An Investigation into the Effect of Ochre Amendments on Soil Carbon Storage, Aggregate Formation and Plant Growth

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Abstract

Ochre is an iron oxide waste that is currently stockpiled pending re-use or disposal via landfill, providing motivation to research an environmentally beneficial application for this accumulating waste product. It is known that iron oxide amendments to soils can stabilise and increase soil organic carbon content, as well as having other potential benefits to soil structure. This thesis researches whether the ochre amendments to soils could be utilised as a land-based CO_2 removal method, aiding in the mitigation of climate change. It is hypothesised that additions of ochre to soils could reduce carbon availability, leading to carbon sequestration. This hypothesis is tested through an adsorption experiment which assesses whether ochre amendments to an Agricultural and Woodland Soil increase organic carbon sorption and affect phosphorus availability, metal release and soil pH. Soil incubation/plant growth experiments were also conducted which assess whether ochre amendments reduce carbon availability and affect plant growth and aggregate formation. The adsorption study concluded that the addition of the ochre to soils enhanced organic carbon sorption, reducing soil carbon availability and lability, based on the assessment of the change in quantity of carbon released into soil solution (5.89 $\pm$ 0.86 to 10.62 $\pm$ 1.29 mg/L and 7.15 $\pm$ 0.42 to 10.75 $\pm$ 3.01 mg/L for ochre amended Agricultural and Woodland Soils, respectively), relative to control soils (10.95 $\pm$ 0.16 mg/L and 13.66 $\pm$ 0.21 mg/L for Agricultural and Woodland Soil controls, respectively) ($p \leq 0.05$: ANOVA and Dunnett's test). Ochres with a high goethite content (67.27 - 97.50 %) and a relatively low - neutral pH (4.45 $\pm$ 0.05 to 8.34 $\pm$ 0.04) were favourable in increasing soil organic carbon sorption. However, ochres with a high Na content (344 $\pm$ 159 to 846 $\pm$ 41.7 mg/kg) were found to perform less favourably. The incubation/plant growth study found a lack of difference in the extractable carbon content of ochre amended soils, compared to control soils, but found a reduction in the cold water extractable carbon content in ochre treated Woodland Incubation Soils (6.30 $\pm$ 0.96 to 14.88 $\pm$ 3.47 mg/L), indicating decreased carbon availability and increased carbon storage, relative to the Woodland Incubation control (20.88 $\pm$ 3.40 mg/L) ($p \leq 0.05$: ANOVA and Dunnett's test). This provides motivation for further experiments to take place, requiring a longer incubation period, with thorough mixing, to ensure there is an increased opportunity of contact between the iron oxide and dissolved organic carbon. In practical applications, a solution for increasing contact chances is to plough the ochre into the soil as a slurry. Despite the addition of ochre causing there to be more fine-grained material in the soils, generally no effects on aggregate fraction size were found, implying that the fine ochres have supported the aggregation of material to form coarse, stable macroaggregates, improving soil structure. Future work should look into the organic carbon content of each aggregate size, as well as measuring changes in carbon pools, as the results of this thesis suggest there is a shift in carbon from a very labile to a less labile pool. Finally, additional experiments should analyse for potential toxic metal uptake in the plant shoot biomass, given that some of the ochre treated soils released more metals into solution than the control in the adsorption study, despite the ochre amendments having no effect on plant growth.

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Declaration

I declare that this thesis is a presentation of original work and I am the sole author. This work has not previously been presented for an award at this, or any other, University. All sources are acknowledged as references.

Chapter 1: Introduction to the Thesis

The basis for this research stems from the recognised importance of utilising land-based CO_2 removal methods to increase soil carbon (C) sinks through land management, as a potential climate change mitigation technique. It is known that iron (Fe) oxide in soil can can stabilise and increase soil organic carbon (SOC) content, as well as having other potential benefits to soil structure, as explored in the literature review of Chapter 2. This knowledge provides the motivation for this thesis, meriting experiments that not only decipher whether Fe oxides have the ability to adsorb dissolved organic carbon (DOC), but also store this DOC long-term, reducing C availability and sequestering it. It is of importance to explore the potential effects of Fe oxide amendments on plant growth, as well as physical and chemical soil properties including aggregate stability, phosphorus availability and soil pH.

Ochre is an Fe oxide waste that accumulates in streams draining from now abandoned coal and metal mines and in settlement ponds or constructed wetlands designed to reduce pollution levels in acid mine drainage. In the UK, the Coal Authority is responsible for 82 (as of December 2020) mine water treatment schemes that remove ca. 4000 tonnes of Fe per year from these water courses, resulting in ochre production. Currently ochre is stockpiled pending re-use or disposal. A significant proportion is landfilled, providing motivation to research an environmentally beneficial application for this accumulating waste product.

Additional to this chapter, this thesis comprises 4 further chapters. The literature review of Chapter 2, exploring the above information in depth, gives rise to the overarching hypothesis that the additions of Fe oxide ochre to soils will reduce C availability, leading to C sequestration. This hypothesis is tested throughout Chapters 3 and 4.

Chapter 3 discusses adsorption experiments which assess whether ochres do have the ability to adsorb organic C, based on the assessment of the change in quantity of C released into soil solution. Chapter 3 also investigates the potential of ochre amendments releasing potentially toxic mobile metals into soils.

Chapter 4 explores soil incubation/plant growth experiments which assess whether C availability is reduced in ochre amended soils in a more realistic experiment than the adsorption study, as well as evaluating whether ochre amendments affect plant growth and whether Fe oxide is the limiting factor in soil aggregate production.

Chapter 5 closes the thesis, summarising the key findings of the adsorption and incubation/plant growth experiments of Chapters 3 and 4, as well as discussing recommendations for future experiments and work within this field.

Chapter 2: Literature Review

1 Importance of Carbon Sequestration

The term 'carbon sequestration' is commonly used to describe any increase in SOC content caused by a change in land management, with the implication that increased soil C storage mitigates climate change (Powlson *et al.*, 2011). Terrestrial ecosystems remove about 3 GtC/yr from the atmosphere through net growth, absorbing about 30 % of CO_2 emissions from fossil fuel burning and net deforestation, while the world's forest ecosystems store more than twice the C in the atmosphere (Canadell *et al.* 2007; Canadell and Raupach, 2008). Terrestrial ecosystems store about 2,100 GtC in living organisms, leaf litter and soil organic matter (SOM), which is almost three times that currently present in the atmosphere (Royal Society, 2009). The Royal Society (2009) report recognises the importance of land-based CO_2 removal methods to increase land C sinks through land management. Beerling *et al.* (2018) discuss the method of biogeochemical improvement of soils by adding crushed, fast-reacting silicate rocks to croplands as one such CO_2-removal strategy; this approach has the potential to improve crop production, increase protection from pests and diseases, and restore soil fertility and structure. Beerling *et al.* (2018) recognise that the issues of public perception, trust and acceptance to methods such as this must also be addressed. As discussed in Sections 1.2 and 1.3, Fe oxide amendments to soils could also stabilise and increase SOC content, acting as another CO_2-removal strategy, as well as having other potential benefits to soil structure, giving motivation for this research area to be studied further. However, Powlson *et al.* (2011) highlight the importance of understanding how the climate change benefit of increased SOC from biogeochemical soil improvement methods must be balanced against any potential greenhouse gas emissions associated with possible transport or processing procedures of such amendments.

2 Iron Oxide–Organic Matter Interactions

The diverse physical and chemical characteristics of organic matter (OM) in soil enable it to influence soil biological, physical and chemical properties (Baldock and Nelson, 1999). OM contributes significantly to soil resilience and provides the chemical energy and nutrients essential to the activity of soil biological systems, as well as contributing directly to soil cation exchange and buffering capacity and both directly and indirectly to soil structural stability, water retention and soil thermal properties (Baldock and Skjemstad, 1999).

Minerals are widely assumed to protect OM from degradation in the environment, promoting the persistence of C in soil (Wang *et al.*, 2019; Kleber *et al.*, 2021). Kleber *et al.* (2021) state that to do so, a broad set of interactions occur, with minerals adsorbing organic compounds to their surfaces and/or acting as catalysts for organic reactions. Decomposition of adsorbed OM is typically substantially slower than the decomposition of the same type of OM in a

freely suspended or dissolved state (Kalbitz *et al.*, 2005; Kleber *et al.*, 2021). Investigations of sediment dynamics demonstrate that fine-grained minerals and mineral–OM assemblages have a controlling influence on the cohesive nature of sediment (Kleber *et al.*, 2021).

A key stabilisation mechanism of SOM is that of SOM sorption onto soil minerals, including clay minerals and Fe and Al oxides, reducing its availability to microorganisms, inhibiting microbial degradation (Nelson *et al.*, 1994; Sollins *et al.*, 1996; Vieublé Gonod *et al.*, 1998; Baldock and Skjemstad, 2000; Gonod *et al.*, 2006). Mineral-associated OM is considered a stable reservoir for soil nutrients that influences long-term soil C and N dynamics, constraining potential CO_2 emissions and cycling reactive N (Jastrow, 1996; Gentsch *et al.*, 2015; Kleber *et al.*, 2015; Jilling *et al.*, 2018). However, the results of Jilling *et al.* (2021) indicate that common root exudates, glucose and oxalic acid, can increase the turnover and potential release of C and N from mineral associated OM through indirect (microbial) and direct (non-microbial) mechanisms. Both C and N were transferred from mineral associated OM into bioavailable pools, with glucose working primarily via the microbial community and oxalic acid directly destabilising metal-organic interactions, demonstrating the potential for simulated root exudates to facilitate mineral associated OM destabilisation (Jilling *et al.*, 2021). It is hence of importance to take this into consideration when determining whether Fe oxides have the ability to adsorb and store soil C long-term.

The importance of considering other controls on C storage is also supported by the results of Moni *et al.* (2010), who observed positive relationships involving amorphous Fe oxides for horizons at small pit scale, suggesting that, at this scale, amorphous Fe oxides may be involved in processes leading to organic C soil storage. However, at the larger field scale, the abundance of controls for organic C storage is increasing, meaning such relationships were no longer observable as the empirical relationships established at pit scale are not powerful enough to predict the behaviour of a whole landscape. The mechanistic knowledge established at pit scale may thus not easily be scaled up in soils where interaction with Fe oxides is not the only control on organic C storage. Wagai *et al.* (2020) state that given that OM is mainly located in meso-density fractions, the capacity of a soil to protect OM may be controlled by the balance of three processes: (1) microbial processing of plant-derived OM, (2) dissolution of metals, and (3) the synthesis of organo-metallic phases and their association with clays to form meso-density microaggregates.

The results of Wen *et al.* (2019) show that vigorous Fe mobilisation can be regulated by long-term application of organic amendments, and that these organically amended soils contained significantly higher concentrations of poorly crystalline Fe that was closely related to SOC storage in their studied upland and paddy soils. The work by Wen *et al.* (2019) implies that continuous organic amendments to soils could initialise a positive feedback loop for the presence and maintenance of poorly crystalline Fe, which in turn helps preserve SOC over longer time scales. The comprehensive understanding of SOC–Fe associations regulated by agricultural management practices is of crucial importance for enhancing soil C sequestration for sustainable management of SOC in modern agroecosystems, and for improving our understanding of C

cycling processes under the global environmental change scenarios (Wen *et al.*, 2019).

Wen *et al.* (2019) state that the potential mechanisms of OM storage are proposed as follows: (1) dissolved organic matter (DOM) from the organically amended soils is more likely to co-precipitate with poorly crystalline Fe, and DOM from the inorganically fertilised soils is to a larger extent adsorbed on poorly crystalline Fe. The co-precipitated Fe–OM complexes are more resistant to desorption than the adsorbed OM. (2) DOM extracts from organic amended soils contain higher concentration of aromatic functional groups, and exhibit stronger inhibitory effects on crystallisation of poorly crystalline Fe compared with inorganically amended soils. (3) Microbially mediated Fe cycling is regulated by long-term inorganic/organic inputs. Greater consumption of poorly crystalline Fe was observed in inorganically fertilised soil than that in organically amended soil, due to a higher relative abundance of well-known Fe(III) reducers; conversely, Fe(II) oxidisers, were more abundant, and produced higher levels of poorly crystalline Fe under organic amendments (Wen *et al.*, 2019).

Kleber *et al.* (2021) recognise that future work must distinguish adsorption as a condition for further reactions instead of as a final destination for organic adsorbates. This merits experiments that not only decipher whether Fe oxides have the ability to adsorb DOC, removing C from solution, but also store this DOC long-term, reducing its availability.

Goethite, one of the most thermodynamically stable Fe oxides, holds a strong ability to adsorb to SOC, subsequently decreasing the release of C from soils and fixing it (Liu *et al.*, 2014). Goethite, found in the pedosphere, hydrosphere and biosphere, resulting from rock weathering, is often poorly crystalline and rich in defects and impurities, giving it good surface activity (Liu *et al.*, 2014). Organic compound adsorption onto goethite usually depends on the solution pH, chemical composition of OM, ionic strength and cation composition in solution; organic acids, such as humic acid, can improve the adsorption capacity of goethite for other organic compounds (Antelo *et al.*, 2007; Liu *et al.*, 2014). Humic substances (humic acids and fulvic acids) may constitute more than 80 % of SOM (Stevenson, 1994). It is well known that phosphate shows a relatively strong affinity for Fe (hydr)oxides, including goethite (Hiemstra and van Riemsdijk, 1996, Manning and Goldberg, 1996). Sibanda and Young (1986) investigated the competitive adsorption of phosphate and humic substances on goethite, concluding that humic substances block some surface sites, thereby reducing the adsorption of phosphate. These results are supported by that of Antelo *et al.* (2007), who found that soil humic acid and phosphate compete for the goethite surface, since the presence of phosphate decreases the adsorption of soil humic acid and the presence of soil humic acid decreases the adsorption of phosphate. These studies highlight the importance of accounting for phosphate when experimenting whether Fe oxides have the ability to adsorb and fix DOC, as phosphorus is often a limiting nutrient (Lajtha and Jarrell, 1999), and so the possibility of reduced P levels could have an effect on plant growth in Fe oxide amended soils.

3 Role of Iron Oxide in Soil Structure

Soil structure is an important soil property which mediates many biological and physical processes in soils; soil structure determines porosity and infiltration, hence regulating water availability to plants and soil erosion susceptibility (Six *et al.*, 2000a). Since soil structure also influences losses of agrochemicals, C sequestration and N gas losses, it is important to maintain soil structure to reduce the environmental impact of agricultural practices (Six *et al.*, 2000a).

The presence of Fe oxides, as well as Al oxides and kaolinite, is an important factor for the stability of a soil (Six *et al.*, 2000a). Much research has revealed that Al and Fe oxides are important aggregating agents, affecting soil structural stability (Barberis *et al.*, 1991; Oades and Waters, 1991; Mbagwu and Schwertman, 2006). Kemper and Koch (1966) and Six *et al.* (2000a) found a positive correlation between Fe oxide content and aggregate stability, with the latter finding a smaller effect of soil management practices on soil stability in soils with a high presence of oxides and kaolinite.

The aggregating effect of oxides is mainly at the microaggregate level (Oades *et al.*, 1989; Igwe *et al.*, 1999; Muggler *et al.*, 1999), but macroaggregation has also been related to oxide content (Arduino *et al.*, 1989; Six *et al.*, 2000b; Imhoff *et al.*, 2002). Six *et al.* (2000a) state that oxides can act as binding agents in three ways: (1) organic materials adsorb on oxide surfaces (Oades *et al.*, 1989); (2) an electrostatic binding occurs between the positively charged oxides and negatively charged clay minerals (El-Swaify and Emerson, 1975); and (3) a coat of oxides on the surface of minerals forms bridges between primary and secondary particles (Fordham and Norrish, 1983; Kitagawa, 1983; Muggler *et al.*, 1999).

Yin *et al.* (2016) found that there was a positive relation between pyrophosphate-extractable Fe and water-stable aggregates but no significant relations were observed between dithionite-citrate or ammonium oxalate extractable Fe oxides and aggregate stability. Igwe *et al.* (2009) state that oxalate extracted oxides are responsible for aggregate formation. Moreover, Arduino *et al.* (1989) found oxalate extractable Fe with the greatest surface area to promote macroaggregate stabilisation. Yin *et al.* (2016) found that oxalate extractable Fe under P fertiliser treatments was greater than those under other N and manure fertiliser combination treatments. This result is in agreement with that of other studies (Jim, 2003), who reported that P was likely to be fixed as a result of Fe oxides. P fertilisation stimulates the precipitation of oxalate extractable Fe and phosphates that act as bonding agents to enhance aggregation. Yin *et al.* (2016) state that N and P fertilisers may reduce soil pH, increasing the mobility of Fe oxides, which are pH-dependent, allowing Fe oxides to interact synergistically with SOC and dispersible clay to improve aggregate stability (Bronick and Lal, 2005). OM can form a complex with Fe and Al oxides at low pH to give rise to mobile, organic-metallic compounds that decrease microbial access to SOC and mineralisation (Bronick and Lal, 2005).

Wang *et al.* (2019) found that long-term manure fertilisation promoted the formation of non-crystalline Fe fractions, which bounds to SOC to form soil macro-aggregates. Thus, the formation of SOC–Fe association in soil and soil aggregates plays a crucial role in SOC preservation. Wang *et al.* (2019) conclude that the understanding of SOC–mineral associations

is of great importance to enhance C sequestration in soils. The results of Yin *et al.* (2016) and Wang *et al.* (2019) are promising in that if Fe oxides were added to agricultural soils in order to enhance SOC storage, added P and manure fertilisers would have not an adverse but a promoting effect on enhancing aggregation and preserving SOC.

4 Proposed Research

Upon assessment of the related literature which has been reviewed here, the following hypothesis can be formulated: additions of ochre to soils could reduce C availability, leading to C sequestration. To test this hypothesis, the following objectives are proposed: (1) conduct adsorption experiments to assess whether ochres do adsorb C and then, if successful in doing so, (2) conduct soil incubation/plant growth experiments to assess whether C availability is reduced in ochre amended soils.

Chapter 3: An Investigation into the Effect of Ochre Amendments on Carbon Release into Solution from Soil

1 Introduction

1.1 Carbon Storage Potential

Nearly 80 % of the total C in terrestrial ecosystems is preserved in soils, with 75 % of this C being in the form of SOC (Lal, 2004; Wen *et al.*, 2019). Köchy *et al.* (2015) report the global stocks of SOC as 1923, 1455 and 720 Pg for the upper 2.3, 1.0 and 0.3 m, respectively. Thus, the quantity of SOC in the 0 - 30 cm layer is about twice the amount of C in atmospheric CO_2 and three times that in global above-ground vegetation (Powlson *et al.*, 2011). Soil minerals, predominantly Fe and Al oxides, hydroxides, and oxyhydroxides, are known to influence the biological stability of SOM, including DOC, via sorption to reactive surface sites due to their small size (down to several nanometres) and high surface reactivity via surface hydroxyl groups, or co-precipitation with the solid phase (McLean and Bledsoe, 1992; Kaiser and Guggenberger, 2003; Kleber *et al.*, 2005; Wagai and Mayer, 2007; Saidy *et al.*, 2012; Kleber *et al.*, 2015; Porras *et al.*, 2017; Fujii *et al.*, 2019; Wen *et al.*, 2019; Wagai *et al.*, 2020). Fe oxides can also promote aggregation (Shang and Tiessen, 1998), which indirectly enhances OM stability (Totsche *et al.*, 2017), without necessarily showing proportionality to metal concentrations (Wagai *et al.*, 2020). A decreasing pH increases the concentration of Fe cations in the soil solution by enhancing the dissolution of Fe (Wagai and Mayer, 2007; Porras *et al.*, 2017). Heckman *et al.* (2009) suggest a regulatory role of pH with respect to the mode of OM stabilisation, with increased Fe complexation leading to enhanced stabilisation of OM, owing to the increased solubility of Fe with decreasing pH in the acidic range (Porras *et al.*, 2017). Fe can hence be a strong predictor of SOC storage and turnover in soils with a relatively high extractable metals content and a moderately acidic to circumneutral pH, protecting SOC against decomposition in these soils. This leads to the hypothesis that Fe oxide additions would enhance organic C sorption and increase soil C storage (Kleber *et al.*, 2005; Porras *et al.*, 2017).

McKnight *et al.* (1992) found that from 1979 - 1986, ∼40 % of the DOC in the river systems studied was removed from solution by sorption onto hydrous Fe and Al oxides, which then settled. Previous empirical studies have reported a positive correlation between Fe content, SOC stocks and ^{14}C-based turnover time, all of which reduce CO_2 emissions (Masiello *et al.* 2004; Torn *et al.* 1997; Rasmussen *et al.* 2005; Heckman *et al.* 2009; Saidy *et al.*, 2012; Porras *et al.*, 2017). SOC stabilisation also has benefits for agricultural use and agronomic crop productivity, due to the positive influence of SOC on a range of soil physical, chemical, and biological properties, stabilising soil structure, enhancing resistance to soil erosion and increasing substrate and supply of nutrients for microbes, thus improving the productive capacity of soil (Bronick and Lal, 2005; Saidy *et al.*, 2012; Peng *et al.*, 2015; Ramesh *et al.*, 2019). Powlson *et al.* (2011) state that any measure that increases SOC content is likely

to have beneficial impacts on soil properties and functioning; thus, even practices that increase SOC but have no direct benefit for climate change mitigation are likely to be beneficial in other ways, with SOC accumulation perhaps therefore being regarded as a 'no regrets' policy.

1.2 Current Ochre Use

Ochre is an Fe oxyhydroxide-rich waste that mainly accumulates in two environments in the UK: (1) in streams draining from now abandoned coal and metal mines and, (2) settlement ponds or constructed wetlands designed to reduce pollution levels in acid mine drainage (Younger *et al.*, 2002). It has been previously reported that treatment of polluting discharges from abandoned coal mines in the UK produces ca. 30000 tonnes of hydrous Fe oxides per year (Dobbie *et al.*, 2009). In the UK, the Coal Authority is responsible for 82 (as of December 2020) mine water treatment schemes that remove ca. 4000 tonnes of Fe per year from water courses resulting in ochre production (UK Government, 2017). Currently ochre is stockpiled pending re-use or disposal; a significant proportion is landfilled.

1.3 Potential Ochre Use

Previous studies have shown the potential for Fe oxides to be sorptive for As and P, giving rise to investigations into the potential use of ochre (Livesey and Huang, 1981; Elkhatib *et al.*, 1984a; Elkhatib *et al.*, 1984b; Bowell, 1994; Sun and Doner, 1996; Matis *et al.*, 1997; Manning *et al.*, 1998; Sun and Doner, 1998; Jain *et al.*, 1999; Jackson and Miller, 2000; Manning and Suarez, 2000; Goldberg and Johnston, 2001; Grafe *et al.*, 2001; Ford, 2002; Garcia-Sanchez *et al.*, 2002; Goldberg, 2002; Smith *et al.*, 2002; Waltham and Eick, 2002; Doi *et al.*, 2005; Heal *et al.*, 2005; Dobbie *et al.*, 2009; Sibrell *et al.*, 2009; Fenton *et al.*, 2012; Olimah *et al.*, 2015).

A potential commercial use for ochre is to remediate arsenic contaminated soil (Boisson *et al.*, 1999; Garcia-Sanchez *et al.*, 1999; Warren *et al.*, 2003; Warren and Alloway, 2003). Dobbie *et al.* (2009) found that ochre also has the potential for removing phosphorus from wastewater and agricultural runoff, providing possible applications for farmers. In their studies, Dobbie *et al.* (2009) diverted secondary-treated wastewater effluent through a trough containing granular and pelletised ochre, finding no detectable release of potentially toxic mobile metals, including Al, Cd, Cr, Cu, Fe, Ni, Pb and Zn, from the ochres. It is evident there exist a number of possible environmentally beneficial uses for ochre. Given the role of Fe in OM stabilisation, another possible, currently unexplored role is the use of ochre amendments to enhance organic C sorption. The hypothesised capability of ochre to enhance organic C sorption and increase SOC storage, would give a great motive for it to be added as a treatment to soils.

