Upland soil functions under organic grazing systems

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Abstract

Upland soils provide a wide range of key ecosystem services such as carbon storage, water quality regulation and flood mitigation, and supporting important trophic interactions. UK upland areas are largely used for low intensity livestock grazing due to low grassland productivity. However, it is not fully understood whether organic grazing management has any impact on key soil functions in upland areas, compared to conventional management. This thesis uses a paired case study approach to investigate the key differences in soil organic matter (OM), bulk density (BD), saturated hydraulic conductivity (K_s), effective porosity, pH, moisture content available N, exchangeable cations, and earthworm populations, between the two management systems. Each pair of field sites comprised of one organic and one conventional farm; site pairs were located in the Forest of Bowland, Yorkshire Dales, and North York Moors, and chosen for their similarity in size and proximity. All sites had a long history of sheep grazing, organic sites had all been under organic management for at least 10 years. This was then followed by an avoidance behaviour experiment to investigate the impact of the veterinary medicines used in conventional grazing management (albendazole, ivermectin, levamisole and moxidectin) on the endogeic earthworm *A. chlorotica*, an earthworm commonly found in upland grasslands. Organically manged sites had less OM than conventional sites, particularly at 0-5 cm depth (means of 14% under organic and 39% under conventional) but differences in OM content were attributed to differences in soil type. Saturated hydraulic conductivity was higher under organic management (medians of 65 mm hr^{-1} for organic and 33 mm hr^{-1} for conventional) while conventional sites had a higher proportion of flow through macropores. Abundance of *A. chlorotica* was significantly higher under organic management (33% of the total earthworm population, 16% under conventional). However, earthworm density was positively correlated with pH across all sites, most likely as a function of soil type. Avoidance behaviour was observed in *A. chlorotica* under exposure of environmentally realistic concentrations of albendazole and ivermectin. Calculations of carbon balance show both management systems to be carbon sinks, with conventional farming having slightly bigger SOC stocks, especially under manure application. Overall, it was difficult to disentangle the treatment effect from environmental factors due to differences in soil type and topography between field sites, but findings suggest that conventional grazing may be slightly better for carbon storage, and that anthelmintics used in livestock farming may be negatively impacting earthworm behaviour.

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Chapter 1: Introduction

1.1 Research background

Soils provide many essential ecosystem services, such as nutrient supply and cycling; water storage, retention, and filtration; carbon storage, and the regulation of greenhouse gasses such as nitrous oxide and methane (Blackwell et al., 2011; Dominati et al., 2014). In recent years there has been growing concern around the impact of agricultural land management practices, particularly livestock grazing, on soil functions (Chantigny, 2003; Marshall et al., 2014; Stavi et al., 2016; Bünemann et al., 2018; Schils et al., 2022). In Europe, over a third of agricultural land is used for livestock grazing (Eurostat, 2020). In the United Kingdom (UK), a large proportion of livestock grazing is in upland areas (normally >150 m above sea level), which are generally unsuitable for crop production, so are mainly used for pasture instead. Upland soils provide vital ecosystem services, such as carbon storage and sequestration, water quality regulation and flood mitigation (Haines-Young and Potschin, 2009). It is therefore crucial that we understand how livestock grazing in upland areas might impact these soil functions, to enhance the ability of upland soils to secure our water supplies, carbon stores, and reduce flood vulnerability, particularly in the face of climate change.

Conventional grazing management has been shown to alter key soil characteristics and functions such as water storage (Christensen et al., 2004), carbon storage and cycling (Eldridge et al., 2017) and nitrogen (N) cycling (Di and Cameron, 2002). Evidence also suggests that the veterinary treatments applied to livestock can harm soil fauna, which can potentially disrupt soil biofunctions (Beynon, 2012a). Organic livestock farms are an alternative to conventional practices, and are strictly regulated with the aim of maintaining soil health and ecosystem services. Factors such as grazing intensity, fertiliser application and the use of veterinary treatments can differ between organic and conventional grazing management (outlined in section 1.3), so there is potential for soil physical and chemical properties, and thus soil function, to vary between the two systems. For example, organic sheep farming standards state that total annual N applications, including those from animal manures, should be no more than 170 kg N ha^{-1} yr^{-1}

(Soil Association, 2021). In contrast, conventional grazing generally has no limits, and it is recommended to apply ~ 300 kg N ha^{-1} yr^{-1} (on the most intensively grazed areas outside of Nitrogen Vulnerable Zones (NVZ) (Defra, 2010). Lower N inputs have not only been associated with lower N leaching levels from soils (Di and Cameron, 2002), but can also result in lower plant productivity and carrying capacity than conventionally managed grassland. This can result in lower grazing densities under organic livestock grazing, and so the soil could potentially be less compacted under organic grazing than under conventional systems, helping to maintain soil hydrological functionality. Furthermore, the restricted use of N fertilisers and antiparasitic treatments under organic farming could mean that soil microbial and faunal communities are different to conventionally managed soils, which can also result in soil structural differences between the two management systems (Marinari et al., 2000; Beynon, 2012a). However, there is a dearth of literature on organic livestock farming, and it is not clear whether organic sheep farming has any impact on key soil functions nor how soil function under organic sheep grazing differs from those in soil under conventional sheep grazing. Therefore, the following sections aim to provide an overview of upland farming systems and organic livestock farming regulations, followed by a literature review of what is known about the impacts of livestock farming on soil functions. This information is then used to develop the project aims and objectives outlined in section 1.5.

1.2 Overview of UK upland farming

The UK landscape is diverse both topographically and ecologically, hence agricultural practices can vary across the landscape as farmers adapt to make the best of different soil types, relief, climates, and precipitation regimes. Livestock farming in the UK is one such farming practice and has adopted a stratified system, where farming is typically split into three tiers: lowland, upland/marginal and hill/mountain farming. Lowland livestock farming is characterised by low-lying fertile land, which generally experiences lower rainfall, slower winds and milder winters than upland/hilly areas (Backshall et al., 2001; Environment Food and Rural Affairs Committee, 2011). Although grassland productivity is largely determined by soil type, the milder climatic conditions at lower altitudes often result in higher grassland productivity and the ability to support a larger number of livestock per hectare (Backshall et al., 2001) Lowland farms therefore tend to more intensively grazed than upland areas as they can support a higher number of livestock units.

Conversely, upland and hill farms are typically located on the arable land margin, are subjected to notably higher precipitation and wind speeds, and temperatures are generally lower with winters being particularly harsh (Backshall et al., 2001; Environment Food and Rural Affairs Committee, 2011). Although upland farms are located above 150 m above sea level, whether a farm is defined as upland is determined less by its altitude and more by its climatic conditions and vegetation. The variable topographical relief in upland and hill areas also creates limitations; flat areas are prone to water-logging, and steep slopes not only restrict machine access but also increase soil, water and nutrient movement downslope (Backshall et al., 2001). These climatic and geographical constraints can lower soil fertility, as high rainfall and low evapotranspiration can potentially increase the leaching of important base cations (calcium (Ca), magnesium (Mg), potassium (K), and sodium (Na)) from the soil. This can result in soil acidity and reduced nutrient availability to plants (Hendershot et al., 2008). Therefore, stocking levels in hill and upland areas tend to be lower than on lowland farms.

Upland farming systems are similar in their climatic and vegetation characteristics, but they can differ slightly in their grazing regimes. In hill farming (~400 m above sea level, Figure 1.1) the farms are located at higher altitudes where it is often unfeasible to fence off large areas of pasture due to constraints such as land ownership (Backshall et al., 2001). Hill farms therefore have little in-bye land (an area of enclosed pasture located lower down in the valley close to the farmhouse) and sheep are instead free to roam over large wild areas of unimproved/rough grassland. Due to these limitations, hill farms almost exclusively graze sheep, although suckler cows may be brought in to graze over winter (Backshall et al., 2001). Upland farms differ in that they are typically sited in the valleys of upland areas, and so generally have a much larger area of in-bye land available. This allows farmers to grow silage or hay instead of needing to buy in

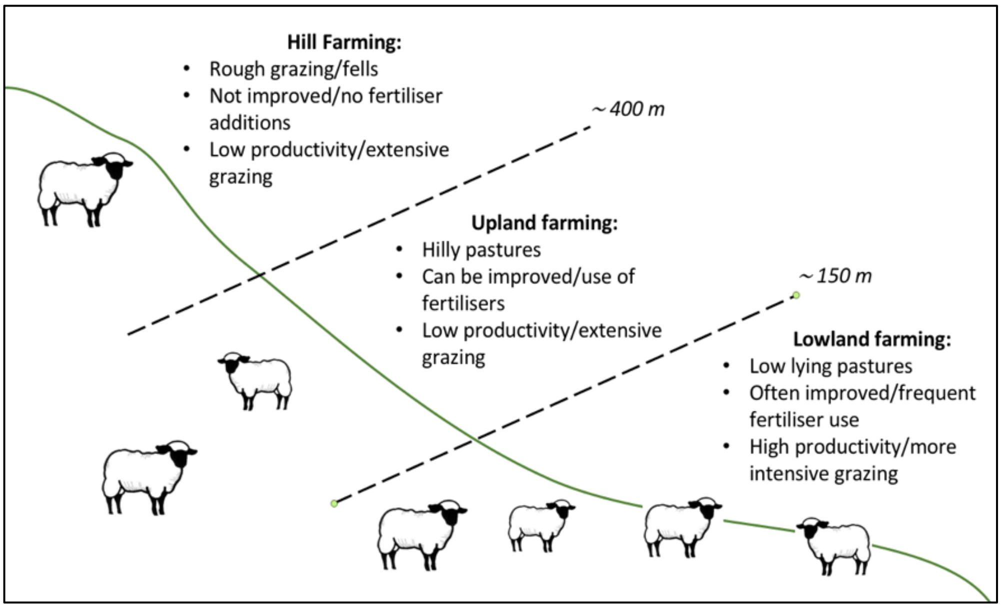

Figure 1.1 Overview of the UK stratified sheep farming system. Boundaries are conceptual.

feed over winter as is often the case in hill farming. It is also common for upland farmers to keep suckler or dairy cows for additional income, which also produces manure, a beneficial by-product.

In-bye land (in both hill and upland farming) is often improved nutritionally by planting species such as *Trifolium pratense* (red clover) and *Lolium* (ryegrasses) (Backshall et al., 2001), or by applying farmyard manures, which can increase the carrying capacity of the grassland. In-bye fields are also often rotated with hay/silage crops as well as rotating between sheep, cow or mixed grazing. Liming materials such as calcium carbonate ($CaCO_3$) can also be applied to soil to maintain an optimum soil pH for nutrient availability. Upland in-bye land is often under organic soil with a pH of around pH 5.5, and where the pH is lower, it is recommended that 4-7 t/ha of liming material applied (AHDB, 2022). The extent of liming practices in upland varies depending on the severity of soil acidification and the cost-effectiveness of liming. The specifics of how upland farmers manage their in-byes and livestock numbers appears to be highly variable and is determined by factors such as field size/shape, availability of livestock housing, staffing numbers, site accessibility and so on (Backshall et al., 2001).

The difficulties in maintaining agricultural land in areas such as the hills and uplands of the UK was recognised by the European Commission (EC) in 1975, with the introduction of Lesser Favoured Areas (LFAs), which have since been renamed as ‘Areas facing Natural or other specific Constraints’ (ANCs) (European Commission, 2018). Farmers within ANCs are eligible for financial assistance in addition to the Common Agricultural Policy (CAP), which includes enhanced rates of grant and special payments for livestock farming (European Commission, 2018). In the UK, ANCs mostly encompass land where agricultural productivity is low, economic results are lower than the national average, and there is a low or dwindling population reliant on agriculture, and these areas encompass 100% of UK upland areas (European Commission, 2018). Due to the ecological importance of UK grasslands, support is available through the Sustainable Farming Incentive (SFI), which provide additional financial income for maintaining their land to standards which work towards fulfilling UK grassland management programs and the UKs 25 year environment plan (GOV.UK, 2021).

UK uplands soil can often be classed as organo-mineral (OM) (Bol et al., 2011). Typically, OM soils consist of a surface organic layer that is no more than 40 cm deep, covering mineral horizons below or directly above the parent material (Holden et al., 2007). OM soils are often waterlogged, slowing down the decomposition of organic matter, enabling the development of a peaty O horizon. The peaty layer is shallow and not permanently saturated, allowing bioturbation to mix some mineral soil into the organic soil. 59% of organo-mineral soils are located in UK upland areas, making this an important soil type to focus on in this study.

1.3 Organic livestock farming

Organic farming can be defined as an integrated system that promote farming methods that are intended to optimise the productivity and fitness of diverse communities within the agroecosystem. Over the past 20 years, organic farming has increased globally by ~40 Mha (Figure 1.2a) and there is almost twice as much organic permanent grassland globally than there is organic arable land (Figure 1.2b) (FiBL and INFOAM Organics International, 2017). The majority of organic land is currently in Oceania (45%) and Europe (25%) (FiBL and INFOAM Organics International, 2017) (Figure 3). In the UK, 507 thousand ha of land is farmed organically, of which 61% is permanent grassland, which accounts for the largest share of organic area in the UK, followed by temporary pasture (20%) and cereals (9.2%) (Defra, 2022a)

To be classed as organic, producers must comply with strict regulations. The International Federation of Organic Agriculture Movements (IFOAM) offers a baseline organic production worldwide, which is then used as a guide for organic regulatory bodies worldwide. For example, in the European Union (EU), organic

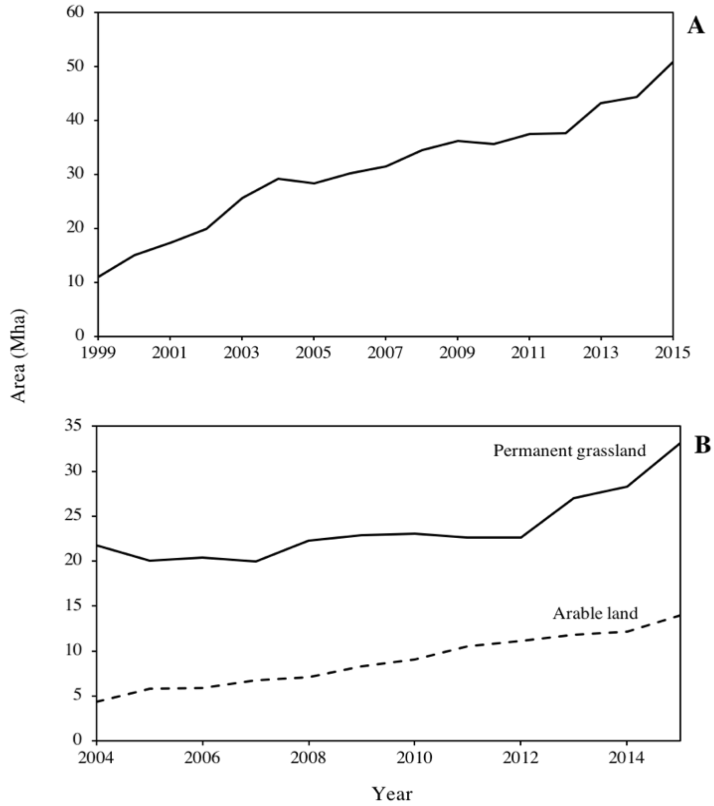

Figure 1.2: Changes in area of organic land: A = total area increase globally; B = increase shown in permanent grassland and arable land fractions, adapted from FiBL and INFOAM Organics International (2017).

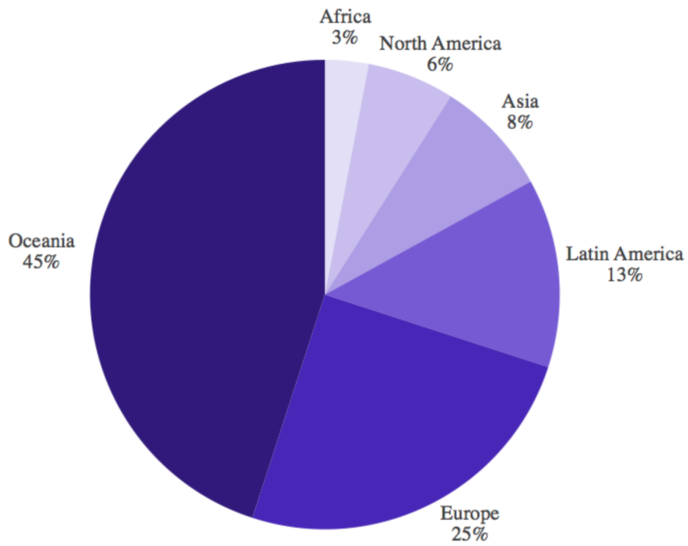

Figure 1.3: Distribution of organic land by region. Adapted from adapted from FiBL and INFOAM Organics International (2017).

farming is governed by EU law, and organic farmers must register with a recognised organic accreditation organisation (European Commission, 2017). In the UK, organic standards are set by DEFRA, and are based on 'The DEFRA UK Compendium of Organic Standards' (Defra, 2006). However, this is currently under review as the UK separates from the EU (Defra, 2022) and each accreditation organisation then owns its own standards based on DEFRA's Compendium. UK organic sheep farmers are certified by different accreditation organisations; however, these organisations all appear to be in partnership with, and follow the standards of, the Soil Association, which is by far the largest organic accreditation organisation in the UK. Further information can also be found in Natural England (2001), European Commission (2017) and Soil Association (2021). The following tables (Table 1.1 and Table 1.2), provide a summary of organic sheep farming standards based on those provided by the Soil Association (2021), which are applicable/relevant to livestock farming in UK upland areas.

