is enriched by human activities via atmospheric deposition and through phosphorous fertilizers and sewage sludge applications. Cadmium mining, production and use all enhance cadmium concentrations in soils. Cadmium in soils is almost always present in the form Cd^{II} oxidation state, which has an electronic configuration of Pd^0 with filled d-shell. Cadmium participates in covalent binding with surfaces resulting in it being less soluble than harder cations. Cd^{II} form inner sphere complexes with high selectivity onto surfaces rather than binding with outer sphere cations. Chemical methods are used to infer cadmium binding mechanism, Christensen (1984) had suggested that sorption rather than precipitation controlled solution cadmium in the environmentally relevant concentration range in soil; precipitation is unlikely to control cadmium solubility unless at excessive cadmium contamination and at pH values >7.0 . Cadmium sorption is affected by pH values, with a factor of 3-5 stronger sorption per unit pH increase (Christensen, 1984), confirming that pH is generally the primary statistical factor associated with cadmium concentrations in solution, followed by soils organic matter content (Boekhold and Van der Zee, 1992). Soil organic matter, iron oxyhydroxides, aluminium and manganese and clay minerals are all main cadmium adsorbents in soil.

4.1.5. Lead

Lead is known to be the first metal extracted by man from its ores (mainly galena), being unessential and unbeneficial for living organisms, it is used only for industrial purposes. Lead mining was recognised as one of the main sources of soil contamination. In the United Kingdom, a mean soil level of 30,000 mg/kg lead was present 100m of an old smelter (Colbourn and Thornton, 1978). However, lead is extremely toxic to any living biota that is in its presence. In recent years, the concerns with the effects of lead to human health have caused a decrease in the application and use of the metal. Lead is known to accumulate in soils where it can remain bioavailable for prolonged periods of time. Another issue posed by lead is its leaching into drinking water sources from soil surfaces. Prapaipong et al. (2008) reported that lead polluted soils can exceed their lead carrying capacity which could have potential impacts on shallow groundwater systems and can pose toxic to humans and the ecosystem.

Lead is found in abundance in the continental crust at 14.8 mg/kg (Wedepohl, 1995). In silicate rocks lead tends to increase with increasing silicate content. Lead's affinity for sulphur leads it to accumulate in sulphur phases in rocks. Shales have higher lead abundance with 22 mg/kg compared to sandstones which have 10 mg/kg.

Lead is continuously emitted from elevated temperature processes such as lead ore smelting, coal burning and up until the 1980s, it was used in petrol. Due to the intensive use of lead in industrial activities in the past, lead is accumulated on terrestrial surfaces, especially on soil surfaces. Lead in soils exists predominantly in the +2-oxidation state, however in reduced soils it occurs mainly as insoluble lead(II) sulphide precipitated by sulphide. Under oxidising conditions lead is present as Pb^{2+} ion which becomes less soluble with increasing pH.

4.1.6. Zinc

Zinc is a naturally occurring element in the earth's crust as constituents of rocks and zinc-rich ores. Zinc ores were formed by natural geological processes and are located worldwide, making it the 24th most abundant element on earth. High concentrations of zinc in parent materials along with application of sewage sludge, mining and smelting have created an abundance of zinc in soils, beyond its micronutrient limit for biota. The prominent level of zinc can be very toxic to plants and soil organisms.

Zinc is present in the +2-oxidation state in soils and the concentration of zinc in solution can be affected by precipitation-dissolution, sorption and solution complexation reactions. When zinc is added to soils, an initial reaction takes place where zinc adsorbs onto the solid phases which is then followed by a process referred to as ageing which is the removal of the metal from the labile pool (Alloway, 2013).

4.1.7. Effect of contaminated soil on the environment and human health

Contaminated soil can adversely affect the environment and human health especially when exposed to lead, cadmium, mercury and arsenic (Järup, L. 2003). Spreading of contamination by leaching and wind allows for the contamination area to grow and even enter water ways.

Many plants find it hard to grow when metal(loid)s effect or change the chemistry of the soil, and requires time to adapt. Agriculture produce can be affected by contaminated soil which can lead to losses in produce during harvest and a great fall in the quality of produce. Agriculture grown on contaminated land has a possibility of being slightly contaminated, so when consumed by animals and humans, the metal(loid)s that were taken in by plants are now entering the bodies of the animals and humans.

Table 2. Effect of several risk elements on human health

Risk element	Effect on human health
Arsenic ¹	<ul style="list-style-type: none">• Inorganic arsenic poisoning through consumption of contaminated water/food• Damage to liver, skin and heart• Gastrointestinal and neurological damage
Cadmium ¹	<ul style="list-style-type: none">• Liver and kidney damage• Low bone density• Itai-itai disease
Lead ¹	<ul style="list-style-type: none">• Neurological damage, lowers IQ and attention span• Bone deterioration• Hypertension, hand-eye coordination impaired• Kidney disease
Zinc ²	<ul style="list-style-type: none">• Focal neuronal deficits• Elevated risk of prostate cancer• Nausea, epigastric pain
Mercury ¹	<ul style="list-style-type: none">• Damage to central nervous system and gastric system• Brain development affected- lower IQ• Co-ordination, sense of touch and eyesight affected• Liver, heart and kidney damage

¹ Science Communication Unit, University of the West of England, Bristol (2013).

² Plum, Rink, & Haase, 2010

4.2. Introduction to the Location

4.2.1. Map



Fig. 1. Location of Kutná Hora (www.mapy.cz)

4.2.2. Introduction to the area and smelting processes

Kutná Hora is one of Czech Republic's oldest and most significant mining regions located in the Central Bohemian region, approximately 85 kilometres from the Czech capital, Prague. A vast area of land, including agricultural in Kutná Hora contain soils that have been contaminated by PTE due to its long history of mining and smelting. In the era between the 13th and 16th century, Kutná Hora was categorised as one of the main centres of mining and smelting in central Europe with its two main products being silver and copper. During the 14th century, Kutná Hora became one of Europe's largest mining towns, producing 5-6 tons of silver and 50-100 tons of copper annually (Pauliš and Mikuš, 1998). The smelting processes involved four stages which consisted of the sorting of ore and treatment to get an initial sulphide concentrate, oxidation of sulphides through roasting, reduction to silver and silver separation and refining.

4.2.3. Site Characterisation

The geological bedrock of Kutná Hora is mainly formed of Mesozoic rocks such as sandstone, siltstone and claystone, that are covered by quaternary sediments. Kank mountain, slightly north

of the main town is formed by several types of gneiss and Mesozoic rocks. The Quaternary cover consists of sand, gravel and loess (Horák, J. and Hejzman, M., 2016).

Mining took place on quartz enriched with arsenic which was formed from metamorphosed igneous substances (Pauliš and Mikuš, 1998). Not only do the quartz veins contain arsenic, but also a wide range of metal(loid)s. The composition and spatial arrangement of the contaminants is not completely known despite the multiple articles published on Kutná Hora. However, the main elements present in Kutná Hora soils have been recorded as As, Be, Cd, Co, Cr, Cu, Hg, Pb, V and Zn, due to the high concentrations they were found in. Elements such as Fe, Mo, Ni and Sb were all also found abundantly, but at lower concentrations (Horák, J. and Hejzman, M., 2016).

4.3. Amendments

Amendments are used to immobilise concentrations of contaminants in soil through sorption and/or precipitation (Basta and McGowen, 2004). The increase in adsorption of metal(loid)s and decrease of solubility reduces the plant available portion, and decreases the possibility of pollutants spreading. Organic and inorganic matter in soils absorb the metal(loid)s present in the soil which results in the reduction of the mobility and availability of metal(loid)s. The absorbing capacity of soils to retain metal(loid)s follow certain properties such as pH, redox potential, water content and salinity. Other properties such as the soils residual properties that attribute to the source of contamination, intrinsic affinity of metal(loid) ions for soil adsorption surfaces, the types of soils and minerals present in the soil (e.g. humus and clay) and the period of contact between the soil and the metal(loid)s (Alloway, 2013).

It has been proven that the strength of metal(loid) binding is influenced by the humus content, thus an increase in humus content can lead to reduced metal bioavailability. Humus that forms through the decomposition of plants is normally situated in high concentration on the upper 20 cm of the soil profile. The distribution of oxygen-substituted acid groups determines the surface chemistry of colloidal and molecular humic and fulvic acids. Hu-COOH and Hu-OH groups tend to create stronger acidity due to their electron withdrawing effects which contrasts the weakening of acidity of humic and fulvic acids due to electrostatic restriction from charged groups (Alloway, 2013). This combination creates a linear increase in negative charge from pH 3 to pH 10, with stronger adsorption of metal(loid)s at a higher pH (Alloway, 2013). Metal(loid)s present in humus rich soils tend to bind with oxygen, nitrogen and sulphur atoms. The binding of metal(loid)s is suggested to

be dominated by the formation of two or three bonds to carboxyl and acidic hydroxyl groups (Alloway, 2013).