1.4 Aims, Objectives and Hypotheses

The aim of this study was to understand the effect of ochre amendments on the availability and lability of SOC, based on the assessment of the change in quantity of C released into soil solution. This was achieved through an adsorption experiment in which ochres were added to

2 soils with varying properties and the quantity of C released into solution was measured. It is hypothesised that the addition of the ochres will enhance organic C sorption, decreasing the release of C into solution.

2 Methodology

2.1 Experimental Materials

The ochre samples were provided by Mr. J Aumonier of the Coal Authority and were collected from the former mine sites of Morlais (SN 57225 02234), Saltburn (NZ 67260 19783), Ynysarwed (SN 80857 01749), Lynemouth (NZ 28372 91616), Blenkinsop (NY 67006 64317), Polkemmet (NS 93550 64050) and Whitworth (SS 79810 96870). Previous analysis for the Coal Authority indicates that the ochres have varying As (and other contaminant metal) concentrations, and they all have relatively high production rates (Aumonier, Pers. Comm., 2020). The Saltburn, Ynysarwed, Blenkinsop, Polkemmet and Whitworth ochres were supplied moist in sealed, plastic containers; the Morlais and Lynemouth ochres were supplied dry. All of the ochres were freeze dried and crushed to $\leq$2 mm prior to characterisation and use in experiments to homogenise the material for reproducibility.

Two soils were used in this study, an Agricultural Soil (SE 62635 49583) with a relatively low OM content and a Woodland Soil (SE 62084 50860) with a greater OM content, provided by Prof. M Hodson. The soils were air-dried, sieved to $\leq$2 mm to remove all visible fine and coarse roots, leaf litter, or any less decomposed plant materials and gravels, and stored prior to characterisation and use in experiments.

The ochres were characterised using standard methods applied to the freeze-dried samples. The pH was determined in water (Rowell, 1994). Mass loss on ignition at 350°C was used as a proxy for total OM and total C (Rowell, 1994). The surface area of the ochres was determined by N_2 adsorption and application of the BET isotherm (Brunauer *et al.*, 1938). Pseudo-total As, Fe and other potentially toxic metals were measured by aqua regia digest (British Standard, 1995). Particle size analysis was carried out using a laser granulometer. Mineralogy of the ochres was determined by X-ray diffraction using a Philips PW1050 with Philips X40 software at the University of Leeds. The ochres were also characterised in terms of extractable Fe using sodium pyrophosphate, acid ammonium oxalate and dithionite-citrate methods which are assumed to extract organically complexed Fe; organically complexed and amorphous inorganic Fe; and organically complexed, amorphous inorganic Fe and Fe oxides, respectively (Loeppert and Inskeep, 1996). The Fe extractions are discussed in more detail in Chapter 4, Section 2.2. The soils were characterised using standard methods applied to the air-dried samples. The methods for pH, total C content, potentially toxic metals and mineralogy were used as outlined above. In addition to those analyses, plant available inorganic phosphorus was measured by the Olsen P extraction and analysis on an autoanalyser (Seal AA3 Autoanalyser) (Sims, 2000) and soil texture was assessed by hand texturing established by Thien (1979). The Olsen P

extraction is discussed in more detail below in Chapter 4, Section 2.2. Results of these analyses are given in Table 1.

Table 1: Mean soil and ochre physical and chemical properties used in the adsorption experiments (n = 3, ± standard deviation). Different letters represent a significant difference in parameter between the soils and ochres ($p \leq 0.05$: ANOVA and Tukey test).

Parameter	Agricultural Soil	Woodland Soil	Morlais Ochre	Saltburn Ochre	Ynysarwed Ochre	Lynemouth Ochre	Whitworth Ochre	Blenkinsop 1 Ochre	Blenkinsop 2 Ochre	Polkemmet Ochre
Textural Class	Sandy clay loam	Sandy clay	ND	ND	ND	ND	ND	ND	ND	ND
pH	6.86 ± 0.030^{a}	6.69 ± 0.010^{b}	7.92 ± 0.020^{c}	8.23 ± 0.030^{d}	8.34 ± 0.040^{e}	8.07 ± 0.020^{f}	4.45 ± 0.050^{g}	7.83 ± 0.010^{h}	9.36 ± 0.050^{i}	8.02 ± 0.020^{f}
Total C (%)	3.69 ± 0.090^{a}	5.63 ± 0.12^{b}	8.41 ± 0.19^{c}	11.24 ± 0.060^{d}	7.52 ± 0.12^{e}	12.38 ± 0.20^{f}	14.31 ± 0.10^{g}	10.68 ± 0.090^{h}	6.67 ± 0.13^{i}	12.45 ± 0.15^{f}
BET Surface Area (m^2/g)	ND	ND	71.2876	163.2901	272.8558	132.9758	105.4271	176.7204	38.8041	139.1668
Particle size	ND	ND	<2µm 1.70 % 2 - 20µm 34.1 % 20 - 2000µm 64.2 %	<2µm 2.60 % 2 - 20µm 23.0 % 20 - 2000µm 74.4 %	<2µm 16.5 % 2 - 20µm 32.7 % 20 - 2000µm 50.8 %	<2µm 15.1 % 2 - 20µm 60.8 % 20 - 2000µm 24.1 %	<2µm 3.99 % 2 - 20µm 38.9 % 20 - 2000µm 57.1 %	<2µm 7.45 % 2 - 20µm 25.9 % 20 - 2000µm 66.7 %	<2µm 7.67 % 2 - 20µm 44.5 % 20 - 2000µm 46.8 %	<2µm 6.06 % 2 - 20µm 36.5 % 20 - 2000µm 57.4%
Ca, mg/kg	720 ± 2.6^{ab}	690 ± 82^{a}	1500 ± 14^{b}	5000 ± 130^{c}	10000 ± 300^{d}	3900 ± 460^{e}	210 ± 63^{a}	2900 ± 27^{g}	29000 ± 630^{h}	2000 ± 16^{b}
Mg, mg/kg	420 ± 0.93^{a}	380 ± 3.2^{b}	210 ± 3.7^{c}	340 ± 8.5^{d}	400 ± 5.4^{ab}	810 ± 25^{e}	61 ± 7.7^{f}	300 ± 3.0^{g}	ND	290 ± 1.5^{g}
K, mg/kg	290 ± 3.0^{a}	210 ± 6.2^{b}	75 ± 21^{c}	6.2 ± 1.4^{d}	16 ± 0.71^{d}	61 ± 7.6^{ce}	63 ± 1.8^{ce}	11 ± 0.97^{d}	7.9 ± 1.3^{d}	52 ± 1.0^{e}
Na, mg/kg	8.1 ± 16^{a}	120 ± 110^{ac}	45 ± 24^{ac}	340 ± 160^{ac}	240 ± 170^{ac}	850 ± 42^{b}	120 ± 96^{ac}	140 ± 150^{ac}	380 ± 150^{c}	100 ± 130^{ac}
Al, mg/kg	2700 ± 19^{a}	1300 ± 7.0^{b}	400 ± 6.9^{c}	23.2 ± 0.98^{d}	370 ± 5.6^{e}	55 ± 2.1^{f}	330 ± 4.9^{g}	260 ± 2.0^{h}	74 ± 1.0^{f}	290 ± 2.6^{i}
Fe, mg/kg	4000 ± 50^{a}	2500 ± 16^{b}	31000 ± 170^{c}	39000 ± 710^{d}	33000 ± 350^{e}	39000 ± 1400^{d}	36000 ± 58^{f}	39000 ± 240^{d}	22000 ± 320^{g}	39000 ± 140^{d}
Mn, mg/kg	110 ± 0.52^{a}	69 ± 0.51^{b}	390 ± 7.1^{c}	82 ± 2.1^{b}	310 ± 4.9^{d}	100 ± 3.1^{a}	17 ± 0.30^{e}	130 ± 0.81^{f}	930 ± 12^{g}	240 ± 0.88^{h}
B, mg/kg	53 ± 0.13^{a}	31 ± 0.24^{a}	520 ± 6.5^{b}	780 ± 18^{c}	600 ± 6.2^{d}	840 ± 23^{e}	720 ± 11^{f}	850 ± 6.7^{e}	340 ± 3.6^{g}	850 ± 2.9^{e}
Ba, mg/kg	51 ± 0.055^{a}	27 ± 0.10^{b}	26 ± 0.72^{c}	6.2 ± 0.40^{d}	6.3 ± 0.69^{d}	11 ± 0.42^{e}	12 ± 0.38^{e}	6.2 ± 0.70^{d}	3.3 ± 0.60^{f}	29 ± 0.19^{g}
Co, mg/kg	1.50 ± 0.0079^{a}	0.83 ± 0.0069^{b}	0.86 ± 0.035^{b}	2.9 ± 0.080^{c}	3.5 ± 0.040^{d}	1.3 ± 0.080^{e}	0.48 ± 0.012^{f}	1.9 ± 0.011^{g}	5.9 ± 0.073^{h}	0.45 ± 0.029^{i}
Li, mg/kg	5.60 ± 0.039^{a}	2.4 ± 0.052^{b}	1.2 ± 0.031^{c}	0.28 ± 0.0087^{d}	0.13 ± 0.0032^{e}	1.0 ± 0.030^{f}	0.54 ± 0.019^{g}	0.37 ± 0.010^{h}	1.5 ± 0.019^{i}	0.63 ± 0.022^{j}
Zn, mg/kg	15 ±0.31^{a}	13 ± 0.12^{b}	11 ± 0.18^{b}	26 ± 0.97^{c}	18 ± 0.35^{d}	18 ± 0.25^{d}	18 ± 1.7^{d}	23 ± 0.82^{g}	13 ± 0.33^{b}	17 ± 0.51^{ad}
Pb, mg/kg	7.5 ± 0.11^{a}	7.9 ± 0.077^{b}	2.9 ± 0.085^{c}	2.2 ± 0.12^{d}	1.7 ± 0.059^{e}	2.5 ± 0.045^{f}	3.7 ± 0.036^{g}	2.4 ± 0.092^{df}	1.0 ± 0.035^{h}	3.2 ± 0.018^{i}
Sr, mg/kg	3.5 ± 0.084^{a}	2.2 ± 0.22^{a}	19 ± 0.37^{b}	100 ± 3.1^{c}	61 ± 0.78d^{e}	120 ± 4.9^{f}	2.5 ± 0.25^{a}	64 ± 0.15^{d}	240 ± 5.5^{g}	56 ± 0.47^{e}
SP Fe (mg/kg)	3240 ± 326^{ab}	4170 ± 5170^{ab}	701 ± 503^{a}	1770 ± 1450^{ab}	6920 ± 2170^{b}	1330 ± 643^{ab}	3180 ± 640^{ab}	3880 ± 2290^{ab}	396 ± 493^{a}	445 ± 347^{a}
AO Fe (mg/kg)	5190 ± 203^{a}	2650 ± 225^{b}	50900 ± 8690^{c}	49600 ± 1210^{cg}	67300 ± 1660^{cd}	84600 ± 15700^{def}	86199 ± 15700^{ef}	97000 ± 3480^{f}	38600 ± 2580^{g}	60600 ± 6650^{c}
DC Fe (mg/kg)	7630 ± 378^{a}	6140 ± 1780^{a}	144000 ± 13900b^{d}	128000 ± 25200^{b}	153000 ± 7700^{bd}	190000 ± 4860^{ceg}	171000 ± 1330^{cde}	191000 ± 10700^{eg}	88700 ± 8520^{f}	213000 ± 8590^{g}
Extractable Phosphorus (mg/L)	1.73 ± 0.23^{a}	0.931 ± 0.04^{b}	ND	ND	ND	ND	ND	ND	ND	ND
Mineralogy	ND	ND	Goethite 67 % Quartz 30 % Calcite 3 %	Goethite 95 % Calcite 4 % Quartz 1 %	Amorphous 100%	Goethite 98 % Calcite 3 %	Goethite 84% Quartz 16 %	Goethite 97 % Calcite 3 %	Calcite 30 % Goethite 27 % Aragonite 23 % Brushite 12 % Magnetite 8 % Quartz 1 %	Goethite 97 % Quartz 2 % Calcite 1 %

ND = not determined; SP = Sodium pyrophosphate extractable Fe; AO = Ammonium oxalate extractable Fe; DC = Dithionite-citrate extractable Fe

2.2 Adsorption Experiment

An adsorption experiment was carried out in order to determine whether the ochres had the capacity to reduce C released from soil into solution. One gram of air-dried soil sieved to ≤2mm was shaken with 0, 0.05 and 0.1 g (dry mass ratios of 0, 0.05 and 0.1:1 ochre:soil) freeze-dried ochre in 20 mL of 0.01 M $CaCl_2$ at 20°C in an end over end shaker for 24 hours at 30 rpm (Houba *et al.*, 2000). Experiments were performed in triplicate. After 24 hours, the pH of each suspension was measured (using a Thermo Orion Star A111 pH meter calibrated with pH 4.01, 7 and 10.01 buffers). Subsequently, samples were centrifuged at 3000 rpm for 10 minutes; the supernatant was then filtered (Whatman 40 filter paper) into separate vials and frozen at -20°C until DOC, metal and phosphorus analyses. Elemental analysis of solutions was carried out by ICP-OES (Thermo iCAP 7000), except for analysis of DOC which was carried out using an elemental analyser (Elementar varioTOC select).

Method blanks were run for ICP-OES extractions to enable the detection limit of each element to be calculated (-0.00116 - 0.563 mg/L) (Table 2 and 3). The coefficient of variation determined from duplicate analysis of 10 % of solutions for elements above detection ranged from 7.62 - 88.1 % for the ICP-OES; precision for elements below detection is not reported (Appendix A: Table A1). The coefficient of variation determined from duplicate analysis of 10 % of solutions was 1.03 % for the elemental analyser (Gill, 1997). Accuracy was calculated using the following equation:

$$\text{accuracy} = \frac{100 \times \text{measured concentration}}{\text{reported value of a certified reference material}} \tag{1}$$

Based on the analysis of an in-house certified reference material of 0.5 mg/L, the accuracy of the ICP-OES analyses ranged from 86.7 - 157 % for metal analysis (Appendix A: Table A1); the CRM was not matrix-matched to the standards. Based on the analysis of an in-house certified reference material of 50 mg/L, the accuracy of the DOC analyses was 103 %.

2.3 Statistical Analysis

Statistical analyses were conducted using R Studio (Version 4.1.0). Data are presented as means ± standard deviation, n = 3. All statistical analyses were tested at a confidence level of 95 %. A Levene and Kolmogorov-Smirnov test were used to assess the equality of variance and normality of the data, respectively. Data sets for each soil comprised triplicate control samples together with triplicate treatments at 0.05:1 and 0.1:1 ochre:soil amendment levels of 8 different ochres. pH was converted to proton concentration for statistical analysis. All values below detection were retained.

The dataset was split between the two soils; a one-way ANOVA was performed on each dataset, with ochre treatment (control, 0.05:1 and 0.1:1) as the factor, subsequently employing a Dunnett's test to compare the differences in DOC, metals and phosphorus released into solution, and the pH of the solution between the control treatment and the ochre treatments.

These analyses were performed in order to confirm an effect of the ochre treatments on the variable stated above.

Following this, a three-way ANOVA was performed, with soil (Agricultural vs Woodland), ochre level (control, 0.05:1 and 0.1:1) and ochre type as factors (Wilcox, 1993). Control treatment data were removed from the dataset to ensure the ANOVA was balanced. If the three-way ANOVA identified significant effects, either a one-way ANOVA, with a subsequent Tukey test, or a Kruskal-Wallis multiple comparison test, adjusted with the Bonferroni method, with a subsequent Dunn test, was performed, depending on whether the data was normally distributed with equal variances. These tests determined whether there were significant differences in DOC, metals and phosphorus released into solution, and the pH of the solution between soils, treatments and ochres. Having identified significant interactions with the three-way ANOVA, differences in DOC, metals, phosphorus and pH of the solutions of ochre treatments of the same concentration treatment between soils, and of the different ochre treatments within the same soils were assessed with a two-way ANOVA, with factors of soil and ochre, and treatment and ochre, respectively, followed by a Tukey test. An independent samples t-test, with soil as the factor, compared differences in DOC, metals and phosphorus in solution, and pH, between the Agricultural and Woodland Soils in the control treatments.

To support the results and allow for the control data to be incorporated into the analysis, two-way ANOVAs, with subsequent Tukey tests, were used to see if the differences in percentage reduction of DOC between soil and ochre types, and treatment and ochre types, were significant. Percentage reduction was determined by taking an average of the control data for each soil and using this value to calculate the percentage reduction for each of the ochre treatments relative to the control. A single sample t-test was also performed to determine whether the mean percentage reduction in DOC due to the ochre treatments was significantly greater than 0.

3 Results

3.1 Soils and Ochres

As seen in Table 1 (Section 2.1), the soils and ochres had varying physical and chemical properties. The Woodland Soil has a greater organic content (Total C 5.63 %) compared to the Agricultural Soil (Total C 3.69 %) ($p \leq 0.05$: ANOVA and Tukey test). The Fe content of all of the ochres exists primarily as finely divided crystalline Fe solid phases, followed by non-crystalline inorganic forms of Fe and organic complexed Fe, as told by the Fe extraction data ($p \leq 0.05$: ANOVA and Tukey test). These results of the Fe extractions are consistent with the mineralogy of the ochres as the majority of the ochres are largely made up of goethite, a finely divided crystalline Fe solid phase.

3.2 Dissolved Organic Carbon

DOC results of the adsorption experiment for the Agricultural and Woodland Soils are shown in Figure 1 and Figure 2, respectively. For both soils, the ochre treatments significantly reduced the C released from Agricultural and Woodland Soils into solution ($p \leq 0.05$), with the exceptions of the 0.05 treatment of Blenkinsop 2 in both soils, both treatments (0.05 and 0.1) of Lynemouth in the Agricultural Soil, the 0.1 treatment of Saltburn in both soils, and the 0.1 treatment of Blenkinsop 2 in the Agricultural Soil (Figures 1 and 2). The t-test between controls showed that the Agricultural Soil released significantly less C into solution than the Woodland Soil ($p > 0.05$). Comparing all of the 0.05 treatments with the 0.1 treatments, the two were found not to be significantly different ($p > 0.05$). No difference was found between the Agricultural and Woodland Soils when comparing the low and high treatments of each soil individually ($p > 0.05$).

These results are supported by the DOC percentage reduction data; there is no significant difference in percentage reduction of C release between the 0.05 and 0.1 treatments when analysing the soils individually. However, within the Agricultural Soil, the two-way ANOVA revealed the 0.1 concentration of Whitworth to have a significantly greater DOC percentage reduction than the Blenkinsop 2 ochre at the same treatment level ($p \leq 0.05$). When comparing the two soils together, the 0.1 Lynemouth treatment in the Woodland Soil had a significantly greater DOC percentage reduction than the same ochre treatment in the Agricultural Soil ($p \leq 0.05$). The single sample t-tests revealed that the average percentage reduction for Agricultural and Woodland Soils were significantly greater than 0 ($p \leq 0.05$).

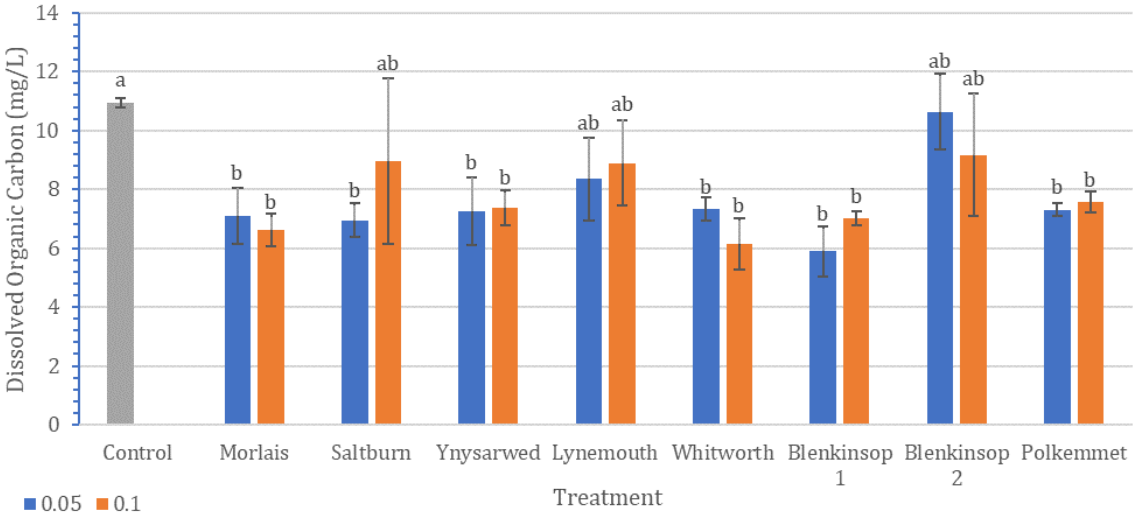

Figure 1: Mean organic carbon released into solution (mg/L) from the Agricultural Soil for control, 0.05 and 0.1 treatments. 0.05 and 0.1 represent the treatments of dry mass ratios 0.05:1 and 0.1:1 ochre:soil, respectively. Error bars represent standard deviation, n=3. Different letters represent a significant difference in DOC concentration between treatments and ochre types ($p \leq 0.05$: ANOVA and Dunnett's test).

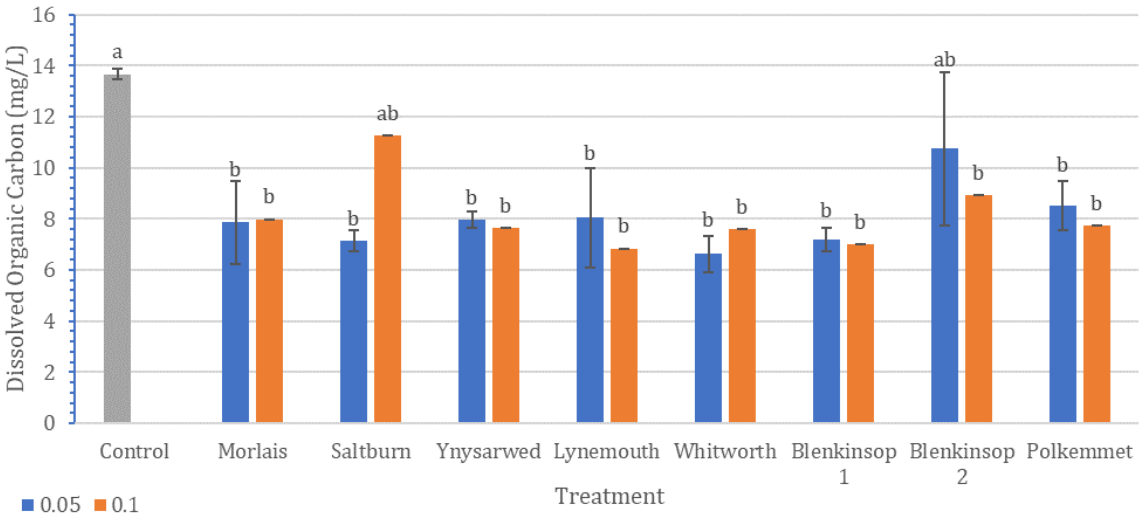

Figure 2: Mean organic carbon released into solution (mg/L) from the Woodland Soil for each control, 0.05 and 0.1 treatments. 0.05 and 0.1 represent the treatments of dry mass ratios 0.05:1 and 0.1:1 ochre:soil, respectively. Error bars represent standard deviation, n=3. Different letters represent a significant difference in DOC concentration between treatments and ochre types ($p \leq 0.05$: ANOVA and Dunnett's test).

3.3 Metals

Metal release into solution from the controls and ochre treated soils was not significantly different for: Zn, Pb, Cd and Fe in the Agricultural Soils ($p > 0.05$). In Agricultural and Woodland Soils, all releases of Cu, Cr, Ni and As were below their detection limits (0.330, 0.00482, 0.00870 and 0.0515 mg/L, respectively). For the Woodland Soil, all releases of Tl were also below detection (0.00380 mg/L). For both soils, in the majority of ochre treatments, releases of Pb, Co, Li, Tl and Ni were below detection (Table 2 and 3).

Table 2: Mean concentration of metals released into solution (mg/L) from the Agricultural Soil for each treatment ± standard deviation, n=3. 0.05 and 0.1 represent the treatments of dry mass ratios 0.05:1 and 0.1:1 ochre:soil, respectively. Different letters represent a significant difference in metal concentration between treatments and ochre types ($p \leq 0.05$). BDL represents treatment means which were below the detection limit. The Na data for Saltburn and Lynemouth are highlighted in bold and are discussed in Section 4.3.