Table 1.1 Overview of organic animal management regulations relevant to upland livestock farming, compared to conventional upland management practices

	Animal management *Conventional*	*Organic*
Flocking and stocking	Stocking densities vary as they depend on the number of staff available, suitability of the environment and capacity of the farm. It should be low enough to maintain sheep welfare, however, there is less focus on environmental sustainability. Rotation systems are generally used, most commonly deferred grazing, and 'leader-follower' grazing. Grazing for parasite control or 'clean grazing' (where whole fields are rested for periods of >12 months) is not often used.	Livestock must be kept at low enough stocking densities so as to prevent: poaching of the soil, over-grazing of vegetation, and the application of more than 170 kg N ha^{-1} yr^{-1}, pollution of watercourses. Clean grazing rotation method is preferred as it prevents worm infestation and allows the soil to recover. Grassland is left to rest for 12 months before livestock are allowed to graze again. Clover or similar is often planted to the resting land to maintain a good soil carbon/nutrient status.
Feeding	In upland areas, feed concentrates tend to only be given for few weeks before lambing. GMO foods are allowed.	Animals must not be fed with grains, concentrates, supplements, vitamins, minerals, feed additives and carriers containing GMOs or their derivatives. Only feeds that are certified by an organic certification body can be used. This automatically confirms their non-GM status.
Sheering	Sheering practices are very uncommon in upland areas. However, if sheering does take place, both conventional and organic sheep are generally sheered once a year and not during winter unless suitable housing is provided. During winter organic sheep must be left with a covering of wool and suitable warm housing must be made available.	
Pesticide management	Internal and external parasites are treated using a range of different endo- and ectoparasiticides. All animals are administered specific anthelmintics against Nematodirus, generally twice a year as a preventative measure. All ewes are also wormed after lambing.	Intestinal worms are controlled by rotational and clean grazing systems so as to limit the use of antiparasitic drugs. Anthelmintics can only be used on individual animals after evidence shows they are infected (e.g. through faecal egg counts), and can only be used on a whole herd as part of a disease control program and only with permission from the Soil Association. The use of organo-phosphorus and organo-chlorine is prohibited in any form for any purpose unless required by law. If required by law to use these substances then the farmer must not use any treated animals for organic produce production, unless they are re-converted to organic. Other endo- and ectoparasiticides can be used providing they don't contain products not allowed by the Soil Association.
Foot problems	Both conventional and organic livestock farming administer vaccines to prevent footrot. Organic farming requires that zinc or copper sulphate and iodine vaccines are only to be used when there is no other alternative. For both farming systems, leftover footbath solution must be disposed of using a licenced waste contractor or by diluting to 1:3 treatment to water ratio and spreading onto own land. In organic, farming animals must not be in contact with the disposal area for at least 1 month.	

Table 1.2: Overview of organic land management regulations relevant to upland livestock farming, compared to conventional upland management practices

	Land management *Conventional*	*Organic*
Soil fertility and nutrient status	In upland areas, organic amendments such as farmyard manure and slurry can be used. This practice allows for higher grassland productivity, allowing for a slightly higher grazing density. How much or how little is applied depends on forage needs and the initial productivity of the grassland. Inorganic fertilisers can be added, however this is very uncommon in upland areas.	Soil must be managed with the aim of developing and protecting an optimum soil structure, biological activity and fertility. It is recommended this is done by applying organic manure or compost and plant remains where needed. Due to the low grazing densities, organic farms in upland areas rarely add manure. The total N applied from all fertilisers (including manure) must not total more than 170 kg N ha^{-1} yr^{-1}. Any manure collected from animal housing should be returned to the soil. The use of fertilisers, composts or manure or other nutrient inputs containing GMOs or their derivatives is prohibited. Compost not licenced by the Soil Association must contain specific maximum metal concentrations, and be sourced from certified organic suppliers.
Boundaries	Fencing can be used in conventional management	Should maintain field boundaries such as hedges, ditches, banks and stone walls (ditches must have clearing regimes) and only used fencing where these options are not available. Permission must be sought from the Soil Association before removing hedges, banks, ditches or walls.
Water management	Common Agricultural Policy (CAP) requires all farmers who receive Single Farm Payments to maintain land in 'good agricultural and environmental condition' and comply with the Nitrates, Groundwater and Sewage Sludge Directives (European Commission, 2017). There are also grant, incentive and voluntary schemes provided by the Environment Agency (EA) that promote environmentally and hydrologically sustainable agricultural practices. For this reason practices vary, but all comply with the CAP as a minimum.	Organic farmers are responsible for operating sustainably within the natural hydrological function of the water catchment area and drainage basin, and must design their systems to use only as much water as the catchment can sustain and have the least impact on water quality and flow downstream of the farm. They must also aim to eliminate water pollutants (pathogens/agrochemicals/ certain nutrients etc.) entering the water catchment by: putting measures in place to separate water polluted by wastes, manures, and silage and compost leachate; bio-filters must be used in dirty water, manure and water management systems; regularly monitor soil, ground and surface water for contamination where irrigation or potential pollutants (allowed by the Soil Association) are used.

In summary, the main differences in management practices we are likely to see in organic compared to conventional livestock farming in upland areas are:

- Lower livestock units per hectare, due to flock rotation, grassland resting periods etc.
- A mostly ‘restorative’/‘reaction’ based approach to using antiparasitic treatments, rather than using them as preventative treatments (excluding vaccines): pharmaceuticals are only applied when absolutely necessary once an animal is afflicted, with some treatments being completely banned (e.g. organo-phosphates and pyrethroids). Instead, clean-grazing systems are implemented to prevent infestations.
- Where grassland soil fertility is maintained through the addition of organic matter, it is only applied as a supplement to nutrient recycling and not as a replacement. Total annual N application is restricted to 170 kg N ha^{-1} yr^{-1}. In conventional farming, for non-Nitrate Vulnerable Zones (NVZ) in the UK on an average grass growth class it is recommended to apply 50-350 kg N ha^{-1} yr^{-1} (Defra, 2010).
- Overall, environmentally sustainable management of the land is required, with farmers also responsible for sustainable soil management, water and fuel use.

1.4 Impacts of grazing on key soil functions: a review of current literature

This section contains an overview of the current literature covering the impacts of livestock grazing on grassland soil function, to highlight areas that would benefit further research. There are currently very few studies covering the effects of organic livestock grazing on soil function, and so instead this review draws on research from studies which cover a broader range of grazing management types, to hypothesise how the soil may respond differently under organic grazing systems, specifically in upland areas. Much of the literature on this topic tends to fall into the following categories, and so this literature review has been organised as such: i) The effects of grazing on soil physical & hydrological properties; ii) The effects of grazing on soil carbon; iii) Fertiliser additions; iv) The impacts of veterinary treatments on soil biological function.

1.4.1 The effects of grazing on soil physical & hydrological properties

1.4.1.1 Soil physical properties as an indicator of soil function

The physical structure of soil refers to the arrangement of solid soil particles and their aggregates (both artificially and naturally formed), and the pore spaces located between them. There are two complementary aspects of soil structure: the solid phase and pore space. The solid phase is based on mechanisms of soil aggregation, and is viewed as a three-stage hierarchical organisation of aggregates, each

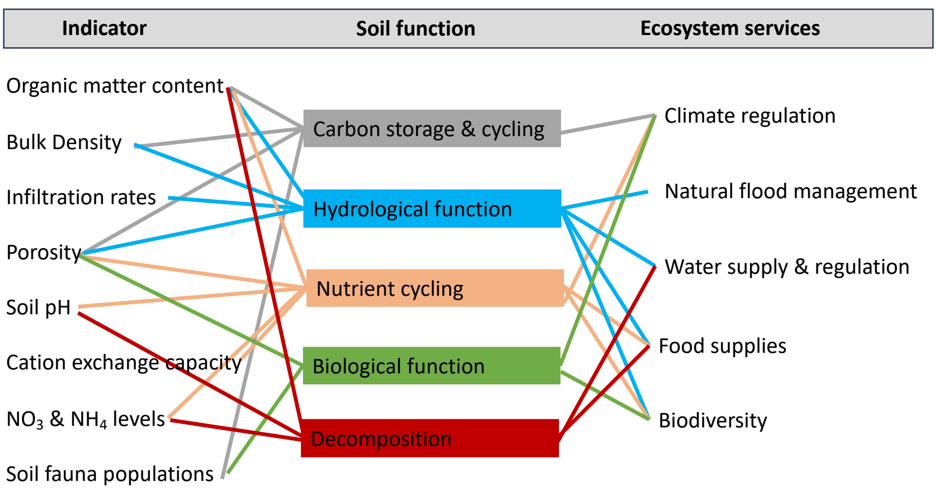

Figure 1.4: Conceptualisation of soil property indicators and their link to soil function and ecosystem services

involving different binding agents (Tisdall and Oades, 1982; Brady and Weil, 2014): Primary particles (<20 µm) consist of clay platelets interacting with Fe or Al oxides and organic polymers (organoclay clusters), and silt particles, which then bind to humus particles and mineral grains. These primary particles then bind together with particulate organic matter to form submicroaggregates which, along with sand grains, are then bound with root hairs, hyphae, and microbial excretions to form microaggregates (20-250 µm). Macroaggregates (250-500 µm) are the fusion of these microaggregates by fungal hyphae and fine roots (Brady and Weil, 2014). This hierarchical order of aggregates is not found in either sandy or oxide-rich soils (Oades and Waters, 1991; Christensen, 2001; Six et al., 2004), but has been identified in soils with higher organic matter content, such as much of the soil in upland areas. Soils with much higher organic matter content such as peaty or organo-mineral soils typically have 20 cm peaty surface layer. Peat typically has a loose and less well-defined structure compared to mineral soil aggregates. The organic matter in peat forms fibrous or amorphous structures, resulting in loosely packed aggregates with irregular shapes, held together by organic substances, such as humic acids and microbial-produced polysaccharides (Brady and Weil, 2014).

The pore space aspect of soil structure can be defined as “the combination of different types of pores”, where soil aggregates and particles are viewed as the walls of the pore space (Pagliai and Vignozzi, 2002). As with aggregates, a similar hierarchical classification system exists to define soil pore size, however there are no generally agreed upon size thresholds. The pore spaces can be separated into approximately two components: intra-aggregate porosity (microscopic pore space between soil particles) and interaggregate porosity (pore space caused by arrangement of aggregates, created by biological activity) (Cameron and Buchan, 2005). Soils high in organic matter such as those in upland areas generally have a higher proportion of large-sized pores compared to mineral soils, due to the higher proportion of partially decomposed plant material. Due to the fibrous nature of this organic material, peat generally has poor interconnectivity between pore spaces, which can lead to higher moisture content and water logging (Brady and Weil, 2014).

Soil structure regulates many processes within the soil such as the retention and infiltration of water, the movement and exchange of gasses, nutrients and organic matter, susceptibility to erosion, and root penetration (Mueller et al., 2009; Brady and Weil, 2014; Rabot et al., 2018). Soil structure also creates the necessary environment for a wide variety of soil fauna which create an important feedback, as they actively shape the structure of the soil, modifying the distribution of water and air (Feeney et al., 2006; Bottinelli et al., 2015). Since soil structure is key to the performance of these soil ecosystem functions, it makes sense to use soil structure as an indicator of these soil functions, yet there is no universally agreed way to characterise soil structure, or how structural measures can be used to indicate soil functioning. Rabot et al. (2018) provided a critical analysis of a wide number of soil structural properties, their methods of measurement and their efficiencies as related to soil functions. They found that characterisation of the pore space in particular can be a good indication of soil function, with evidence suggesting that processes occurring in the soil are controlled by properties such as the pore shape, size distribution, surface density and connectivity, which agrees with previous research on the topic (e.g.: Young et al., 2001; Pagliai and Vignozzi, 2002). Rabot et al. (2018) also found that aggregate structure alone did not provide a significant link between soil structure and functions or processes, explaining that aggregate analysis is related more to the actual mechanical stability of the aggregates, rather than the soil structure itself. This indicates that investigating soil physical properties, especially the pore space, could be a beneficial approach to assessing multiple soil functions, and so could be of use in this project.

1.4.1.2 Impacts of grazing intensity on soil physical properties

Soil pores are essential in the transport of surface water to groundwater supplies, preventing excessive overland flow, which can exacerbate soil surface erosion and downstream flooding (Kutilek and Nielsen, 1994). Added surface pressures from agricultural practices such as farm machinery and the hoof action of grazing livestock has been shown to result in soil compaction (often measured as bulk density) (Hamza and Anderson, 2005; Drewry et al., 2008; Iglesias et al., 2014; Eldridge et al., 2017; Pilon et al., 2017). This pressure creates a denser configuration of aggregates and a reduction in the connectivity of pores spaces, often through a reduction in macropore space, which can reduce the permeability of the soil, leading to lower infiltration rates (Drewry et al., 2008). For example, Houlbrooke and Laurenson (2013) found a reduction in total porosity and an increase in microporosity (in silt loam), with a 5.7% decline in macroporosity in soils under cattle grazing, when compared to soil under lower surface pressures of sheep grazing. Similarly, Drewry et al. (2000) found that a range of mineral soils exposed to dairy farming had reduced macroporosity (by 3.6%) than sheep farms (1.5%), higher bulk density (0.16 mg m^{-3}) compared to sheep farms (0.12 mg m^{-3}), and lower saturate hydraulic conductivity (32 mm hr^{-1}) compared to sheep farms (86 mm hr^{-1}). This increases the chance that the intensity of rainfall is higher than the permeability of the soil, causing soil surface water run-off, known as infiltration excess overland flow (OLF) (Ludvíková et al., 2014; Russell and Bisinger, 2015), which can lead to a higher risk of downstream flooding as water flow into groundwater supplies becomes restricted (Steinfeld et al., 2006). Compaction can also form a dense hardpan which can result in the early initiation of rapid subsurface lateral throughflow in the upper soil layer during precipitation events (O'Connell et al., 2004). The infiltration rate of soils is influenced by a large number of factors such as soil structure, soil density, soil type, macroporosity from biological activity, the supply of surface water, and vegetation type and density. Leonard and Andrieux (1998) found that between each of these factors, the infiltration behaviour of soils depends primarily on soil type. In general, coarse-textured gravels and sands have higher infiltration capacities than fine-textured clays and loamy soils, such as those commonly found in upland areas (Table 1.3).

Soil can also undergo erosion as the OLF erodes and transports soil particles downstream, resulting in the loss of soil and essential soil components such as carbon and nitrogen (Steinfeld et al., 2006). Run-off can increase with increasing and gradient, such as the steep hills commonly found in upland areas in the UK. Soil

Table 1.3. Soil type and infiltration rate, as described by Hillel (1980)

Soil Type	**Steady-state Infiltration Rate (mm hr^{-1})**
Gravels and sands	>20
Sandy and silty loams	10-20
Loams	5-10
Clayey soils	1-5

erosion from overland flow can also be exacerbated by vegetation removal and exposure of the soil surface, which has been shown to happen due to over-grazing (James and Alexander, 1998). However, upland sheep farming has a much more extensive grazing regime than lowland and intensive farms, and so surface erosion due to over-grazing is unlikely.

Marshall et al. (2009) evidenced that infiltration excess OLF due to compaction tends to occur under the most intensive grazing regimes, and that extensive grazing common to upland areas is not generally high enough to cause dense enough compaction that results in infiltration excess OLF (although it may occur where livestock traffic is concentrated, for example near feeding areas, field entrances, camping grounds). Instead, they found that saturation-excess OLF was more common in upland soils due to their shallow nature and the slow draining of the B horizon preventing rapid drainage of the A horizon. The study was conducted at permanent sheep grazed pastures in the upland areas of Pontbren, Wales, and found that the A horizons remained near to saturation for prolonged periods, despite the relatively low precipitation amounts, typical of that area. B horizons (illuvial horizon) develop from the accumulation of leached materials and fine particles from the upper organic horizons, and often contains a high amount of densely packed clay particles, with consequently low permeability. In acidic soils common to UK uplands, the low pH can also lead to the mobilization and transport of iron ions downwards through the soil profile. When these ions encounter different (higher) pH levels or textural differences within the soil, precipitation out of solution occurs. Overtime, this precipitation of iron can lead to the development of a hard iron pan, further limiting percolation of water through the lower horizons (Lundström et al., 2000). Land history can also play an important role, if the site was previously more intensively grazed, or subject to heavy machinery use, then higher bulk density may be apparent.

In general, the literature agrees that the more intensive the grazing is, the more compact the soil is likely to be (Pande et al., 2000; Pilon et al., 2017). However, it is very difficult to compare studies on the impacts of grazing on soil structure due to inconsistencies in quantifying intensive grazing. Most studies describe the intensity as either high, medium or low, and do not use the same definitions. Furthermore, estimates are often created using different methods which are usually subjective and vary depending on local practices. Studies on the impacts of sheep grazing are also outnumbered by articles covering the impact of cattle grazing. However, cattle exert twice the static pressure on the soil as sheep (Noble and Tongway, 1986), therefore the results cannot be assumed the same for sheep grazing. Abdalla et al. (2018) addressed these issues when comparing the literature by calculating a continuous variable to represent grazing intensity, taking into account the carying capacity of the land (the amount of forage produced by the land and how many months this will support the livestock based on the energy requirements of the particular animal in question). They then used this to produce meta-analyses of the impacts of grazing intensity on several soil quality indicators, including bulk density. The analyses included studies from different climatic zones and, unlike similar previous meta-analyses, grouped the sites according to their similarities in temperature and moisture, giving insight into how climate may impact the response of soil to grazing. Sites located in dry-cool or dry-warm environments had significantly different bulk densities under grazing to those situated in moist-cool or moist-warm environments. However, there were too few sites in moist-cool environments with bulk density measurements to be able to draw any major conclusions from the data, highlighting a need for more grazing studies in these climates. Nevertheless, their results do agree with Wang and Wesche (2016), who collected data from 100 sites with varying annual temperatures and precipitation regimes. They found that bulk density tends to increase with grazing intensity in moist soils with smaller particles, whereas dryer soils, especially those with larger particles (i.e. higher sand content), tend to be more resistant to compaction. This indicates that upland soils located in cool/wet upland areas may be susceptible to structural damage from livestock grazing. Furthermore, due to the lower stocking densities and therefore lower surface pressures used in organic farming, there could be less impact on bulk density under this grazing management system compared to conventional grazing regimes. However, the few studies currently available on the impacts of grazing on soil structure in UK soils mainly focus on lowland sites (Ward et al., 2016a) and the effects of grazing on UK upland soils still remains poorly understood.