Soil consists of manganese oxides in multiple forms ranging from $Mn^{2+}O$ to $Mn^{4+}O_2$ as well as iron oxides due to weathering in soils (Alloway, 2013). Manganese and iron hydrous oxides tend to have high adsorption affinities for metal(loid)s which bound as inner-sphere-mono and bidentate surface complexes. Extended X-ray adsorption fine structure spectrometry and X-ray adsorption near edge technology was used by Arcon et al. (2005) to understand that arsenic adsorption in UK soils contaminated by mine spoils was present as arsenates which were bound to iron oxides and aluminium hydroxides. Extended X-ray adsorption fine structure spectrometry was also used by Fendorf et al. (1997), who investigated the bonding of arsenate and chromate to goethite, and concluded that three types of binding mechanisms were present: monodentate, bidentate-mononuclear and bidentate-binuclear (see fig. 2).

The likeliness of a metal(loid) to be adsorbed by specific oxides is determined by the speciation of the metal(loid) ion in solution and by the oxide charge relations, morphology and pore sizes within the oxide surface (Gerth, J., 2005). When testing the effect of pH on metal adsorption, it was seen that increase in pH leads to increase of metal(loid) adsorption due to reduced competition with H^+ ions and increased negative surfaces.

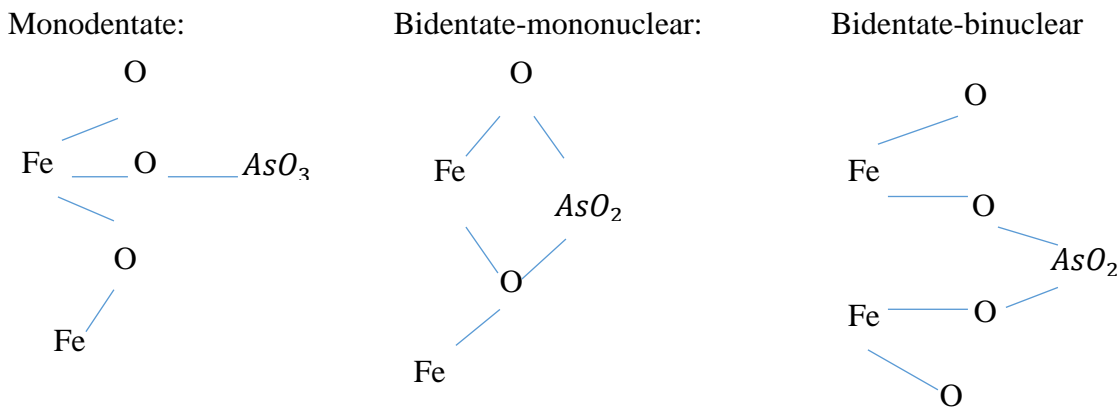


Fig. 2. Mondentate, bidentate-mononuclear and bidentate-binuclear binding mechanisms

Strong associations have been found between manganese and iron oxides and cobalt, nickel, arsenic, lead, chromium and zinc (Arcon et al., 2005; Kumpiene et al. 2008; McKenzie, R. M., 1977). Reduction- oxidation coupling is thought to occur during certain sorption reactions. Co^{2+} is

found to oxidise to Co^{3+} on Mn^{4+}O_2 surfaces as well as that manganese oxides oxidise Cr^{3+} to toxic Cr^{6+} (Kumpiene et al., 2008). It has been noted that it may be possible for As^{3+} to oxidise to As^{5+} on oxide surfaces which would contribute to adsorption and reduce potential toxicity (Alloway, 2013).

Clay is found naturally in many soils and has been tested multiple times as an amendment due to its structure and CEC capacity. It has been suggested that clay minerals may be involved in arsenic sorption in soils.

4.3.1. Arsenic amendments

The content and nature of iron oxy/hydroxides has been proven to be the main controller of arsenic (arsenate and arsenite) sorption. Indian soils containing higher amounts of sesquioxides showed arsenate more efficiently adsorbed; addition of Fe oxy/hydroxides to the Indian soils proved to enhance arsenate sorption (Misra and Tiwari, 1963).

Autoradiography using arsenate-73, electron microscopy and microprobe analysis were used to study arsenate adsorption in various soil components in acidic Australian soils, with results indicating that iron oxy/hydroxides are more effective than other soil components in taking up arsenate on a unit mass base (Fordham and Norrish, 1979). Speciation of arsenate and arsenite on iron oxy/hydroxide surfaces include inner-sphere, bidentate (mono- and bi- nuclear) and monodentate surface complexes, with higher share of monodentate complexes at low arsenic loads and increasing pH and predominance of binuclear bidentate complexes at higher arsenic loads (Fendorf et al., 1997; Lumsdon et al., 1984; Manning and Goldberg, 1996). Arsenate adsorption involves an initial ligand exchange reaction of the aqueous arsenate species with OH ligands at the oxy/hydroxide surface which forms an inner-sphere monodentate surface complex, followed by a second ligand exchange reaction, resulting in the formation of an inner-sphere bidentate surface complex (Grossl et al., 1997), which is then followed by diffusion of arsenic to interior surface sites (Fuller et al., 1993).

4.3.2. Cadmium amendments

Protons are the main competing cations for Cd^{2+} sorption on organic matter or oxyhydroxides because Cd^{2+} binds to the surface oxygen atoms of carboxylic or phenolic groups of humic substances or to surface hydroxyl groups on oxyhydroxides.

Speciation modelling predicted that most of the cadmium is bound to organic matter in soils with pH <6.5, whereas Fe-oxides become most important adsorptive constituent at pH > 6.5 (Buekers et al., 2008). Cadmium can form complexes in solution with soluble ligands such as inorganic ligands or dissolved organic matter, which reduce charges and mobilise cadmium in soil. Clay minerals are unlikely to be important adsorbents for cadmium as the selectivity for sorption on clay minerals is typically less than five at relevant conditions (Farrah et al., 1980).

4.3.3. Lead amendments

Lead in soils highly depends on the organic matter content as lead is strongly adsorbed on humic matter at pH 4 or more (Bunzl et al., 1976; Kerndoff and Schnitzer, 1980). In the absence of humic matter, lead is found to be strongly adsorbed on clay minerals (Hildebrand and Blum, 1974) and iron oxides (Kinniburgh et al., 1976). However, phosphorous compounds have been proven to be the most effective amendment to immobilise lead (Shi and Erickson, 2001; Geebelen et al., 2002). Hooda and Alloway (1998) found that lead sorption in agricultural soils correlated with pH, CEC, organic matter content, clay content and calcium carbonate. However, organic matter in soils are reported to absorb more lead than clay minerals (Sipos et al., 2005) nevertheless, phosphorous compounds are the most effective amendments for immobilising lead. X-ray adsorption studies on interactions of Pb^{2+} with ferrihydrite (Trivedi et al., 2003) confirmed that lead sorption on oxisols was greater than the amount of negative surface charge, thus indicating the participation of lead in inner-sphere surface reactions (Appel and Ma, 2002). At pH>5 bidentate complexes on the oxide surfaces dominated, whereas at pH 4.5 a mixture on monodentate and bidentate complexes were present.

4.3.4. Zinc amendments

Zinc can be adsorbed non-specifically by ion exchange on clay mineral surfaces or on ionised groups of organic matters. The reactions are weakly selective, reversible and weakly pH dependent (Alloway, 2013). The ageing processes that take place are thought to be due to an intraparticle diffusion of divalent metals in amorphous oxyhydroxides, which depend on electrostatic forces of attraction. Zinc fixation increases with pH due to the increasing number of reactive sites on the oxides (Buekers et al., 2007), reducing the zinc bioavailability. Acid soils contaminated with zinc would require liming to increase the soil pH to reduce metal solubility. Addition of bauxite residues

or beringite have a high metal fixing capacity and can effectively fix metals in the long term, even if the soil acidifies (Lombi et al., 2003).

4.4. Clay and Clay Minerals

4.4.1. Properties

Clays are composed of layers of silica tetrahedral aluminium octahedra in ratios of 1:1 with a thickness of 0.7 nm and 2:1 with a thickness of 1 nm. The most common clay minerals found in worldwide soils are illite, smectite, vermiculite, kaolinite and chlorite. They are associated with a weathering sequence characterised by decreasing isomorphous substitution and variable accessibility of the interlayer space (Alloway, 2013). Illite is a by-product of mica; vermiculite is further weathering of illite or mica. Alteration or re-crystallisation from solution produce smectite. Deposition of non-exchangeable aluminium-hydroxy polymers forms chlorite. The most common 1:1 clay is kaolinite, is formed because of geological inheritance or severe weathering (Alloway, 2013). Other properties include the clay's plasticity and hardening abilities, the diverse types of surfaces, anisotropy of the layers/particles and adsorption and ion exchange abilities.