	Control	Morlais		Saltburn		Ynysarwed		Lynemouth		Whitworth		Blenkinsop 1		Blenkinsop 2		Polkemmet		Detection Limit
		0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	
Zn	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	**0.022 ± 0.026^{a}**	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	**0.013 ± 0.0025^{a}**	**0.021 ± 0.0030^{a}**	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	**0.0184**
Pb	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	0.0031 ± 0.0065^{a}	BDL^{a}	0.00320
Al	**0.052 ± 0.034^{a}**	**BDL^{b}**	**0.017 ± 0.015^{b}**	**BDL^{b}**	**0.025 ± 0.018^{b}**	**BDL^{b}**	**0.016 ± 0.0041^{b}**	**0.012 ± 0.0029^{b}**	**0.015 ± 0.0074^{b}**	**0.011 ± 0.0055^{b}**	**0.016 ± 0.0036^{b}**	**BDL^{b}**	**0.012 ± 0.0034^{b}**	**BDL^{b}**	**0.012 ± 0.0041^{b}**	**BDL^{b}**	**0.014 ± 0.0031^{b}**	**0.0137**
B	0.082 ± 0.059^{a}	0.067 ± 0.026^{abc}	0.056 ± 0.035^{abc}	0.056 ± 0.0074^{abc}	0.054 ± 0.0065^{abc}	0.022 ± 0.0037^{bc}	0.023 ± 0.0050^{bc}	0.084 ± 0.0066^{abc}	0.12 ± 0.021^{ab}	0.038 ± 0.0024^{abc}	0.042 ± 0.0012^{abc}	0.036 ± 0.0055^{abc}	0.024 ± 0.0036^{bc}	0.017 ± 0.013^{bc}	0.018 ± 0.0016^{c}	0.020 ± 0.0040^{bc}	0.027 ± 0.0017^{bc}	0.00942
Ba	**0.72 ± 0.035^{a}**	**0.59 ± 0.052^{ab}**	**0.54 ± 0.097^{ab}**	**0.49 ± 0.038^{ab}**	**0.38 ± 0.034^{b}**	**0.40 ± 0.047^{b}**	**0.36 ± 0.060^{b}**	**0.34 ± 0.046^{b}**	**0.31 ± 0.016^{b}**	**0.49 ± 0.017^{ab}**	**0.40 ± 0.010^{b}**	**0.34 ± 0.0068^{b}**	**0.27 ± 0.015^{b}**	**0.28 ± 0.24^{b}**	**0.36 ± 0.044^{b}**	**0.44 ± 0.38^{ab}**	**0.58 ± 0.075^{a}**	**0.00960**
Cd	0.0014 ± 0.00012^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	0.0020 ± 0.00010^{a}	BDL^{a}	BDL^{a}	0.0011 ± 0.000069^{a}	BDL^{a}	0.0043 ± 0.0052^{a}	BDL^{a}	0.00134
Ag	**0.011 ± 0.0032^{a}**	**0.012 ± 0.0015^{ab}**	**0.037 ± 0.013^{ab}**	**0.0091 ± 0.00086^{ab}**	**0.038 ± 0.030^{ab}**	**0.0081 ± 0.0018^{ab}**	**0.074 ± 0.028^{b}**	**0.0094 ± 0.0038^{ab}**	**0.043 ± 0.028^{ab}**	**0.014 ± 0.0051^{ab}**	**0.11 ± 0.041^{b}**	**0.0047 ± 0.0027^{ab}**	**0.049 ± 0.042^{ab}**	**0.0068 ± 0.0079^{ab}**	**0.024 ± 0.027^{ab}**	**0.026 ± 0.025^{ab}**	**0.0045 ± 0.0020^{ab}**	**0.0249**
Co	BDL^{a}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	0.00029 ± 0.00036^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	0.0024 ± 0.0052^{b}	BDL^{ab}	-0.000176
Fe	**0.020 ± 0.011^{a}**	**BDL^{a}**	**0.017 ± 0.018^{a}**	**BDL^{a}**	**0.022 ± 0.012^{a}**	**BDL^{a}**	**BDL^{a}**	**0.014 ± 0.012^{a}**	**BDL^{a}**	**BDL^{a}**	**BDL^{a}**	**BDL^{a}**	**BDL^{a}**	**BDL^{a}**	**BDL^{a}**	**BDL^{a}**	**BDL^{a}**	**0.0225**
K	9.2 ± 2^{a}	8.4 ± 1.9^{ab}	8.4 ± 1.3^{ab}	7.3 ± 0.76^{ab}	6.8 ± 0.74^{ab}	6.1 ± 1.3^{ab}	6.0 ± 1.6^{ab}	6.3 ± 0.41^{ab}	6.5 ± 1.0^{ab}	6.5 ± 0.43^{ab}	6.9 ± 0.17^{ab}	7.7 ± 1.2^{ab}	5.9 ± 0.96^{ab}	4.2 ± 3.4^{b}	5.4 ± 1.1^{ab}	4.4 ± 3.6^{b}	5.5 ± 0.25^{ab}	0.563
Li	**-0.000081 ± 0.0025^{a}**	**0.0019 ± 0.0028^{ab}**	**BDL^{ab}**	**0.0022 ± 0.0048^{ab}**	**BDL^{b}**	**0.0023 ± 0.0004^{ab}**	**BDL^{b}**	**0.0021 ± 0.00098^{ab}**	**BDL^{b}**	**0.0022 ± 0.0022^{ab}**	**BDL^{b}**	**0.0047 ± 0.0014^{ab}**	**BDL^{b}**	**-0.00015 ± 0.0041^{ab}**	**BDL^{b}**	**-0.00063 ± 0.0029^{ab}**	**BDL^{b}**	**-0.00116**
Mg	8.3 ± 0.33^{a}	11 ± 2.5^{abc}	12 ± 1.8^{abc}	11 ± 1.2^{abc}	10 ± 2.4^{abc}	12 ± 4.2^{abc}	12 ± 3.3^{abc}	11 ± 1.1^{abc}	12 ± 2.4^{abc}	9.1 ± 0.85^{abc}	9.3 ± 0.087^{abc}	14 ± 3.5^{abc}	10 ± 1.5^{abc}	12 ± 11^{abc}	25 ± 4.7^{c}	5.2 ± 4.5^{ab}	7.6 ± 0.20^{ab}	0.0338
Mn	**0.75 ± 0.14^{a}**	**0.16 ± 0.028^{ab}**	**0.45 ± 0.13^{ab}**	**0.15 ± 0.073^{ab}**	**0.15 ± 0.024^{ab}**	**0.27 ± 0.066^{ab}**	**0.25 ± 0.066^{ab}**	**0.13 ± 0.018^{ab}**	**0.072 ± 0.022^{b}**	**2.4 ± 0.059^{b}**	**3.6 ± 0.16^{b}**	**0.045 ± 0.023^{b}**	**0.18 ± 0.038^{ab}**	**0.17 ± 0.15^{ab}**	**0.27 ± 0.047^{ab}**	**1.2 ± 1.0^{ab}**	**0.097 ± 0.0061^{b}**	**0.000493**
Tl	BDL^{a}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	0.0048 ± 0.00046^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	BDL^{ab}	0.0058 ± 0.0056^{b}	BDL^{ab}	0.00401
Na	**2.7 ± 2.0^{a}**	**1.9 ± 0.24^{abcd}**	**2.7 ± 0.24^{abcd}**	**6.8 ± 0.74^{cd}**	**10 ± 1.7^{cd}**	**1.3 ± 0.19^{abcd}**	**1.8 ± 0.53^{abcd}**	**14 ± 1.8^{d}**	**14 ± 2.8^{d}**	**1.1 ± 0.23^{abcd}**	**1.0 ± 0.089^{abcd}**	**2.4 ± 0.36^{abcd}**	**2.7 ± 0.46^{abcd}**	**1.6 ± 1.4^{abcd}**	**3.1 ± 0.63^{abcd}**	**0.67 ± 0.56^{abc}**	**1.2 ± 0.069^{abcd}**	**0.0955**
Ca	BDL^{a}	9.7 ± 2.2^{a}	11 ± 1.6^{a}	9.6 ± 1.1^{a}	8.2 ± 1.7^{a}	9.2 ± 3.3^{a}	8.0 ± 2.0^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	12 ± 2.8^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	BDL^{a}	9.31

Table 3: Mean concentration of metals released into solution (mg/L) from the Woodland Soil for each treatment ± standard deviation, n=3. 0.5 and 0.1 represent the treatments of dry mass ratios 0.05:1 and 0.1:1 ochre:soil, respectively. Different letters represent a significant difference in metal concentration between treatments and ochre types ($p \leq 0.05$). BDL represents treatment means which were below the detection limit. The Na data for Saltburn and Lynemouth are highlighted in bold and are discussed in Section 4.3.

	Control	Morlais		Saltburn		Ynysarwed		Lynemouth		Whitworth		Blenkinsop 1		Blenkinsop 2		Polkemmet		Detection Limit
		0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	0.05	0.1	
Zn	**0.020 ± 0.0078^{a}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**0.017 ± 0.0013^{ab}**	**BDL^{ab}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**0.0184**
Pb	0.0021 ± 0.0026^{a}	BDL^{ab}	BDL^{b}	BDL^{b}	BDL^{ab}	BDL^{ab}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	0.00320
Al	**0.034 ± 0.025^{a}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**0.0093 ± 0.0059^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**0.0086 ± 0.0056^{b}**	**0.010 ± 0.0041^{b}**	**BDL^{b}**	**0.012 ± 0.0037^{b}**	**0.010 ± 0.0059^{b}**	**0.0137**
B	0.037 ± 0.0042^{a}	0.059 ± 0.047^{abc}	0.050 ± 0.029^{abc}	0.045 ± 0.0086^{abc}	0.049 ± 0.0^{abc}	0.029 ± 0.0064^{abc}	0.031 ± 0.0023^{abc}	0.089 ± 0.0062^{bc}	0.12 ± 0.016^{b}	0.062 ± 0.009^{abc}	0.046 ± 0.0028^{abc}	0.040 ± 0.0018^{abc}	0.032 ± 0.0033^{abc}	0.030 ± 0.0011^{abc}	0.027 ± 0.0037^{ac}	0.035 ± 0.0083^{abc}	0.040 ± 0.0039^{abc}	0.00942
Ba	**0.62 ± 0.041^{a}**	**0.49 ± 0.18^{abc}**	**0.40 ± 0.064^{bc}**	**0.46 ± 0.055^{bc}**	**0.29 ± 0.0^{bc}**	**0.30 ± 0.016^{bc}**	**0.18 ± 0.012^{b}**	**0.29 ± 0.023^{bc}**	**0.22 ± 0.030^{bc}**	**0.60 ± 0.027^{ac}**	**0.30 ± 0.026^{bc}**	**0.41 ± 0.055^{bc}**	**0.27 ± 0.014^{bc}**	**0.37 ± 0.035^{bc}**	**0.27 ± 0.046^{bc}**	**0.50 ± 0.020^{abc}**	**0.37 ± 0.085^{bc}**	**0.00960**
Cd	0.0014 ± 0.000095^{a}	BDL^{b}	0.0012 ± 0.00014^{ab}	BDL^{b}	BDL^{ab}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{ab}	0.0016 ± 0.000098^{ab}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	0.0012 ± 0.00013^{ab}	0.0012 ± 0.00018^{ab}	0.00134
Ag	**BDL^{a}**	**BDL^{ab}**	**0.026 ± 0.0038^{ab}**	**BDL^{ab}**	**0.025 ± 0.0^{ab}**	**0.049 ± 0.036^{ab}**	**0.021 ± 0.0034^{ab}**	**0.048 ± 0.024^{ab}**	**BDL^{ab}**	**0.083 ± 0.061^{b}**	**BDL^{ab}**	**0.028 ± 0.0099^{ab}**	**BDL^{ab}**	**0.051 ± 0.031^{ab}**	**BDL^{ab}**	**0.059 ± 0.029^{ab}**	**BDL^{ab}**	**0.0249**
Co	-0.00043 ± 0.00025^{a}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{ab}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	-0.00033 ± 0.00014^{ab}	BDL^{ab}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	BDL^{b}	-0.000176
Fe	**0.026 ± 0.011^{a}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{ab}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**0.014 ± 0.0081^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**0.0225**
K	4.1 ± 0.60^{a}	4.3 ± 2.0^{ab}	4.5 ± 0.34^{ab}	4.4 ± 0.87^{ab}	4.0 ± 0.0^{ab}	4.6 ± 1.5^{ab}	4.3 ± 0.22^{ab}	4.3 ± 0.42^{ab}	4.9 ± 0.77^{ab}	4.5 ± 0.12^{ab}	4.4 ± 0.55^{ab}	4.4 ± 0.27^{ab}	4.3 ± 0.30^{ab}	3.9 ± 0.62^{ab}	4.1 ± 0.66^{ab}	4.7 ± 0.56^{ab}	6.7 ± 1.2^{b}	0.563
Li	**0.0031 ± 0.0011^{a}**	**0.00028 ± 0.0058^{ab}**	**BDL^{b}**	**0.0018 ± 0.003^{ab}**	**BDL^{ab}**	**0.0051 ± 0.00064^{ab}**	**BDL^{b}**	**0.0041 ± 0.0030^{ab}**	**BDL^{ab}**	**0.0042 ± 0.00011^{ab}**	**BDL^{b}**	**0.0038 ± 0.00062^{ab}**	**BDL^{b}**	**-0.0033 ± 0.0052^{ab}**	**BDL^{b}**	**BDL^{b}**	**BDL^{b}**	**-0.00116**
Mg	12 ± 0.90^{a}	14 ± 5.9^{ab}	14 ± 0.26^{ab}	16 ± 4.4^{ab}	14 ± 0.0^{ab}	20 ± 7.2^{ab}	20 ± 2.9^{ab}	16 ± 1.1^{ab}	20 ± 3.0^{ab}	18 ± 2.1^{ab}	19 ± 2.2^{ab}	18 ± 1.5^{ab}	18 ± 1.7^{ab}	24 ± 3.6^{b}	39 ± 4.4^{b}	17 ± 1.8^{ab}	20 ± 2.5^{ab}	0.0338
Mn	**0.68 ± 0.056^{a}**	**0.36 ± 0.15^{bc}**	**0.17 ± 0.021^{bc}**	**0.085 ± 0.017^{bc}**	**0.031 ± 0.0^{bc}**	**0.21 ± 0.057^{bc}**	**0.097 ± 0.017^{bc}**	**0.050 ± 0.0091^{bc}**	**0.026 ± 0.0066^{b}**	**1.1 ± 0.13^{c}**	**1.0 ± 0.088^{c}**	**0.21 ± 0.025^{bc}**	**0.096 ± 0.025^{bc}**	**0.32 ± 0.039^{bc}**	**0.14 ± 0.032^{bc}**	**0.29 ± 0.058^{bc}**	**0.098 ± 0.015^{bc}**	**0.000493**
Na	1.8 ± 0.19^{a}	2.8 ± 1.3^{ab}	3.0 ± 0.2^{ab}	7.4 ± 2.0^{b}	10 ± 0.0^{b}	2.8 ± 0.77^{ab}	3.1 ± 0.41^{ab}	7.3 ± 2.9^{ab}	14 ± 6.3^{b}	2.5 ± 0.23^{ab}	2.1 ± 0.29^{ab}	3.5 ± 0.15^{ab}	4.3 ± 0.45^{ab}	4.0 ± 0.35^{ab}	4.9 ± 0.87^{ab}	2.6 ± 0.48^{ab}	4.5 ± 1.1^{ab}	0.00401
Ca	**BDL^{a}**	**BDL^{a}**	**BDL^{a}**	**8.9 ± 2.2^{a}**	**BDL^{a}**	**8.8 ± 2.5^{a}**	**BDL^{a}**	**BDL^{a}**	**8.8 ± 1.1^{a}**	**9.2 ± 1.2^{a}**	**8.9 ± 0.93^{a}**	**9.1 ± 1.1^{a}**	**9.1 ± 0.70^{a}**	**BDL^{a}**	**BDL^{a}**	**8.4 ± 1.3^{a}**	**BDL^{a}**	**0.0955**

Na release from the Saltburn and Lynemouth treated Agricultural Soil was significantly greater at both treatment levels than it was from the Agricultural control ($p \leq 0.05$); Na release from the Saltburn treated Woodland Soil at both treatment levels, and the Lynemouth treated Woodland Soil for the 0.1 treatment was significantly greater than it was from the Woodland control ($p \leq 0.05$). Generally, ochre treated soils reduced the release of Al, Ba, Li and Zn into solution, relative to the controls (Table 2 and 3). For the Woodland soil, ochre amendments generally reduced Zn, Pb, Co and Fe release into solution, relative to the Woodland Soil control (Table 3).

The 0.05 Lynemouth ochre treatment, in the Agricultural Soil, released significantly more Na into solution than the Polkemmet ochre of the same treatment concentration ($p \leq 0.05$). The 0.1 Lynemouth ochre treatment also had significantly lower Mn release from the Woodland Soil than the high concentration of the Whitworth ochre ($p \leq 0.05$). The Blenkinsop 2 treated soils generally released more B and Mg into solution than some of the other ochre treatments (Table 2 and 3).

The t-test comparing the controls of both soils revealed that the Agricultural Soil released significantly more Ba and K, with the Woodland Soil releasing significantly more Mg ($p \leq 0.05$). When comparing the same ochre level treatment across both soils, Ba, Cd and K release was significantly greater in Morlais, Whitworth and Polkemmet treated Agricultural Soils than Woodland Soils for the 0.1 treatment level ($p \leq 0.05$). The 0.05 Lynemouth ochre treatment had significantly lower Na concentrations released into solution in the Woodland Soil than the Agricultural Soil ($p \leq 0.05$). At the 0.1 treatment level, there was a significantly greater Mn release in Lynemouth, Blenkinsop 1, Blenkinsop 2 and Polkemmet treated Woodland Soils than that in the Agricultural Soil ($p \leq 0.05$). It was found that for both treatment levels, the Mn release from the Whitworth treated Agricultural Soil was significantly greater than that in the Woodland Soil; for the 0.1 treatment, the Mn release from the Morlais treated Agricultural Soil was also significantly greater than that in the treated Woodland Soil ($p \leq 0.05$).

3.4 Phosphorus

In both Agricultural and Woodland Soils, all ochres significantly reduced the P released into solution, compared to the control treatments ($p \leq 0.05$) (Figures 3 and 4). No differences were found when comparing P release between different ochres within the same soils and treatment levels, nor were there significant differences between the 0.05 and 0.1 treatments when all the data were considered ($p > 0.05$). No difference was found between the Agricultural and Woodland Soils when comparing individual treatments ($p > 0.05$).

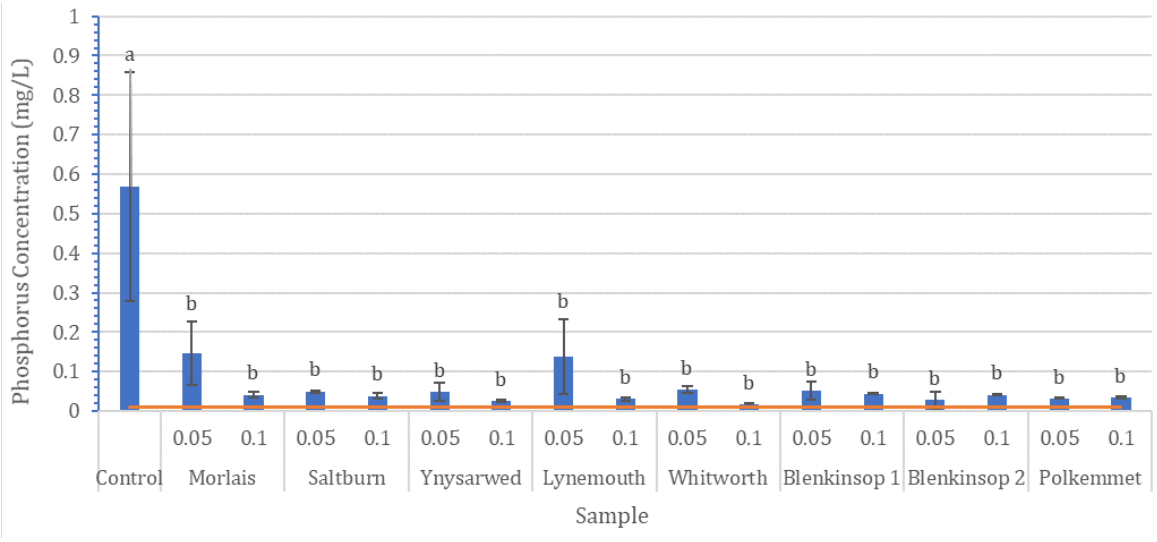

Figure 3: Mean concentration of phosphorus released into solution (mg/L) from the Agricultural Soil for each treatment. Error bars represent standard deviation, n=3. 0.05 and 0.1 represent the treatments of dry mass ratios 0.05:1 and 0.1:1 ochre:soil, respectively. The red line represents the detection limit of phosphorus on the ICP-OES; all values lie above the detection limit (0.00904 mg/L).

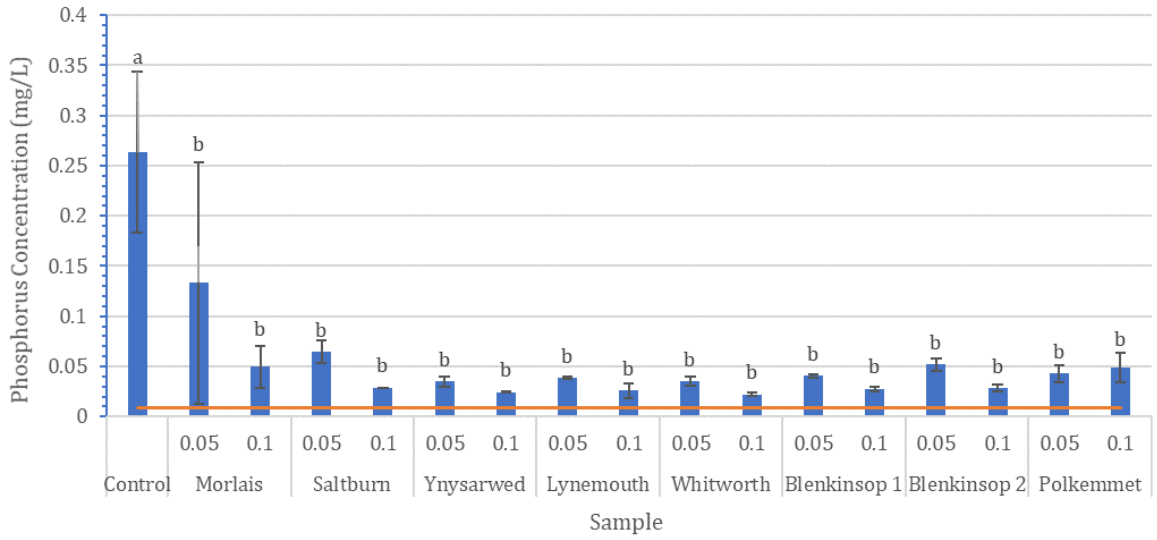

Figure 4: Mean concentration of phosphorus released into solution (mg/L) from the Woodland Soil for each treatment. Error bars represent standard deviation, n=3. 0.05 and 0.1 represent the treatments of dry mass ratios 0.05:1 and 0.1:1 ochre:soil, respectively. The red line represents the detection limit of phosphorus on the ICP-OES; all values lie above the detection limit (0.00904 mg/L).

3.5 pH

The ochre treatments had a significantly higher pH than the controls in Agricultural and Woodland Soils ($p \leq 0.05$), with the exception of the Whitworth treatments in both soils (Table

4). The Whitworth treated Woodland Soil had a significantly higher pH in both treatments than the Whitworth treated Agricultural Soil; the Polkemmet ochre also followed this trend in the 0.05 treatment ($p \leq 0.05$). The Blenkinsop 2 treated Agricultural Soil had a significantly higher pH than the Whitworth treated Agricultural Soil for the 0.1 ochre treatment ($p \leq 0.05$) (Table 4). Comparing all of the 0.05 treatments with the 0.1 treatments, the two were found not to be significantly different to each other ($p > 0.05$). No difference was found between the Agricultural and Woodland Soils when comparing the individual treatments ($p > 0.05$).