1.4.2 The effects of grazing on soil carbon

Soil organic carbon (SOC) plays a major role in many important soil functions including nutrient cycling, soil structure formation and water retention, and as such has been recognised as one of the most important indicators of soil health (Palm et al., 2007; Lal, 2010). SOC stocks are dependent on inputs of biological matter and release of carbon from the soil system (e.g. through CO_2 respiration and dissolved carbon leaching). SOC stocks refer to the total amount of organic carbon stored in a given area or volume of soil, whereas soil organic concentration refers to the percent of organic carbon withing a given mass of soil. SOC stocks can be calculated from values of soil organic matter (SOM) concentration and bulk density values using the relation:

$$SOC \text{ stocks (kg m}^{-3}) = C \times BD \times D \qquad (2.1)$$

where C = SOC concentration (%) calculated using SOM × 0.54, BD = bulk density (kg m^{-3}), D = soil depth (cm).

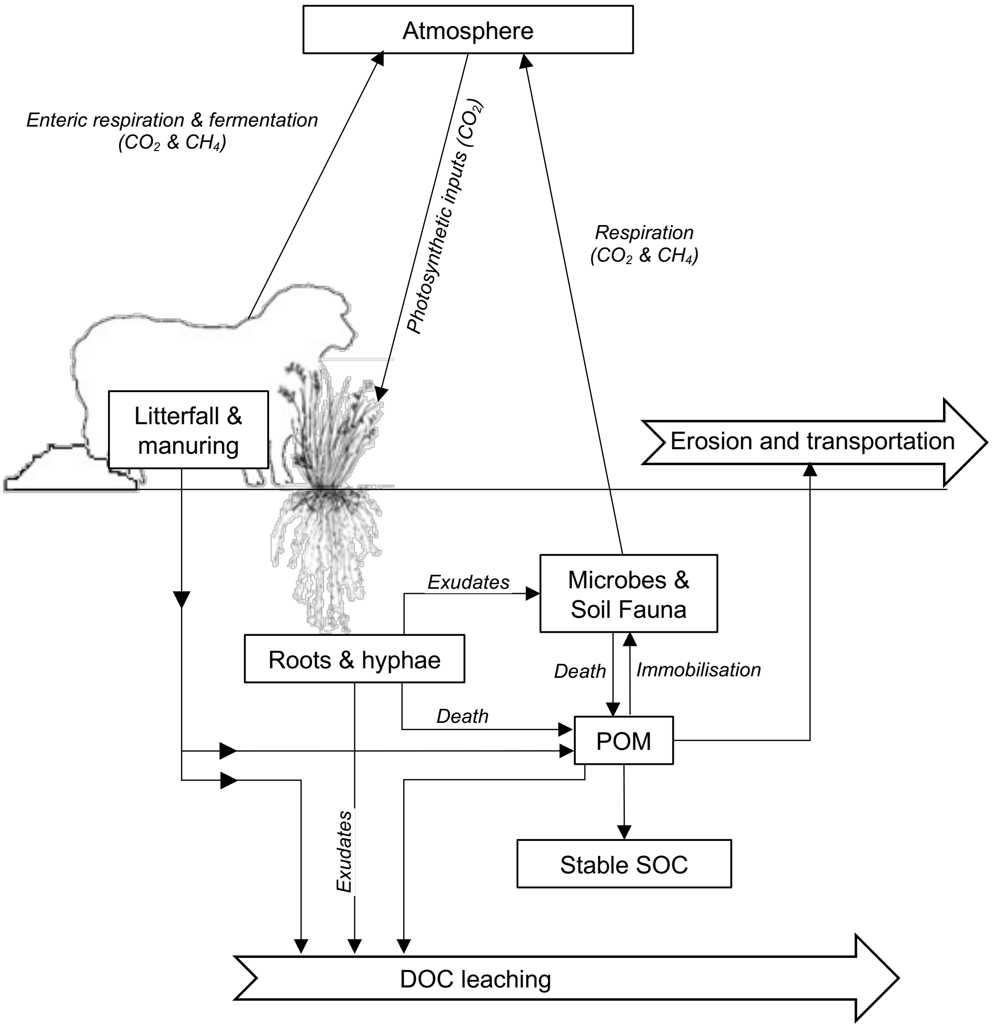

Figure 1.5: Carbon cycle in an upland grassland

Land management practices such as grazing management can alter these C inputs through removal of aboveground biomass from herbivory and cutting, addition of fertilisers to pastures, and the mechanical disturbance of the soil through hoof action. Grazing has been shown to generate changes in the SOC stocks of grassland soil, with many studies reporting that concentrations of SOC decrease as grazing intensifies (Su et al., 2004; Gao et al., 2008; Klumpp et al., 2009; Martinsen et al., 2011; Mofidi et al., 2012; Mcsherry and Ritchie, 2013a; Wang and Wesche, 2016; Waters et al., 2017a; Eldridge et al., 2017b). This has mostly been attributed to a reduction in CO_2 fixation through loss of photosynthetic material as the animal consumes the plant matter, leading to lower aboveground inputs from reduced leaf fall and lower below ground C inputs through reduced root production. Loss of vegetation can also exacerbate C losses through exposing the soil surface, increasing erosion rates (Skapetas et al., 2004; Li et al., 2008; Steffens et al., 2008). Other studies have found both neutral and positive effects of grazing on SOC stocks (Reeder and Schuman, 2002; Martinsen et al., 2011; Li et al., 2011; Lu et al., 2015; Eze et al., 2017a) as compaction reduces the oxygen content of the soil, slowing down decomposition rates of organic material (Li et al., 2011). Grazing can also alter grassland species composition to one with a higher root to shoot ratio, increasing organic matter contribution from the root (Jean D Reeder et al., 2004).

Contrasting findings on the effects of grazing on SOC have been attributed to differences in factors such as site productivity, climate, soil type and land management history. For example, Waters et al. (2017) found that the response of SOC to grazing intensity was mostly dependant on the species composition and productivity of the vegetation community, with lower productivity exhibiting lower SOC. This was due to both a decrease in above and below ground organic inputs from plant material, and reduced vegetation cover, exposing the soil surface causing increased soil erosion. In warmer, semi-arid climates, not only have grassland soils been shown to have lower initial SOC pools (Derner et al., 2006; Mcsherry and Ritchie, 2013a), but grazing can cause a shift in grassland composition to a more C_4 grass dominated ecosystem (Derner et al., 2006) which can result in a larger transfer of fixed carbon below ground to roots (Derner and Schuman, 2007). In contrast, grasslands situated in wetter, cooler environments where C_3 grasslands dominate, higher grazing intensities have been associated with lower SOC (Mcsherry and Ritchie, 2013a). This is thought to be due partly to a higher fraction of SOC stocks being labile in cool, wet climates, which may increase the turnover of C under grazing (Derner and Schuman, 2007; Eze et al., 2017a). These wet and cool grasslands are typical to UK upland ecosystems, which are often used for livestock grazing as they are unsuitable for crop cultivation. This could mean that SOC stocks in UK upland grasslands are under threat from grazing. However, research into the effects of grazing on UK soil is limited, especially on upland soils, with most recent studies focusing on lowland grasslands (Ward et al., 2016a). Furthermore, studies on upland soils also tend to focus on peatlands, highlighting a need for more studies on organo-mineral soils, of which 59% are located in upland areas (Bol et al., 2011).

Microclimates within grassland ecosystems, influenced by topographical variations such as slope, aspect, and elevation, play a crucial role in shaping the distribution of moisture and organic matter accumulation (Raghubanshi, 1992). In areas with shallower slope gradients, organic matter tends to accumulate due to slower infiltration rates and higher moisture content. The reduced water movement slows down the decomposition processes, resulting in a build-up of organic matter. Conversely, steeper slope gradients facilitate rapid downslope movement of soil water, leading to a more aerated soil and faster decomposition rates, resulting in lower organic matter concentrations. Assessing the saturation levels and understanding upslope contributing areas can be done using the topographical wetness index. This tool provides insights into the likelihood of an area being saturated based on slope and elevation data, and helps evaluate its influence on organic matter decomposition rates downslope (Sørensen et al., 2006).

The majority of SOC in grasslands has been shown to be stored in the stable or inert pool, which is both physically protected (contained within soil aggregates or sorbed to clay minerals and oxides) and chemically protected (as humus) (Eze et al., 2017a). Stable carbon stocks are largely unavailable to microorganisms and have very slow turnover rates of days to millennia (von Lützow et al., 2007). Contrastingly, labile carbon which comprises particulate organic matter (POM), microbial biomass C (MBC) and dissolved organic carbon (DOC) makes up only a small fraction of SOC and has a much faster turnover rate. Labile carbon plays an important role in the soil carbon cycle, and given its high mobility, fast turnover rate and broad reactivity in the soil (Boddy et al., 2007), labile carbon can be an important source of the occurrence of stabilised carbon (Schmidt et al., 2011). It is also responsible for the transport of nutrients throughout the system and influences both the activity and mass of the soil microbial population. The high turnover rate of labile carbon makes it much more sensitive to changes in land management practices than SOC as a whole and can therefore be used as a key indicator of soil natural functions (Silveira, 2005; Jokubauskaite et al., 2015). Grazing is one such land management practice that has been shown to cause changes in the labile C pool, mostly through a decrease in the MBC fraction (Zhou et al., 2017), and increased CO_2 respiration (e.g. Cao et al. (2013)). However, there are much fewer studies on the response of soil DOC fluxes to grazing. DOC is made up of low molecular weight particles such as amino and organic acids, along with higher molecular weight materials such as enzymes and humic substances (Zsolnay, 2003; Bi et al., 2013). It plays an important role in the transport of nitrogen and phosphorus, as well as toxic metals & other contaminants (Kaiser, 2001), facilitating their transport throughout the soil profile towards streams and groundwater (Worrall et al., 2009; Kaiser and Kalbitz, 2012). Since DOC carries these key nutrients and metals throughout the soil, it has a major influence on the balance of these elements within the soil system. The extent to which DOC is involved in these nutrient transport mechanisms depends strongly on soil characteristics (Qualls and Richardson, 2003; Silveira, 2005). The few studies that have focused on the impact of grazing on DOC losses have so far shown little to no significant effects from grazing (Ward et al., 2007; Chapman et al., 2010). However, grazed grasslands are often subjected to the application of synthetic fertilisers and manure to ensure grass yields meet the needs of grazing livestock, and recent studies have suggested that the application of organic fertilisers to soil can increase water-extractable organic carbon by a factor of 2.7-3.2 (Kalbitz et al., 2000). This has been attributed to the increased direct addition of plant derived water-extractable organic carbon, such as root exudates, due to increased plant growth (Kalbitz et al., 2000; McTiernan et al., 2001; Wang et al., 2016). Therefore, grazing may not be having a direct impact on DOC concentrations, but pasture management practices such as the application of fertilisers may be impacting DOC fluxes.

1.4.3 Fertiliser additions

1.4.3.1 Nitrate leaching

Nitrogen is an essential nutrient for plant growth. In order for plant growth to meet the needs of grazing livestock, additional nitrogen is often applied to grazed pastures in the form of organic fertiliser, such as farmyard manure (FYM). N can also be added to the soil by planting legumes such as clover within the pasture; legumes contain rhizobium bacteria within their root nodules that biologically fix N. Not all soil N is taken up by plants; a large part of the N is incorporated into soil organic matter (SOM), lost to the atmosphere, or leached into ground or surface waters (Figure 1.6). Ammonium (NH_4^+) is readily converted into nitrate (NO_3^-) within the soil during a two-step process named nitrification (Figure 1.6). The first stage of nitrification involves the oxidation of ammonium by Nitrosomonas, which convert (NH_4^+) to nitrite (NO_2^-) via:

$$2NH_4^+ + 3O_2 = 2NO_2^- + 2H_2O + 4H^+$$

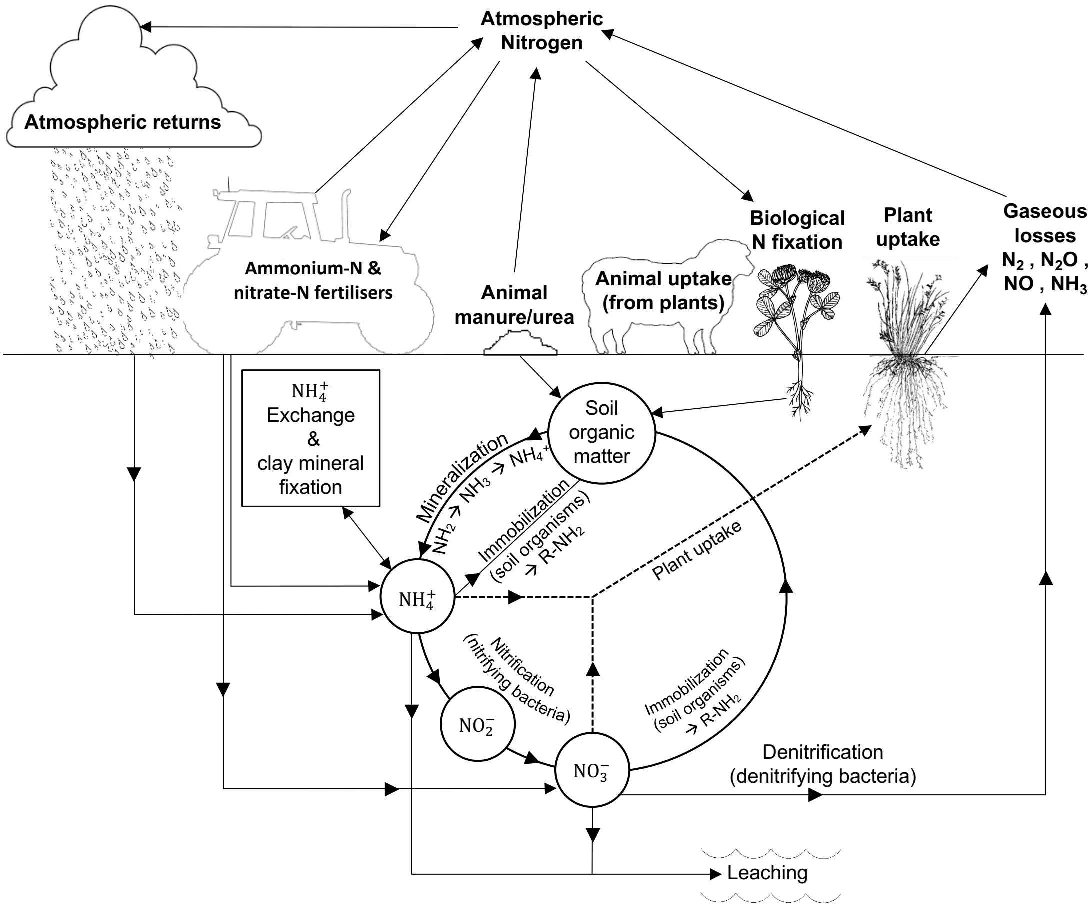

Figure 1.6: Nitrogen cycle in a grazed grassland system

This is followed by a second step by Nitrobacter, which are responsible for further oxidation of NO_2 to NO_3^- via $2NO_2^- + O_2 = 2NO_3^-$.

Both stages of nitrification are pH-sensitive, but the second stage, nitrite oxidation, is particularly affected by pH. Nitrite-oxidizing bacteria are more sensitive to low pH conditions, and their activity decreases as pH decreases. This can lead to a slowdown or inhibition of the nitrite-to-nitrate conversion (Norton and Stark, 2011). NH_4^+ concentrations are usually low (Di and Cameron, 2002), and the negative charge of NO_3^- in most soils means that it is not easily retained by the soil, and is therefore usually the dominant form of leached N (Di and Cameron, 2002). It was originally thought nitrification was limited in acidic soils such as those found in UK upland areas, due to the limited availability of NH_3 required for the growth of bacterial ammonia oxidisers (Boer and Kowalchuk, 2001). However, the recent discovery of obligately acidophilic ammonia oxidising archaea (AOA) has revealed that nitrification can occur in low pH soils (Li et al., 2018). AOA organisms possess specific adaptations allowing them to grow at low pH (Lehtovirta-Morley et al., 2011; Lehtovirta-Morley et al., 2018), and they have been found to be particularly functionally important in acidic soil ecosystems (Leininger et al., 2006; Prosser and Nicol, 2008; Yao et al., 2011). The recent literature now shows that nitrification can occur at pH values as low as 3.0 such as those found in UK upland areas, and that rates of nitrification in acidic soils can be equal or even greater than that typically found in neutral pH soils (Li et al., 2018).