Table 3. Comparison of clay properties (White, R, E., 1987)

	Kaolinite	Vermiculite	Illite	Smectite
Tetrahedral	0	10% Al(3+)	20% Al(3+)	2.5% Al(3+)
Octahedral	0	15% Mg(2+)	0	15% Mg(2+)
Shrink-swell	Low	Medium-High	None	High
Interlayer	H-bonds	Exchangeable cations	Fixed K(+)	Exchangeable cations
Origin	Recrystallization under intense acid weathering	Intermediate alteration of micas	Early alteration of micas	Recrystallization under alkaline weathering

4.4.2. Structure

Clay minerals can be characterised by the SiO₄ tetrahedral and AlO₄ octahedra that form sheets with the chemical composition of (AlSi)₃O₄ (Yaya et al., 2012). Each tetrahedron shares three of

its vertex oxygens with another tetrahedral to form a 2-dimensional hexagonal array. Tetrahedral sheets in clays are bonded to octahedral sheets formed from small cations (eg. aluminium or magnesium) and are coordinated by six oxygen atoms. The remaining vertex forms a part of the octahedral sheet, with an additional oxygen atom that is bonded to a hydrogen atom located above the gap in the tetrahedral sheet at the centre of six tetrahedrals, to form an OH group in the clay structure (Schaetzl and Anderson, 2005).

The way the tetrahedral and octahedral sheets are composed into layers provides the basis of clay categorisation. The 1:1 clay is formed when there is only one tetrahedral and one octahedral group in each layer; likewise, the 2:1 clay is formed when there are two tetrahedral sheets with an unshared vertex of each sheet pointing towards each other and forming each side of the octahedral sheet (Schaetzl and Anderson, 2005). Tetrahedral sheets become twisted when bonding occurs between tetrahedral and octahedral sheets which cause a ditrigonal distortion to the hexagonal array and makes the octahedral sheet flat, minimising the bond-valence distortions of the crystalline (Yaya et al., 2012). The composition of the tetrahedral and octahedral sheets determines the charge of the layer (the layer tends to have no charge or a net negative charge). Interlayer cations (Na^+ and K^+) help balance any charge in the interlayer. The crystal structure is formed from the stacks of layers' interspaces with the interlayers.

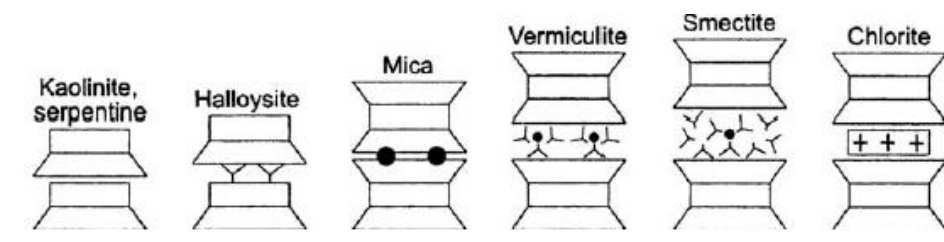


Fig. 3. Clay structure (Schaetzl and Anderson, 2005)

Tetrahedral sheets follow the formula $\text{Si}_4\text{O}_{10}^{4-}$, consisting of only negative charges and exists only with cations and additional oxygens.

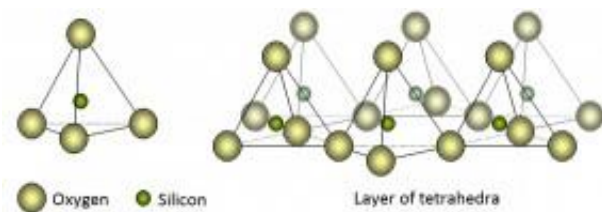


Fig. 4. Tetrahedral sheet structure (Jordán, A., 2014)

Octahedral follow one of the two formulas: brucite $Mg_3(OH)_6$ where all the octahedrons are occupied and forms a trioctahedral and gibbsite $Al_2(OH)_6$ where two thirds of the octahedrons are occupied and forms a dioctahedral.

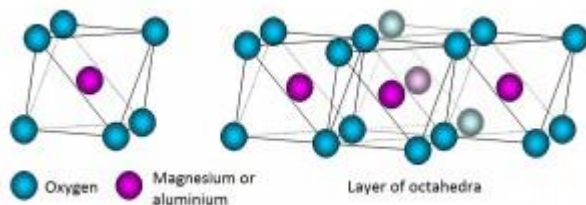


Fig. 5. Octahedral sheet structure (Jordán, A., 2014)

4.4.3. Surface Chemistry

The surface area of phyllosilicates such as smectites can reach almost up to $600m^2/g$. In the tetrahedral or octahedral layers the substitution of Al^{3+} or Si^{4+} forms a positive charge deficit which then gets balanced out by cations in the inter-layer space of clay minerals (Alloway, 2013). The negative charge of the layers along with the accessibility of the inter-layer sets the ground for the cation exchange capacity (CEC) of the clay. Illite is known to have the largest surface charge but limited access to its inter-layer space as it is usually filled with dehydrated potassium ions that are balancing the isomorphous substitution. Vermiculite on the other hand, is completely opposite, with less available isomorphous substitution and the largest CEC. Kaolinite, being a pure aluminosilicate, has very little isomorphous substitution and limited CEC (Alloway, 2013).

Table 4. Cation exchange capacity values for kaolinite and vermiculite (Alloway, 2013).

Vermiculite	Kaolinite
100-160 $cmol_c kg^{-1}$	3-15 $cmol_c kg^{-1}$

4.4.4. Bonding

In the inter-layer space of phyllosilicates, cationic metal(loid)s are held as exchangeable cations, but are poorly represented due to the high concentrations of calcium, magnesium and potassium in soils. EXAFS (extended X-ray adsorption fine structure (spectrometry)) and XANES (X-ray adsorption near edge structure) were used by Furnare et al. (2005) to study Cu^{2+} adsorption on vermiculite and smectite, which suggested that copper was covantly bound through inner-sphere complexes to unbounded oxygens at edges of clay minerals.

Highly specific binding can also occur within the inter-layer with nickel, cobalt, zinc to be thought to form multi-nuclear species which bind irreversibly. It was found that zinc and copper were specifically bound to AlHO in the inter-layer space of clays. Cadmium and lead on the other hand were found to be held exchangeably, through outer-sphere linkages and could be displaced by other cations. The relationship between adsorption and pH is similar to that of oxides; there was found to be a steep rise in affinity over a single pH unit.

4.5. Clay Amendments

Kaolinite and Vermiculite have been chosen for this experiment due to their different structures and bonding abilities. Kaolinite is found abundantly in soils worldwide and is one of the most common 1:1 layered minerals, which is formed through advanced weathering processes. One layer consists of an alumina octahedral sheet and a silica tetrahedral sheet that share a common plane of oxygen atoms, with layers of the mineral being bonded by hydrogen (Bear, 1965). Due to its structure, the silica/oxygen and alumina and hydroxyl sheets in kaolinites are left exposed and can interact with different components in the soil (Grim, 1968). Kaolinite particles cannot easily be broken down; thus, the majority of sorption activity occurs on the edges and surfaces of the structure. Kaolinite has the ability to create barriers that cannot be broken down easily, making sediments that have an abundance of kaolinite and minerals effective in controlling any migration of dissolved elements (Devidal et al, 1996). Kaolinite is a non-expanding clay making isomorphous substitution very limited or impossible (Mitchell, 1993).

Vermiculite is a 2:1 hydrated magnesium aluminium silicate clay that is found in various parts of the world. It is formed through natural weathering and percolating ground waters. The 2:1 structure of vermiculite is composed of two flat layers of silica and aluminium tetrahedra which is joined together with a layer of apical oxygen atoms along with magnesium, iron and hydroxyl molecules, creating the octahedral layer (White, R, E., 1987). The weak bonding of vermiculite allows it to expand and shrink upon wetting and drying.

5. Methodology

5.1. Study area



Fig. 6. Location of soil samples: Ulice Fučíkova, Kutná Hora

The samples were collected from a private garden on the street Fučíkova in Kutná Hora. The samples were suspected to be contaminated due to the vast history of mining and smelting processed that occurred in Kutná Hora during the 13th to 16th century.

5.2. Determination of soil properties

To prepare the soils for the experiment soil samples were sieved into 2mm particles and homogenised into one sample of soil. To determine the soil properties, experiments such as pH and CO_X were performed on the homogenised soil.

5.2.1. Soil pH

Soil $\text{pH}_{\text{H}_2\text{O}}$ was determined by weighing 10g of soil and shaking it for 30 minutes with 20 ml of deionised water and measuring it with a pH meter. Potential soil pH was determined by weighing 5g of soil and shaking it for 30 minutes with 20 ml of KCL solution and measuring it with a pH meter.