Table 4: Mean pH of treatment for Agricultural and Woodland soils ± standard deviation, n=3. 0.05 and 0.1 represent the treatments of dry mass ratios 0.05:1 and 0.1:1 ochre:soil, respectively. Different letters represent a significant difference in pH between treatments and ochre types within the same soil groups ($p \leq 0.05$).

Sample		Agricultural Soil	Woodland Soil
Control		5.67 ± 0.78^{a}	6.09 ± 0.31^{a}
Morlais	**0.05**	6.71 ± 0.11^{bc}	6.73 ± 0.01^{b}
	0.1	7.01 ± 0.11^{bc}	6.98 ± 0.01^{b}
Saltburn	**0.05**	7.16 ± 0.03^{bc}	7.33 ± 0.02^{b}
	0.1	7.21 ± 0.02^{bc}	7.41 ± 0.02^{b}
Ynysarwed	**0.05**	7.28 ± 0.04^{bc}	7.46 ± 0.02^{b}
	0.1	7.33 ± 0.02^{bc}	7.48 ± 0.04^{b}
Lynemouth	**0.05**	7.09 ± 0.07^{bc}	7.30 ± 0.06^{b}
	0.1	7.24 ± 0.03^{bc}	7.38 ± 0.05^{b}
Whitworth	**0.05**	5.91 ± 0.09^{abc}	6.27 ± 0.05^{ab}
	0.1	5.32 ± 0.07^{ab}	5.98 ± 0.09^{ab}
Blenkinsop 1	**0.05**	6.80 ± 0.04^{bc}	6.86 ± 0.03^{b}
	0.1	6.95 ± 0.06^{bc}	7.15 ± 0.03^{b}
Blenkinsop 2	**0.05**	7.34 ± 0.05^{bc}	7.50 ± 0.04^{b}
	0.1	7.48 ± 0.04^{c}	7.34 ± 0.30^{b}
Polkemmet	**0.05**	6.43 ± 0.13^{bc}	6.76 ± 0.08^{b}
	0.1	6.94 ± 0.04^{bc}	7.40 ± 0.21^{b}

4 Discussion

The DOC results of the adsorption experiment largely support the hypothesis that the addition of Fe oxides to soils will enhance organic C sorption, reducing soil C availability and lability, decreasing the release of C into solution (Figures 1 and 2, Section 3.2). Differences between ochres appear to be related to their physical (surface area, mineralogy) and chemical (pH, Na content) properties which will be discussed in turn.

4.1 Ochre Surface Area and Mineralogy

The Blenkinsop 2 ochre has the smallest BET surface area (38.8 m^2/g), nearly half of the next smallest surface area (Morlais: 71.3 m^2/g) and substantially lower than that of the other ochres (Table 1, Section 2.1). This is a likely reason why the Blenkinsop 2 ochre treatment does not

result in a significant decrease of C released into solution in the majority of treated Agricultural and Woodland Soils. The low surface area causes a low number of adsorption sites available for OM.

Furthermore, unlike the other ochres which have high goethite contents ranging from 67.27 - 97.50 %, as mentioned in Section 3.1, the Blenkinsop 2 ochre has little goethite in its mineralogy (26.7 %) (Table 1, Section 2.1). Goethite has a high adsorption capacity for SOC, decreasing the release of C into solution, as mentioned in Chapter 2 (Ohno *et al.*, 2007; Liu *et al.*, 2014). The mineralogy of the Blenkinsop 2 ochre is dominated by calcite (Table 1, Section 2.1): there has been relatively little direct research on the adsorption of SOC by different forms of calcite in soils (Rowley *et al.*, 2018), however Randtke (1988) showed that calcium carbonate is precipitated as a dense crystalline solid with very low surface area, resulting in minimal sites for natural OM adsorption. This analysis is consistent with the concentration of DC extractable Fe in the Blenkinsop 2 ochre, which is significantly lower than that of the other ochres (Table 1, Section 2.1), meaning the Blenkinsop 2 ochre has fewer finely divided crystalline Fe solid phases, such as goethite, as estimated by the methods in Loeppert and Inskeep (1996) (explored further in Chapter 4, Section 4.3).

4.2 Solution pH

The solutions for the ochre treated Agricultural and Woodland Soils generally had a significantly higher pH than the control soils (Table 4, Section 3.5). Of the different ochres, Blenkinsop 2 has the greatest pH (9.36 ± 0.05) (Table 1, Section 2.1), this results in the Blenkinsop 2 treated soil solutions having a significantly higher pH than the control treatments in both soils, and the high concentration of the Whitworth treated Agricultural Soil. The relatively low pH of the Whitworth ochre (4.45 ± 0.05) resulted in the Whitworth treated soils showing no significant difference when compared to the control soil treatments. All of the ochres, excluding Whitworth ochre, contain calcite, with Blenkinsop 2 also containing aragonite (Table 1, Section 2.1) – these $CaCO_3$ minerals dissolve in water, consuming protons and increasing the pH of the solution. The low pH of the Whitworth treated soils is consistent with the absence of calcium carbonate.

Zhang *et al.* (2020) found that a higher soil pH and/or lower levels of CEC might cause greater levels of DOC in solution due to strong desorption of SOM (Oste *et al.*, 2002). Some studies suggest that a high soil pH, and hence, high levels of OH^-, can increase desorption of SOC (thus more DOC in soil water) due to increased negative charges on SOM (Oste *et al.*, 2002; Whittinghill and Hobbie, 2011). The pH trends seen in the ochres support this: Blenkinsop 2 has no significant effect on C released into solution in the majority of treated Agricultural and Woodland Soils, whereas the Whitworth treated Agricultural and Woodland Soils had significant decreases of C released into solution in both treatments (Figures 1 and 2). However, this does not explain the insignificant differences in C released into solution in the Saltburn and Lynemouth treated soils.

4.3 Exchangeable Sodium

The majority of the Saltburn and Lynemouth treated soils released significantly higher concentrations of Na into solution than the control treatments (Tables 2 and 3, Section 3.3), corresponding to the high Na contents of these ochres (344 ± 159 to 846 ± 41.7 mg/kg for Saltburn and Lynemouth, respectively) (Table 1, Section 2.1). The effect of these ochres on C release into solution is consistent with the known impacts of Na on soils. Both treatments (0.05 and 0.1) of Lynemouth in the Agricultural soil, and the high concentration treatments (0.1) of Saltburn in both soils did not significantly reduce C released into solution, compared to the control treatments of each soil (Figures 1 and 2, Section 3.2).

Exchangeable Na has adverse effects on soil dispersion: when Na-induced soil dispersion causes loss of soil structure, aggregate stability is reduced (Gupta *et al.*, 1984; Suarez *et al.*, 1984; Levy and Torrento, 1995). Exchangeable Na^+ ions promote dispersion for two reasons: (1) because of their single charge and large hydrated size, they have a weak attraction to soil colloids, forming loose outer-sphere complexes around the colloids. (2) Compared to divalent cations, twice as many monovalent Na^+ ions are needed to provide enough positive charges to counter the negative charges on a clay surface, producing well-spread Na-saturated colloids. This limits the forces of cohesion between colloids, with the poorly balanced electronegativity of each colloidal surface repelling other electronegative colloids, dispersing the soil (Weil and Brady, 2016). In the presence of $CaCO_3$, excess exchangeable Na results in high pH of sodic soils (Gupta *et al.*, 1984; Suarez *et al.*, 1984); the high pH levels in sodic soils may cause SOM to disperse and dissolve, as explained in Section 4.2. Gupta *et al.* (1984) found that additions of OM at a sodium adsorption ratio greater than 15 - 20 also increased soil dispersion. Increased pH and Na in solution leads to increased desorption of SOC and increased DOC in soil solution (Zhang et a.., 2021; Setia *et al.*, 2013).

4.4 Metals

Given the known presence of potential toxic metals in ochre deposits, there is a concern that their addition to soils may lead to harmful increases in metal concentrations. In soils, it is key to look at how available metals are for uptake by soil organisms (Smith and Huyck, 1999; Kabata-Pendias, 2004). Within the adsorption experiments, a good proxy for this is the release of metals into solution. The comparison of these metal concentrations in solution to water quality standards could determine the level of risk and potential for harm. However, this data would act as a worst-case scenario as in the field, it is unlikely that all of the available metals will be released into the soil due to sorption-contact constraints. In this study, the majority of metals released into solution were either below detection or no greater than those released by the controls (Table 2 and 3). These results are largely supported by those of the aqua regia digestions measuring pseudo total metals in the ochres and soils, of which concentrations of Cd, Cr, Cu and Tl were below detection and concentrations of Mg, K, Al, Ba, Li and Pb were greater in soils than in the ochres (Table 1, Section 2.1). Cd, Ni and Pb, some of the metals

which were below the detection limit of the ICP-OES in the adsorption experiments, do not breach limits set by the Environmental Quality Standards Directive for these priority substances (2008/105/EC) (European Commission, 2013). Furthermore, concentrations of As, Cu, Fe, Mn and Zn largely lie below the standards for specific pollutants set by the Water Framework Directive (Water Framework Directive, 2015). However, for some ochre treated Agricultural and Woodland Soils, the concentration of Mn release from soil into solution breaches this limit (0.123 mg/L) (Tables 2 and 3, Section 3.3). The concentration of Mn release into solution in the Agricultural and Woodland controls also breached this standard; however, only the Whitworth treated soils at both treatment levels produces concentrations with a cause for concern as these solution Mn concentrations are significantly higher (2.38 ± 0.0586 mg/L, 3.64 ± 0.157 mg/L, 1.05 ± 0.134 mg/L and 1.00 ± 0.0881 mg/L for the 0.05 and 0.1 Whitworth treated Agricultural Soil and 0.05 and 0.1 Whitworth treated Woodland Soil, respectively) than the respective controls (0.747 ± 0.135 mg/L and 0.678 ± 0.0555 mg/L for the Agricultural and Woodland control) (Tables 2 and 3, Section 3.3). However, as mentioned, these values are most likely an overestimation of the concentration of Mn that could be released into soil solution in the field. The low solubility of Mn at a neutral and alkaline pH prevents excessive uptake by plants, with Mn toxicity associated with acid soils (Sims, 1986); the low pH of the Whitworth treated soils merits constant monitoring in the field.

4.5 Conclusion

The addition of the ochre to soils enhances organic C sorption, reducing soil C availability and lability, decreasing the release of C into solution. Ochres with a high goethite content and a relatively low - neutral pH performed the best in significantly decreasing the concentration of C released from the soils into solution. This is consistent with the knowledge of the behaviour of Fe and OM due to sorption: goethite has a high adsorption capacity for SOC and a higher soil pH, signifies higher levels of OH^- and an increase in desorption of SOC due to increased negative charges on SOM. However, ochres generally contained $CaCO_3$ which caused an increase in pH, potentially causing desorption of SOM and a release of C into solution. Furthermore, the ochres with a high Na content, released more Na into solution, performing less well due to their adverse effects on organic C sorption and storage, resultant from an increase in soil dispersion and a decrease in aggregate stability.

Chapter 4: An Investigation into the Effect of Ochre Amendments on Carbon Storage, Soil Aggregate Formation and Plant Growth

1 Introduction

1.1 Carbon Storage Potential

As stated in Chapter 3, Section 1.1, soil minerals, predominantly Fe oxides, hydroxides, and oxyhydroxides, are known to influence the biological stability of SOM, including DOC, via sorption to reactive surface sites due to their small size (down to several nanometres) and high surface reactivity via surface hydroxyl groups, or co-precipitation with the solid phase (McLean and Bledsoe, 1992; Kaiser and Guggenberger, 2003; Kleber *et al.*, 2005; Wagai and Mayer, 2007; Saidy *et al.*, 2012; Kleber *et al.*, 2015; Porras *et al.*, 2017; Fujii *et al.*, 2019; Wen *et al.*, 2019; Wagai *et al.*, 2020). The results of Chapter 3 conclude that the addition of Fe oxides to soils enhances organic C sorption, reducing soil C availability and lability, decreasing the release of C into solution.

1.2 Aggregation Potential

Soil structure has important influences on edaphic conditions and the environment; the structure is often measured by the stability of soil aggregates (Bronick and Lal, 2005). Soil aggregation sustains soil fertility by reducing erosion and mediating soil aeration, as well as water infiltration and retention (Zhao *et al.*, 2017). Large pores in the soil generally favour high infiltration rates, good tilth, and adequate aeration for plant growth (Kemper and Rosenau, 1986). Soil aggregation protects SOM from mineralising because it physically reduces the accessibility of organic compounds for microorganisms, extracellular enzymes, and oxygen (Oades, 1984; Six *et al.*, 2002; von Lützow *et al.*, 2006). Studies on the stability of soil aggregates under different land use rarely focus on the effects of various soil organic components, Fe oxides in different size fractions of soil aggregates, and their interactions. However, Zhao *et al.* (2017) found that the interaction of Fe oxides and SOM, and the differential roles of Fe oxides in formation or break-down of soil aggregates of middle sizes, were key to the stability and size distribution of soil aggregates in their studied red soil in southern China.

1.3 Plant Growth Potential

Fe is among the essential nutrients in plants, being involved in metabolic functions (Zuo and Zhang, 2011). In plants, Fe participates in many physiological processes including chlorophyll biosynthesis, respiration, and redox reactions (Mimmo *et al.*, 2014; Ye *et al.*, 2015; Zargar *et al.*, 2015; Rui *et al.*, 2016). Rizwan *et al.* (2019) found that Fe oxide nanoparticles improved the plant growth and reduced the oxidative stress and cadmium concentration in wheat, as

measured by plant height and dry weights of shoots and roots. Lebrun *et al.* (2021) discovered that ochre amendments to soil, alone and combined with biochar and manure, improved soil conditions, which in turn allowed better plant growth of *Agrostis capillaris*, providing motivation for it to be added to agricultural soils. Mertz *et al.* (2021) found similar results, with additions of ochre with manure preventing metal contaminants leaching during infiltration by modifying the physical, chemical and biological properties of soil, consequently ameliorating plant growth.

1.4 Aims, Objectives and Hypotheses

The aim of this study was to understand the effect of ochre amendments on C lability, in a more realistic experiment than the adsorption study (Chapter 3), as well as plant growth, and to determine whether Fe oxide is the limiting factor in soil aggregate production. This was achieved through a plant growth experiment in which ochres were added to 2 soils with varying properties, which were incubated for 8 weeks and then used in plant growth experiments. The soil aggregate size distribution, quantities of cold and hot water extractable C, extractable phosphorus, extractable Fe and soil pH was measured on the soil post incubation and post plant growth. Changes in these variables were used to determine the effects of ochre amendments on C lability and plant growth. It is hypothesised that the addition of Fe oxide ochres will reduce C availability and increase the proportion of macroaggregates produced, having no effect on plant growth.

2 Methodology

2.1 Experimental Materials

The Agricultural and Woodland Soils and Morlais, Lynemouth and Polkemmet ochres were used, as previously described in Chapter 3, Section 2.1. The soils and ochres were characterised using standard methods, as described in Chapter 3, Section 2.1. Results of these analyses are given in Table 1: Chapter 3, Section 2.1.

2.2 Incubation Experiment

Four hundred grams of moist soil were mixed with moist ochre equivalent to dry masses of 0, 2 and 20 g (dry mass ratios of 0, 0.005 and 0.05:1 ochre:soil); five replicates were established per treatment. Fifty millilitres of deionised water were added to the samples containing Woodland Soil and the components were well mixed by hand; no deionised water was added to the Agricultural Soil samples due to its wetness. Treatments and controls were incubated for 8 weeks at 30°C in sealed plastic bags in darkness. Bags were opened weekly and the soils and ochres were mixed by hand.

After 8 weeks of incubation, analysis of hot and cold-water extractable C, plant available inorganic P, extractable Al, Fe, Mn and Si, soil pH in water and soil aggregate size was conducted.

The extraction of hot-water C was conducted in two steps: the first involving the removal of readily soluble cold-water extractable C (CWEC) from the soils that may have come from recent liming of the soil or from animal excreta and soluble plant residues; the second involving the extraction of labile components of soil C at 80°C for 16 h; this is subsequently referred to as hot-water extractable C (HWEC). The supernatants of each extraction were filtered through 0.45 µm cellulose nitrate membrane filters and analysed for DOC using an elemental analyser (Elementar varioTOC select).

Plant available inorganic P levels were measured by the Olsen P extraction, at pH 8.5; 5 g of sample was shaken at 20°C with 100 mL of sodium hydrogen carbonate and 0.05 % m/v polyacrylamide solution for 30 minutes and filtered through Whatman no. 2 filter paper, with analysis on an autoanalyser (Seal Analytical Nutrient Autoanalyser) (Olsen, 1954).

Quantities of extractable Al, Fe, Mn and Si were determined through the dissoluting methods of sodium pyrophosphate (SP), ammonium oxalate (AO) and dithionite-citrate (DC) (Loeppert and Inskeep, 1996). These results allow for quantities of organic complexed Fe, non-crystalline inorganic forms of Fe and finely divided crystalline Fe solid phases to be estimated. The AO and DC extraction samples were filtered through Whatman no. 40 filter paper, with the SP samples filtered through a 0.2-micron nylon syringe filter. Elemental analysis of all samples was carried out by ICP-OES (Thermo iCAP 7000). Data for extractable Al, Mn and Si for each extraction are presented in Appendix B (Figures B1, B2 and B3) and will not be discussed further.

Water stable soil aggregates were measured by wet sieving to 5 aggregate size fractions: large macro-aggregates (>2000 µm), medium macro-aggregates (1000 - 2000 µm), small macro-aggregates (250 - 1000 µm), micro-aggregates (53 - 250 µm) and silt/clay micro-aggregates and minerals (<53 µm) (Cambardella and Elliott, 1993). The soil was dried overnight at 105°C, and subsequently weighed.

For all analyses, accuracy was calculated using Equation 1 (Chapter 3, Section 2.2). Method blanks were run for ICP-OES extractions to enable the detection limits of Al, Fe, Mn and Si to be calculated (0.000313 - 0.115 mg/L). The coefficient of variation determined from duplicate analysis of 10 % of solutions for elements above detection ranged from 4.52 - 13.4 %, 2.42 - 9.67 % and 6.31 - 10.5 % for the ICP-OES for the SP, AO and DC extractions, respectively (Gill, 1997). Based on the analysis of an in-house certified reference material of 0.5 mg/L, the accuracy of the ICP-OES analyses ranged from 94.7 - 117 %, 105 - 133 % and 80.0 - 117 % for the ICP-OES for the SP, AO and DC extractions, respectively; the CRM was not matrix-matched to the standards. For the C analysis by elemental analyser, the coefficient of variation was 2.26 %. Based on the analysis of matrix-matched in-house certified reference material of 50 mg/L, the accuracy of the DOC analyses was 102 %. For the P analysis by autoanalyser, the coefficient of variation for a solution with a mean concentration of 1.99 mg/L

was 0.241 % (n = 10). Based on the analysis of a matrix-matched in-house certified reference material of 1 mg/L, the accuracy of the Olsen P analyses was 118 %.

2.3 Plant Growth Experiment

Wheat (*Triticum aestivum*) is among the world's major cereals and is the staple food for over 50 % of the global population, with 713 million tons per annum production (FAO, 2014; McVetty *et al.*, 2016; Rady *et al.*, 2019; Lu *et al.*, 2020). A plant growth experiment was set up through the addition of pre-germinated common wheat (*Triticum aestivum*; 3 seeds per pot) to a 150 g subsample of the soil-ochre mix after a week of them being removed from incubation. The plants were watered weekly, with approximately 25 mL water added to each sample in order for the sample to maintain as weight of 175 g with the water added; the plant pots were randomly assorted and rotated at random to ensure uniformity in growth. The plants were grown for 40 days under natural light conditions and harvested. Shoots were cut 1 cm above ground level; shoots were oven-dried at 70°C for 24 hours and weighed for biomass determination. Soil pH was determined in water to understand the effects of ochre addition on pH (Rowell, 1994). After plant harvest, analysis of CWEC and HWEC, plant available inorganic P, soil aggregate size, extractable Al, Fe, Mn and Si and soil pH in water was repeated on the Plant Growth Soil. Soil respiration rates were also measured following plant harvest, to determine whether ochre amendments to soils had an effect on this, relative to the control. Moist soil was incubated with NaOH for 1 week at 20°C. $BaCl_2$, deionised water and phenolphthalein was added to the NaOH and titrated against HCl. Soil respiration rates were calculated following methods in Rowell (1994).

2.4 Statistical Analysis

Statistical analyses were conducted using R Studio (Version 4.1.0). Data are presented as means ± standard deviation, n = 5, unless otherwise stated. All statistical analyses were tested at a confidence level of 95 %. A Levene and Kolmogorov-Smirnov test were used to assess the equality of variance and normality of the data, respectively. pH was converted to proton concentration for statistical analysis. Incubation Soil and Plant Growth Soil data were analysed separately. The dataset was split between the two soils; a one-way ANOVA was performed on each dataset, with ochre treatment (control, 0.005:1 and 0.05:1) as the factor, subsequently employing a Dunnett's test to compare the differences in CWEC, HWEC, extractable P, extractable Fe, aggregate fraction sizes and the pH of the solutions for both the Incubation and the Plant Growth Soil, as well as biomass and soil respiration for Plant Growth Soil, between the control treatment and the ochre treatments. These analyses were performed in order to confirm any effects of the ochre treatments on these variables. Following this, a three-way ANOVA was performed, with soil (Agricultural vs Woodland), ochre level (control, 0.005:1 and 0.05:1) and ochre type as factors (Wilcox, 1993). Control treatment data were removed from the dataset to ensure the ANOVA was balanced. If the three-way ANOVA

identified significant effects, either a one-way ANOVA, with a subsequent Tukey test, or a Dunn (1964) Kruskal-Wallis multiple comparison test, adjusted with the Bonferroni method, was performed, as per the previous chapter, depending on whether the data was normally distributed with equal variances. These tests determined whether there were significant differences in CWEC, HWEC, extractable P, extractable Fe, aggregate fraction sizes and the pH of the solutions for Incubation Soil, as well as biomass and soil respiration for Plant Growth Soil, as per above, between soils, treatments and ochres. Having identified significant interactions with the three-way ANOVA, differences in these variables for ochre treatments of the same concentration between soils, and for the different ochre treatments within the same soils were assessed with a two-way ANOVA, with factors of soil and ochre, and treatment and ochre, respectively, followed by a Tukey test. An independent samples t-test, with soil as the factor, compared differences in CWEC, HWEC, extractable P, extractable Fe, aggregate fraction sizes and the pH of the solutions for Incubation Soil, as well as biomass and soil respiration for Plant Growth Soil between the Agricultural and Woodland Soils in the control treatments.

Correlations between extractable C (CWEC and HWEC) and Fe extractions and pH were identified for each soil group, with Pearson correlations used on normal data, identified using a Kolmogorov-Smirnov test, and Spearman correlations used on non-normal data. A Welch two sample t-test was performed to identify significant differences between the Fe extractions of the Incubation samples and the Plant Growth samples.

3 Results

The results of the incubation and plant growth study are discussed in turn in Sections 3.1 and 3.2, with differences in Fe extractions between the incubation and the plant growth experiments discussed in Section 3.3. Figures 1, 2, 3 and 4 present the CWEC and HWEC, extractable P, extractable Fe and aggregate size distribution, respectively, and Table 1 the pH results of the incubation and plant growth experiments. Figure 5 and Table 2 present the shoot biomass and soil respiration results of the plant growth experiment.