NO_3^- leaching has become a major global concern due to the large volumes of N applied as fertiliser and organic wastes to agricultural land, as leached NO_3^- can contaminate surface waters, groundwater and drinking water supplies. NO_3^- drained into surface water bodies can also result in eutrophication, algal bloom, and fish poisoning. For this reason, the Water Framework Directive (WFD) limits NO_3^- in groundwater supplies to 37.5 mg NO_3^- L^{-1} (Defra, 2015). There are two main controls on the amount of NO_3^- leached from grassland soil into the groundwater: i) drainage volume and ii) the amount of NO_3^- in the soil that is more than what is required for plant uptake (Di and Cameron, 2002). Excess NO_3^- is often due to increased N inputs from fertiliser application; fertilisers are usually applied in spring and autumn when temperatures are lower and pasture growth is slow, when there is a demand for more feeds. Annual NO_3^- leaching has been shown to be worsened if fertiliser is applied at times of year when demands are lower: e.g. Silva et al. (1999) showed that when organic fertiliser was applied at >200 kg N ha^{-1} yr^{-1} split into four equal applications, NO_3^-—N leaching losses were in the range of 6-7 kg N ha^{-1} yr^{-1}. However, when applications were split into two equal applications that did not synchronise with N demands, NO_3^-—N leaching losses more than doubled to 13-49kg N ha^{-1} yr^{-1}. For this reason, there are restrictions on the time of year fertiliser can be applied in organic farming.

NO_3^- leaching losses tend to be worse from grazed grassland systems than from cut grassland or cropping systems as during grazing only 10-40% of the N is ingested, the rest is returned to the soil pasture system in the form of urine and faeces (Jarvis et al., 1995). Many studies show that the higher the N input into grassland soil system, the greater the NO_3^- leaching losses are, as often the applied N exceeds the needs for plant uptake (Di and Cameron, 2002). Additionally, many studies provide evidence that grazing intensity does not impact N leaching and that leaching losses are primarily controlled by N input (Hoogendoorn et al., 2016; Hoogendoorn et al., 2017). Despite the evidence that higher N inputs often result in higher NO_3^- leaching there are no strict limits on the amount of N allowed to be applied to conventionally managed agricultural fields, except in NVZs, where the application of both organic and inorganic N fertilisers are restricted depending on crop type. For grasslands located in NVZs, total N application from both inorganic and organic fertilisers (including from grazed animals) is limited to 170 kg N ha^{-1} yr^{-1} (Defra, 2017). Grasslands that are not located in NVZs are instead expected to apply 'best practice' standards where the application of N should be matched to the required plant yield, ensuring that N application does not exceed the requirements needed for the necessary forage growth required to support the livestock (Defra, 2010). For non-NVZ conventionally managed moderate to intensive livestock farming in the UK on an average grass growth class, it is recommended to apply a total of 50-350 kg N ha^{-1} yr^{-1}. Whereas UK organic farming standards limit N application to 170 kg N ha^{-1} yr^{-1} for the entire farm. Lowering total N input has been suggested as one of the main strategies to reduce NO_3^- leaching in grazed pastures (Di and Cameron, 2002). Upland farms are generally extensively grazed and N application rates are much lower than those of lowland intensively grazed pastures. However, there is currently no literature looking at how N leaching differs under upland organic sheep grazing compared to conventional management in upland areas, therefore further research on this topic could be beneficial to further our understanding of NO_3^- losses under different grazing management systems.

1.4.3.2 Impacts of fertiliser additions on soil physical properties

Much of the literature surrounding the effects of N inputs from both synthetic and organic fertilisers agree that soil structure improves under their application, with many evidencing increases in porosity and water holding capacity, and decreases in bulk density up to 30 cm depth (e.g.: Marinari et al., 2000; Pagliai et al., 2004; Hati et al., 2008). One highly cited article is that by Marinari et al. (2000), who found that porosity increased significantly under both organic and mineral N applications of 200 kg N ha^{-1}. Soil with organic fertiliser inputs had 46% higher porosity compared to unfertilised control soils, and soils with mineral fertiliser had 18 % more porosity compared to control samples. Pore shape was also different between the two treatments: mineral N treatments had 51% more rounded pores and 31% elongated pores of 100-200 µm compared to control soils, whereas soils with organic fertiliser treatments had more than twice the amount of elongated pore space at 100-200 µm. Research has shown that pores ranging from 100-200 µm are essential for feeding roots to grow through (Tippkotter, 1983). Much of the literature agrees with these results, and attributes increases in porosity to a growth in the soil microbial biomass and an increase in CO_2 production within the soil (Pagliai et al., 2004; Giacometti et al., 2013; Tao et al., 2015; Bei et al., 2018). An accepted explanation is that N inputs have a priming effect on the soil organic matter, releasing C and N, making it available as a nutrient source to the soil microbial population (Marinari et al., 2000). The higher biological activity under organic fertiliser inputs compared to mineral N inputs has been attributed to the fact that organic fertilisers offer a more balanced nutritional status than mineral fertiliser, supplying phosphorus to the soil (Marinari et al., 2000). Studies have also shown that organic fertilisers are more beneficial than mineral fertiliser for soil fauna communities (Axelsen and Kristensen, 2000; Murray et al., 2006; Birkhofer et al., 2008; S. Wang et al., 2016). Wang et al. (2016) found that although the number of soil fauna did not differ between the different fertiliser treatments, the community composition did, with species indicative of a higher soil quality becoming more abundant under organic fertiliser applications. This change in community composition was also attributed to the improvement in the soil nutrient environment under organic fertiliser, which provided a more complete range of nutrients than the mineral fertiliser treatment (Wang et al., 2016).

Given that organic sheep farming restricts the use of fertilisers for improving pastures, there may be differences in soil microbial and faunal communities, and therefore soil physical properties, between the two management systems. Again, I could not identify any research on how soil structure differs between these two pasture management systems in upland areas, and so further research into this topic is needed.

1.4.4 Use of antiparasitic veterinary treatments in sheep and cattle

An important part of conventional sheep husbandry is the administration of veterinary treatments, the most common two groups being the endoparasiticides and the ectoparasiticides, which are used for the control of internal and external parasites respectively. The list of sheep diseases and their veterinary treatments used in conventional sheep management is extensive, so for the purpose of this review the focus will remain on the common endo- and ectoparasites often found in sheep and their most common treatments. An outline of these endo- and ectoparasiticides is presented in Table 1.4.

The importance of soil and dung communities on pasture fertility, soil health and as prey for higher vertebrates have been well documented and reviewed (Beynon, 2012a; Beynon, 2012c). However, antiparasitic medicines such as endo- and ectoparasiticides have been shown in many studies to have detrimental effects on soil and dung fauna (Boxall et al., 2002; Beynon, 2012a; Beynon, 2012c; Bai and Ogbourne, 2016; Rath et al., 2016; Goodenough et al., 2019), and could therefore be harming the health and functionality of grazed soils. The most commonly used group of endoparasiticides used to treat sheep are the anthelmintics, a group of broad-spectrum antiparasitic compounds used to control parasites such as gastro-intestinal helminthes, liver fluke, lungworms and sheep scab (Stubbings et al., 2020). They are normally administered orally but can also be applied topically, in-feed or by injection. Once applied, anthelmintics then enter into the environment in excretion through faeces, irrespective of its method of administration (Beynon, 2012a; de Souza and Guimarães, 2022). This is particularly true of ivermectin, a commonly used anthelmintic: Halley et al. (1989) found that >98% of ivermectin was excreted in faeces for all administration routes.

Table 1.4. Common endo- and ectoparasites found in sheep and their treatments. Data taken from SCOPS (2012).

	Parasite	Common treatments	Treatment group	Administration route	Half life (DT_{50}) range	Soil–water partition coefficient (KOC) ranges
Ectoparasites	Gastro-intestinal helminthes	Over 20 types	Anthelmintics	Oral, injection, topical	2-200 days	Macrocyclinc lactones: 100-2000 or higher All other groups range from 100 - 5000
	Liver fluke	Over 20 types	Anthelmintics	Oral, injection, topical	2-200 days	
	Lungworms	Over 20 types	Anthelmintics	Oral, injection, topical	2-200 days	
Endoparasites	Sheep Scab	Ivermectin, moxidectin, doramectin	Anthelmintics	Injection	Ivermectin: 2-15 days Moxidectin: 10-120 days Doramectin: 35-85 days	Ivermectin: 1000-20000 or higher Moxidectin: 1000-10000 or higher Doramectin: 1000-20000 or higher
	Sheep Scab	Diazinon	Organophosphate	Topical (sheep dip)	12-25 days	200-10000
	Lice	Cypermethrin	Pyrethoid	Topical (pour on)	6-24 days	1000-10000
	Lice	Deltamethrin	Pyrethoid	Topical (spot on)	30-120 days	10000-100000
	Lice	Diazinon	Organophosphate	Topical (sheep dip)	12-25 days	200-10000
	Blowfly strike	Dicyclanil	Pyramidine	Topical (spray on)	14-60 days	100-1000
	Blowfly strike	Cypermethrin	Pyrethoid	Topical (pour on)	6-24 days	1000-10000
	Blowfly strike	Deltamethrin	Pyrethoid	Topical (spot on)	30-120 days	10000-100000
	Blowfly strike	Cyromazine	Triazine	Topical (spray on)	10-100 days	100-1000

Sheep and cattle dung hosts many small fauna and is an important resource for dung beetles (Hempel et al., 2006; Davis et al., 2010; Manning et al., 2018; Ambrožová et al., 2021). Anthelmintics excreted in sheep dung have been shown to have lethal effects on dung fauna such as coprophagous flies and dung beetles, with their larvae being particularly sensitive (Hempel et al., 2006; Römbke et al., 2007). The decomposition of dung is an important input of carbon and nutrients into the soil. Early studies found little effect of anthelmintics on dung decomposition (Hirschberger and Bauer, 1994), but a more recent study by Sommer and Bibby (2002) found that dung decomposition was severely stunted by the presence of both levamisole and fenbendazole, two commonly used anthelmintics, highlighting that anthelmintics could impact the carbon and nutrient status of grazed soil. Other studies have reported negative impacts of anthelmintics on earthworms (Diao et al., 2007; Jensen et al., 2007; Gao et al., 2007; Gao et al., 2015; Bai and Ogbourne, 2016; de Souza and Guimarães, 2022), whilst others have reported negative impacts on the soil fungus *Fusarium oxysporum*, reducing both the production and germination of spores, with these effects being observed long after short-term exposure (Kollmann et al., 2003).

There are very few studies on the fate of endoparasiticides in the environment. However, one study by Mougin et al. (2003) found that ivermectin could be stored in the soil for long periods of time. They reported that the half-life (DT_{50}) of ivermectin was 240 days under dark conditions. However, this reduced to 21 days when exposed to sunlight. Krogh et al. (2009) found that ivermectin DT_{50} was dependent on soil conditions, ranging 16-36 days under aerobic soils. However, when under anaerobic conditions this increased dramatically, with the extractable amount remaining unchanged between 14-120 days. They also report that temperatures were a significant factor in the found that DT_{50} of ivermectin, and that lower temperatures significantly increased DT_{50} values. This is important for upland soils, which are prone to waterlogging and cold temperatures. Furthermore, contamination is not limited to soil and dung; anthelmintics have been found in groundwater supplies (Beynon, 2012a) which could potentially reach surface-water bodies through run-off, in addition to the surface water contamination from washed-off treated animals. Residues of endoparasiticides are also known to be toxic to a number of aquatic invertebrates (Burridge and Haya, 1993) and have been found in water sediments (Prasse et al., 2009), which raises concerns over the impact of endoand ectoparasiticides in the surrounding environments, and how far these contaminants may travel.

Ectoparasiticides are another group of commonly used parasitic treatments, used to control and treat external parasites in livestock such as ticks, lice, and flies (Table 1.4). These are often applied topically as sheep dip or spray, but may also be administered orally, injected, or applied using impregnated ear-tags (Wall, 2007). Like endoparasiticides, ectoparasiticides also enter the soil through excretion of faeces and urine by grazing animals, and less so by spillage or waste management due to strict regulations surrounding its use and disposal (Beynon, 2012c). However, there is very little literature on the impacts of ectoparasiticides on the environment or on soil health. Furthermore, the vast majority of the literature focuses on either cattle or horses; with few studies on the impact of ectoparasiticides on soil health through their use on sheep. The physical structure and faunal communities of sheep, cattle and horse dung are very different (Beynon, 2012b and references therein), and so it is difficult to draw conclusions on the effect of ectoparasiticides on soil function through sheep dung from studies on cattle and horses. There is some evidence that a particular ectoparasiticide, organophosphate can remain in the environment for long periods of time: Ragnarsdottir, (2000) reports that these compounds are often detected in soil years after application. These compounds have relatively high KOCs (Table 1.4): A higher KOC value indicates a greater tendency for the chemical to adsorb to soil particles rather than remain in the water phase. Although their strong adherence to soil particles suggest that they are immobile, they could be transported through the environment through the transport of particulates. This could lead to a greater transport distance which could pose a threat to the wider ecosystem. A review by Beynon (2012b) reported that the few studies available on the impact of ectoparasiticides on dung fauna were inconclusive. However, there is evidence that endoparasiticides are lethal to some earthworm species when applied topically at manufacturer-recommended doses (Potter et al., 1994), which could also then negatively impact the structure of the soil.

The literature is also inconclusive as to the effects of either endo- or ectoparasiticides on soil microbial communities. Förster et al. (2011) found no significant effect on soil microbial biomass or respiration when ivermectin was applied to cow slurry. Lagos et al. (2022) found that micro-organisms were not impacted by the compounds, but instead played a significant role in the dissipation of albendazole and ivermectin. Soil microbial communities are important in maintaining ecosystem functioning, and are responsible for the mineralisation of organic matter and in breaking down organic contaminants (De Schrijver and De Mot, 1999). Further research into the effects of both groups of antiparasitic treatments on the effects of microbial communities is therefore needed.

There is clearly a need for new research into the effects of antiparasitic treatments on soil health, given that earlier studies have reported negative impacts of their use on soil and dung fauna and microbial communities, and that these communities play essential roles in the functioning of soil ecosystem services, particularly the development and maintenance of a healthy soil structure. Furthermore, many organic farms were previously conventionally managed, and so it could be beneficial to investigate the presence of antiparasitic medicines in the soil after converting to organic to see if there are signs of recovery and establish whether the lower application rates used in organic farms are proving to be beneficial to soil faunal communities.

1.5 Synthesis

It is clear from the literature that grazing can impact many key soil functions, and that these effects could potentially differ under organic grazing management compared to conventional grazing, due to their differences in practices such as fertiliser inputs and the use of veterinary treatments. However, studies on the effects of organic grazing on upland soil functions were missing from the literature, highlighting a large research gap. Soil structural differences, in particular, could benefit from further study, as this could potentially differ between the two management systems due to their differences in N input and the use of veterinary treatments. The review of the literature also suggested a research gap on the fate of veterinary treatments in the upland soil system. The application of veterinary treatments in conventional livestock management differs markedly to organic livestock management and so further research into the fate of veterinary treatments in the soil and how it could differ between the two systems is needed. UK organo-mineral soils in particular were missing from the literature in all grazing-related subject areas, and few studies exist for soils located in cool-wet climates, such as

UK grassland soils. This was especially true for studies regarding the effects of grazing on soil physical properties and hydrological functions. Since many UK grazing sites are located in upland areas, more research is needed to understand how the different grazing management systems may be impacting UK upland organo-mineral soils.

1.6 Aims and objectives

1.6.1 Research questions

The central aim of this project is to investigate the impacts of livestock grazing in upland areas on soil function, examining how this impact might differ under organic grazing management compared to conventional grazing management, and where possible, to develop understanding of the controls behind these differences. This aim can be separated into three research questions:

Table 1.5: Thesis research questions and the soil functions they aim to investigate.

Research Question	Soil function(s)	Chapter(s)
1. How do soil physical and chemical properties differ under organic grazing management compared to conventional grazing management?	- Hydrological function - Carbon storage & cycling - Nutrient cycling	2, 3
2. Do earthworm populations differ under organic grazing management compared to conventional grazing management?	- Biological function - Nutrient cycling	3, 4
3. Do anthelmintic treatments used in conventional livestock farming impact the avoidance behaviour of earthworms?	- Biological function	4

1.6.2 Research approach & thesis structure

A case-study approach was used to answer the first three research questions, and the final research question was investigated using a laboratory experiment, based on findings from the case study. The same field sites were used in two of the research chapters. Therefore, to avoid repetition, site selection and site descriptions are outlined below (section 1.6.3). The rest of this thesis is organised into three research chapters which are described below, ending with a final synthesis chapter, which reflects on the research as a whole, considering the central aim of the research project.

- **Chapter 2: Impact of organic sheep grazing on upland soil functioning**

Field measurements and field samples were taken to measure a variety of soil physical and hydrological properties at organic and conventional field sites to answer research question 1.

- **Chapter 3: Earthworm populations in upland soil under organic and conventional livestock grazing**
  Earthworm surveys were conducted at all field sites. Results from the two management systems were then compared in order to answer research question 2.
- **Chapter 4: Avoidance behaviour of *Allolobophora chlorotica* under exposure to the anthelmintic drugs albendazole, moxidectin, ivermectin and levamisole**
  This chapter aims to answer research question 4, based on earthworm survey results from Chapter 3. The avoidance behaviour of *Allolobophora chlorotica* was investigated when exposed to anthelmintics known to have been used at conventional field sites.

1.6.3 Field site selection and description

Three pairs of sites were chosen, whereby each pair comprised one organic and one conventional upland sheep farm located in the same area Figure 1.7. Triplicate pairs were chosen to ensure good statistical power within a feasible sampling timeframe.