5.2.2. Oxidisable carbon

Tyurin's method was used to determine the organic carbon in the soil by oxidation with $\text{K}_2\text{Cr}_2\text{O}_7$ in a strong acid medium until the formation of CO_2 . The sample was sieved through a 0.25 mm sieve and 0.3g was weighed into a 150ml beaker. 10ml of $\text{K}_2\text{Cr}_2\text{O}_7$ into the beaker, stirred, covered

with a watch glass and placed into an oven at 125°C for 30 minutes. Titration with $(\text{NH}_4)_2\text{Fe}(\text{SO}_4)_2$ was conducted to calculate Factor of Mohr salt, oxidisable carbon and humus content.

5.2.3. Lowering soil pH

Half of the homogenised soil (750g) was acidified to reduce the pH of the soil. The soil was mixed with 1.2 litres of water and treated with HNO_3 over a period of 3 days till the pH reached a value below pH 6. In total an amount of 60 ml of HNO_3 was added to soil and the end pH value remained at 5.93.

5.2.4. Pseudo total concentration of PTE

The pseudo total content of PTE in the original soil as well as the now acidified soil was established after digestion by *aqua regia*, following the method used by Sabienė et al. (2004). Pseudo total digestion required the weighing of 1g of dry sample to be weighed into Teflon bottles and adding 7ml of 35% HCL and 3ml of 65% HNO_3 . The bottles were closed slightly with Teflon caps and left overnight to react. In the morning, the samples were heated on a heating plate at 120°C for 2 hours. The contents in the Teflon bottles were then washed into 50ml volumetric flasks and filled to the 50ml mark with deionised water and then filtered through filter paper into clean PVC tubes. The samples were diluted 10 times and filtered through a nylon filter for analysis.

5.2.5. Plant available PTE

The plant available fraction of PTE of both the original and acidified soil was determined through NH_4NO_3 extractions as done by Sabienė et al. (2004). The experiment required 20g of soil to be weighed and shaken for 2 hours with 50ml of 1M NH_4NO_3 solution. The solution was filtered through filter paper, diluted 10 times and filtered through a nylon filter for analysis.

5.2.6. BCR sequential extraction

A modified four step BCR sequential extraction procedure described by Tokalioglu et al. (2003) was used to determine the reducible, exchangeable, oxidisable and reducible fractions of the PTE in the soil of the original and acidified soil.

5.2.7. Experimental Plan & Methodology

The experiment was set for a duration of eight weeks, with samples being analysed after two, four and eight weeks. Each analysis consisted of fourteen beakers with seven samples of normal soil and seven samples of acidified soil; each soil type had a control and 3 replicates per clay amendment. Altogether the experiment consisted of forty-two reaction vessels that had 20g of soil weighed into them. The samples were covered with a watch glass and kept in room temperature. The clays used were kaolinite and vermiculite which were applied at a 10% application rate per 10 grams. The 10% application rate was deduced from an experiment conducted by Yin & Zhu (2015) who applied two amendments at application rates of 0%, 2.5%, 5% and 10%. Since twenty grams of soil was used per reaction vessel, with a 10% application rate of clay, two grams of each clay was added to the reaction vessels. The kaolinite used in the experiment was in powder form, while vermiculite was left in small ‘chunks’.

Water regime: twenty millilitres of water was added to the soil samples every two weeks and slightly mixed with a glass rod. The amount of water was determined by conducting a small experiment that involved applying different volumes of water to ten grams of the soil. The one to one ratio of water to soil proved to be the best water application rate.

A mini experiment was then later conducted to determine if the form of vermiculite used affected the results. To provide a more accurate analysis, the vermiculite was grounded into a powder form, to fit the same consistency of the kaolinite used. The experiment maintained the same structure as the main experiment.



Fig. 7. Kaolinite, vermiculite (chunks) and vermiculite (crushed/powdered)

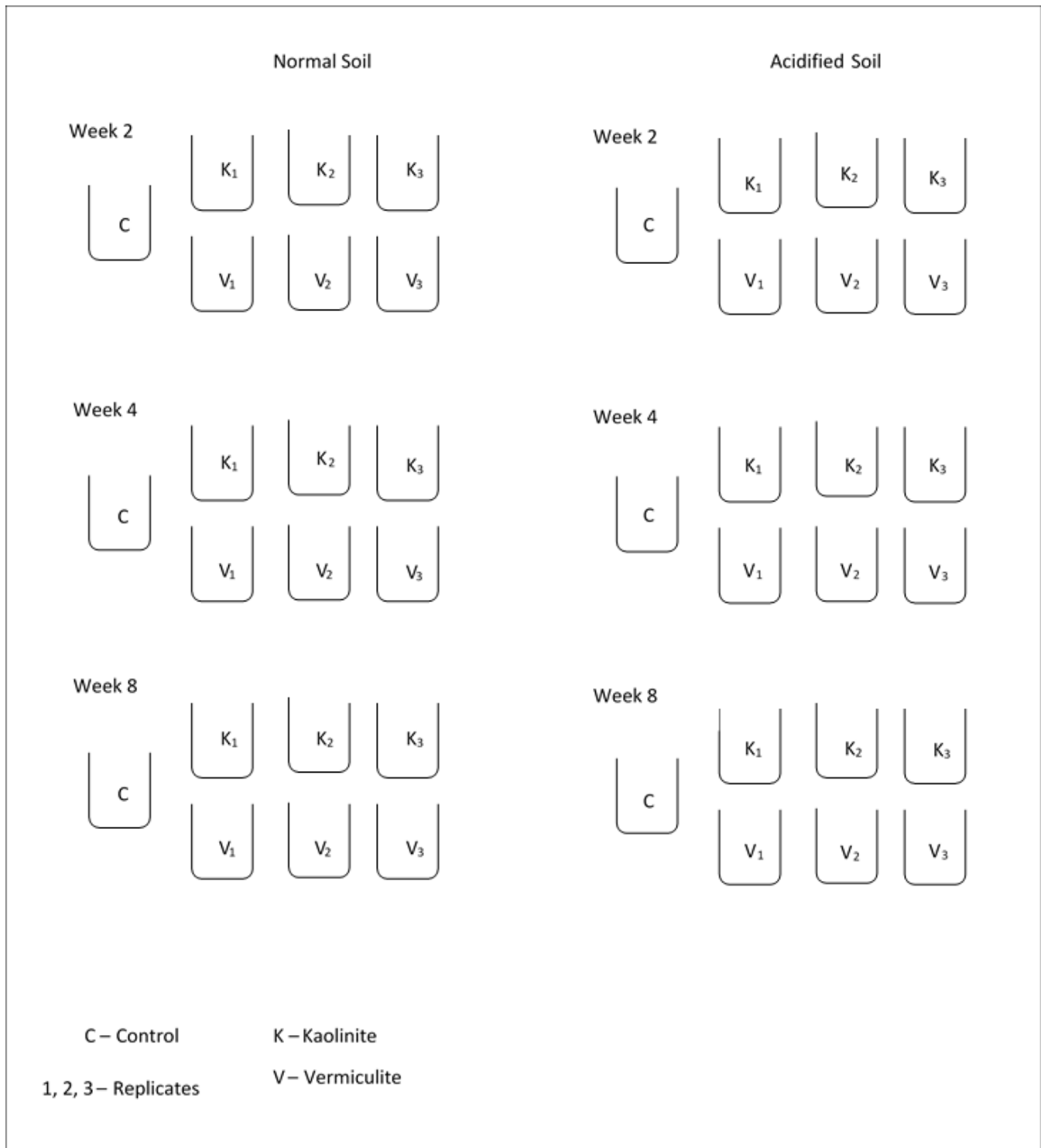


Fig. 8. Experimental plan set for the experiment

6. Results

6.1. Soil and clay characterisation

Table 5. pH values of the soil before experimentation

pH (H ₂ O)	pH (KCL)
7.48	7.30
7.52	7.35
7.58	7.37
7.52	7.34

Table 5 indicate that the soil in its original state before experimentation remained close to a neutral value. The oxidisable carbon as well as the humus were investigated and the values of the oxidisable carbon was 3.6% while the humus content came to 6.21%.

Table 6. Percentage of the four metal(loid)s present in the clay measured using XRF (X-Ray Fluorescence) Spectroscopy (Delta Professional, Olympus).

	Arsenic	Cadmium	Zinc	Lead
Kaolinite	11 * 10 ⁻⁴	15 * 10 ⁻⁴	9 * 10 ⁻⁴	60 * 10 ⁻⁴
Vermiculite	3.7 * 10 ⁻⁴	ND	44 * 10 ⁻⁴	8 * 10 ⁻⁴

Table 6 presents the percentages of arsenic, cadmium, zinc and lead that was present in the clay before it was added into the soils. The values are relatively minimal, however could affect any changes in PTE values.