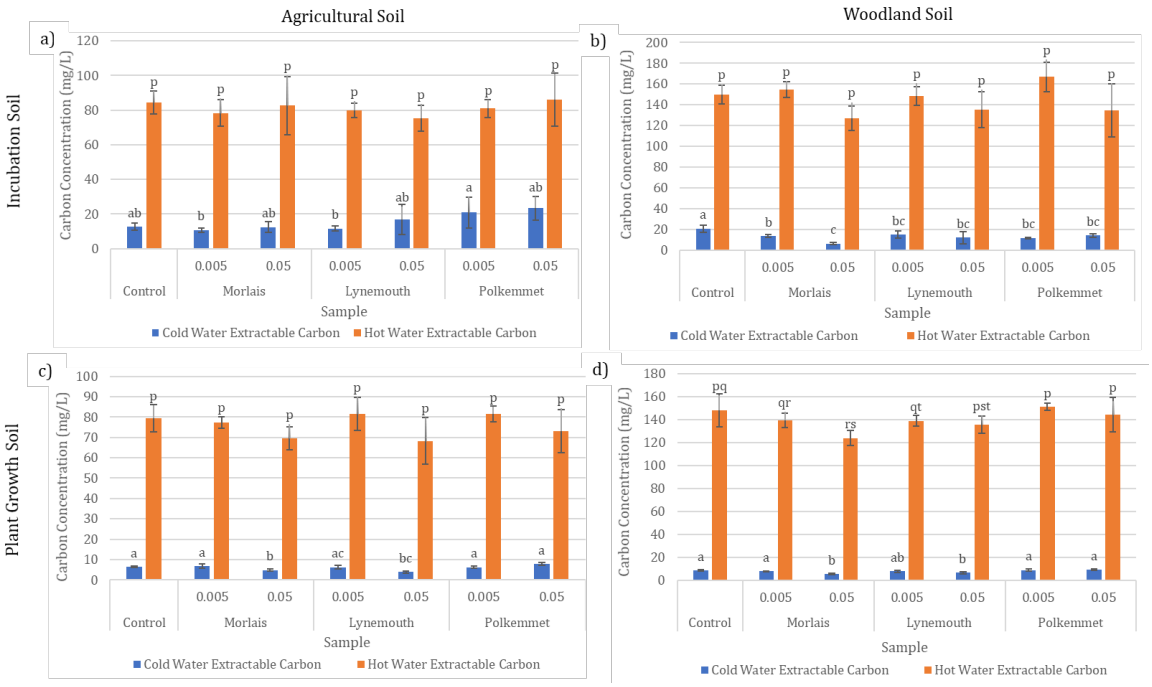

Figure 1: Mean carbon concentration (mg/L) of CWEC and HWEC of (a) Agricultural Incubation samples, (b) Woodland Incubation samples, (c) Agricultural Plant Growth samples and (d) Woodland Plant Growth samples. Error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively. Different letters represent significant differences in carbon concentration within each carbon extraction type (CWEC: a - c; HWEC: p - t) for three different comparisons, between the control and the ochre treatments (one-way ANOVA and Dunnett's test;), between ochres of the same treatment level (two-way ANOVAs and Tukey tests) and between different treatment levels of the same ochre (two-way ANOVAs and Tukey tests), all at $p \leq 0.05$.

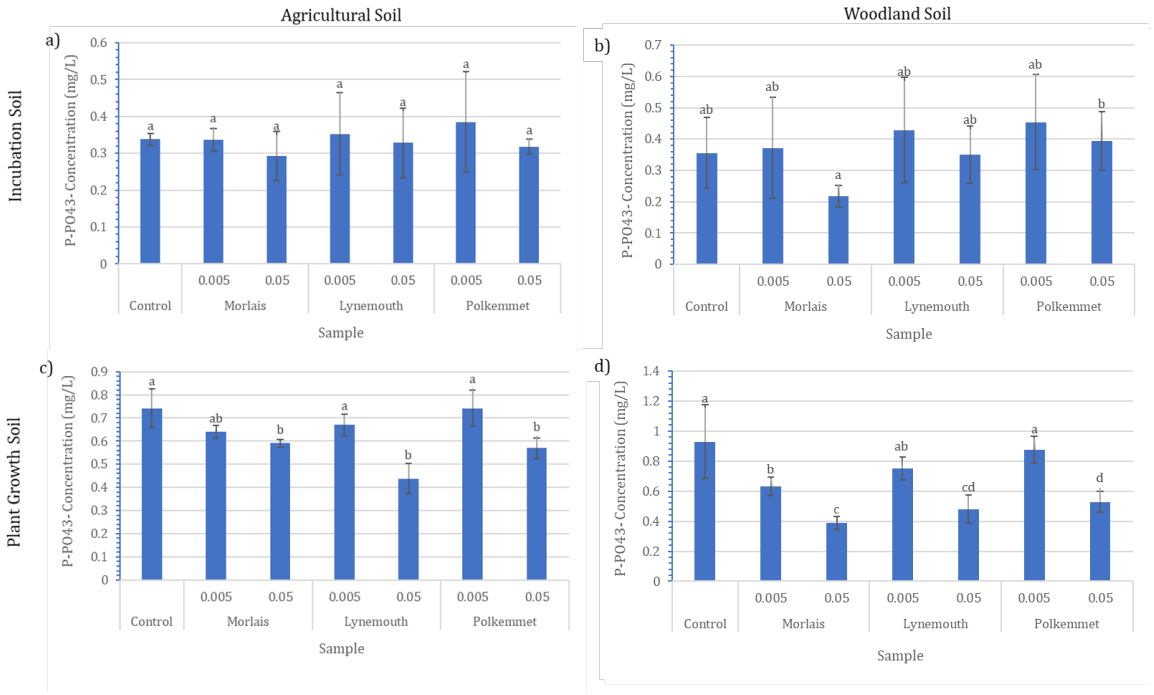

Figure 2: Mean concentration of P present as phosphate (mg/L) of (a) Agricultural Incubation samples, (b) Woodland Incubation samples, (c) Agricultural Plant Growth samples and (d) Woodland Plant Growth samples, as determined by the Olsen P extraction method. Error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively. Different letters (a - d) represent a significant difference in P concentration for three different comparisons, between the control and the ochre treatments (one-way ANOVA and Dunnett's test;), between ochres of the same treatment level (two-way ANOVAs and Tukey tests) and between different treatment levels of the same ochre (two-way ANOVAs and Tukey tests), all at $p \leq 0.05$.

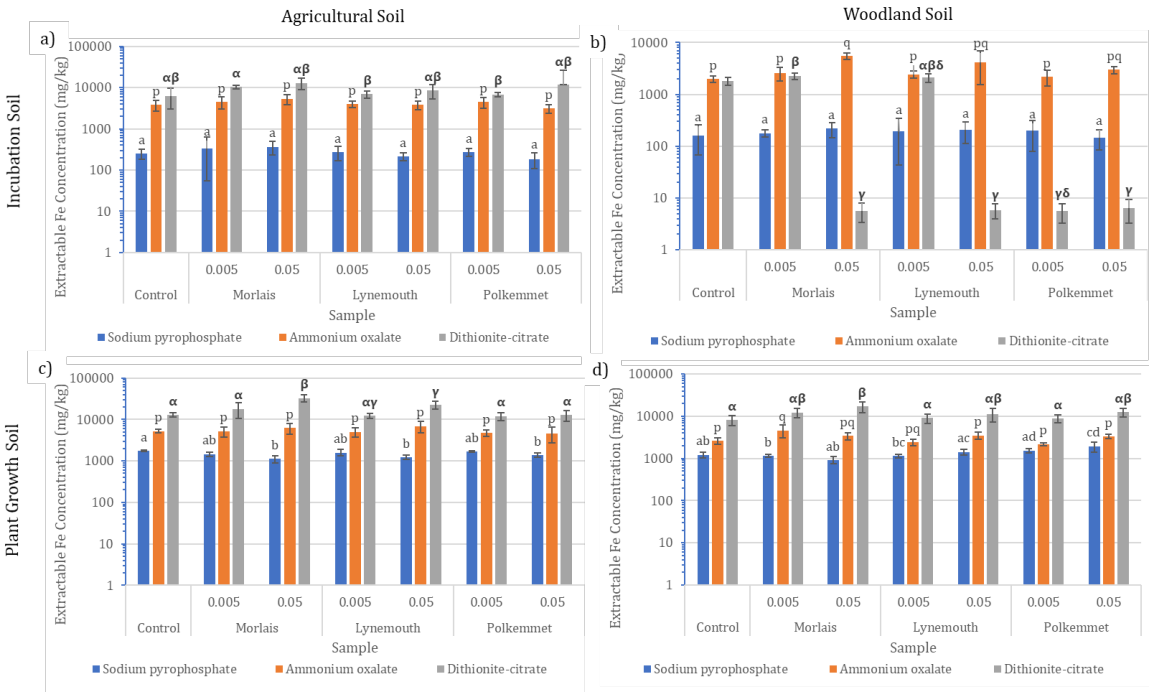

Figure 3: Mean concentrations (mg/kg) of extractable Fe for sodium pyrophosphate (SP) (organically complexed Fe), ammonium oxalate (AO) (organically complexed + amorphous inorganic Fe) and dithionite citrate (DC) (organically complexed, amorphous inorganic Fe + Fe oxides) extractions of (a) Agricultural Incubation samples, (b) Woodland Incubation samples, (c) Agricultural Plant Growth samples and (d) Woodland Plant Growth samples. Error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively. Different letters represent a significant difference in Fe concentration within each extraction type (SP: a - d; AO: p - q; DC: α - δ) for three different comparisons, between the control and the ochre treatments (one-way ANOVA and Dunnett's test;), between ochres of the same treatment level (two-way ANOVAs and Tukey tests) and between different treatment levels of the same ochre (two-way ANOVAs and Tukey tests), all at $p \leq 0.05$.

Table 1: pH of Agricultural and Woodland Incubation and Plant Growth samples. Error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively. Different superscript letters within each soil column (Incubation Agricultural Soil: a; Incubation Woodland Soil: p - r; Plant Growth Agricultural Soil: α - β; Plant Growth Woodland Soil: π - σ) represent a significant difference in pH for three different comparisons, between the control and the ochre treatments (one-way ANOVA and Dunnett's test;), between ochres of the same treatment level (two-way ANOVAs and Tukey tests) and between different treatment levels of the same ochre (two-way ANOVAs and Tukey tests), all at $p \leq 0.05$.

		INCUBATION		PLANT GROWTH	
		Agricultural Soil	**Woodland Soil**	**Agricultural Soil**	**Woodland Soil**
Control		7.47 ± 0.08^{a}	7.54 ± 0.04^{p}	7.26 ± 0.15$^{\alpha}$	7.15 ± 0.03$^{\pi}$
Morlais	**0.005**	7.45 ± 0.03^{a}	7.29 ± 0.05^{q}	7.42 ± 0.08$^{\beta}$	7.20 ± 0.04$^{\pi}$
	0.05	7.46 ± 0.18^{a}	7.21 ± 0.04^{r}	7.41 ± 0.08$^{\beta}$	7.15 ± 0.07$^{\pi}$
Lynemouth	**0.005**	7.49 ± 0.05^{a}	7.39 ± 0.05^{q}	7.47 ± 0.05$^{\beta}$	7.36 ± 0.02$^{\rho}$
	0.05	7.49 ± 0.24^{a}	7.61 ± 0.09^{p}	7.19 ± 0.05$^{\alpha}$	7.73 ± 0.08$^{\sigma}$
Polkemmet	**0.005**	7.46 ± 0.11^{a}	7.65 ± 0.05^{p}	7.40 ± 0.03$^{\beta}$	7.37 ± 0.06$^{\rho}$
	0.05	7.60 ± 0.05^{a}	7.67 ± 0.05^{p}	7.21 ± 0.08$^{\alpha}$	7.48 ± 0.06$^{\rho\sigma}$

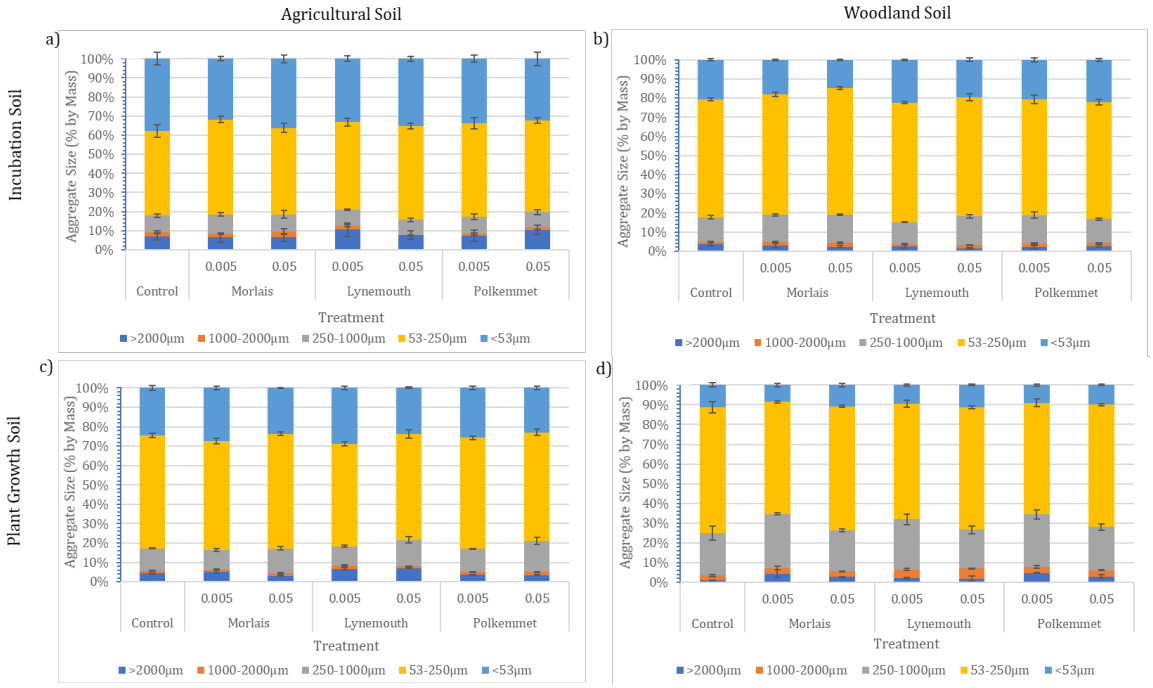

Figure 4: Aggregate percentages by total mass for >2000 µm; 1000 - 2000 µm; 250 - 1000 µm; 53 - 250 µm and <53µm size fractions of (a) Agricultural Incubation samples, (b) Woodland Incubation samples, (c) Agricultural Plant Growth samples and (d) Woodland Plant Growth samples. Error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively.

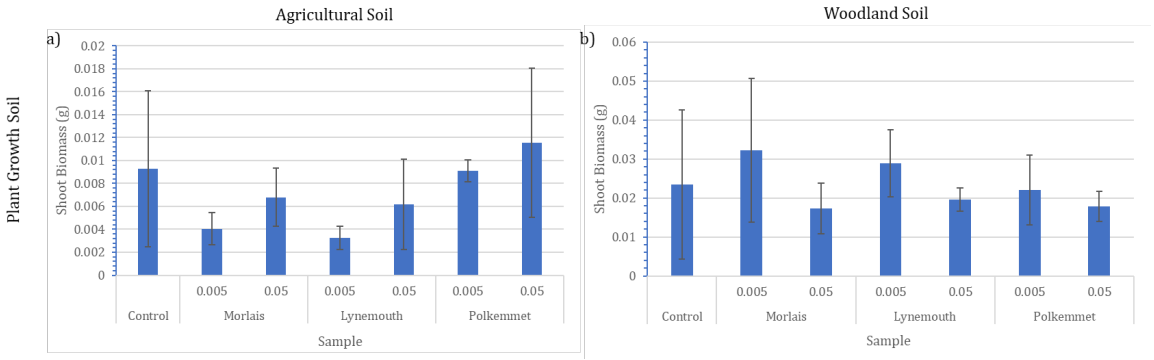

Figure 5: Mean shoot biomass of *Triticum aestivum*, as measured by oven dried shoot weight (g) for (a) Agricultural and (b) Woodland soil samples. For Agricutural samples, error bars represent standard deviation, n=5 for Lynemouth 0.005 treatment; n=4 for Morlais 0.005 and 0.05, Polkemmet 0.005 and Lynemouth 0.05 treatments; n=3 for control, Polkemmet 0.05 treatments. For Woodland samples, error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively. Significance letters are not included as no significant difference in shoot biomass was found between the control and the ochre treatments, between ochre types of the same treatment level or between different treatment levels of the same ochre type ($p>0.05$: one-way ANOVA and Dunnett's test; two-way ANOVAs and Tukey tests).

Table 2: Soil respiration rates ($gCO_2/g/s$) for Agricultural and Woodland soil samples. Error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively. Significance letters are not included as no significant difference in soil respiration was found between the control and the ochre treatments, between ochre types of the same treatment level and between different treatment levels of the same ochre type ($p>0.05$: one-way ANOVA and Dunnett's test; two-way ANOVAs and Tukey tests).

		Agricultural Soil	Woodland Soil
Control		$1.90 \times 10^{-9} \pm 1.08 \times 10^{-9}$	$2.22 \times 10^{-9} \pm 9.65 \times 10^{-10}$
Morlais	**0.005**	$9.95 \times 10^{-10} \pm 1.11 \times 10^{-9}$	$2.15 \times 10^{-9} \pm 1.34 \times 10^{-9}$
	0.05	$1.85 \times 10^{-9} \pm 1.08 \times 10^{-9}$	$2.71 \times 10^{-9} \pm 1.50 \times 10^{-9}$
Lynemouth	**0.005**	$1.46 \times 10^{-9} \pm 1.24 \times 10^{-9}$	$2.27 \times 10^{-9} \pm 1.06 \times 10^{-9}$
	0.05	$1.92 \times 10^{-9} \pm 1.12 \times 10^{-9}$	$2.07 \times 10^{-9} \pm 9.69 \times 10^{-10}$
Polkemmet	**0.005**	$1.75 \times 10^{-9} \pm 1.12 \times 10^{-9}$	$2.25 \times 10^{-9} \pm 1.07 \times 10^{-9}$
	0.05	$1.79 \times 10^{-9} \pm 1.03 \times 10^{-9}$	$1.88 \times 10^{-9} \pm 7.51 \times 10^{-10}$

3.1 Incubation Analysis

3.1.1 Comparisons to the controls

The ochre amendments to the Agricultural Soil generally had no significant effects on any of the variables (CWEC, HWEC, extractable P, extractable Fe, aggregate fraction sizes and

the pH of the solutions), relative to the Agricultural control ($p>0.05$). The ochre amended Woodland Soil also generally showed no significant effect on these variables, as compared to the Woodland control ($p>0.05$), with the exception of the CWEC and DC extractable Fe data. All ochre treated Woodland Soils had significantly lower concentration of CWEC, the labile SOC fraction, relative to the control (Figure 1), with the majority of the treated Woodland Soils having a significantly higher DC extractable Fe concentration than that of the control ($p\leq0.05$) (Figure 3).

3.1.2 Comparisons between ochres

Comparing the ochre treatments at the same treatment levels across soils, there were some differences within variables but no noticeable trends across variables. The 0.005 treatment of the Polkemmet ochre in Agricultural Soil has significantly greater CWEC concentrations than the 0.005 treatments of Morlais and Lynemouth ($p\leq0.05$) (Figure 1). The 0.005 Morlais treated Agricultural Soil had a significantly greater DC extractable Fe concentration than the Lynemouth and Polkemmet treatments ($p\leq0.05$) (Figure 3). At the 0.005 treatment level, the Polkemmet treated Agricultural Soil had a significantly smaller percentage mass of the 1000 - 2000 µm size fraction than the Morlais and Lynemouth treated soils (Figure 4). At the 0.05 treatment level, the Morlais treated Agricultural Soil had a significantly smaller percentage mass of the >2000 µm size fraction but a significantly greater percentage mass of the <53 µm size fraction than the Polkemmet treatment, as well as a significantly greater percentage mass of the 1000 - 2000 µm size fraction than the Lynemouth treatment ($p\leq0.05$) (Figure 4).

The 0.005 Morlais treated Woodland Soil had a significantly greater DC extractable Fe content than the Polkemmet treatment ($p\leq0.05$) (Figure 3). The Polkemmet treated Woodland Soil had a significantly greater pH than the Morlais and Lynemouth ochres at the 0.005 treatment level, with the Morlais treatment having a significantly lower pH than the Lynemouth and Polkemmet treatments at the 0.05 treatment level ($p\leq0.05$) (Table 1). The Lynemouth treated Woodland Soil had a significantly smaller percentage mass of the 250 - 1000 µm size fraction than the Morlais and Polkemmet treated soils, as well as a significantly greater percentage mass of the <53 µm size fraction than the Morlais treated Woodland Soil ($p\leq0.05$) (Figure 4). In the 0.05 ochre treated Woodland Soil, the Polkemmet treatment had a significantly smaller percentage mass of the 250 - 1000 µm size fraction than the Lynemouth treatment; the Morlais treatment had a significantly greater percentage mass of the 53 - 250 µm size fraction but a significantly smaller percentage mass of the <53 µm size fraction than the Lynemouth and Polkemmet ochres ($p\leq0.05$) (Figure 4).

3.1.3 Comparisons between treatment levels

Comparing all of the 0.005 treatments with the 0.05 treatments in each soil, there were some differences within variables but no noticeable trends across variables. The 0.05 ochre treated Agricultural Soils tended to have greater DC extractable Fe concentrations than the corresponding 0.005 treatments ($p\leq0.05$) (Figure 3). No difference was found in the SP and AO

extraction types ($p>0.05$) (Figure 3). At the 0.05 treatment level, the Morlais and Lynemouth treated Woodland Soils had a significantly lower and greater pH than at the 0.005 treatment level, respectively ($p\leq0.05$) (Table 1). In the Agricultural Soil, the 0.05 treatments of Morlais and Lynemouth had significantly greater percentage masses of <53 µm and 1000 - 2000 µm size fractions, respectively, than the 0.005 treatments of the same ochres ($p\leq0.05$) (Figure 4). In the Woodland Soil, the 0.05 treatment of Morlais had a significantly greater percentage mass of the 53 - 250 µm size fraction, and a significantly smaller percentage mass of the <53 µm size fraction than the 0.005 Morlais treated Woodland Soil ($p\leq0.05$) (Figure 4). Furthermore, in the Woodland Soil, the 0.05 Lynemouth treatment had significantly greater percentage masses of 1000 - 2000 µm and 250 - 1000 µm size fractions, and a significantly smaller percentage mass of the <53 µm size fraction, with the Polkemmet treatment at the 0.05 level having a significantly smaller percentage mass of the 250 - 1000 µm size fraction, as compared to the 0.005 treatments of the same ochres ($p\leq0.05$) (Figure 4).

3.1.4 Comparisons between soils at control level

The t-test comparing the controls of both soils revealed that Woodland Soil had significantly greater HWEC than the Agricultural Soil but the Agricultural control had a significantly greater AO extractable Fe concentration than the Woodland control ($p\leq0.05$) (Figure 1 and 3). The Agricultural Soil had significantly greater percentage masses of >2000 µm and <53 µm and smaller percentage masses of 250 - 1000 µm and 53 - 250 µm size fractions than the Woodland Soil ($p\leq0.05$) (Figure 4).

3.1.5 Comparisons between soils at ochre treatment level

When comparing the same ochre level treatment across both soils, it was found that at the 0.05 treatment level, all ochre treated Agricultural Soils had a significantly lower HWEC concentration than in the Woodland Soils ($p\leq0.05$) (Figure 1). This corresponds to the Fe extraction data: at the 0.005 treatment level, the ochre treated Agricultural Soils had significantly higher DC extractable Fe concentration than the Woodland Soil ($p\leq0.05$) (Figure 3). The 0.005 and 0.05 Morlais treated Agricultural Soils had a significantly higher pH, with Polkemmet having a significantly lower pH than the same treatment in the Woodland Soil ($p\leq0.05$) (Table 1). The ochre treated Agricultural Soils, at both treatment levels, tended to have significantly greater percentage masses of >2000 µm and <53 µm, and significantly smaller percentage masses of 250 - 1000 µm and 53 - 250 µm size fractions, compared to the Woodland Soil ($p\leq0.05$) (Figure 4).

3.1.6 Correlations between variables

There were no significant correlations between CWEC or HWEC and the different extractable Fe fractions or pH in the Agricultural samples ($p>0.05$), with the exception of a significant moderate negative correlation between HWEC and pH ($p\leq0.05$; $r=-0.51$). In the Woodland

samples, a significant moderate positive correlation was found between CWEC and DC extractable Fe ($p \leq 0.05$; $r=0.47$), with moderate negative correlations found between CWEC and AO extractable Fe and pH ($p \leq 0.05$; $r=-0.46$ and -0.40, respectively), as well as between HWEC and AO extractable Fe ($p \leq 0.05$, $r=-0.42$). No significant interactions were found on the Woodland samples between CWEC, HWEC and the other variables ($p>0.05$).