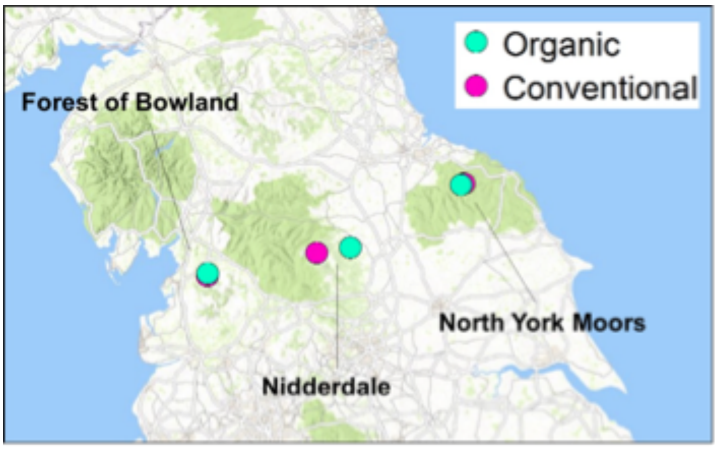

Figure 1.7: Map depicting the location of field sites in the north of England

The sites were chosen after identifying all organic sheep farms in northern England. Three organic sites were then chosen from this list where there was a comparative conventional site within 20 km which had similar climate and grazing regime. One field was chosen at each site for sample collection, based on: a) size, ensuring similar sized sampling fields across all sites; b) the field was predominantly grazed by sheep, and c) the field had been grazed in the last 12 months. A single field was chosen at each site to ensure sampling was feasible within the project timeframe. The sites used in this study can be classified as being organo-mineral (organic content of surface material >20% (Joint Nature Conservation Committee, 2011; Forestry Commission, 2019). Further details on the field sites are provided in Table 1.5.

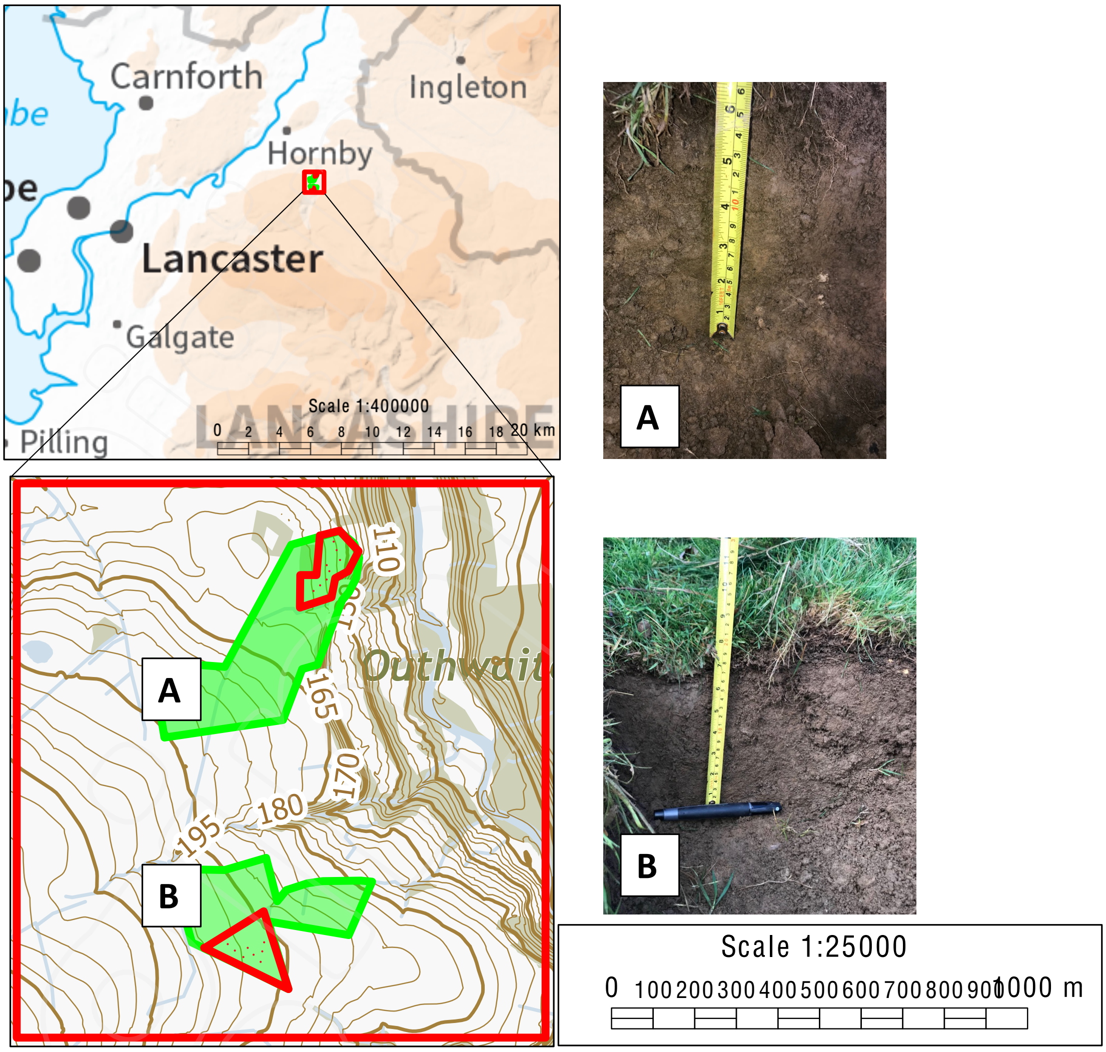

Figure 1.8: Location of field sampling sites at Forest of Bowland, sample plots (red dots), and soil profile at Forest of Bowland. A= organic site (FoB-O), B = Conventional site (B). See table 1.5 for grid references.

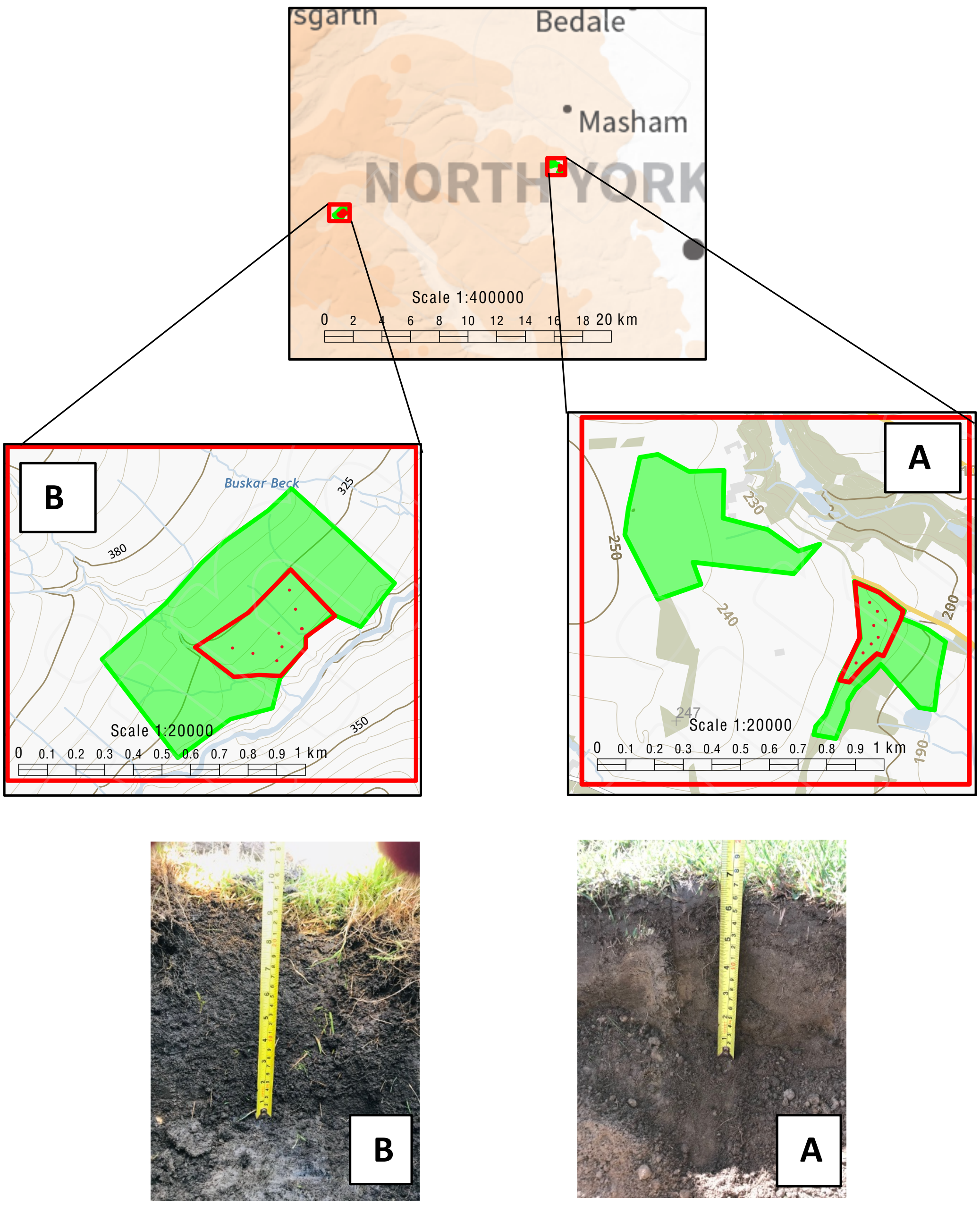

Figure 1.9: Location of field sites at Yorkshire Dales, location of sampling points (red dots) and soil profiles. A= Organic (YD-O), C=Conventional (YD-C). See table 1.5 for grid references

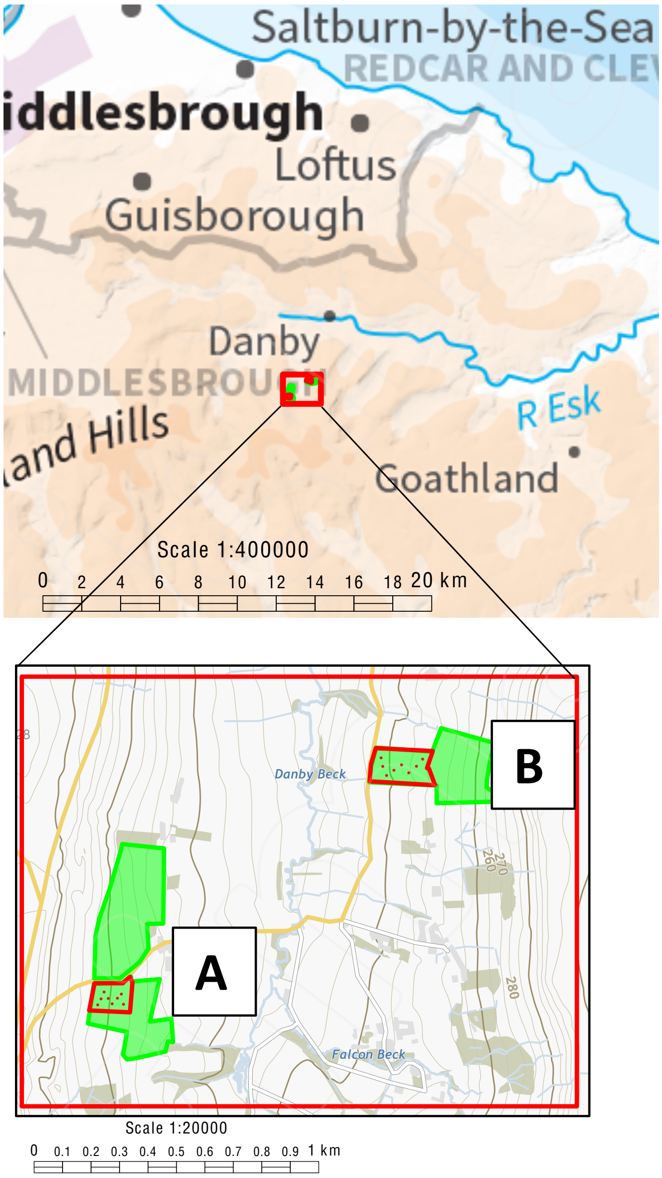

Figure 1.10: Location of field sites and sampling points at North York Moors. A=Organice (NYM-O), B=Conventional (NYM-C)

Table 1.6: Description of study sites. Annual mean rainfall obtained from Met. Office (2020), Soil types are classified by WRB, and obtained from Cranfield University (2023) *1 LU is equivalent to adult dairy cow. One upland ewe is 0.08 LU ha^{-1}, one beef cow = 0.75 LU ha^{-1}, one suckler cow = 0.65 LU ha^{-1} (Nix, 2003)

Area	**Forest of Bowland (FoB)**		**Yorkshire Dales (YD)**		**North York Moors (NYM)**	
Site	**FoB-C**	**FoB-O**	**YD-C**	**YD-O**	**NYM-C**	**NYM-O**
Management	Conventional	Organic for >50 years (Soil Association)	Conventional	Organic for >10 years (Soil Association)	Conventional	Organic & Biodynamic organic for >80 years (Biodynamic Agricultural Association)
Grid reference	54º05'N 2º36'W	54º05'N 2º36'W	54º09'N 1º53'W	54º11'N 1º40'W	54º26'N 0º55'W	54º25'N 0º56'W
Elevation range (m above sea level)	175-215	165-202	321-347	216-224	194-219	238-262
Annual mean rainfall (mm) 1991-2020 (Met. Office, 2020)	1315		1587		980	
Soil type & parent material	Predominantly Wilcocks 721c (Umbric Stagnosol) with Brickfield (Br) 713g (Eutric Stagnosol) at lower altitudes, Parent material: Drift Palaeozoic sandstone, mudstone and shale. (Cranfield University, 2023a)	Brickfield (Br) 713g (Eutric Stagnosol) Parent material: Palaeozoic sandstone and shale (Cranfield University, 2023b)	Wilcocks 721c (Umbric Stagnosol) Parement material: Drift from Palaeozoic sandstone mudstone and shale (Cranfield University, 2023c)	Predominantly Rivington 1 0541f (Eutric Endoleptic Cambisols) with some Dunkeswick 711p (Eutric Albic Luvic Stagnosols) at higher altitudes. Parent material: Till from Palaeozoic and Mesozoic sandstone and shale (Cranfield University, 2023d)	Predominantly Stow 421a (Stagnic Cambisols) with Onecote 721b (Clayic Dystric Histic Stagnosols) at higher altitudes) Parent material: Jurassic mudstone and siltstone (Cranfield University, 2023e)	Stow 421a (Stagnic Cambisols) Parent material: Jurassic mudstone and siltstone (Cranfield University, 2023f)
Soil texture (%)	Sand: 23.7 ± 1.9 Silt: 42.1 ± 2.7 Clay: 34.2 ± 4.2 Texural Class: Clay Loam	Sand: 31.7 ± 0.6 Silt: 54.1 ± 4.8 Clay:: 14.2 ± 4.4 Texural Class: Sandy Silt loam	Sand: 15.1 ± 3.2 Silt: 60.1 ± 2.8 Clay: 24.8 ± 1.1 Texural Class:Silty Clay Loam	Sand: 37.5 ± 1.4 Silt: 51.5 ± 0.9 Clay: 11.0 ± 1.9 Texural Class: Sandy Silt Loam	Sand: 16.6 ± 0.9 Silt: 64.2 ± 2.4 Clay: 19.2 ± 1.8 Texural Class: Silty Clay Loam	Sand: 28.0 ± 1.3 Silt: 37.5 ± 3.0 Clay: 34.5 ± 3.5 Texural Class: Clay loam
Farmyard manure application	Surface application of 6 t ha^{-1} year^{-1} of FYM in May	None	None	None	None	None
Anthelmintic usage	All animals treated with: Moxidectin and triclabendazole April 2018; Albendazole September 2018; Moxidectin and triclabendazole April 2019	Whole flock treated with levamisole in Jan 2017; individual ewes given ivermectin or moxidectin in June 2017, no treated sheep in the field since this date	Whole flock treated annually with moxidectin, albendazole and triclabendazole, last treated April 2018	Individuals given albendazole April 2018	Unknown due to a change in farm management	Anthelmintics not used in 10 years, instead they use a 'biodynamic' treatment of garlic and vinegar
Stocking & pasture area information	~7 sheep per ha plus ~1.3 cattle April-Oct (~50 ewes and ~10 cattle grazed over ~7.5 ha)	~7 sheep per ha all year round, no cattle (~70 ewes grazed over ~10.5 ha area)	~6 sheep per ha plus ~1 cattle per ha April-Sept (~60 sheep and ~10 cattle grazed over ~10 ha area)	~3 sheep per ha plus ~1 cattle per ha April-Oct (~50 ewes & & ~15 cattle grazed over ~16 ha area)	~9 sheep per ha, Apr-Oct, no cattle (~60 sheep grazed over ~7 ha area)	~5 sheep ha^{-1} and 0.5 cattle per ha all year round (~50 sheep and ~6 cattle grazed over ~10.5 ha area)
*Annual stocking density (LU ha^{-1})**	1.4	0.5	1.2	0.9	0.7	0.8

Chapter 2: Impact of organic sheep grazing on organo-mineral soil functioning

2.1 Introduction

In recent decades there has been increasing concern about the impacts of livestock grazing on essential soil functions. (Chantigny, 2003; Marshall et al., 2014; Stavi et al., 2016; Bünemann et al., 2018; Schils et al., 2022). Surface pressures from grazing livestock have been shown to result in surface erosion and carbon losses (Mcsherry and Ritchie, 2013b; Eldridge et al., 2017c) and increase the risk of compaction and overland flow (Drewry et al., 2018). In the UK, around half of all breeding ewes and beef cattle are grazed in upland areas (Defra, 2011), where soils tend to contain high levels of organic matter. Indeed, 59% of organo-mineral soils in the UK are located in upland areas (Bol et al., 2011). Upland soils provide vital ecosystem services such as carbon storage, water quality regulation and flood mitigation (Haines-Young and Potschin, 2009). However, we are yet to fully understand the functioning of organo-mineral soils in upland areas, or how they respond to different management practices such as livestock grazing.