6.2. Pseudo total concentrations

Table 7. Pseudo total maximum/preventive values for metal(loid)s in agricultural soils set by the Ministry of Environment in the Czech Republic (Vyhláška č. 13/1994 Sb.) (Vyhláška č. 153/2016 Sb.) (mg/kg)

	1993	2016
Arsenic	30	20
Cadmium	1	0.5
Lead	140	60
Zinc	200	120

Table 8. Pseudo total concentrations of elements before the addition of the clay amendments (mg/kg)

	Arsenic	Cadmium	Lead	Zinc
Normal Soil	81.3	2.19	159	454
Acidified Soil	79.8	2.17	138	466

Table 9. Pseudo total concentrations of PTE in the soil after two weeks in contact with clay (mg/kg)

	Arsenic	Cadmium	Lead	Zinc
Control	86.3	2.63	149	508
Control (acidified soil)	81.9	2.36	158	456
Kaolinite	76.1	3.17	195	439
Kaolinite (acidified soil)	75.8	2.14	125	1973
Vermiculite	65.1	1.94	117	386
Vermiculite (acidified soil)	73.1	2.08	124	410

Table 10. Pseudo total concentrations of PTE in the soil after eight weeks in contact with clay (mg/kg)

	Arsenic	Cadmium	Lead	Zinc
Control	85.7	2.54	157	540
Control (acidified soil)	80.3	2.32	137	474
Kaolinite	84.2	2.38	139	453
Kaolinite (acidified soil)	75.2	2.17	140	438
Vermiculite	84.9	2.57	156	512
Vermiculite (acidified soil)	75.3	2.37	130	441

Table 9 presents the controls and the effect of the clays after two weeks. It is quite evident that overall both kaolinite and vermiculite proved to reduce the pseudo total concentrations of the metal(loid)s, with an exception to lead and cadmium amended with kaolinite in the original soil. Vermiculite, under both normal soil conditions as well as acidified soil conditions seems to have reduced the pseudo total concentrations of all the four metal(loid)s. Lowering the pH enabled kaolinite to adsorb more of the metal(loid)s than kaolinite placed in the original soil conditions. Vermiculite on the other hand, adsorbs more metal(loid)s in the original soil conditions after two weeks but then adsorbs more metal(loid)s in the acidified soil after eight weeks.

The controls for zinc from table 9 and 10 are higher than the control before the experiment displayed in table 8. The result for zinc after two weeks (table 9) amended with kaolinite in acidified soil is also significantly higher than control (acidified soil) and results for zinc amended with kaolinite (acidified soil) after eight weeks. These higher values indicate heterogenic zinc distribution in the soil. Table 10 presents identical data as that of table 9, as the pseudo total concentrations of metal(loid)s decreased below the control values. However, after eight weeks, majority of the values are greater than the results after two weeks. The only values that reduced from week two to week eight were the values for cadmium and zinc when amended with kaolinite. The values prove that vermiculite and kaolinite both can reduce the pseudo total concentrations of the four metal(loid)s and that lowering soil pH increased metal adsorption with kaolinite.

6.3. Plant available PTE concentrations

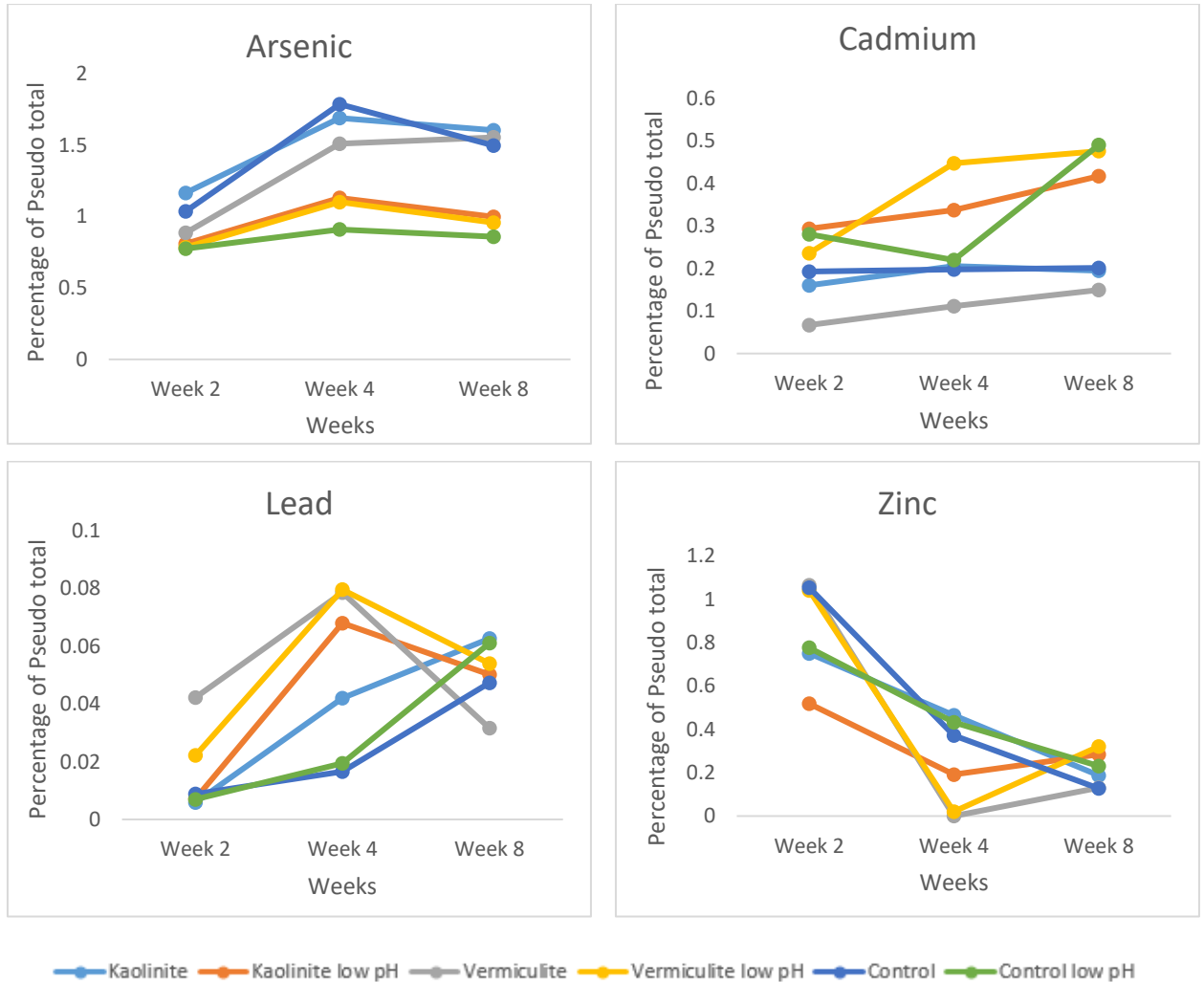


Fig. 9. Changes in plant available concentrations over eight weeks

The results shown in fig. 9 shows the plant extractable percentage of pseudo total arsenic in the original soil increased over time compared to the arsenic in the acidified soil, which remained constant. The values for kaolinite and vermiculite in acidified soil were similar to the control low pH at week two but increased over the period of eight weeks. In week four kaolinite and vermiculite dropped lower than the control, only to increase by week eight. The graph for cadmium indicates straightforward values for the original soil when compared to values obtained for the acidified soil. The control and kaolinite values are quite identical, with slight change occurring over time. Vermiculite managed to remain under the control values throughout the eight weeks, which could

indicate that vermiculite is a suitable amendment for cadmium. However, vermiculite in the acidified soil started off under the control low pH value at week two, then increased by approximately 0.2% and then maintained a similar value to the control low pH. Kaolinite and vermiculite in both soil conditions appear to slightly increase over the eight-week time frame. In graph for lead it is evident that the values for vermiculite, vermiculite in acidified soil and kaolinite in acidified soil spiked from week two to week four, remaining over both the controls, before decreasing in week eight. Kaolinite differed from the other three samples, as instead of any spikes, there was a gradual increase from week two to week eight. It is clear from the graph that neither of the clays or soil conditions managed to effectively reduce the plant available portion of lead. The graph for zinc shows that both clays under both conditions significantly reduced from week two to week four, only to increase by week eight. However, the increase in week eight remained lower than the original values of plant available zinc, indicating that the clays were effective at reducing plant available zinc in the soil tested. Nevertheless, despite the graphs depicting drastic changes over time, the absolute values are quite small, indicating that there are no drastic changes in concentrations of the metal(loid)s.

6.4. BCR Sequential extraction



Fig. 10. Evaluation of the fraction distribution of the soil sample amended with kaolinite

In fig. 10 the graphs for arsenic, cadmium and zinc shows that kaolinite had little to no effect in reducing the reducible and oxidisable fraction in the soil over the eight-week period but managed to reduce the reducible fraction of lead. Kaolinite did manage to reduce the exchangeable fraction for all the metals except for arsenic. Lead seemed to have almost no exchangeable fraction to begin with.