3.2 Plant Growth Analysis

3.2.1 Comparisons to the controls

The ochre amended soils generally had no significant effects on any of the variables, relative to the controls ($p>0.05$). The ochre amendments generally had no significant effects on CWEC and HWEC ($p>0.05$); however, at the 0.05 treatment level, the Morlais and Lynemouth ochre amendments to both soils significantly reduced concentrations of CWEC, relative to their respective controls ($p \leq 0.05$) (Figure 1). In both soils, the majority of the ochre treatments significantly reduced the levels of extractable P, as compared to the Agricultural and Woodland controls ($p \leq 0.05$) (Figure 2). The ochre amendments to both soils generally had no significant effects on extractable Fe, relative to the controls ($p>0.05$), with the exception that the 0.05 ochre treatments in both soils tended to have significantly greater SP and DC extractable Fe concentrations than the controls ($p \leq 0.05$) (Figure 3). In both Agricultural and Woodland Soils, the ochre amendments generally increased the pH of the soil solution, relative to the respective controls ($p \leq 0.05$) (Table 1). The ochre amendments to both soils generally showed no significant effects on the distribution of aggregate size fractions, as compared to the respective controls ($p>0.05$), with the exception of all ochres at the 0.005 treatment level. These ochre treatments had a significantly greater percentage mass of 53 - 250 µm and <53 µm size fractions and significantly smaller percentage mass of the 1000 - 2000 µm size fraction ($p \leq 0.05$) (Figure 4). The ochre amended Agricultural and Woodland Soils showed no significant effect on shoot biomass or soil respiration, relative to the controls ($p>0.05$) (Figure 5; Table 2).

3.2.2 Comparisons between ochres

Comparing the ochre treatments at the same treatment levels across soils, there were some differences within variables but no noticeable trends across variables. In the Agricultural Soil, the 0.05 Morlais treatment had a significantly higher pH than the Lynemouth and Polkemmet treatments ($p \leq 0.05$) (Table 1). At the 0.005 treatment level, the Lynemouth treated Agricultural Soil had significantly greater percentage masses of the >2000 µm and <53 µm size fractions than the Polkemmet treatment, but a significantly smaller percentage mass of the 53 - 250 µm size fraction than the Morlais and Polkemmet ochres ($p \leq 0.05$). At the 0.05 treatment level, the Lynemouth treated Agricultural Soil had a significantly greater percentage mass of the >2000 µm size fraction but a significantly smaller percentage mass of the 1000 - 2000 µm size fraction than the Morlais and Polkemmet, and the Polkemmet treatments, respectively ($p \leq 0.05$) (Figure 4). The 0.05 Morlais treated Agricultural Soil had a significantly

smaller percentage mass of the 250 - 1000 µm size fraction than the Polkemmet treatment, but a significantly greater percentage mass of the 53 - 250 µm size fraction than both the Lynemouth and Polkemmet ochres ($p \leq 0.05$) (Figure 4).

In both soils, at the 0.05 level, the Polkemmet treatment had a significantly greater CWEC concentration than the Morlais and Lynemouth ochres, as well as having a significantly greater HWEC concentration than the Morlais ochre in the treated Woodland Soil ($p \leq 0.05$) (Figure 1). At the 0.005 treatment level, the Polkemmet ochre also had a significantly higher HWEC concentration than the Morlais and Lynemouth ochres ($p \leq 0.05$) (Figure 1). At both treatment levels, the Polkemmet treated Woodland Soils had significantly greater extractable P concentrations than Morlais and Lynemouth treatments ($p \leq 0.05$) (Figure 2). At both treatment levels, the Morlais treated Woodland Soils had a significantly lower pH than the Lynemouth and Polkemmet treatments ($p \leq 0.05$) (Table 1). The 0.005 Lynemouth treated Woodland Soil had a significantly smaller percentage mass of the >2000 µm and a greater percentage mass of the 1000 - 2000 µm size fractions than the Polkemmet and Morlais ochres, respectively ($p \leq 0.05$) (Figure 4). In the 0.05 ochre treated Woodland Soil, the Lynemouth treatment had a significantly greater percentage mass of the 1000 - 2000 µm size fraction than the Morlais and Polkemmet treatments ($p \leq 0.05$) (Figure 4).

3.2.3 Comparisons between treatment levels

Comparing all of the 0.005 treatments with the 0.05 treatments in each soil, it was found that the Morlais and Lynemouth treated Agricultural Soils had significantly lower CWEC concentrations at the 0.05 level than the 0.005 level; the same significant trend was found in the Morlais ochre in the Woodland Soil ($p \leq 0.05$), however no differences were found in the Fe extractions ($p > 0.05$) (Figure 1). Furthermore, in both soils, the majority of the 0.05 ochre treatments had a significantly lower extractable P concentration than the corresponding 0.005 treatments ($p \leq 0.05$) (Figure 2). At the 0.05 treatment level, the Lynemouth and Polkemmet treated Agricultural Soils had a significantly lower pH than at the 0.005 treatment level, with the 0.05 Lynemouth treatment in the Woodland Soil having a significantly greater pH than its 0.005 counterpart ($p \leq 0.05$) (Table 1). The 0.05 Morlais, Lynemouth and Polkemmet treated Agricultural Soils had significantly smaller percentage masses of <53 µm size fraction and greater percentage masses of 250 - 1000 µm size fractions than the corresponding 0.005 treatments ($p \leq 0.05$) (Figure 4). The 0.05 Morlais treatment had a significantly smaller percentage mass of >2000 µm size fraction, with the 0.05 Lynemouth treatment having a significantly smaller percentage mass of the 1000 - 2000 µm size fraction than the 0.005 treatments ($p \leq 0.05$) (Figure 4). In the Woodland Soil, the 0.05 treatment of Morlais, Lynemouth and Polkemmet had significantly smaller percentage masses of 250 - 1000 µm size fraction and significantly greater percentage masses of the 53 - 250 µm size fraction than the 0.005 treatments, with the Morlais and Lynemouth treated soils having significantly greater percentage masses of the <53 µm size fraction at the 0.05 treatment level, as compared to the 0.005 treatments of the same ochres ($p \leq 0.05$) (Figure 4).

3.2.4 Comparisons between soils at control level

The t-test comparing the controls of both soils revealed that the Agricultural Soil had significantly lower concentrations of CWEC and HWEC than the Woodland Soil, corresponding with the Agricultural control having a significantly greater SP, AO and DC extractable Fe concentrations than the Woodland control ($p \leq 0.05$) (Figure 1 and 3). The Agricultural Soil had significantly greater percentage masses of >2000 µm, 1000 - 2000 µm and <53 µm and smaller percentage masses of 250 - 1000 µm and 53 - 250 µm size fractions than the Woodland Soil ($p \leq 0.05$) (Figure 4). No significant difference was found in the biomass and soil respiration rate between the controls of the two soils ($p > 0.05$) (Figure 5; Table 2).

3.2.5 Comparisons between soils at ochre treatment level

When comparing the same ochre level treatment across both soils, it was found that the Polkemmet ochre at both treatments, and the Lynemouth ochre at the 0.05 treatment, had significantly lower CWEC concentrations in the Agricultural Soil than in the Woodland Soil, with the Morlais, Lynemouth and Polkemmet ochres at both treatment levels, having significantly lower HWEC concentrations in the Agricultural Soil, as compared to the Woodland Soil ($p \leq 0.05$) (Figure 1). This corresponds to the Fe extraction data: at the 0.005 and 0.05 levels, the ochre treated Agricultural Soils had significantly greater AO and DC extractable Fe levels than the Woodland Soil, respectively ($p \leq 0.05$) (Figure 3). However, the 0.05 Morlais treatment had a significantly greater extractable P concentration in the Agricultural Soil than in the Woodland Soil ($p \leq 0.05$) (Figure 2). At both treatment levels, the Morlais ochre had a significantly higher pH in the Agricultural Soil than in the Woodland Soil, however, at the 0.05 treatment level, the Lynemouth and Polkemmet ochres had a significantly greater pH in the Woodland Soil, as compared to the Agricultural Soil ($p \leq 0.05$) (Table 1). The ochre treated Agricultural Soils, at both treatment levels, tended to have significantly greater percentage mass of <53 µm size fraction and significantly smaller percentage masses of 1000 - 2000 µm, 250 - 1000 µm and 53 - 250 µm size fractions, compared to the Woodland Soil ($p \leq 0.05$) (Figure 4). At both treatment levels, the Morlais and Lynemouth amended Woodland Soils had significantly greater shoot biomass than the treated Agricultural Soils ($p \leq 0.05$) (Figure 5).

3.2.6 Correlations between variables

In the Agricultural samples, significant weak and moderate positive correlations were found between CWEC and HWEC, respectively, and SP ($p \leq 0.05$; $r = 0.35$ and 0.54, respectively), with moderate negative correlations found between CWEC and HWEC, respectively, and DC extractable Fe ($p \leq 0.05$; $r = -0.49$ and -0.43, respectively), as well as between HWEC and pH ($p \leq 0.05$; $r = -0.42$). No significant interactions were found in the Agricultural samples between CWEC, HWEC and the other variables ($p > 0.05$). In the Woodland samples, a significant moderate positive correlation was found between CWEC and SP extractable Fe ($p \leq 0.05$; $r = 0.55$), with a moderate negative correlation found between HWEC and DC extractable Fe

($p \leq 0.05$; r=-0.40). No significant interactions were found on the Woodland samples between CWEC, HWEC and the other variables ($p > 0.05$).

3.3 Iron Extraction Analysis

The t-test comparing the Fe extractions of the Incubation samples and the Plant Growth samples revealed that the majority of the ochre treated Plant Growth samples had significantly greater SP and DC extractable Fe concentrations than the ochre treated Incubation samples ($p \leq 0.05$) (Figure 3). No difference was found in the AO extractable Fe between the ochre treated samples ($p > 0.05$) (Figure 3). For the Agricultural control, it was found that the Plant Growth samples had significantly greater SP and DC extractable Fe concentrations, with the Woodland Plant Growth control samples having significantly greater Fe concentrations for all extractions than the corresponding Incubation samples ($p \leq 0.05$) (Figure 3).

4 Discussion

As hypothesised, Fe oxide amendments to soils did not have any adverse effects on plant growth, as measured by shoot biomass (Figure 5, Section 3). The majority of the Fe oxide amendments to soils had no effect on C availability, as measured by CWEC and HWEC, with the exception of the ochre treated Woodland Soils which reduced the CWEC concentration, relative to the control, decreasing labile C availability (Figure 1, Section 3). In the majority of treatments, the ochres had no effect on the distribution of aggregate sizes, somewhat supporting the hypothesis that the Fe oxide amendments would increase the proportion of macroaggregates produced, given that no increase in finer sized particles was observed, despite the addition of the ochres which are fine grained (Figure 4, Section 3).

4.1 Plant Growth

The additions of Fe oxides to soils did not have any improving or adverse effects on plant growth, relative to the control treatments, as measured by wheat shoot biomass (Figure 4, Section 3), despite the majority of the ochre treatments reducing the levels of soil extractable P, as measured by the Olsen P extraction, relative to the controls (Figure 2, Section 3). Ochres are known to have an affinity for P adsorption, having been found effective in P removal in constructed wetlands, wastewater and agricultural runoff (Heal *et al.*, 2003; Heal *et al.*, 2005; Dobbie *et al.*, 2009). The reduced release of P into solution in the adsorption experiment (Chapter 3, Section 3.3) and the reduced Olsen P extractable P in the incubation and plant growth experiments are both consistent with sorption of P by Fe oxides which is known to occur on the basis of previous studies (Dobbie *et al.*, 2009). Phosphorus is often a limiting nutrient (Lajtha and Jarrell, 1999), and so the reduced P levels seemingly having no effect on plant growth in the ochre amended soils is an unexpected but promising result.

The ochre amendments could have added some potentially harmful elements to the soils, given the known presence of potential toxic metals in ochre deposits (Table 1: Chapter 3, Section 2.1) and the findings of the adsorption experiments, that some of the ochre treated soils released more metals into solution than the controls (Chapter 3, Section 3.2). Subsequently, there is a concern that the addition of ochre to soils may lead to harmful increases in the concentrations of metals. However, this does not seem to have impacted plant growth in the ochre treated soils, relative to the controls, but it does possibly merit additional experiments to analyse for potential toxic metal uptake in the plant shoot biomass.

In the experiments carried out by Olimah *et al.* (2015), studying whether ochre has the potential to be a remedial treatment for arsenic contaminated soils, it was also found that ochre amendments to soils had no effect on plant growth, as measured by rye grass biomass. However, some studies have found that additions of Fe oxide have improved plant growth: Rizwan *et al.* (2019) found that Fe oxide nanoparticles improved the plant growth, increasing the lengths of shoot and roots of wheat (*Triticum aestivum*) with plant height, spike length, and dry weights of shoots, roots, spikes, and grains increased in particular with the higher rates of Fe oxide nanoparticles. A possible reason for the differing results found Rizwan *et al.* (2019) is that the soil was Fe poor (198 mg/kg (Hussain *et al.*, 2018)) and therefore Fe was a limiting nutrient, whereas for the soils used in the present study were not Fe poor (4040 ± 50.1 mg/kg and 2470 ± 15.8 mg/kg for the Agricultural Soil and Woodland Soil, respectively).

Briat *et al.* (2007) and Liu and Lal (2015) provide a possible explanation of increased plant growth with Fe oxide nanoparticles: the nanoparticles provide a great potential for the supply of nutrients to the plants, these nutrients, such as Fe, stimulate the biosynthesis of chlorophyll and redox process in plants which may positively affect the plant growth. A possible reason for the Fe oxide amendments having no beneficial effects on plant growth is that they are not small enough to enter plant cells; it is possible for nanoparticles to directly enter plant cells through the sieve-like cell wall structures if the particle sizes are smaller than the sizes of cell wall pores (5 to 20 nm) (Liu and Lal, 2015). The ochres have only 1.70 - 15.1 % particles in the size range of <2 µm (Table 1: Chapter 3, Section 2.1); nanoparticles have particle sizes ranging between 0.001 and 0.1 µm in size (Liu and Lal, 2015). Furthermore, the nanoparticles have a higher surface area:mass ratio and are therefore more readily dissolved for uptake of aqueous Fe.

4.2 Available Carbon

The majority of the Fe oxide amendments had no effect on extractable C content of the soil, relative to the control samples (Figure 1, Section 3). This is supported by the fact that, in general, few significant correlations were found in both the Incubation and Plant Growth Soils between extractable C and extractable Fe, with the significant correlations having low r values. However, the ochre treated Woodland Incubation Soils all reduced CWEC, the labile SOC fraction, relative to the control (Figure 1, Section 3), indicating that the amendments to these soils decreased C availability, increasing C storage (Ghani *et al.*, 2003). No difference was found

in soil respiration between the ochre treated soils and the respective controls (Table 2, Section 3), meaning that in general, all the soils respired at the same rate, suggesting that CO_2 was released at the same rate from all treatments and indicating that the biological functionality of the soil has not been impacted. In general, the ochre amendments had no effect on soil C availability, however in Woodland Soils, these two measures, extractable C and soil respiration, give differing results, suggesting that the C moves from a very labile to a less labile pool, whilst still being sufficiently available for soil bacteria respiration to not be reduced.

A possible reason for the treated Woodland Soil reducing CWEC, decreasing C availability is that it has a greater organic content (Total C 5.63 %) compared to the Agricultural Soil (Total C 3.69 %) (Table 1: Chapter 3, Section 2.1). Kalbitz and Kaiser (2008) found DOM-derived C to significantly contribute to stable OM accumulation and organic C storage in their studied woodland mineral soils. Cotrufo *et al.* (2019) found that the share of C between mineral-associated and particulate OM affects soil C stocks and mediates the effects of other variables on soil C stocks. In the Plant Growth Soils, the Woodland control had more CWEC than the Agricultural control, as well as having more HWEC than the Agricultural control in the Incubation and Plant Growth Soils – this indicates that as well as having more C than the Agricultural Soil, the C was more mobile and therefore possibly more able to interact with the ochre. The Woodland samples had additional deionised water added to them prior to incubation – this could have increased the mixing of soil and ochre, further providing extra chances of contact between the Fe oxides and the DOC, as discussed next.

Possible reasons for the ochre amendments to the Agricultural Soil having no effect on C availability are associated with time and mixing. The adsorption experiments (Chapter 3) had a high liquid to solid ratio, giving the Fe oxides a good chance of coming into contact with the DOC, whereas in the incubation and plant growth experiments, there was a much lower liquid to solid ratio, limiting these chances of contact. A possible solution for these problems in practical applications would be to plough the ochre into the soil as a slurry allowing the chances of contact to increase, however, this would increase the transport costs, as well as the water costs. Ploughing continues to be a widely used farming practice in most agricultural soils, but it is known that ploughing is destructive to soils and SOM (Six *et al.*, 1999; West and Post, 2002; Baker *et al.*, 2007).

However, in both the Incubation and the Plant Growth Soils, the ochre treated Agricultural Soils tended to have lower extractable C concentrations than the Woodland Soils, which was found to correspond with the Agricultural Soils having greater extractable Fe concentrations than the Woodland Soils. There are some significant correlations between extractable C and DC extractable Fe in the Agricultural Plant Growth Soils, with more significant correlations in the Agricultural Plant Growth Soils than the Woodland Plant Growth Soils, supporting this idea. These correlations are consistent with the goethite mineralogy of the ochres which, as mentioned in Chapter 3, Section 4.1, has a high adsorption capacity for SOC, enhancing soil C storage (Ohno *et al.*, 2007; Liu *et al.*, 2014). These results suggest that overall, the greater quantity of extractable Fe in the ochre amended Agricultural Soils are adsorbing more DOC to

their surfaces, decreasing C lability in solution (Kleber *et al.*, 2021).

4.3 Extractable Fe

Quantities of finely divided crystalline Fe solid phases and non-crystalline inorganic forms of Fe can be estimated by subtracting the concentrations of AO Fe from DC Fe and SP Fe from AO Fe, respectively, with organic complexed Fe represented by the concentration of SP Fe (Loeppert and Inskeep, 1996). As it was found that the majority of the ochre treated Plant Growth samples had greater SP and DC extractable Fe concentrations than the ochre treated Incubation samples, it can be estimated that the form of Fe changes, with an increase in finely divided crystalline Fe solid phases and organic complexed Fe, but a decrease in non-crystalline inorganic forms of Fe. This means the ochre is either crystallising or dissolving and associating with the organic C. The finely divided crystalline Fe solid phases may be present as goethite which, as mentioned in Chapter 3, Section 4.1 has a high adsorption capacity for SOC, which could enhance C storage (Ohno *et al.*, 2007; Liu *et al.*, 2014).

4.4 Aggregate Size

Soils having a high content of Fe oxides are usually characterised by very stable soil structure (Russel, 1971). Microaggregates are bound together into stable macroaggregates of >250 µm diameter (Tisdall and Oades, 1982). Chesters *et al.* (1957) found that Fe oxide showed a marked effect on soil aggregation, with a tendency to be more important in the smaller aggregate size range. It was hypothesised that the ochre amendments to soils would improve soil structure as seen by an increased proportion of macroaggregates within aggregate fraction sizes. Figure 6 depicts the possible aggregation scenarios of the ochre amendments, as discussed below.

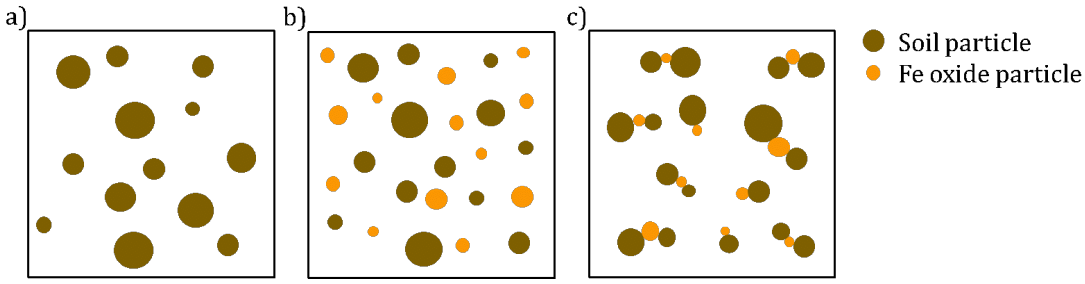

Figure 6: Aggregate distributions in (a) soils alone, (b) soil and ochre mixtures with no interactions and (c) soil and ochre mixtures where ochres have supported the aggregation of material to form macroaggregates.

The ochre amendments to Incubation and Plant Growth Soils generally showed no effects on aggregate fraction size, as compared to the respective controls, with the exception of the 0.005 ochre treatments in the Plant Growth Soils, in which the ochre treatments had a greater proportion of 53 - 250 µm and <53 µm size fractions and a smaller proportion of the 1000 - 2000 µm size fraction (Figure 4, Section 3). Comparisons between the 0.005 and 0.05, low and

high, ochre treatments generally showed no consistent pattern in aggregate size proportions. A possible explanation for the difference within the 0.005 ochre treated Plant Growth Soils is that the addition of the ochre has caused there to be more fine-grained material in these samples (Figure 6b), relative to the control samples (Figure 6a). This increase in the proportion of smaller aggregate size fractions possibly indicates that there has been no interaction between the soils and the ochres (Figure 6b). However, as this pattern was only seen in the Plant Growth Soils and not in the Incubated Soils, it could be inferred that this difference exists as a result of the presence of plant roots in the Plant Growth samples. Kumar *et al.* (2017) reported that aggregate stability tended to decrease with increasing vegetation density, perhaps because the large macroaggregates were fragmented by growing roots.

As the majority of the treatments showed no significant differences within aggregate fraction size proportions, it could be inferred that the fine ochres have supported the aggregation of material to form coarse, stable macroaggregates (Figure 6c). In the ochre treated Incubation samples, in the absence of plant roots, the fine-grained ochres promoted aggregation; in the Plant Growth ochre treated samples, these aggregates proved to be stable in the 0.05 treatment, however there was potentially insufficient ochre in the 0.005 treatments to have a lasting effect. The ochres have only 1.70 - 15.1 % and 34.1 - 60.8 % particles in the size ranges of <2 µm and 2 - 20 µm (Table 1: Chapter 3, Section 2.1), providing evidence of their fine-grained clay-silt properties. Approximately 54.7 %, 92.0 % and 66.5 % of the particles in the Morlais, Lynemouth and Polkemmet ochres, respectively, are in the size range of <53 µm, with approximately 37.7 %, 7.99 % and 31.2 % in the size range of 53 - 250 µm. Averages of these approximations were used to calculate the expected <53 µm and 53 - 250 µm percentage size fractions when accounting for the addition of the fine-grained ochres, at 0.005 and 0.05 treatment levels, to the soils. A single samples t-test revealed that there was largely a significant difference between the expected percentage of the <53 µm and 53 - 250 µm size fractions and the actual percentage size fractions, achieved from the water stable aggregate sieving analysis ($p \leq 0.05$), as seen in Table 3, which highlights the ochre amended soils that were found to be significantly different to their corresponding expected size fraction at the same treatment level. In the majority of cases, the value of the expected percentage size fraction was larger than the actual percentage size fraction value (Table 3), reinforcing the idea that, in most ochre amended soils, the ochres may have supported the aggregation of material to form macroaggregates (Figure 6c), rather than having no interaction with the soils and increasing the proportion of fine-grained microaggregates (Figure 6b). However, as not all ochres treatments followed this trend, this analysis warrants for further experiments which assess the effect of ochre amendments on soil aggregation; it would also be of key interest to analyse the organic C content of each aggregate size fraction.

Table 3: Comparison of actual ochre amended soil aggregate size fraction (% by mass) against estimated expected size fraction. Ochre amendments at the 0.005 treatment level are compared to the expected 0.005 ochre amended size fraction and ochre amendments at the 0.05 treatment level are compared to the expected 0.05 ochre amended size fraction. Different letters (a - b) represent a significant difference in actual size fraction of the ochre amended soils and the corresponding expected size fraction (single samples t-test), at $p \leq 0.05$. Data highlighted in bold are significant results where the percentage size fraction of the ochre amended soils is significantly lower than the expected size fraction ($p \leq 0.05$).