Organic livestock farming and pasture management has been recognised as a sustainable alternative to more intensive grazing systems (Tuomisto et al., 2012; Soil Association, 2021). There are key differences between organic and conventional livestock management (outlined in section 1.3) which could have a potentially beneficial impact on upland soil function. Stocking rates in upland areas are already relatively low, with farms usually stocking around 1 livestock unit per hectare (LU ha^{-1}). However, many organic sites tend to have more extensive stocking rates than their conventional alternatives (Table 1.6). Lower livestock units per hectare could reduce surface pressures, helping to maintain soil and pore structure thereby maintaining good hydrological function (Marshall et al., 2014). Moreover, veterinary antiparaciticides used in conventional livestock management can negatively impact soil fauna communities (Beynon, 2012b), therefore, the limited use of antiparasitic veterinary medicines in organic farming might be more beneficial for communities of important soil bioengineers such as earthworms, which help maintain good soil and pore structure, and retention of organic matter, by creating stable aggregates (Hallam and Hodson, 2020).

As outlined in the review of the literature in the previous chapter, soil physio-hydrological and chemical properties can be an effective approach to assessing a variety of soil functions (Figure 1.4). Therefore, given the need to broaden our understanding of upland soil functioning under organic grazing management, this chapter uses a case study approach to examine and compare the differences in key soil property indicators to assess the soil hydrological functioning, carbon storage and cycling and nutrient cycling functions between the organic and conventional upland sites described in section 1.6.

2.2 Methodology

2.2.1 Sampling and field measurements

Soil samples were taken from each field from field sites outlined in Section 1.6.3 and Table 1.6. In each field, eight plots were chosen using a W sampling pattern, also known as the modified diagonal (Peters et al., 2007). A 20 x 20 x 20 cm pit was dug in each plot. From the side of each pit, an intact soil sample was taken at each 5 cm depth increment (0-5, 5-10, 10-15 and 15-20 cm) using a bulk density ring (5 cm diameter and 5 cm length, Eijkelkamp, The Netherlands) hammered horizontally into the soil. Samples were collected from eight plots at each site in autumn (October 2018) to analyse soils that had been continually grazed for at least 6 months prior to sampling. The same number of samples were taken again from different pits in spring at the start of the grazing season (April 2019). The intact soil cores were then returned to the laboratory on the day of sampling and refrigerated at 4°C until analysis of soil hydrological (soil permeability and proportion of flow through different pore size classes) and physical properties (bulk density, organic matter)

In situ soil strength was measured at 5 cm depth increments down to 50 cm using an Eijkelkamp cone penetrometer at 20 locations using a W sampling pattern in each field in October 2018. The conical point was 1 cm^2 in area, and the measurement range was 1000 MPa. The cone was inserted into the soil at an approximately constant rate of 2 cm s^{-1}, while recording the changes in pressure and at which depth these occurred, until the penetrometer reached either its depth or pressure limit.

Single loose soil samples from 0-10 and 10-20 cm depth intervals were also taken on the same day as the soil core sampling in October 2018 and April 2019, and then from additional pits dug in August 2019. These samples were used for determination of soil texture and soil chemical properties (soil pH, available N, exchangeable cations).

2.2.2 Laboratory measurements

As an indicator of nutrient cycling functioning, available N (NH_4-N, NO_3-N) was extracted from 10 g field moist subsamples of the loose soil from 0-10 and 10-20 cm horizons using 1 M potassium chloride (KCl) and analysed within 24 h using a Skalar SAN++ auto analyser. Further 10 g subsamples were oven dried at 105 °C for 24 hours and weighed to determine soil moisture content. Exchangeable basic cations (calcium (Ca), magnesium (Mg), potassium (K) and sodium (Na)) were also extracted from air dried 5 g subsamples from 0-10 and 10-20 cm horizons using Silver Thiourea (Pleysier and Juo, 1980) and analysed using a Thermo Scientific iCAP 7600 Duo ICP-OES Analyzer. Soil pH for 0-10 and 10-20 cm horizons was determined in a 1:2.5 soil to deionized water ratio (Van Reeuwijk, 1993)

The saturated hydraulic conductivity (K_s) (indicator of hydrological functioning) of each intact soil core sample was measured using an Eijkelkamp 25 place laboratory permeameter. Intact core samples were first saturated with water, then inserted into the permeameter which forms a difference in head across the sample. Samples were taken from the permeameter, removed from their rings and oven dried at 105°C for 24 hours then weighed. Large stones were removed using a 2 mm sieve, weighed and dry bulk density determined (indicator of hydrological functioning, & used to assess soil carbon contents), assuming that stone density is equal to 2.6 g cm^{-3} (Don et al., 2007; Poeplau et al., 2013; Eze et al., 2017b). A subsample from each soil core was taken to measure organic matter content (indicator of soil carbon storage and cycling) using the loss on ignition method (Ball, 1964; Rowell, 1994). The remaining soil from each core was then reweighed and washed on sieves of different mesh sizes (50 µm to 2 mm) to separate the roots. Roots were oven dried at 65°C for 48 hr and then weighed to determine root biomass. SOC stocks (kg m^{-3}) for each depth increment (0-5 cm, 5-10 cm, 10-15 cm, 15-20 cm) (indicator of carbon cycling and storage) were calculated from values of SOM concentration and bulk density using the relation:

$$SOC \text{ stocks } (\mathrm{kg\ m^{-3}}) = C \times BD \times D \qquad (2.1)$$

where C = SOC concentration (%) calculated using SOM × 0.54, BD = bulk density (kg m^{-3}), D = soil depth (cm).

A mini disk infiltrometer (METER group inc., USA) was used to measure the flow through macropores of different pore size classes (<0.5 mm, 0.5-1 mm, 1-3 mm, >3 mm) (indicator of hydrological functioning) in each of the field-moist intact soil core samples collected in spring, prior to permeameter measurements. The infiltrometer was held in place on top of the core samples using a clamp-stand to prevent the weight of the instrument compressing the soil surface. Infiltration measurements were taken until a steady-state condition was reached using supply heads of -6, -3, -1 and 0 cm, which enabled flow through pore sizes of equivalent diameter equal to or less than 0.5 mm, 1 mm, 3 mm, and all pores respectively (Reynolds and Elrick, 2010). The minimum effective porosity (θ) volume was estimated from the tension infiltrometer data using Poiseuille's equation (2.1) (Holden, 2009), where μ is the viscosity of water, ρ is the density of water, g is the acceleration due to gravity, r is the minimum radius for the size class, and K_m is the pore flow at a given pressure head and the next higher pressure head (Azevedo et al., 1998):

$$\theta = (8\mu K_m / \rho g r^2) \qquad (2.1)$$

2.2.3 Statistical analyses

The normality of all data was tested using the Anderson-Darling test and, where left skewed (K_s, effective porosity, organic matter, root biomass, moisture content, available N and exchangeable cations), the data were log10 transformed. Data were re-tested using the Anderson-Darling test which confirmed that the log10 transformation had resulted in the normalization of the data. An ANOVA general linear model was then used to test for differences ($p < 0.05$) in soil properties by season, management, location, and depth, with post-hoc Tukey tests determining differences between pairs. The non-parametric Kruskal-Wallis test was used on soil strength data as they did not meet the requirements for an ANOVA test. All statistical analysis was completed using Minitab 19.

2.3 Results

Soil bulk density under conventionally grazed sites was generally lower than under organically managed sites (Figure 2.1) ($p < 0.001$) (mean of 0.85 and 1.06 g cm^{-3} for conventional and organic respectively). Bulk density also varied significantly with depth ($p < 0.001$), with the biggest differences being under conventional sites (mean difference of 0.42g cm^{-3} between 0-5 cm and 15-20 cm under conventional and 0.32 g cm^{-3} under organic) (Figure 2.1). Both autumn and spring results show a pattern of increasing bulk density with depth under both management systems. However, mean values of bulk

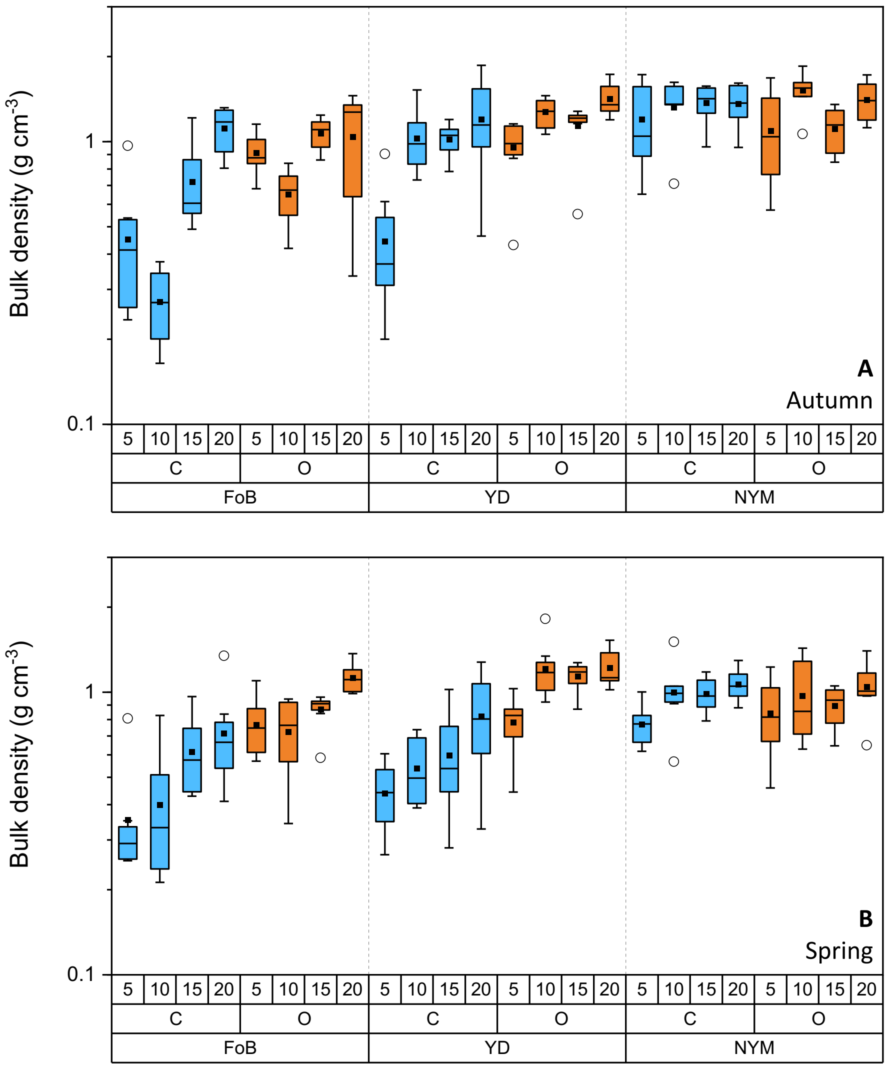

Figure 2.1: Bulk density values in (A) Autumn, and (B) Spring, for 0-5 cm (5), 5-10 cm (10), 10-15 cm (15) & 15-20 cm (20) depth intervals for all sites. C = Conventional, O = Organic, FoB = Forest of Bowland, YD = Yorkshire Dales, NYM = North York Moors. Box lines indicate the median, boxes the 25th and 75th percentile, square markers indicate the mean, whiskers the minimum and maximum values (excluding outliers), circles (more than 1.5 interquartile ranges) represent outliers.

density were significantly lower in spring for all sites (mean difference of 0.32 g cm^{-3} for all depths under conventional and 0.19 g cm^{-3} under organic) ($p < 0.001$).

Soils at conventional sites on the whole contained more organic matter (OM) than organically managed sites (mean value of 27 % under conventional and 13 % under organic), particularly at 0-5 cm (mean value of 39 % under conventional and 14 % under organic) with the exception of both sites at NYM, where OM content was very similar under all depths and seasons (mean of 12 % under conventional and 13 % under organic) (Figure 2.2). ANOVA showed OM varied significantly with management ($p < 0.001$), depth ($p < 0.001$) and season ($p < 0.001$) (Figure 2). OM and bulk density were strongly negatively correlated over all data ($R^2 = -0.61$, $p = <0.001$) therefore, similarly to bulk

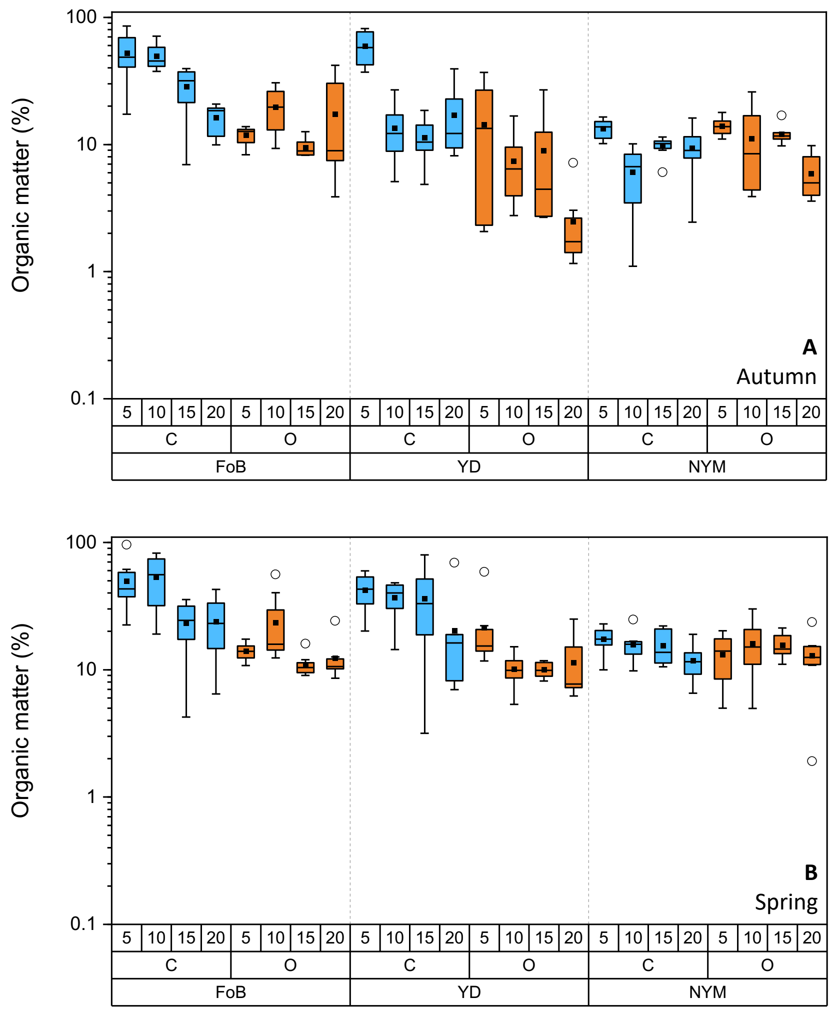

Figure 2.2: Organic matter content in (A) Autumn, and (B) Spring, for 0-5 cm (5), 5-10 cm (10), 10-15 cm (15) & 15-20 cm (20) depth intervals for all sites. C = Conventional site, O = Organic site, FoB = Forest of Bowland, YD = Yorkshire Dales, NYM = North York Moors. Box lines indicate the median, boxes the 25th and 75th percentile, square markers indicate the mean, whiskers the minimum and maximum values (excluding outliers), circles (more than 1.5 interquartile ranges) represent outliers.

density, OM varied much more with depth at conventional sites (mean difference of 22.6 % between 5 and 20 cm) than at organic sites (mean difference of 4.1 % between 5 and 20 cm) (Figure 2.2). Carbon stocks were overall significantly higher under conventional management compared to organic sites ($p < 0.001$) (Figure 2.3) (means of 4.141 kg m^{-3} for conventional and 3.109 kg $m^{-3)}$ for organic), and being significantly higher at 0.5 cm ($p<0.001$). The range of values varied much more widely under organic sites (0.043-11.345 kg m^{-3}) compared to conventional site (0.401-9.056 kg m^{-3}).

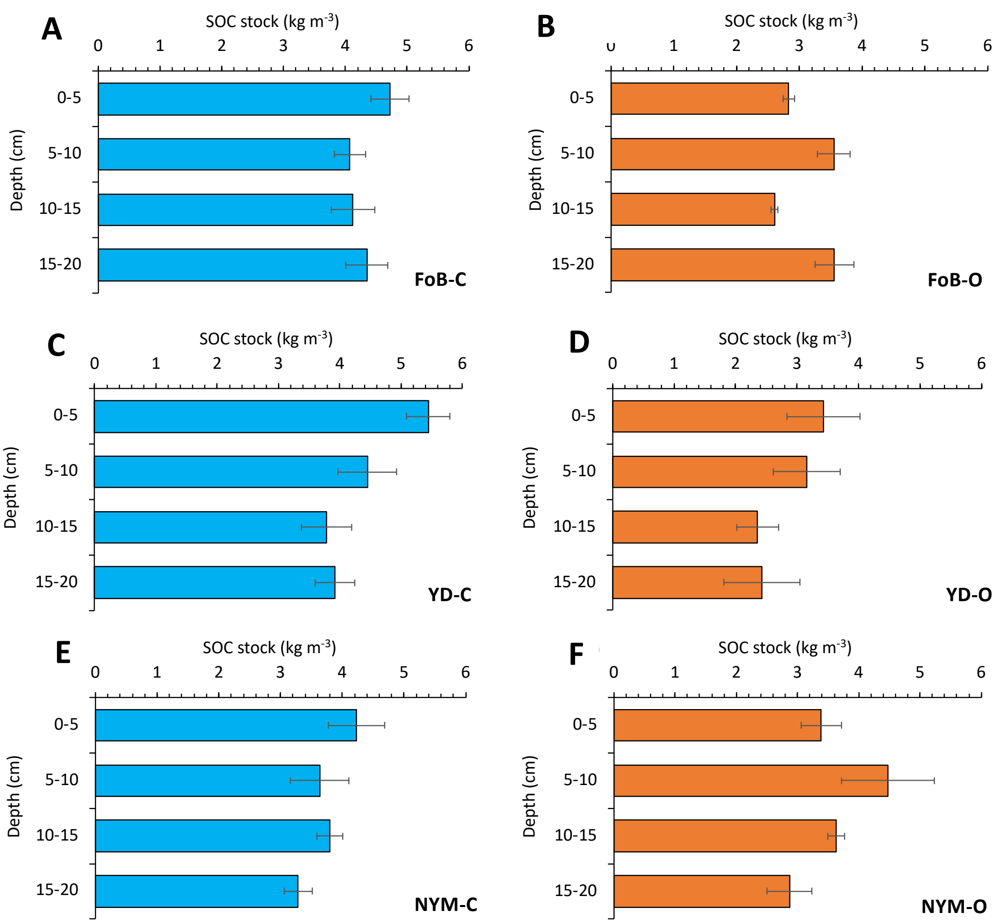

Figure 2.3: Soil organic carbon stock (SOC) content for each depth (0-5, 5-10, 10-15, 15-20) at each site. FoB-C = Forest of Bowland conventional, FoB-O = Forest of Bowland organic, YD-C = Yorkshire Dales conventional, YD-O = Yorksire Dales organic, NYM-C=North York Moors conventional, NYM-O=North York Moors organic. Error bars are one standard error of the mean, *n*=8 for all depths.