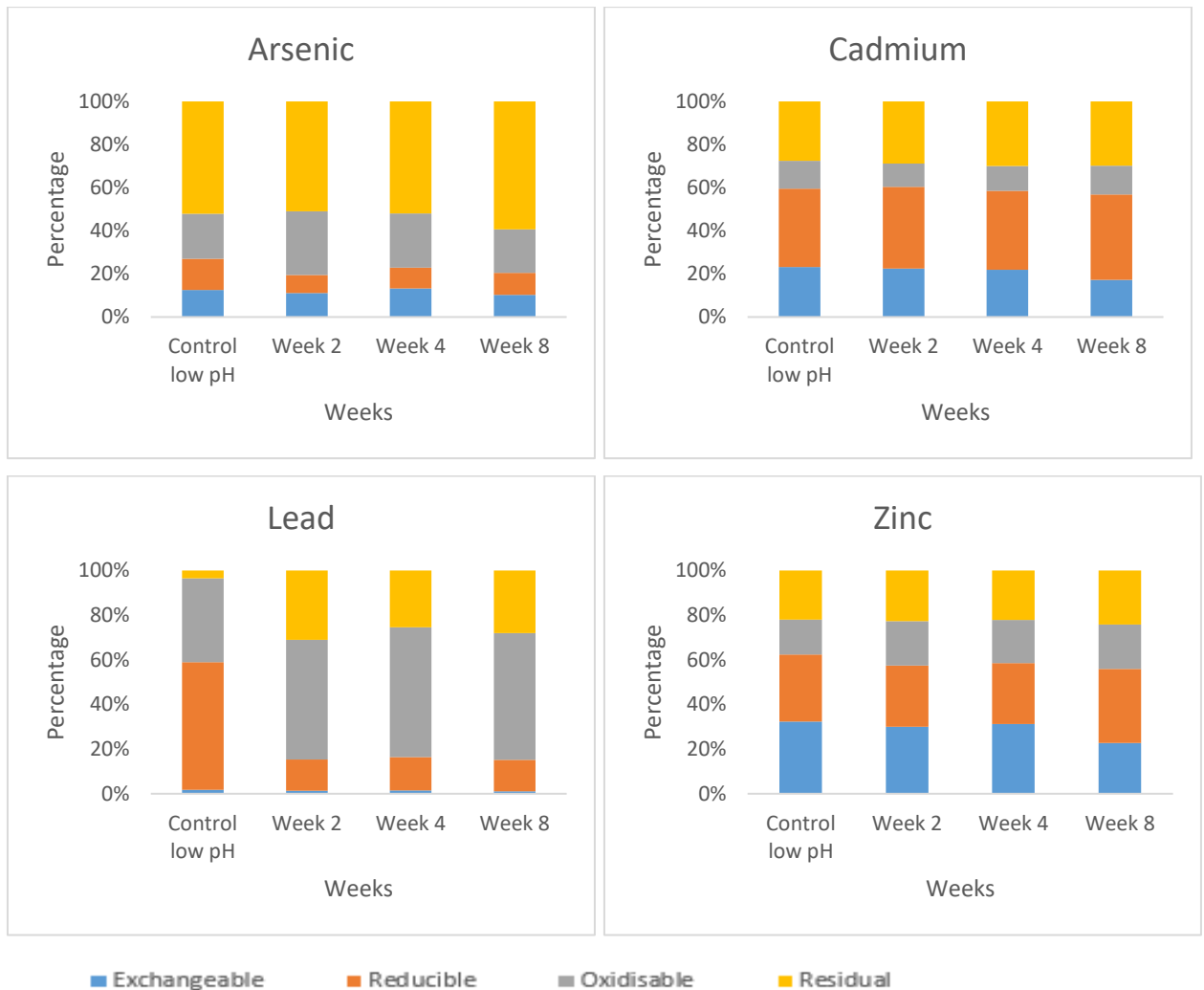


Fig.11. Evaluation of the fraction distribution of the acidified soil sample amended with kaolinite

The residual and oxidisable fraction for arsenic, cadmium and zinc show minor changes over time. The residual fraction of lead increases and decreases over the eight weeks, but the value after eight weeks remains greater than the control at the start. In the graph displaying the values for lead it is also possible to notice a significant difference in the reducible fraction between the control and weeks two, four and eight; dropping from 60% to under 20% after two weeks, remaining constant over the next six weeks. All metals show a slight decrease in the exchangeable fraction when week eight is compared to the control. Lead once again shows very low amounts of exchangeable fraction, similar to lead in the original soil amended with kaolinite shown in figure 11.



Fig.12. Evaluation of the fraction distribution of the soil sample amended with vermiculite.

Graphs for arsenic, cadmium and zinc suggests that no significant changes occurred in the residual and exchangeable fractions. The reducible fractions of all four metals indicate a slight reduction in the reducible fraction. The graph for lead suggest no change occurred in the oxidisable fraction and increases in the residual fraction of lead.

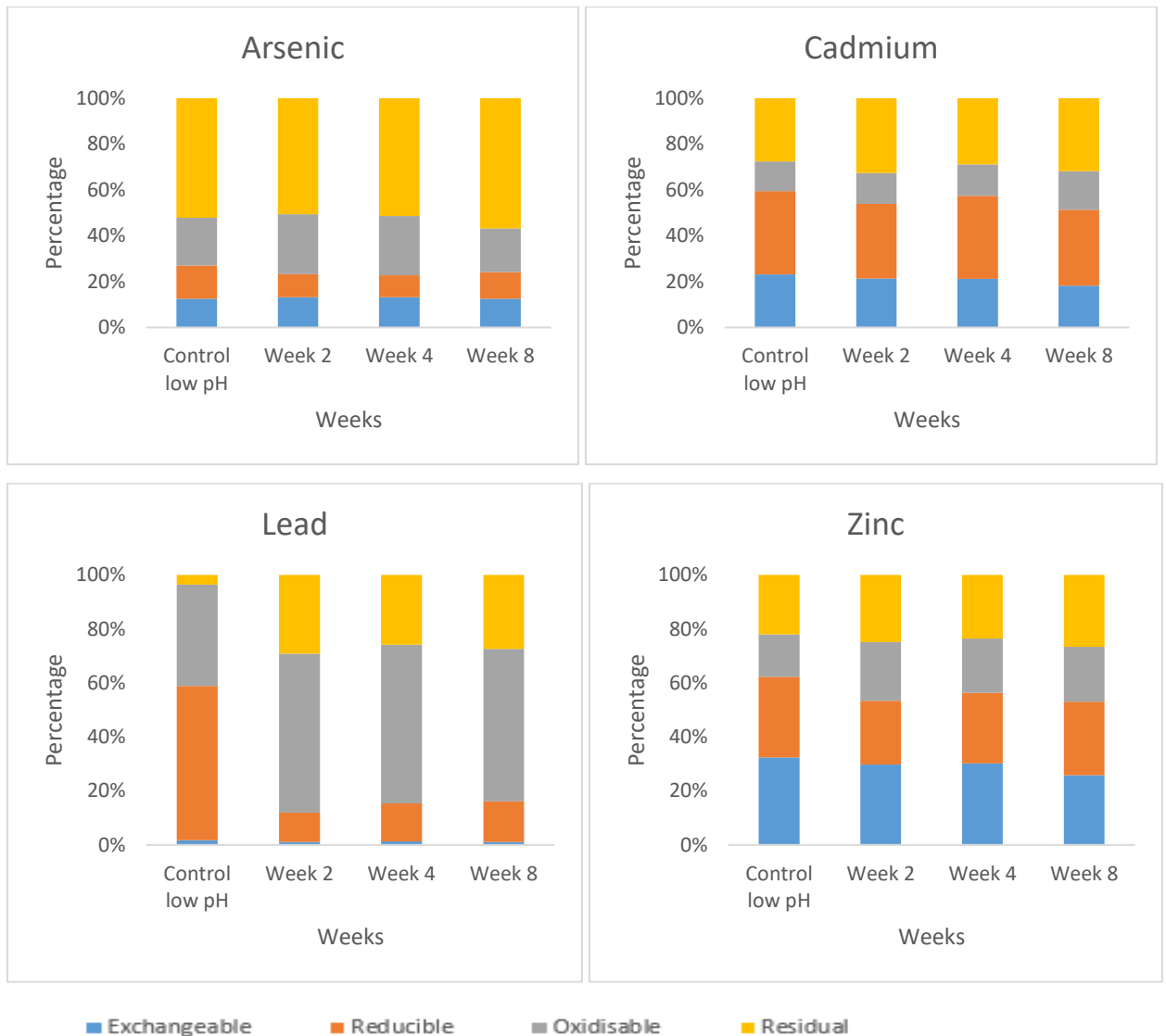


Fig.13. Evaluation of the fraction distribution of the acidified soil sample amended with vermiculite.

The residual fraction remained almost constant for arsenic, cadmium and zinc, while lead shows an increase from the control to week eight. Graphs for cadmium, lead and zinc show an increase in the oxidisable fraction compared to arsenic which shows a decrease in the oxidisable fraction. The exchangeable fraction show slight or no changes for all the four elements, with only a slight decrease of exchangeable cadmium and zinc, and unnoticeable changes in exchangeable arsenic and lead.