		Agricultural Incubation Soil		**Woodland Incubation Soil**		**Agricultural Plant Growth Soil**		**Woodland Plant Growth Soil**	
		Aggregate Size (% by Mass)							
		53 - 250 µm	<53 µm	53 - 250 µm	<53 µm	53 - 250 µm	<53 µm	53 - 250 µm	<53 µm
Control	**Actual Size Fraction (%)**	44.39	37.75	61.49	20.85	58.24	24.51	63.67	11.27
Expected 0.005 Ochre Amended Size Fraction (%)		*44.45*[a]	*37.89*[a]	*61.57*[a]	*20.92*[a]	*58.31*[a]	*24.59*[a]	*63.76*[a]	*11.31*[a]
Expected 0.05 Ochre Amended Size Fraction (%)		*44.96*[a]	*39.09*[a]	*62.28*[a]	*21.59*[a]	*58.98*[a]	*25.38*[a]	*64.49*[a]	*11.67*[a]
Morlais	**0.005**	49.71[b]	**31.74[b]**	62.88[ab]	**18.19[b]**	56.30[a]	27.33[b]	**56.71[b]**	**8.59[b]**
	0.05	45.23[a]	**36.27[b]**	66.29[b]	**14.76[b]**	59.16[a]	**23.67[b]**	**62.80[b]**	10.81[a]
Lynemouth	**0.005**	45.53[a]	**32.72[b]**	62.46[b]	22.36[b]	**52.78[b]**	28.98[b]	**58.59[b]**	**9.52[b]**
	0.05	49.11[b]	**35.18[b]**	62.14[a]	**19.70[b]**	**54.77[b]**	**23.66[b]**	**62.14[b]**	11.31[a]
Polkemmet	**0.005**	49.20[b]	**33.61[b]**	60.38[a]	20.77[a]	57.39[a]	25.68[a]	**56.59[b]**	**9.06[b]**
	0.05	47.97[b]	**32.26[b]**	61.07[a]	22.23[a]	**55.95[b]**	**22.91[b]**	**62.17[b]**	**9.87[b]**

4.5 Conclusions

The lack of significance within the extractable C results, but the promising reduction in CWEC in the ochre treated Woodland Soils, warrants for further experiments to take place; these experiments require a longer incubation period, with thorough mixing, to ensure there is an increased opportunity of contact between the Fe oxide and DOC. However, it is promising that the ochre amendments have had no effect on plant growth, implying that the addition of Fe oxide to soil would cause no damage to crop yield. Furthermore, the idea of the potential incorporation of the ochres into aggregates as evidenced by the analysis of the aggregate data is hopeful; the fine-grain materials of the ochres have not caused an increase in the proportions of the smaller aggregate size fractions, implying that the fine ochres may have supported the aggregation of material to form coarse, stable macroaggregates. It would be of interest to look into the organic C content of each aggregate size fraction.

Chapter 5: Summary and Recommendations

This thesis concludes that the addition of the ochre to soils enhances organic C sorption, reducing soil C availability and lability, as measured by the decrease of the release of C into solution in ochre amended soils, relative to control soils. It was found that ochres with a high goethite content and a relatively low - neutral pH were favourable in increasing SOC sorption as goethite has a high adsorption capacity for SOC and a lower soil pH signifies lower levels of OH^- and a decrease in desorption of SOC due to decreased negative charges on SOM. However, ochres generally contain $CaCO_3$ which was found to cause an increase in pH, leading to destabilisation of soil aggregates and a subsequent release of SOM. Further to this, ochres with a high Na content were found to perform less favourably due to the adverse effects of Na on soil dispersion and a decrease in aggregate stability.

As the adsorption experiments hence proved the additions of ochre to soil to be successful in adsorbing organic C, incubation/plant growth experiments were undertaken, understanding the effect of ochre amendments on C lability in a more realistic experiment than the adsorption study. The results of these experiments found an overall lack of difference in the extractable C content of ochre amended soils in comparison to control soils. However, the promising reduction in the cold water extractable C content in ochre treated Woodland Soils, relative to the Woodland Soil control, indicating decreased C availability and increased C storage, warrants for further experiments to take place. These experiments require a longer incubation period, with thorough mixing, to ensure there is an increased opportunity of contact between the Fe oxide and dissolved organic C. However, in practical applications, a possible solution for ensuring contact chances increase would be to plough the ochre into the soil as a slurry, however, this would increase the transport costs, as well as water costs. It is therefore important to understand the balance between the climate change benefit of increased SOC from ochre amended soils and any potential greenhouse gas emissions associated with possible these transport and water costs.

It was found that CO_2 was released at the same rate from ochre amended soils and control soils, indicating that ochre additions have not affected the biological functionality of the soil. The reduction in cold water extractable C content in the ochre treated Woodland Soils is not consistent with these respiration results however, suggesting that the C moves from a very labile to a less labile pool, whilst still being sufficiently available for soil bacteria respiration to not be reduced. Future experiments should measure these changes in the labile and less labile C pools.

An issue with the addition of ochre to soil is that it may be perceived as a waste disposal method. There is also a concern that ochre amendments to soils may lead to harmful increases in metal concentrations, given the known presence of potential toxic metals in ochre deposits and that in the adsorption experiments, some of the ochre treated soils released more metals into solution than the control. This does not seem to have impacted plant growth in the ochre treated soils, relative to the controls, but it does merit additional experiments that analyse for potential toxic metal uptake in the plant shoot biomass.

It is encouraging that the ochre amendments have had no effect on plant growth, implying that if added to agricultural and woodland soils at field scale, the addition of Fe oxide to soil would cause no damage to crop yield and flora growth. It is also promising that a potential incorporation of ochre into aggregates was found, implying that the fine ochres may have supported the aggregation of material to form coarse, stable macroaggregates, improving soil structure. It would be of interest to look into the organic C content of each aggregate size.

Appendices

A Accuracy and precision for ICP-OES elemental analysis

Table A1: Accuracy and precision for ICP-OES elemental analysis of metals and phosphorus. Precision is not reported for elements that were below detection.

	Accuracy (%)	Precision (%)
Cu	110.06	/
Zn	101.27	/
Pb	116.46	/
Al	116.53	/
B	92.74	72.22
Ba	119.06	75.86
Cd	101.53	/
Ag	157.21	/
Co	108.87	/
Cr	102.76	/
Fe	117.54	/
K	150.47	7.62
Li	92.67	/
Mg	105.59	25.84
Mn	111.70	76.07
Tl	116.51	/
P	ND	88.13
Na	86.70	78.65
Ni	101.60	/
As	ND	/

ND = not determined

B Extractable Al, Mn and Si

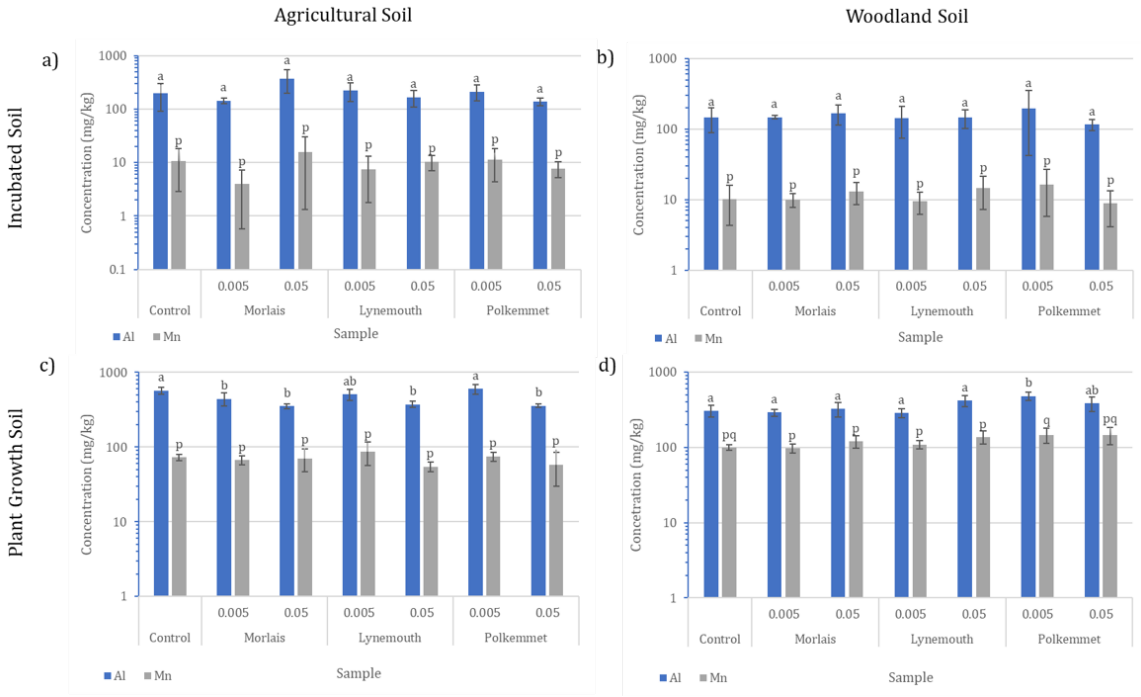

Figure B1: Mean concentrations (mg/kg) of extractable Al and Mn for sodium pyrophosphate (SP) (organically complexed Fe) extractions of (a) Agricultural incubation samples, (b) Woodland incubation samples, (c) Agricultural plant growth samples and (d) Woodland plant growth samples. Error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively. Different letters represent a significant difference in Al and Mn concentration (Al: a - b; Mn: p - q) for three different comparisons, between the control and the ochre treatments (one-way ANOVA and Dunnett's test;), between ochres of the same treatment level (two-way ANOVAs and Tukey tests) and between different treatment levels of the same ochre (two-way ANOVAs and Tukey tests), all at $p \leq 0.05$.

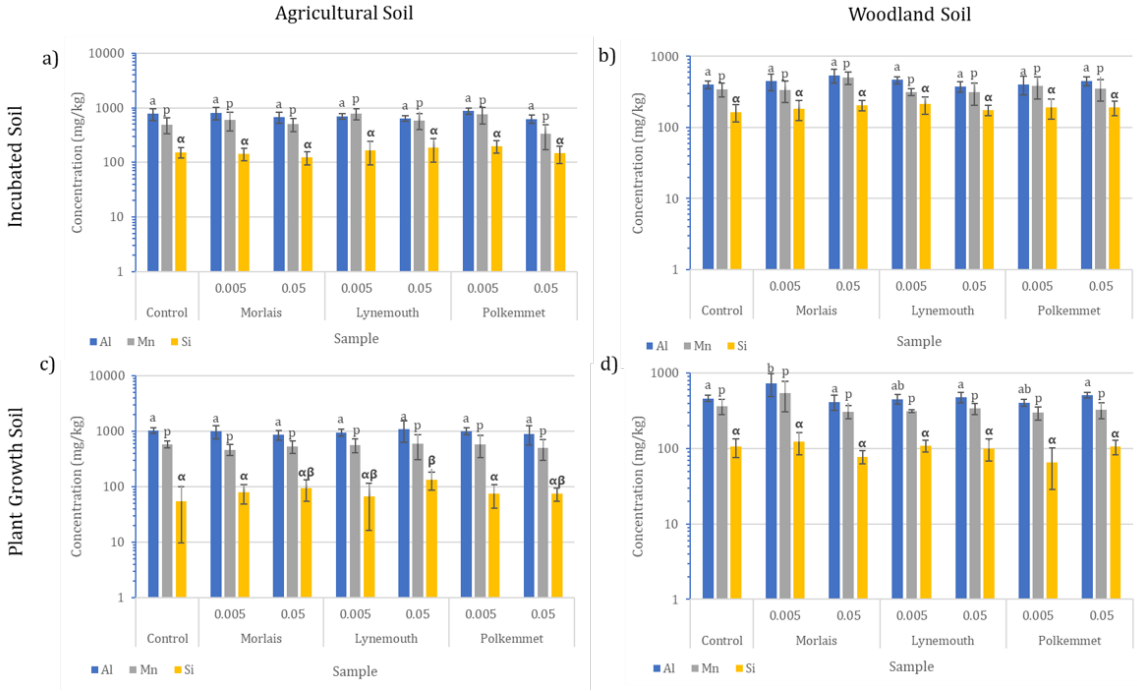

Figure B2: Mean concentrations (mg/kg) of extractable Al, Mn and Si for ammonium oxalate (AO) (organically complexed + amorphous inorganic Fe) extractions of (a) Agricultural incubation samples, (b) Woodland incubation samples, (c) Agricultural plant growth samples and (d) Woodland plant growth samples. Error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively. Different letters represent a significant difference in Al, Mn and Si concentration (Al: a - b; Mn: p; Si: α - β) for three different comparisons, between the control and the ochre treatments (one-way ANOVA and Dunnett's test;), between ochres of the same treatment level (two-way ANOVAs and Tukey tests) and between different treatment levels of the same ochre (two-way ANOVAs and Tukey tests), all at $p \leq 0.05$.

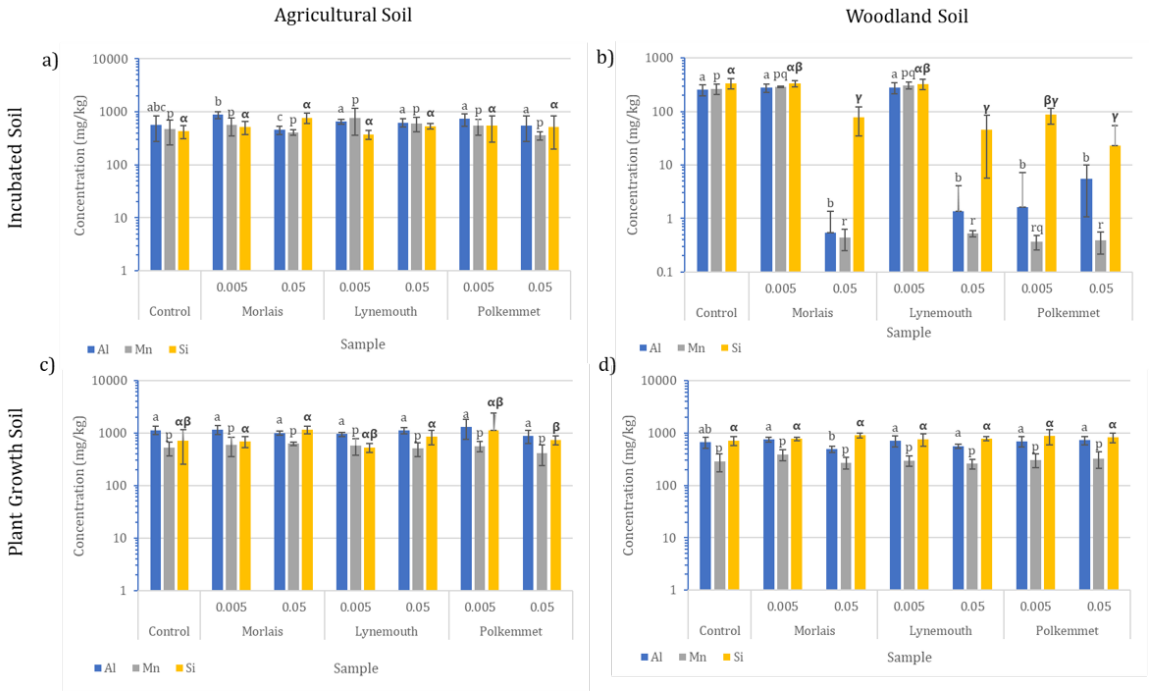

Figure B3: Mean concentrations (mg/kg) of extractable Al, Mn and Si for dithionite citrate (DC) (organically complexed, amorphous inorganic Fe + Fe oxides) extractions of (a) Agricultural incubation samples, (b) Woodland incubation samples, (c) Agricultural plant growth samples and (d) Woodland plant growth samples. Error bars represent standard deviation, n=5. 0.005 and 0.05 represent the treatments of dry mass ratios 0.005:1 and 0.05:1 ochre:soil, respectively. Different letters represent a significant difference in Al, Mn and Si concentration (Al: a - c; Mn: p - q; Si: α - γ) for three different comparisons, between the control and the ochre treatments (one-way ANOVA and Dunnett's test;), between ochres of the same treatment level (two-way ANOVAs and Tukey tests) and between different treatment levels of the same ochre (two-way ANOVAs and Tukey tests), all at $p \leq 0.05$.

References

Antelo, J., Arce, F., Avena, M., Fiol, S., López, R. and Macías, F., 2007. Adsorption of a soil humic acid at the surface of goethite and its competitive interaction with phosphate. Geoderma, 138(1-2), pp.12-19.

Arduino, E., Barberis, E. and Boero, V., 1989. Iron oxides and particle aggregation in B horizons of some Italian soils. Geoderma, 45(3-4), pp.319-329.

Aumonier, J., 2020. Personal communication with Coal Authority. [Accessed 21 August 2020].

Baker, J.M., Ochsner, T.E., Venterea, R.T. and Griffis, T.J., 2007. Tillage and soil carbon sequestration—What do we really know?. Agriculture, ecosystems & environment, 118(1-4), pp.1-5.

Baldock, J.A. and Nelson, P.N., 2000. Soil organic matter. CRC press. Baldock, J.A. and Skjemstad, J.O., 2000. Role of the soil matrix and minerals in protecting natural organic materials against biological attack. Organic geochemistry, 31(7-8), pp.697-710.

Barberis, E., Marsan, F.A., Boero, V. and Arduino, E., 1991. Aggregation of soil particles by iron oxides in various size fractions of soil B horizons. Journal of Soil Science, 42(4), pp.535-542.

Beerling, D.J., Leake, J.R., Long, S.P., Scholes, J.D., Ton, J., Nelson, P.N., Bird, M., Kantzas, E., Taylor, L.L., Sarkar, B. and Kelland, M., 2018. Farming with crops and rocks to address global climate, food and soil security. Nature Plants, 4(3), pp.138-147.

Boisson, J., Mench, M., Vangronsveld, J., Ruttens, A., Kopponen, P. and De Koe, T., 1999. Immobilization of trace metals and arsenic by different soil additives: evaluation by means of chemical extractions. Communications in soil science and plant analysis, 30(3-4), pp.365-387.

Bowell, R.J., 1994. Sorption of arsenic by iron oxides and oxyhydroxides in soils. Applied geochemistry, 9(3), pp.279-286.

Briat, J.F., Curie, C. and Gaymard, F., 2007. Iron utilization and metabolism in plants. Current opinion in plant biology, 10(3), pp.276-282.

British Standard, 1995. BS 7755: section 3.9: 1995 ISO 11466: 1995 Soil quality Part 3. Chemical methods section 3.9 Extraction of trace elements soluble in aqua regia.

Bronick, C.J. and Lal, R., 2005. Soil structure and management: a review. Geoderma, 124(1-2), pp.3-22.

Brunauer, S., Emmett, P.H., Teller, E., 1938. Adsorption of gases in multimolecular layers. J. American Chemical Society, 60, 309-319.

Cambardella, C.A. and Elliott, E.T., 1993. Methods for physical separation and characterization of soil organic matter fractions. In Soil Structure/Soil Biota Interrelationships (pp. 449-457). Elsevier.

Canadell, J.G. and Raupach, M.R., 2008. Managing forests for climate change mitigation. science, 320(5882), pp.1456-1457.

Canadell, J.G., Le Quéré, C., Raupach, M.R., Field, C.B., Buitenhuis, E.T., Ciais, P., Conway, T.J., Gillett, N.P., Houghton, R.A. and Marland, G., 2007. Contributions to

accelerating atmospheric CO_2 growth from economic activity, carbon intensity, and efficiency of natural sinks. Proceedings of the national academy of sciences, 104(47), pp.18866-18870.

Chesters, G., Attoe, O.J. and Allen, O.N., 1957. Soil aggregation in relation to various soil constituents. Soil Science Society of America Journal, 21(3), pp.272-277.

Cotrufo, M.F., Ranalli, M.G., Haddix, M.L., Six, J. and Lugato, E., 2019. Soil carbon storage informed by particulate and mineral-associated organic matter. Nature Geoscience, 12(12), pp.989-994.

Dobbie, K.E., Heal, K.V., Aumonier, J., Smith, K.A., Johnston, A. and Younger, P.L., 2009. Evaluation of iron ochre from mine drainage treatment for removal of phosphorus from wastewater. Chemosphere, 75(6), pp.795-800.

Doi, M., Warren, G. and Hodson, M.E., 2005. A preliminary investigation into the use of ochre as a remedial amendment in arsenic-contaminated soils. Applied Geochemistry, 20(12), pp.2207-2216.

Elkhatib, E.A., Bennett, O.L. and Wright, R.J., 1984a. Arsenite sorption and desorption in soils. Soil Science Society of America Journal, 48(5), pp.1025-1030.

Elkhatib, E.A., Bennett, O.L. and Wright, R.J., 1984b. Kinetics of arsenite sorption in soils. Soil Science Society of America Journal, 48(4), pp.758-762.

El-Swaify, S.A. and Emerson, W.W., 1975. Changes in the physical properties of soil clays due to precipitated aluminum and iron hydroxides: I. Swelling and aggregate stability after drying. Soil Science Society of America Journal, 39(6), pp.1056-1063.

European Commission, 2013. Directive 2008/105/EC of the European Parliament and of the Council of 16 December 2008 on environmental quality standards in the field of water policy, amending and subsequently repealing Council Directives 82/176/EEC, 83/513/EEC, 84/156/EEC, 84/491/EEC, 86/280/EEC and amending Directive 2000/60/EC of the European Parliament and of the Council. Current consolidated version: 13/09/2013, 15(006), pp.84-97. Available at: http://data.europa.eu/eli/dir/2008/105/oj [Accessed: 20/07/2021].

FAO (2021). ProdStat. Core Production Data Base, Electronic Resource http://faostat.fao.org/ (accessed 13.05.21).

Fenton, O., Kirwan, L. and Healy, M.G., 2012. The effectiveness and feasibility of using ochre as a soil amendment to sequester dissolved reactive phosphorus in runoff. Water, Air, & Soil Pollution, 223(3), pp.1249-1261.

Ford, R.G., 2002. Rates of hydrous ferric oxide crystallization and the influence on coprecipitated arsenate. Environmental Science & Technology, 36(11), pp.2459-2463.

Fordham, A.W. and Norrish, K., 1983. The nature of soil particles particularly those reacting with arsenate in a series of chemically treated samples. Soil Research, 21(4), pp.455-477.

Fujii, K., Hayakawa, C., Inagaki, Y. and Ono, K., 2019. Sorption reduces the biodegradation rates of multivalent organic acids in volcanic soils rich in short-range order minerals. Geoderma, 333, pp.188-199.

Garcıa-Sa´nchez, A., Alastuey, A. and Querol, X., 1999. Heavy metal adsorption by

different minerals: application to the remediation of polluted soils. Science of the total environment, 242(1-3), pp.179-188.

Garcia-Sanchez, A., Alvarez-Ayuso, E. and Rodriguez-Martin, F., 2002. Sorption of As (V) by some oxyhydroxides and clay minerals. Application to its immobilization in two polluted mining soils. Clay Minerals, 37(1), pp.187-194.

Gentsch, N., Mikutta, R., Shibistova, O., Wild, B., Schnecker, J., Richter, A., Urich, T., Gittel, A., Šantrůčková, H., Bárta, J. and Lashchinskiy, N., 2015. Properties and bioavailability of particulate and mineral-associated organic matter in Arctic permafrost soils, Lower Kolyma Region, Russia. European Journal of Soil Science, 66(4), pp.722-734.

Ghani, A., Dexter, M. and Perrott, K.W., 2003. Hot-water extractable carbon in soils: a sensitive measurement for determining impacts of fertilisation, grazing and cultivation. Soil biology and biochemistry, 35(9), pp.1231-1243.

Gill, R., 1997. Modern Analytical Geochemistry. Longman, Harlow, UK. Goldberg, S. and Johnston, C.T., 2001. Mechanisms of arsenic adsorption on amorphous oxides evaluated using macroscopic measurements, vibrational spectroscopy, and surface complexation modeling.