	C flux at each site (g/C/m^2/yr)					
Processes	***FoB-C***	***FoB-O***	***YD-C***	***YD-O***	***NYM-C***	***NYM-O***
Inputs						
Uptake via photosynthesis (GPP)[1]	1756	1756	1756	1756	1756	1756
Manure application[2]	270	0	0	0	0	0
Internal cycling						
Above ground biomass (ANPP) at 50% C[3]	125	125	125	125	175	175
Consumed by animals[4]	37.5	37.5	37.5	37.5	52.5	52.5
Returned to litter[5]	87.5	87.5	87.5	87.5	122.5	122.5
Excreted to land[6]	35.85	35.85	35.85	35.85	50.19	50.19
Outputs						
Removed in animal products[7]	1.65	1.65	1.65	1.65	2.31	2.31
Enteric respiration from Cattle (CH_4-C)[8]	7.43	0.00	34.31	5.72	0.00	2.86
Enteric respiration from Sheep (CH_4-C)[8]	3.73	3.73	3.73	3.73	3.73	3.73
Cattle dung decomposition (CH_4-C)[8]	1.05	0.00	4.86	0.81	0.00	0.40
Sheep dung decomposition (CH_4-C)[8]	0.10	0.10	0.10	0.10	0.10	0.10
Ecosystem respiration (CO_2-C)[1]	1538	1538	1538	1538	1538	1538
DOC[9]	33.6	33.6	33.6	33.6	33.6	33.6
Total inputs	*2026*	*1756*	*1756*	*1756*	*1756*	*1756*
Total outputs	*1585.56*	*1577.08*	*1616.25*	*1583.61*	*1577.74*	*1581.00*
Inputs minus outputs	*440.44*	*178.92*	*139.75*	*172.39*	*178.26*	*175.00*

[1] Jones et al. (2017)

[2] Applied at FoB-C only. Calculated from applicatiopn of 6t/ha/yr and 15:1 C:N ratio)

[3] Calculated from Newbould (1985) and site pH values

[4] Calculated from vegetation utilization (Scotlands Nature Agency, 1996), C concentration and stocking rates

[5] Calculated from the difference between annual vegetation production and that consumed by livestock

[6] Calculated from the difference between C consumed by livestock and that removed by animal products

[7] Calculated using Scotlands Nature Agency (1996)

[8] Calculated from Brown et al. (2022)

[9] Calculated from McTiernan et al. (2000)

Table 2.1: Estimated carbon balance (g/C/m^2/yr) for each site, calculated from literature search values and known site information (see Table 1.6) for each site. FoB-C=Forest of Bowland conventional, FoB-O=Forest of Bowland organic, YD-C=Yorkshire Dales conventional, YD-O=Yorkshire Dales organic, NYM-C=North York Moors conventional, NYM-O=North York Moors organic.

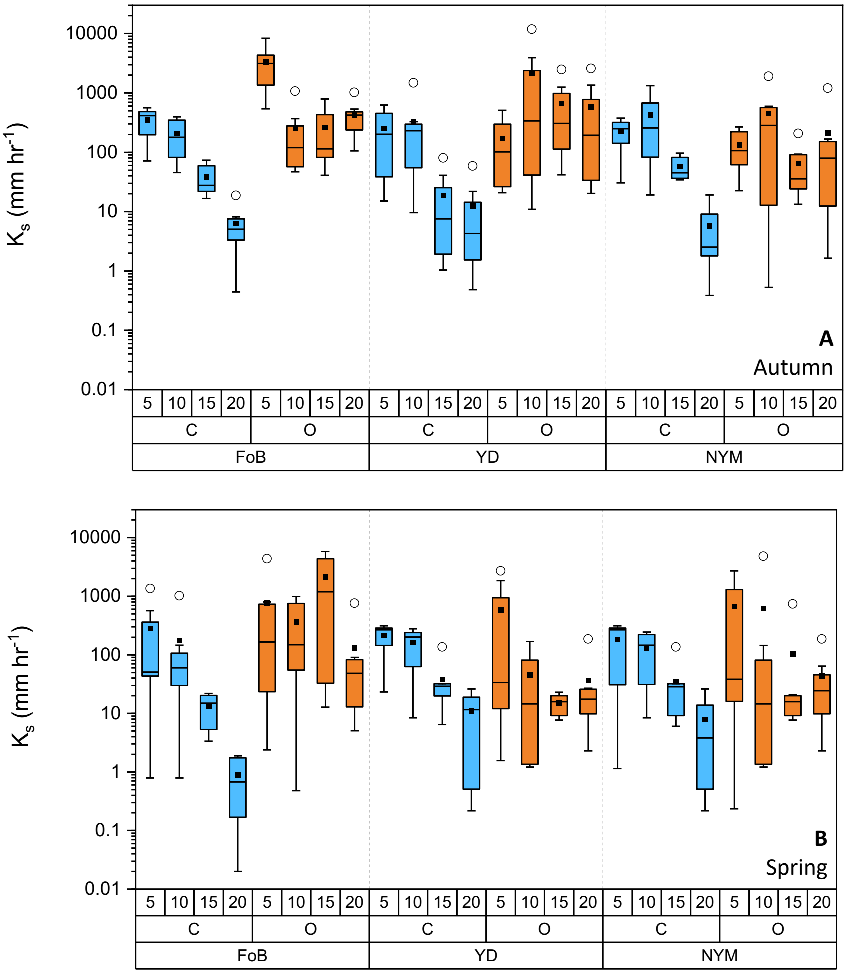

Figure 2.4: Saturated hydraulic conductivity in (A) Autumn, and (B) Spring, for 0-5 cm (5), 5-10 cm (10), 10-15 cm (15) & 15-20 cm (20) depth intervals for all sites. C = Conventional site, O = Organic site, FoB = Forest of Bowland, YD = Yorkshire Dales, NYM = North York Moors. Box lines indicate the median, boxes the 25th and 75th percentile, square markers indicate the mean, whiskers the minimum and maximum values (excluding outliers), circles (more than 1.5 interquartile ranges) represent outliers.

Saturated hydraulic conductivity (K_s) data varied over six orders of magnitude, ranging from 2.0 x 10^{-2} mm hr^{-1} to 1.8 x 10^{4} mm hr^{-1} across the whole dataset. K_s of soils under organic sites was significantly higher than under conventional sites ($p < 0.001$) (median of 33 mm hr^{-1} under conventional and 65 mm hr^{-1} under organic) (Figure 2.4). K_s decreased significantly with depth under both management systems ($p < 0.05$). Median K_s under conventional systems became much smaller with depth, reaching 3.3 mm hr^{-1} at 15-20 cm compared to 53 mm hr^{-1} at organic sites (Figure 2.4); a pattern which occurred across all sites within the same management regime. There was also a wider range of values at each depth under organic grazing. Season was also an important controlling factor ($p < 0.001$), with K_s decreasing by an average of 160 mm hr^{-1} in spring for both management systems. No correlations were found between OM and K_s or BD and K_s.

The majority of the flow under both management systems was through macropores greater than 1 mm in diameter (mean of 82% of flow) (Figure 2.4a). The proportion of flow through the largest macropores was higher at 0-5 cm depths under organic farming (66%) than conventional sites (44%) at the same depth. Organic sites overall had more flow through pores < 0.5 mm (10 %) compared to conventional sites (5%) which had slightly more flow through pore diameters ranging between 1-3 mm (22 % compared to 18% under organic). Mean effective porosity was higher under conventional sites (0.06 m^3 m^{-3}) than organic sites (0.04 m^3 m^{-3}) ($p = 0.04$) (Figure 2.4b), with effective porosity being highest at 5-10 cm depth, significantly more so under conventional sites (0.13 and 0.01 m^3 m^{-3} for conventional and organic sites respectively). Pores with 1 – 3 mm diameter

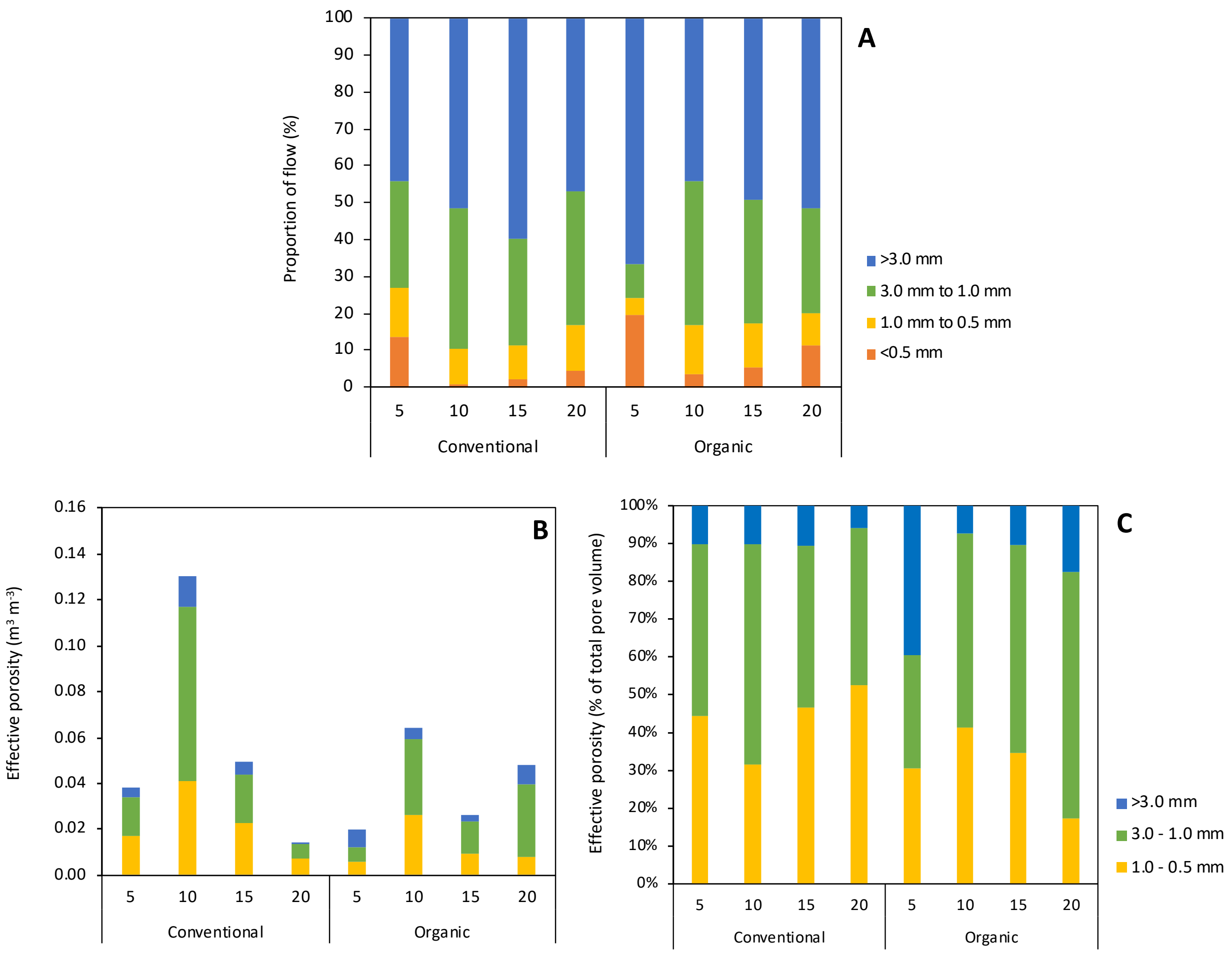

Figure 2.4: Proportion of flow (A) effective porosity (B) and effective porosity expressed as % of total pore volume (C) through different pore size classes for 0-5, 5-10, 10-15 & 15-20 cm depth intervals for conventional and organic sites.

accounted for the largest effective porosity under both management systems (mean of 0.03 and 0.02 $m^3\ m^{-3}$ for conventional and organic sites respectively).

There was little difference in soil strength between the two management systems (Figure 2.5) (p = 0.456) except for YD-C (p < 0.001), which showed a sudden decrease in soil strength at 25 cm depth (Figure 2.5b). Soil strength generally increased gradually with depth at all sites (p < 0.001) except at FoB where it remained fairly consistent with depth, keeping within 2.1-2.5 MPa (Figure 2.5a).

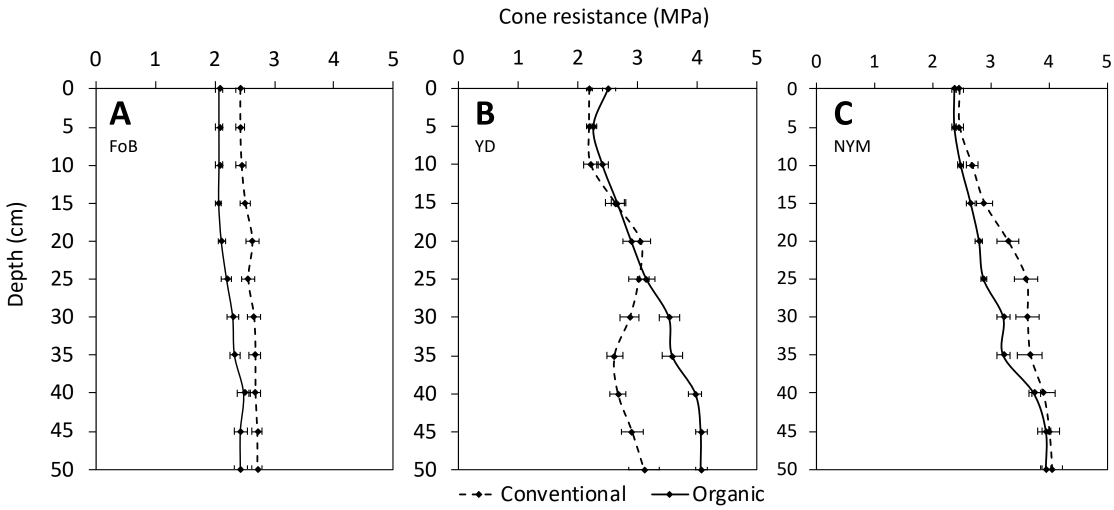

Figure 2.5: Changes in cone resistance with depth for all sites. Error lines represent 1 standard error of the mean (n = 20) for MPa at each depth point. FoB = Forest of Bowland, YD = Yorkshire Dales, NYM = North York Moors.

Soil moisture and volumetric water content were significantly different between management systems (p <0.001) (Table 2.2), There was no significant difference in pH between the two management systems (p = 0.101) although there was a significant difference in pH between locations (p <0.001) (Table 2.2). The mean pH for NYM was 5.85, higher than both FoB and YD which were 5.04 and 4.95 respectively. The pH did not differ significantly with depth at any location (p = 0.885). Available N varied slightly between sites but neither NH_4-N or NO_3-N differed between management systems (p >0.05). Management was a controlling factor for all exchangeable cations (p <0.001) except Ex-K (p = 0.544), with conventionally sites having higher concentrations. However, the difference in cation concentrations between the two management systems was very small (Table 2.2), and is probably of little environmental importance.

Root biomass was not affected significantly by either management (p = 0.462) or season (p >0.875), but both depth and location were controlling factors (p <0.001 for both factors). NYM had significantly lower root biomass (2.06 kg m^{-3}) than at both FoB (5.96

kg m^{-3}) and YD (6.85 kg m^{-3}) areas. Root biomass declined with depth, the biggest difference being between 0-5 cm and 5-10 cm depth (mean of 11.91 and 5.66 kg m^{-3} respectively) (Table 2.2).

Table 2.2: Mean values of soil physio-chemical properties. Values in parentheses are one standard error of the mean (*n* values for each soil property are stated in column headers and are the same for each row in the column). FoB = Forest of Bowland, YD = Yorkshire Dales, NYM = North York Moors. Ex = exchangeable.