6.5. Plant Available PTE results of vermiculite crushed

As mentioned in the experimental plan and methodology, the vermiculite used in the experiment was in chunks. To determine if this influenced the levels of PTE, the vermiculite was grounded to a powder form and was tested in both the normal soil as well as the reduced pH soil. The results below compare the effects of vermiculite in chunks and crushed vermiculite. The vermiculite used in the main experiment is stated as “vermiculite” when placed in the original soil and “acidified soil” for the vermiculite placed in the acidified soil. The grounded vermiculite used for the extra mini experiment is referred to as “vermiculite crushed” when placed in the original soil and “crushed acidified soil” for the powdered vermiculite placed in the acidified soil.

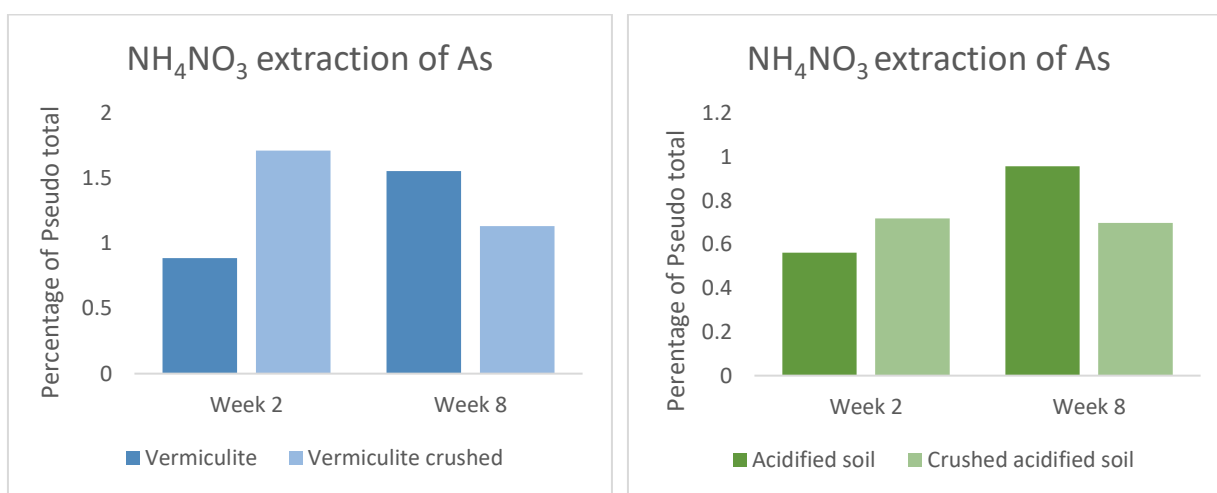


Fig.14. Comparison of the effect of vermiculite crushed and chunks on plant available arsenic.

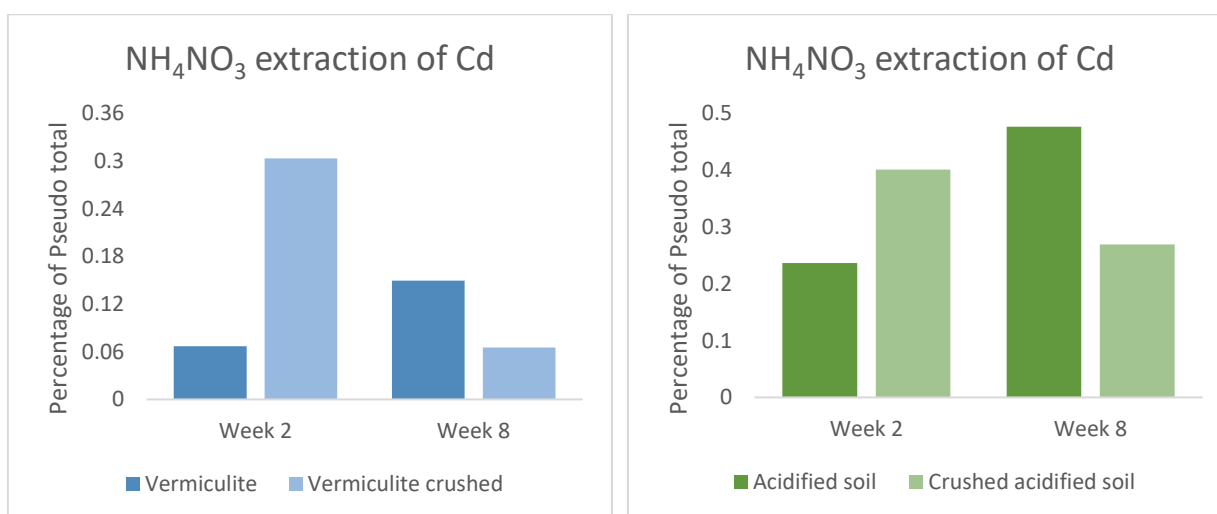


Fig.15. Comparison of the effect of vermiculite crushed and chunks on plant available cadmium.

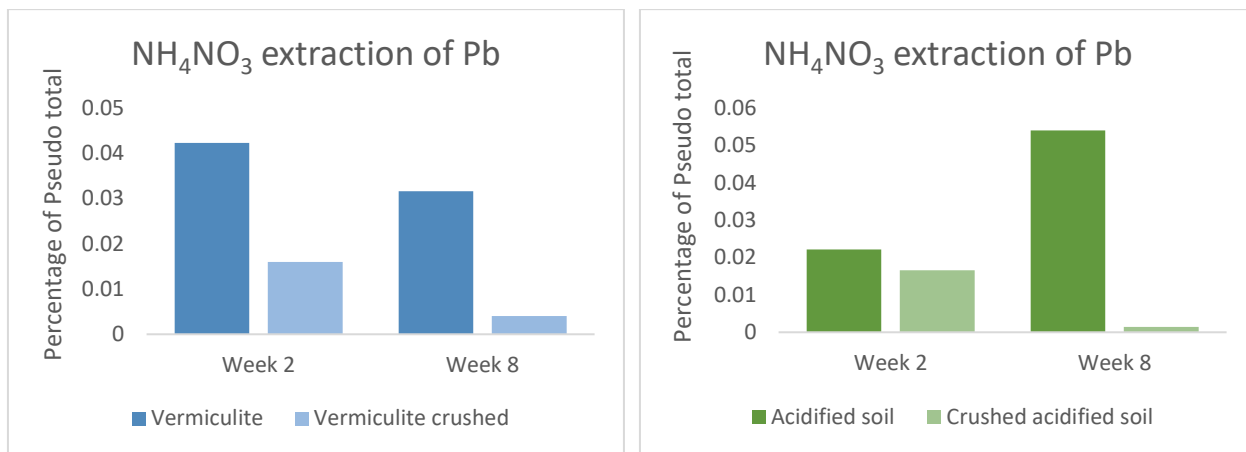


Fig.16. Comparison of the effect of vermiculite crushed and chunks on plant available lead.

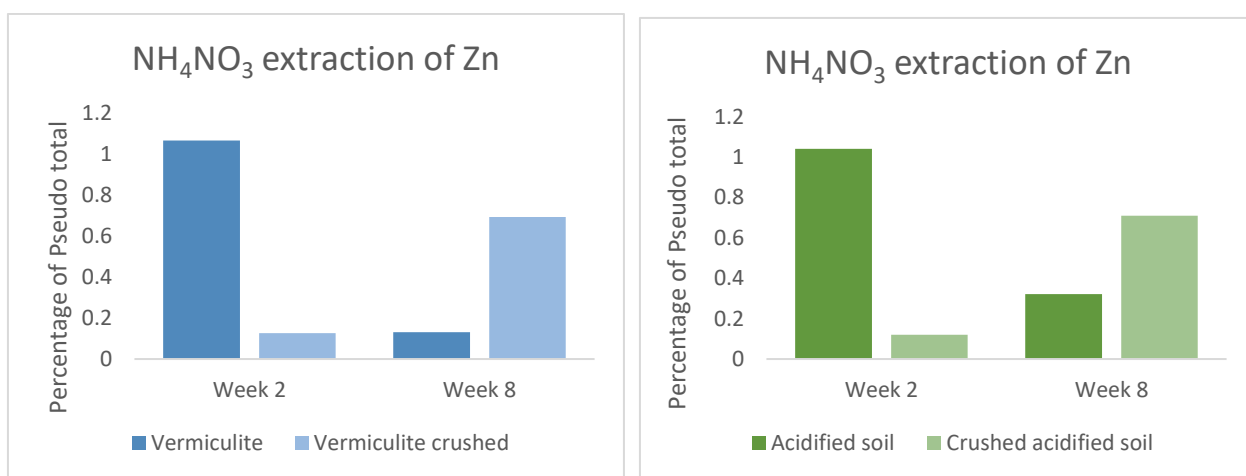


Fig.17. Comparison of the effect of vermiculite crushed and chunks on plant available zinc.