Goldberg, S., 2002. Competitive adsorption of arsenate and arsenite on oxides and clay minerals. Soil Science Society of America Journal, 66(2), pp.413-421.

Grafe, M.J.E.A.M., Eick, M.J. and Grossl, P.R., 2001. Adsorption of arsenate (V) and arsenite (III) on goethite in the presence and absence of dissolved organic carbon. Soil Science Society of America Journal, 65(6), pp.1680-1687.

Gupta, R.K., Bhumbla, D.K. and Abrol, I.P., 1984. Effect of sodicity, pH, organic matter, and-calcium carbonate on the dispersion behavior of soils. Soil Science, 137(4), pp.245-251.

Heal, K., Younger, P.L., Smith, K., Glendinning, S., Quinn, P.F. and Dobbie, K., 2003. Novel use of ochre from mine water treatment plants to reduce point and diffuse phosphorus pollution. Land Contamination & Reclamation.

Heal, K.V., Dobbie, K.E., Bozika, E., McHaffie, H., Simpson, A.E. and Smith, K.A., 2005. Enhancing phosphorus removal in constructed wetlands with ochre from mine drainage treatment. Water Science and Technology, 51(9), pp.275-282.

Heckman, K., Welty-Bernard, A., Rasmussen, C. and Schwartz, E., 2009. Geologic controls of soil carbon cycling and microbial dynamics in temperate conifer forests. Chemical Geology, 267(1-2), pp.12-23.

Hiemstra, T. and Van Riemsdijk, W.H., 1996. A surface structural approach to ion adsorption: the charge distribution (CD) model. Journal of colloid and interface science, 179(2), pp.488-508.

Houba, V.J.G., Temminghoff, E.J.M., Gaikhorst, G.A. and Van Vark, W., 2000. Soil analysis procedures using 0.01 M calcium chloride as extraction reagent. Communications in soil science and plant analysis, 31(9-10), pp.1299-1396.

Hussain, A., Ali, S., Rizwan, M., ur Rehman, M.Z., Javed, M.R., Imran, M., Chatha, S.A.S. and Nazir, R., 2018. Zinc oxide nanoparticles alter the wheat physiological response and reduce the cadmium uptake by plants. Environmental Pollution, 242, pp.1518-1526.

Igwe, C.A., Akamigbo, F.O.R. and Mbagwu, J.S.C., 1999. Chemical and mineralogical properties of soils in southeastern Nigeria in relation to aggregate stability. Geoderma, 92(1-2), pp.111-123.

Igwe, C.A., Zarei, M. and Stahr, K., 2009. Colloidal stability in some tropical soils of southeastern Nigeria as affected by iron and aluminium oxides. Catena, 77(3), pp.232-237.

Imhoff, S., Da Silva, A.P. and Dexter, A., 2002. Factors contributing to the tensile strength and friability of Oxisols. Soil Science Society of America Journal, 66(5), pp.1656-1661.

Jackson, B.P. and Miller, W.P., 2000. Effectiveness of phosphate and hydroxide for desorption of arsenic and selenium species from iron oxides. Soil Science Society of America Journal, 64(5), pp.1616-1622.

Jain, A., Raven, K.P. and Loeppert, R.H., 1999. Arsenite and arsenate adsorption on ferrihydrite: Surface charge reduction and net OH-release stoichiometry. Environmental Science & Technology, 33(8), pp.1179-1184.

Jastrow, J.D., 1996. Soil aggregate formation and the accrual of particulate and mineral-associated organic matter. Soil Biology and Biochemistry, 28(4-5), pp.665-676.

Jilling, A., Keiluweit, M., Contosta, A.R., Frey, S., Schimel, J., Schnecker, J., Smith, R.G., Tiemann, L. and Grandy, A.S., 2018. Minerals in the rhizosphere: overlooked mediators of soil nitrogen availability to plants and microbes. Biogeochemistry, 139(2), pp.103-122.

Jilling, A., Keiluweit, M., Gutknecht, J.L. and Grandy, A.S., 2021. Priming mechanisms providing plants and microbes access to mineral-associated organic matter. Soil Biology and Biochemistry, 158, p.108265.

Jim, C.Y., 2003. Conservation of soils in culturally protected woodlands in rural Hong Kong. Forest ecology and Management, 175(1-3), pp.339-353.

Jones, D.L. and Edwards, A.C., 1998. Influence of sorption on the biological utilization of two simple carbon substrates. Soil Biology and Biochemistry, 30(14), pp.1895-1902.

Kabata-Pendias, A., 2004. Soil–plant transfer of trace elements—an environmental issue. Geoderma, 122(2-4), pp.143-149.

Kaiser, K. and Guggenberger, G., 2003. Mineral surfaces and soil organic matter. European Journal of Soil Science, 54(2), pp.219-236.

Kalbitz, K. and Kaiser, K., 2008. Contribution of dissolved organic matter to carbon storage in forest mineral soils. Journal of Plant Nutrition and Soil Science, 171(1), pp.52-60.

Kalbitz, K., Schwesig, D., Rethemeyer, J. and Matzner, E., 2005. Stabilization of dissolved organic matter by sorption to the mineral soil. Soil Biology and Biochemistry, 37(7), pp.1319-1331.

Kemper, W.D. and Koch, E.J., 1966. Aggregate stability of soils from Western United States and Canada: Measurement procedure, correlations with soil constituents (No. 1355). Agricultural Research Service, US Department of Agriculture.

Kemper, W.D. and Rosenau, R.C., 1986. Aggregate stability and size distribution. Agronomy (USA).

Kitagawa, Y., 1983. Goethite and hematite in some solls from the amazon region. Soil

Science and Plant Nutrition, 29(2), pp.209-217.

Kleber, M., Bourg, I.C., Coward, E.K., Hansel, C.M., Myneni, S.C. and Nunan, N., 2021. Dynamic interactions at the mineral–organic matter interface. Nature Reviews Earth & Environment, 2(6), pp.402-421.

Kleber, M., Eusterhues, K., Keiluweit, M., Mikutta, C., Mikutta, R. and Nico, P.S., 2015. Mineral–organic associations: formation, properties, and relevance in soil environments. In Advances in agronomy (Vol. 130, pp. 1-140). Academic Press.

Kleber, M., Eusterhues, K., Keiluweit, M., Mikutta, C., Mikutta, R. and Nico, P.S., 2015. Mineral–organic associations: formation, properties, and relevance in soil environments. Advances in agronomy, 130, pp.1-140.

Kleber, M., Mikutta, R., Torn, M.S. and Jahn, R., 2005. Poorly crystalline mineral phases protect organic matter in acid subsoil horizons. European Journal of Soil Science, 56(6), pp.717725.

Köchy, M., Hiederer, R. and Freibauer, A., 2015. Global distribution of soil organic carbon–Part 1: Masses and frequency distributions of SOC stocks for the tropics, permafrost regions, wetlands, and the world. Soil, 1(1), pp.351-365.

Kumar, A., Dorodnikov, M., Splettstößer, T., Kuzyakov, Y. and Pausch, J., 2017. Effects of maize roots on aggregate stability and enzyme activities in soil. Geoderma, 306, pp.50-57.

Lajtha, K. and Jarrell, W.M., 1999. Soil phosphorus. Standard soil methods for long-term ecological research. Oxford University Press, New York, pp.115-142.

Lal, R., 2004. Soil carbon sequestration impacts on global climate change and food security. science, 304(5677), pp.1623-1627.

Lebrun, M., Nandillon, R., Miard, F., Le Forestier, L., Morabito, D. and Bourgerie, S., 2021. Effects of biochar, ochre and manure amendments associated with a metallicolous ecotype of Agrostis capillaris on As and Pb stabilization of a former mine technosol. Environmental Geochemistry and Health, 43, pp.1491-1505.

Levy, G.J. and Torrento, J.R., 1995. Clay dispersion and macroaggregate stability as affected by exchangeable potassium and sodium. Soil science, 160(5), pp.352-358.

Liu, H., Chen, T. and Frost, R.L., 2014. An overview of the role of goethite surfaces in the environment. Chemosphere, 103, pp.1-11.

Liu, R. and Lal, R., 2015. Potentials of engineered nanoparticles as fertilizers for increasing agronomic productions. Science of the total environment, 514, pp.131-139.

Livesey, N.T. and Huang, P.M., 1981. Adsorption of arsenate by soils and its relation to selected chemical properties and anions. Soil Science, 131(2), pp.88-94.

Loeppert, R., Inskeep, W.P., 1996. Iron. In: Sparks, D.L. (Ed.), Methods of Soil Analysis: Chemical Methods. Part 3. Book Series 5, Soil Science Society of America, Wisconsin, USA, pp. 639-664.

Lu, M., Cao, X., Pan, J., Gurajala, H.K., He, Z., Yang, X. and Khan, M.B., 2020. Genotypic variations in zinc accumulation and bioaccessibility among wheat (Triticum aestivum L.) genotypes under two different field conditions. Journal of Cereal Science, 93, p.102953.

Lützow, M.V., Kögel-Knabner, I., Ekschmitt, K., Matzner, E., Guggenberger, G., Marschner, B. and Flessa, H., 2006. Stabilization of organic matter in temperate soils: mechanisms and their relevance under different soil conditions–a review. European journal of soil science, 57(4), pp.426-445.

Manning, B.A. and Goldberg, S., 1996. Modeling competitive adsorption of arsenate with phosphate and molybdate on oxide minerals. Soil Science Society of America Journal, 60(1), pp.121-131.

Manning, B.A. and Suarez, D.L., 2000. Modeling arsenic (III) adsorption and heterogeneous oxidation kinetics in soils. Soil Science Society of America Journal, 64(1), pp.128-137.

Manning, B.A., Fendorf, S.E. and Goldberg, S., 1998. Surface structures and stability of arsenic (III) on goethite: spectroscopic evidence for inner-sphere complexes.

Masiello, C.A., Chadwick, O.A., Southon, J., Torn, M.S. and Harden, J.W., 2004. Weathering controls on mechanisms of carbon storage in grassland soils. Global Biogeochemical Cycles, 18(4).

Matis, K.A., Zouboulis, A.I., Malamas, F.B., Afonso, M.R. and Hudson, M.J., 1997. Flotation removal of As (V) onto goethite. Environmental Pollution, 97(3), pp.239-245.

Mbagwu, J.S.C. and Schwertmann, U., 2006. Some factors affecting clay dispersion and aggregate stability in selected soils of Nigeria. International agrophysics, 20(1).

McKnight, D.M., Bencala, K.E., Zellweger, G.W., Aiken, G.R., Feder, G.L. and Thorn, K.A., 1992. Sorption of dissolved organic carbon by hydrous aluminum and iron oxides occurring at the confluence of Deer Creek with the Snake River, Summit County, Colorado. Environmental science & technology, 26(7), pp.1388-1396.

McLean, J.E. and Bledsoe, B.E., 1992. Ground water issue, Behavior of metals in soils. Washington, DC: US Environmental Protection Agency, Office of Solid Waste and Emergency Response. EPA/540/S-92/018.

McVetty, P.B.E., Lukow, O.M., Hall, L.M., Rajcan, I. and Rahman, H., 2016. Grain production and consumption: Oilseeds in North America.

Mertz, S., Le Forestier, L., Bataillard, P. and Devau, N., 2021. Leaching of trace metals (Pb) from contaminated tailings amended with iron oxides and manure: New insight from a modelling approach. Chemical Geology, p.120356.

Mimmo, T., Del Buono, D., Terzano, R., Tomasi, N., Vigani, G., Crecchio, C., Pinton, R., Zocchi, G. and Cesco, S., 2014. Rhizospheric organic compounds in the soil–microorganism–plant system: their role in iron availability. European Journal of Soil Science, 65(5), pp.629-642.

Moni, C., Chabbi, A., Nunan, N., Rumpel, C. and Chenu, C., 2010. Spatial dependance of organic carbon–metal relationships: A multi-scale statistical analysis, from horizon to field. Geoderma, 158(3-4), pp.120-127.

Muggler, C.C., van Griethuysen, C., Buurman, P. and Pape, T., 1999. Aggregation, organic matter, and iron oxide morphology in Oxisols from Minas Gerais, Brazil. Soil Science, 164(10), pp.759-770.

Oades, J.M. and Waters, A.G., 1991. Aggregate hierarchy in soils. Soil Research, 29(6), pp.815-828.

Oades, J.M., 1984. Soil organic matter and structural stability: mechanisms and implications for management. Plant and soil, 76(1), pp.319-337.

Oades, J.M., 1989. An introduction to organic matter in mineral soils. Minerals in soil environments, 1, pp.89-159.

Ohno, T., Chorover, J., Omoike, A. and Hunt, J., 2007. Molecular weight and humification index as predictors of adsorption for plant-and manure-derived dissolved organic matter to goethite. European Journal of Soil Science, 58(1), pp.125-132.

Olimah, J.A., Shaw, L.J. and Hodson, M.E., 2015. Does ochre have the potential to be a remedial treatment for As-contaminated soils?. Environmental Pollution, 206, pp.150-158.

Olsen, S.R., 1954. Estimation of available phosphorus in soils by extraction with sodium bicarbonate (No. 939). US Department of Agriculture.

Peng, X., Yan, X., Zhou, H., Zhang, Y.Z. and Sun, H., 2015. Assessing the contributions of sesquioxides and soil organic matter to aggregation in an Ultisol under long-term fertilization. Soil and Tillage Research, 146, pp.89-98.

Porras, R.C., Pries, C.E.H., McFarlane, K.J., Hanson, P.J. and Torn, M.S., 2017. Association with pedogenic iron and aluminum: effects on soil organic carbon storage and stability in four temperate forest soils. Biogeochemistry, 133(3), pp.333-345.

Powlson, D.S., Whitmore, A.P. and Goulding, K.W., 2011. Soil carbon sequestration to mitigate climate change: a critical re-examination to identify the true and the false. European Journal of Soil Science, 62(1), pp.42-55.

Rady, M.M., Ahmed, S.M., El-Yazal, M.A.S. and Taie, H.A., 2019. Alleviation of cadmium stress in wheat by polyamines. In Cadmium Tolerance in Plants (pp. 463-496). Academic Press.

Ramesh, T., Bolan, N.S., Kirkham, M.B., Wijesekara, H., Kanchikerimath, M., Rao, C.S., Sandeep, S., Rinklebe, J., Ok, Y.S., Choudhury, B.U. and Wang, H., 2019. Soil organic carbon dynamics: Impact of land use changes and management practices: A review. Advances in agronomy, 156, pp.1-107.

Randtke, S.J., 1988. Organic contaminant removal by coagulation and related process combinations. Journal-American Water Works Association, 80(5), pp.40-56.

Rasmussen, C., Torn, M.S. and Southard, R.J., 2005. Mineral assemblage and aggregates control carbon dynamics in a California conifer forest. Soil Science Society of America Journal, 69(6), pp.1711-1721.

Rizwan, M., Ali, S., Ali, B., Adrees, M., Arshad, M., Hussain, A., ur Rehman, M.Z. and Waris, A.A., 2019. Zinc and iron oxide nanoparticles improved the plant growth and reduced the oxidative stress and cadmium concentration in wheat. Chemosphere, 214, pp.269-277.

Rowell, D.L., 1994. Soil Science: Methods and Applications. Prentice Hall, UK.

Rowley, M.C., Grand, S. and Verrecchia, É.P., 2018. Calcium-mediated stabilisation of soil organic carbon. Biogeochemistry, 137(1), pp.27-49.

Royal Society (2009) Geoengineering the climate: science, governance and uncertainty.

Rui, M., Ma, C., Hao, Y., Guo, J., Rui, Y., Tang, X., Zhao, Q., Fan, X., Zhang, Z., Hou, T. and Zhu, S., 2016. Iron oxide nanoparticles as a potential iron fertilizer for peanut (Arachis hypogaea). Frontiers in plant science, 7, p.815.

Russell, E.W., 1971. Soil structure: its maintenance and improvement 1. Journal of soil science, 22(2), pp.137-151.

Saidy, A.R., Smernik, R.J., Baldock, J.A., Kaiser, K., Sanderman, J. and Macdonald, L.M., 2012. Effects of clay mineralogy and hydrous iron oxides on labile organic carbon stabilisation. Geoderma, 173, pp.104-110.

Setia, R., Rengasamy, P. and Marschner, P., 2013. Effect of exchangeable cation concentration on sorption and desorption of dissolved organic carbon in saline soils. Science of the Total Environment, 465, pp.226-232.

Shang, C. and Tiessen, H., 1998. Organic matter stabilization in two semiarid tropical soils: size, density, and magnetic separations. Soil Science Society of America Journal, 62(5), pp.1247-1257.

Sibanda, H.M. and Young, S.D., 1986. Competitive adsorption of humus acids and phosphate on goethite, gibbsite and two tropical soils. Journal of Soil Science, 37(2), pp.197-204.

Sibrell, P.L., Montgomery, G.A., Ritenour, K.L. and Tucker, T.W., 2009. Removal of phosphorus from agricultural wastewaters using adsorption media prepared from acid mine drainage sludge. Water research, 43(8), pp.2240-2250.

Sims, J.T., 1986. Soil pH effects on the distribution and plant availability of manganese, copper, and zinc. Soil Science Society of America Journal, 50(2), pp.367-373.

Sims, J.T., 2000. Soil test phosphorus: Olsen P. Methods of phosphorus analysis for soils, sediments, residuals, and waters, p.20.

Six, J., Conant, R.T., Paul, E.A. and Paustian, K., 2002. Stabilization mechanisms of soil organic matter: implications for C-saturation of soils. Plant and soil, 241(2), pp.155-176.

Six, J., Elliott, E.T. and Paustian, K., 1999. Aggregate and soil organic matter dynamics under conventional and no-tillage systems. Soil Science Society of America Journal, 63(5), pp.1350-1358.

Six, J., Elliott, E.T. and Paustian, K., 2000a. Soil structure and soil organic matter II. A normalized stability index and the effect of mineralogy. Soil Science Society of America Journal, 64(3), pp.1042-1049.

Six, J., Paustian, K., Elliott, E.T. and Combrink, C., 2000b. Soil structure and organic matter I. Distribution of aggregate-size classes and aggregate-associated carbon. Soil Science Society of America Journal, 64(2), pp.681-689.

Smith, E., Naidu, R. and Alston, A.M., 2002. Chemistry of inorganic arsenic in soils: II. Effect of phosphorus, sodium, and calcium on arsenic sorption. Journal of Environmental Quality, 31(2), pp.557-563.

Smith, K.S. and Huyck, H.L., 1999. An overview of the abundance, relative mobility,

bioavailability, and human toxicity of metals. The environmental geochemistry of mineral deposits, 6, pp.29-70.

Sollins, P., Homann, P. and Caldwell, B.A., 1996. Stabilization and destabilization of soil organic matter: mechanisms and controls. Geoderma, 74(1-2), pp.65-105.

Stevenson, F.J., 1994. Humus chemistry: genesis, composition, reactions. John Wiley & Sons.

Suarez, D.L., Rhoades, J.D., Lavado, R. and Grieve, C.M., 1984. Effect of pH on saturated hydraulic conductivity and soil dispersion. Soil Science Society of America Journal, 48(1), pp.50-55.

Sun, X. and Doner, H.E., 1996. An investigation of arsenate and arsenite bonding structures on goethite by FTIR. Soil Science, 161(12), pp.865-872. Sun, X. and Doner, H.E., 1998. Adsorption and oxidation of arsenite on goethite. Soil Science, 163(4), pp.278-287.

Thien, S.J., 1979. A flow diagram for teaching texture-by-feel analysis. Journal of Agronomic education, 8(1), pp.54-55.

Tisdall, J.M. and Oades, J.M., 1982. Organic matter and water-stable aggregates in soils. Journal of soil science, 33(2), pp.141-163.

Torn M.S., Trumbore S.E., Chadwick O.A., Vitousek P.M. and Hendricks D.M. (1997) Mineral control of soil organic carbon storage and turnover. Nature, 389, pp.170-173.

Totsche, K.U., Amelung, W., Gerzabek, M.H., Guggenberger, G., Klumpp, E., Knief, C., Lehndorff, E., Mikutta, R., Peth, S., Prechtel, A. and Ray, N., 2018. Microaggregates in soils. Journal of Plant Nutrition and Soil Science, 181(1), pp.104-136.

UK Government (2020). https://www.gov.uk/government/news/ochre-from-mine-water-gets-new-lease-of-life-as-artists-pigment Government website (accessed 04.01.21).

Vieublé Gonod, L., Jones, D.L. and Chenu, C., 2006. Sorption regulates the fate of the amino acids lysine and leucine in soil aggregates. European Journal of Soil Science, 57(3), pp.320-329.

Wagai, R. and Mayer, L.M., 2007. Sorptive stabilization of organic matter in soils by hydrous iron oxides. Geochimica et Cosmochimica Acta, 71(1), pp.25-35.

Wagai, R., Kajiura, M. and Asano, M., 2020. Iron and aluminum association with microbially processed organic matter via meso-density aggregate formation across soils: organometallic glue hypothesis. SOIL Discussions, pp.1-42.

Waltham, C.A. and Eick, M.J., 2002. Kinetics of arsenic adsorption on goethite in the presence of sorbed silicic acid. Soil Science Society of America Journal, 66(3), pp.818-825.

Warren, G.P. and Alloway, B.J., 2003. Reduction of arsenic uptake by lettuce with ferrous sulfate applied to contaminated soil. Journal of Environmental Quality, 32(3), pp.767-772.

Warren, G.P., Alloway, B.J., Lepp, N.W., Singh, B., Bochereau, F.J.M. and Penny, C., 2003. Field trials to assess the uptake of arsenic by vegetables from contaminated soils and soil remediation with iron oxides. Science of the Total Environment, 311(1-3), pp.19-33.

Water Framework Directive, 2015. Water Framework Directive (Standards and Classification) Directions (England and Wales) 2015.

Weil, R.R. and Brady, N.C., 2016. Nature and Properties of Soils, Global Edition, Harlow, United Kingdom: Pearson Education UK.

Wen, Y., Xiao, J., Goodman, B.A. and He, X., 2019. Effects of Organic Amendments on the Transformation of Fe (Oxyhydr) Oxides and Soil Organic Carbon Storage. Frontiers in Earth Science, 7, p.257.

West, T.O. and Post, W.M., 2002. Soil organic carbon sequestration rates by tillage and crop rotation: a global data analysis. Soil Science Society of America Journal, 66(6), pp.1930-1946.

Whittinghill, K.A. and Hobbie, S.E., 2011. Effects of landscape age on soil organic matter processing in Northern Alaska. Soil Science Society of America Journal, 75(3), pp.907-917.

Wilcox, R.A.N.D., 1993. Robustness in ANOVA. Applied analysis of variance in behavioral science, 514.

Ye, L., Li, L., Wang, L., Wang, S., Li, S., Du, J., Zhang, S. and Shou, H., 2015. MPK3/MPK6 are involved in iron deficiency-induced ethylene production in Arabidopsis. Frontiers in Plant Science, 6, p.953.

Younger, P.L., Banwart, S.A. and Hedin, R.S., 2002. Mine water: hydrology, pollution, remediation (Vol. 5). Springer Science & Business Media.

Zargar, S.M., Agrawal, G.K., Rakwal, R. and Fukao, Y., 2015. Quantitative proteomics reveals role of sugar in decreasing photosynthetic activity due to Fe deficiency. Frontiers in plant science, 6, p.592.

Zhang, K., Wang, X., Wu, L., Lu, T., Guo, Y. and Ding, X., 2021. Impacts of salinity on the stability of soil organic carbon in the croplands of the Yellow River Delta. Land Degradation & Development, 32(4), pp.1873-1882.

Zhang, W., Wang, X., Lu, T., Shi, H. and Zhao, Y., 2020. Influences of soil properties and hydrological processes on soil carbon dynamics in the cropland of North China Plain. Agriculture, Ecosystems & Environment, 295, p.106886.

Zhao, J., Chen, S., Hu, R. and Li, Y., 2017. Aggregate stability and size distribution of red soils under different land uses integrally regulated by soil organic matter, and iron and aluminum oxides. Soil and Tillage Research, 167, pp.73-79.

Zuo, Y. and Zhang, F., 2011. Soil and crop management strategies to prevent iron deficiency in crops. Plant and Soil, 339(1), pp.83-95.