Figures 14, 15 and 16 show that vermiculite used as chunks in the experiment increased the concentrations of the metals from week two to week eight, while vermiculite crushed into a powdered form managed to reduce the total concentrations of the metals over the eight-week period, as was expected. The results for cadmium indicate that there was greater decrease in metal concentration in the original soil compared to the results of cadmium concentrations in the acidified soil. This is probably since an increase in pH increases negative surface charge, increasing cation binding. Lead seems to approximately similar lowered concentration in the original soil as well the acidified soil.

Zinc provided results that were unexpected. Zinc concentration decreased when amended with vermiculite chunks, but increased when amended with powdered vermiculite in both soil conditions.

7. Discussion

Humans have for a long time been exposed to metal(loid)s, some of which are also beneficial for human health when consumed in moderate quantities. If consumed or exposed in high quantities it could have adverse effects on human and plant health. Hence, as Järup, L. (2003) stated, there must be an association between the carrier and the human body. The carrier being air, water and soil, all of which are pathways that lead risk elements into the human body. Indicator values obtained through pseudo total extraction, which can compromise human health and animals set by the Ministry of Environment (Vyhláška č. 153/2016 Sb.) state maximum values of arsenic to be 40 mg/kg, cadmium to be 20 mg/kg and lead to be 400 mg/kg.

Despite the high indicator values for human exposure, the pseudo total concentrations exceed limits set for agricultural soils (garden soils could be classified to some extent as agricultural, in cases where consumable products are being grown in the garden). Table 7 displays the limits set by the Ministry of Environment in the Czech Republic in 1993 and 2016 and can be compared to results obtained from the pseudo total concentrations in table 8-10. The increase in metal(loid) concentration from 1993/2016 was by 50/60 mg/kg for arsenic, 10-20/80-100 mg/kg for lead, 1.10/1.70 mg/kg for cadmium and 250/330 mg/kg. The values for all four risk metals are considerably higher than the limits, and neither of the clay amendments achieved lowering the concentration to a safe limit for plant growth.

The results obtained indicate that both vermiculite and kaolinite have the adsorbing abilities that can reduce the concentration of PTE in the soil. Vermiculite was expected to reduce PTE in the soil to a larger extent than kaolinite, as a result of vermiculites higher CEC and adsorbing abilities. This is observed in the results obtained from the pseudo total extraction, where certain metal(loid)s have lower values when amended with vermiculite in normal and acidified soil. Nevertheless, the decrease in the concentrations of the metal(loid)s amended with vermiculite was not as significant as expected, with values being no more than 10 mg/kg less than the soil amended with kaolinite. The changes in pseudo total values is believed to be from addition of PTE that was introduced into the soil by the clay or by the dilution of soil volume due to the addition of the clay.

Values for the plant available PTE displayed in figure 9, allow to conclude that neither of the clays managed to steadily decrease concentrations of plant available metal(loid)s over time, but rather displays results of values drastically dropping and then increasing or vice versa. Such values for plant available PTE when amended with clay have, as far as one can tell from the literature reviewed, not been recorded by anyone. Similar to the results observed for the pseudo total concentrations, the changes in absolute values of plant available PTE amended with both clays were significantly small.

The results prove that vermiculite in its powdered form reduce the plant available portion of the metal(loid)s to a greater proportion than the vermiculite chunks and kaolinite used, except for zinc. The results from the 'extra' experiment have been concluded to be due to higher surface area, however to our best knowledge, this has not been tested. It is believed that the higher surface area creates more adsorption sites for the metals to bind with the clay particulates, thus decreasing the metal(loid)s concentration. The results of zinc increasing were suspected to be due to the presence of other cations capable of displacing zinc as observed by Kiikkila (2003) and Cao et al. (2004). As vermiculite in a powdered form has more surface area, and is not as tightly compact when in chunks, this allows for other cations to displace zinc more easily. This would explain why zinc was more effectively retained by the vermiculite chunks rather than the vermiculite powder. As the results in fig. 17 show a decrease in lead and cadmium amended with powdered vermiculite, the possibility of the two metals to out-complete zinc by adsorbing to the clay, could be the reason for the increase in zinc.

The ability for a metal to bind to clay is determined by the clay type. The amounts of metals(loid)s adsorbed have been noted to vary according to the several types of the same clay mineral as well as different clay minerals. Garcia-Miragaya et al. (1986) discovered that kaolinite adsorbed low concentrations of cadmium and zinc more strongly than montmorillonite, stating that it could have been due to the higher proportion of weakly acidic edge site on kaolinite. Illite was found to be thirty times more selective of lead than montmorillonite (Hildebrand and Blum, 1974). It has also been noted by Garcia- Miragaya and Page (1977) that cadmium adsorbed to different montmorillonite differed with the exchangeable cation in the clay. It was observed that Na-montmorillonite had the highest adsorption and Al-montmorillonite had the lowest adsorption.

The diversity of metal(loid)s to adsorb to different clay structures, could be a possible explanation for why kaolinite/vermiculite managed to adsorb higher amounts of risk elements than the other,

and can also explain why neither of the clays were able to create a distinct difference in absolute values.

Kaolinite and Vermiculite successfully reduced total concentrations of arsenic, while no noteworthy results occurred for the plant available portion of arsenic. Garcia-Sanchez et al. (2002) tested limonite and bentonite at a 10% application rate with results showing limonite reducing arsenic leaching by 80%, while bentonite reduced arsenic by 50%.

Our experiment has allowed us to find that vermiculite can reduce the total concentrations of all four risk elements analysed in the soil. Kaolinite on the other hand, appeared to be more successful on arsenic and zinc compared to the results obtained for cadmium and lead. The findings from vermiculite agree with several other studies focused on 2:1 structured clays as no literature was found focusing solely on vermiculite as an amendment. Sepiolite a 2:1 clay was used by Sun et al. (2015) to conduct a study which evaluated sepiolite as an amendment to treat cadmium polluted soil in pot and field studies. The results proved sepiolite to be an effective amendment in immobilizing cadmium; reducing plant uptake 14.4%-84.1% in pot trials and 22.8%-61.4% in field trials.

The results obtained from the experiment proved kaolinite and vermiculite (chunks) to be somewhat suitable amendments for zinc, managing to decrease both total and plant available zinc. The results for zinc from this experiment are similar (but not to same extent) to the results obtained by Alvarez-Ayuso and Garcia-Sanchez, (2003a) and Alvarez-Ayuso and Garcia-Sanchez, (2003b). Whose results showed a decrease of the exchangeable fraction of zinc by 76% when amended with palygorskite and 99% when amended with sepiolite, proving magnesium silicate clays can efficiently reduce zinc concentrations.

The inconsistent results for cadmium and lead could be a result of being held exchangeably through the outer-sphere linkages which can be displaced by other cations (Alloway et al., 2013) in a case of vermiculite (chunks). The vermiculite powder seems to work perfectly for cadmium, arsenic and lead as shown in figure 14, 15 and 16. However, it did not work for zinc probably because of other cations displacing zinc to larger extent at the sorbent with higher surface area - the powdered vermiculite, as suggested before.

8. Conclusions

The initial aim when planning the experiment was to describe the fraction distribution of PTE according to the BCR sequential extraction before and after acidification and the addition of the amendments. However, during the experiment it seemed more logical to evaluate plant available content of PTE before and after acidification and addition of amendments. This was due to the fact that the soil sample was from a garden that was used to plant fruit and vegetable, so the aims focused more towards reducing plant available PTE to prevent contamination of the fruits and vegetables.

Batch leaching was changed to focus more on the amendments being used and their ability to adsorb risk elements to decrease plant available content.

Concentrations of total as well as plant available metal(loid)s present in garden soils contaminated by mining were monitored over a two-month period when amended with two structurally different clays; vermiculite and kaolinite.

Over the two-month period, the total concentration of the four metal(loid)s (arsenic, cadmium, lead and zinc) managed to decrease lower than the control when amended with vermiculite (chunks) and the vermiculite powder worked best for cadmium, arsenic and lead. On the other hand, kaolinite was more successful in reducing arsenic and zinc concentrations compared to its ability to reduce concentrations of cadmium and lead. The plant available results prove to be quite inconsistent, with values steadily rising, decreasing then increasing and vice versa. The one result that proved to be successful was the graph for zinc depicted in figure 9. Although all the values decrease in week four only to increase by week eight, the concentration of zinc is significantly lower than the concentration in week two. However, the literature proves sepiolite, illite and limonite to be the most popular and effective amendments tested to treat certain metal(loid)s in contaminated soil.

The pH influence was not as significant as it was expected for our experiment. Nevertheless, content of studied elements in acidified soil has also decreased over the eight-week time frame. However, ultimately maintained higher values than the control ones, pointing on the potential danger of soil acidification process.

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