Location	Management	Soil depth	Moisture content (kg kg^{-1}) (n = 24)	Volumetric water content (cm^3 cm^{-3}) (n = 16)	pH (n = 6)	Stoniness (%) (n = 32)	Root biomass (kg m^{-3}) (n = 16)	NH_4-N (mg kg^{-1}) (n = 24)	NO_3-N (mg kg^{-1}) (n = 24)	Ex-Ca (cmol kg^{-1}) (n = 16)	Ex-Na (cmol kg^{-1}) (n = 16)	Ex-K (cmol kg^{-1}) (n = 16)	Ex-Mg (cmol kg^{-1}) (n = 16)
FoB	Conventional	<10	1.65 (1.57)	0.19 (0.02)	4.59 (0.18)	5.56 (0.59)	19.20 (1.96)	12.01 (2.07)	0.64 (0.37)	30.76 (4.12)	2.32 (0.53)	11.28 (3.06)	6.40 (1.13)
		10-20	1.04 (0.12)	0.40 (0.05)	4.86 (0.19)	9.25 (1.54)	1.04 (0.07)	3.44 (0.68)	0.15 (0.08)	18.95 (1.68)	1.10 (0.20)	2.11 (0.42)	2.77 (0.29)
	Organic	<10	0.47 (0.04)	0.33 (0.03)	5.30 (0.08)	5.86 (0.94)	2.52 (0.33)	3.35 (0.57)	0.33 (0.14)	21.11 (0.64)	0.59 (0.08)	2.80 (0.70)	2.63 (0.34)
		10-20	0.31 (0.03)	0.82 (0.13)	5.44 (0.04)	12.40 (1.83)	1.07 (0.12)	2.07 (0.22)	0.24 (0.12)	18.05 (0.82)	0.40 (0.34)	1.82 (0.29)	1.80 (0.10)
YD	Conventional	<10	1.24 (0.14)	0.66 (0.10)	4.88 (0.04)	5.23 (0.89)	2.54 (0.32)	4.36 (0.57)	0.29 (0.15)	19.77 (2.24)	1.12 (0.18)	3.98 (1.09)	4.88 (0.59)
		10-20	0.52 (0.19)	0.90 (0.19)	4.43 (0.14)	9.25 (1.55)	0.92 (0.14)	1.78 (0.24)	0.03 (0.02)	16.54 (1.57)	0.33 (0.53)	0.71 (0.12)	3.03 (0.33)
	Organic	<10	0.29 (0.37)	1.09 (0.03)	5.22 (0.07)	5.70 (1.04)	19.05 (3.50)	4.81 (0.55)	1.18 (1.14)	12.33 (1.52)	0.46 (0.16)	3.90 (1.45)	3.71 (0.35)
		10-20	0.26 (0.34)	0.97 (0.03)	5.30 (0.39)	11.50 (1.37)	4.88 (1.64)	5.59 (1.82)	0.23 (0.19)	11.97 (1.17)	0.25 (0.03)	2.78 (0.67)	3.31 (0.42)
NYM	Conventional	<10	0.40 (0.67)	0.34 (0.04)	6.08 (0.21)	7.86 (1.05)	2.48 (0.44)	3.39 (0.85)	0.53 (0.30)	21.89 (1.94)	0.91 (0.14)	2.03 (0.56)	2.47 (0.16)
		10-20	0.42 (0.06)	0.31 (0.02)	6.16 (0.03)	13.29 (1.22)	1.02 (0.12)	2.05 (0.27)	0.38 (0.22)	24.87 (2.59)	0.84 (0.12)	0.69 (0.06)	1.75 (0.26)
	Organic	<10	0.57 (0.05)	0.36 (0.04)	5.71 (0.27)	6.10 (0.83)	3.91 (0.43)	3.64 (0.20)	0.18 (0.08)	23.45 (1.33)	0.74 (0.09)	2.59 (0.83)	2.11 (0.16)
		10-20	0.52 (0.07)	0.25 (0.02)	5.46 (0.03)	9.52 (1.34)	0.85 (0.10)	2.69 (0.34)	0.11 (0.05)	21.54 (1.89)	0.70 (0.11)	1.81 (0.37)	1.93 (0.26)

2.4 Discussion

2.4.1 Physical and chemical properties

There was large variability between and within sites, likely due to the natural spatial variability of soils and the differences in microtopography between sites (Beven et al., 1993; Guzman et al., 2019; Bagarello et al., 2021). However, differences were found in bulk density (BD), organic matter (OM), and hydraulic conductivity (K_s) between organically and conventionally managed sites indicating that organic livestock management may have a different impact on upland soil properties than conventional grazing management systems. Although there was a statistically significant difference in chemical properties between the two management systems, the effect size was very small, and unlikely to have any significant environmental impact.

Conventionally grazed sites had significantly more OM and lower BD at the surface than their organic partner sites. Conventional sites in our study had higher grazing densities than their organic partner sites, and so may be receiving more dung deposits at the surface. However, this is unlikely to result in higher OM content, due to internal cycling of photosynthetic C within the grazing system. OM from dung is already present as above-ground biomass, the remainder of which will then be either returned as litter to the soil surface, released into the atmosphere through decomposition and enteric respiration, or removed in animal produce (lambs, wool etc.). Indeed, all sites were estimated to have similar carbon balance, all being classed as carbon sinks, gaining 140-179 g/C/ha/yr, with FoB-C gaining 440 g/C/ha/yr, primarily due to annual farmyard manure additions (Table 2.1). Higher OM content at 0-5 cm at FoB-C is also likely to be due to annual applications of FYM (Table 1.6). All grazing densities in this study can be classed as extensive-intermediate (<1.0-1.5 LU ha^{-1}). In more intensive grazing systems, lower OM content and higher BD are usually found in the upper soil layers due to surface pressures and erosion from hoof action (Pietola et al., 2005; Meyles et al., 2006; Pilon et al., 2017b). Here, under the more extensive-intermediate grazing densities of upland grazing management, the negative impacts of OM degradation from surface pressures are not occurring, and so instead OM, and therefore C is able to accumulate in both systems. This is in line with findings from a national-scale UK study, that found that total carbon stocks at 0-7.5 cm depth where highest under intermediate grazing, and only started to decrease under more intensive grazing systems (>2.0 LU ha^{-1}), where OM levels were lower than those under extensive farming (Ward et al., 2016b). Accumulation of OM under more extensive-intermediate grazing systems in upland areas may also be exacerbated by the slower decomposition rates which are expected in wetter and cooler climatic conditions found in these areas (Medina-Roldán et al., 2012; Ward et al., 2016b). Low pH levels can also slow down decomposition rates of OM due to reduced microbial and soil fauna activity. This could in part explain the differences in soil OM and carbon stock concentrations (SOC) between management systems at FoB and YD, as conventional sites had significantly lower pH at these locations compared to their organic partner sites (Table 2.2). Historical liming activity can also have an impact on current pH levels. Liming history is unknown beyond the past 50 years (based on farmers' knowledge). Liming was fairly common practice earlier in the early 20th century, and it is plausible that some sites have had liming applications to increase their pH and boost productivity. Both sites at NYM have higher Ex-Ca than at other locations, which might be an indication of historical liming practices, which have caused the increase in pH at these sites.

Although stocking densities were generally lower under organic grazing systems in this study, the difference in LU ha^{-1} between the two management systems is relatively small. Topographical differences between the sites are likely to be a key factor behind the differences in OM (and therefore BD) and SOC stocks. The influence of topography on soil water movement and the accumulation of soil organic matter (SOM) is widely recognized (Raghubanshi, 1992; Burke, 1999; Chen and Chiu, 2000; Lopez et al., 2003; Peri et al., 2016; Duan et al., 2023). Shallower slope gradients tend to facilitate the accumulation of organic matter, primarily due to slower infiltration rates higher moisture content, which can slow down OM decomposition processes. Conversely, steeper gradients promote more rapid downslope movement of soil water, and a more aerated soil, resulting in generally more rapid decomposition rates and comparatively lower levels of OM buildup (Raghubanshi, 1992). This was particularly apparent at FoB-C, which had a lower slope gradient and a higher moisture content than its organic partner site. While soil at both sites is classed as poorly drained stagnosols, FoB-C is predominantly Wilcocks 1 (721c) (Cranfield, 2023a), which has a much deeper (20 cm depth) organic horizon, consisting predominantly of peat. Conversely, FoB-O site had a slightly steeper slope gradient, with the soil type being classed as Brickfield 3 (713g) (Cranfield, 2023b), which comprises a loamy Ap horizon at the top 20 cm. YD-C and -O were situated in different catchment areas, and had very different topography and soil type as a result. YD-C has a peaty-surface layer (Wilcocks 1 (721c) (Cranfield, 2023c) which is classed as 'Slowly permeable seasonally waterlogged fine loamy over clayey upland soil with a peaty horizon' (Cranfield. 2023c). Conversely, YD-O is classed as Rivington 1 (541f) a 'well drained coarse loamy soil over sandstone'. Given that conventional sites at both FoB and YD both have a 20 cm deep peaty horizon, it is expected that OM content within this horizon is very high and would vary very little vertically. The water holding capacity of peat is also much higher, and so a higher moisture content at these conventional sites is also to be expected. The results in this study are therefore unsurprising given the differences in soil types at these locations, which are most likely driving the differences in OM content, rather than the differences in grazing management practices.

The change in BD and OM values with depth was not as prominent at NYM sites, where both sites had a much higher pH throughout the upper soil profile. The higher pH could result in an increased solubility of soil organic matter, with a rise in microbial activity increasing the rate at which organic matter is decomposed (Michalzik et al., 2001; Oste et al., 2002; Whittinghill and Hobbie, 2012). Furthermore, it is well known that soil fauna activity tends to be lower in more acidic soils (Lavelle et al., 1995; Edwards and Bohlen, 1996), therefore bioturbation/soil mixing due to soil fauna activity may be lower at FoB and YD where pH levels were lower. The higher pH at NYM could be a more favourable environment for soil fauna activity, resulting in increased bioturbation and better soil mixing, resulting in a difference in OM vertical distribution when compared to the other two locations.

2.4.2 Hydrological functioning

Although organic sites had a much higher K_s, both management systems had high infiltration rates (Figure 2.4). K_s values for both organic and conventional grazing were similar to other studies measuring K_s in upland areas: 142.9 mm hr^{-1} (Marshall et al., 2014), and upland rough grazing or ungrazed hay meadows, particularly on organo-mineral soils: 5.4 – 6.3 x 10^2 mm hr^{-1} (Bond et al., 2021); 3.6 mm hr^{-1} (Monger et al., 2022). Bond et al (2021) showed similarly high K_s values and spatial heterogeneity in organo-mineral soils under extensively grazed grassland. K_s under organic sites was significantly higher and appears to vary less with depth compared with the vertical stratification at conventional sites. However, the range of values was much bigger at all organic sites when compared to conventional, indicating that there is perhaps much stronger spatial heterogeneity under organic sites. Higher moisture content at 0-10 cm under conventional sites could indicate that these sites are more susceptible to saturation-excess overland flow, especially when considering that infiltration rates at 10-20 cm under conventional management (median of 10.4 mm hr^{-1}) were much lower than under organic sites (median 48.3 mm hr^{-1}). Saturation-excess overland flow has been shown to dominate organic soils due to the high saturation state of the upper organic layers and lower infiltration rates of the lower soil layers (J. Holden et al., 2007; Miles R. Marshall et al., 2009; Bond et al., 2021).

No key differences were found in pore size distribution or effective porosity between the two management systems, with the vast majority of flow (mean of 82% across all data) being through macropores greater than 1 mm, which accounted for 3.2% of the soil volume (Figure 2.4). This highlights the dominance of macropore flow and high surface infiltration rates in these upland organo-mineral soils. High proportional macropore flow values at 0-5 cm depths are also comparable to previous values obtained for organic soils and peatlands (Carey et al., 2007; Holden, 2009; Rezanezhad et al., 2016), further adding to the evidence base about the functional importance of macropore systems in upland soils.

Both physical and hydrological properties varied significantly between spring and summer under both management systems. Samples taken in spring had slightly higher OM content, lower BD, and lower K_s. This agrees with previous findings on temporal fluctuations in soil physio-hydrological properties: Cournane et al. (2011) found macroporosity varied throughout the year, with greater macroporosity observed during the winter; James et al. (2003) found soil moisture was higher at the start of the growing season; and Aksakal et al. (2019) found variations in bulk density and aggregate stability, owed to the disruption of macroaggregate stability during winter freezing, with an increase in cohesion during late spring.

2.5 Limitations and recommendations for future studies

Although this study has found differences in physio-hydrological properties between organic and conventional management, these differences were easily explained by the variations in soil type between the conventional and organic sites, particularly at FoB and YD, where the top 20 cm of the soil profile was predominantly a peaty organic horizon, which is very different to their organic partner sites. These differences in soil types also explain the vertical distribution of the soil physio-hydrological properties. All field sites were carbon sinks, with conventional sites containing higher SOC stocks. However, due to the deeper O horizons at two of the conventional sites, owed to their soil type, this is to be expected. It is therefore important that future studies consider the soil type in more detail, and that the topography of the immediate and surrounding catchment area be considered, to gain a better understanding of the influences of contributing upslope areas. Protecting our carbon stores is a key priority and one of the UNs Sustainable Development Goals (SDG13) (Keesstra et al., 2016). It would therefore be beneficial for future studies to also examine the stability and fractionation of carbon under organic grazing compared to conventional grazing, to further understand the origin and stability of carbon stores under these management systems. This study has also shown differences in hydrological properties between the two management systems through increased infiltration rates. However, it is difficult to infer the full impact of these infiltration rates on overland flow, especially given that this could again be due to differences in soil type rather than management practices. Therefore, it is recommended that future studies investigate this further by conducting more in-field infiltration measurements, particularly during rainfall events, and to consider the effect of seasonality, vegetation composition and surface roughness.

A New Zealand study, Schon et al. (2011) found no differences in soil physical properties between upland conventional and organic grazing systems when identical stocking densities were compared. However, differences were found between soil fauna species groups, with a higher proportion of epigeic earthworms and oribatid mites (Oribatida) under organic grazing, and a higher percentage of predatory mesofauna under conventional grazing. Soil fauna are known to impact soil physical and hydrological properties (Bottinelli et al., 2010; Jouquet et al., 2012; Blouin et al., 2013; Fischer et al., 2014; Hallam et al., 2020; Hallam et al., 2021). It might therefore be beneficial for future studies to also consider the impact of these two grazing management systems on soil fauna and soil biological function.

2.6 Conclusions

In conclusion, it is unlikely that organic management systems at these study sites are impacting soil hydrological function and carbon storage any differently to conventional management. The key differences observed in soil properties under organic grazing management, notably lower overall organic matter content and different vertical distribution of soil properties, is likely due to differences in soil types and not driven by management systems. Spatial variation was high for under both management systems, but there was noticeably more spatial heterogeneity under organic management, which may be due more to differences in site topography. Both management systems were estimated to be carbon sinks, and conventional sites had higher SOC stocks, although SOC stocks could also be explained by differences in soil types. It is therefore recommended that future studies ensure comparable field systems in terms of soil type and topography, and to also examine the impacts on these management systems on soil fauna and vegetation communities.

Chapter 3: Earthworm populations and anthelmintic residues in upland soil under organic and conventional livestock grazing

3.1 Introduction

This chapter seeks to answer research questions 2: '*Do earthworm populations differ under organic upland livestock farming compared to conventional upland livestock farming?'* by investigating whether there are differences in earthworm populations and earthworm biomass between organic and conventional sites.

The contributions of soil fauna in maintaining soil ecosystem functions are well recognised (Donald A. Davidson and Grieve, 2006; Wall et al., 2013; Blouin et al., 2013). Essential soil biological functions include the release and recycling of organic matter and nutrients from above- and belowground, the maintenance of good soil structure through helping to develop and maintain the soil pore system, thereby contributing to good hydrological function (Wall et al., 2013; Hallam and Hodson, 2020). Earthworms play key role nutrient cycling by breaking down organic matter, releasing nutrients into the soil. They ingest partially decomposed plant litter, ingest it, and transport and deposit it throughout the soil profile. Furthermore, earthworm casts tend to be much more microbially active than the surrounding soil (Edwards and Bohlen, 1996). Land management practices such as livestock grazing have been shown to compromise soil biological function, e.g.: surface pressures from grazing livestock can cause soil compaction and higher bulk density, resulting in poor infiltration and waterlogging of the soil. This condition restricts the oxygen availability to soil microorganisms, inducing anaerobic conditions and triggering the emission of greenhouse gases, namely nitrous oxide (N_2O) and methane (CH_4). Soil compaction can also have detrimental effects for earthworm populations (Bottinelli et al., 2014), as earthworms require more energy to burrow through the soil, slowing down burrowing activity (Ruiz et al., 2016; Capowiez et al., 2021). Much of the literature surrounding the impact of upland land management on earthworm populations has focused primarily on liming, and how the increase in pH from this practice can be beneficial to earthworm populations (D. A. Davidson and Grieve, 2006; Bishop et al., 2008; McCallum et al., 2016). However, it is still unclear how more environmentally sensitive grazing systems, such as organic livestock management, might impact soil organisms, or whether these management practices might be beneficial for soil fauna communities such as earthworm populations.

Earthworms are generally grouped into three different functional groups (Bottinelli et al., 2020): i) Epigeic earthworms are small-medium in size which live in, and consume, decomposing leaf litter on the soil surface, producing little to no burrows; ii) endogeic species are medium in size and consume mineral soil with a preference for material rich in organic matter, and burrow continuously, and mostly horizontally, within the upper 5-15 cm of the soil; and iii) anecic species are large earthworms which create large permanent vertical burrows, surfacing to consume decomposing leaf litter, some of which is pulled down into burrows (Bottinelli et al., 2020). The behaviour of earthworms in response to changes in land management can vary between earthworm functional groups, primarily due to the differences in their burrowing behaviours and food preferences. For example, epigeic species may be more responsive to surface disturbances as they inhabit surface plant litter, but may not be as ecologically important in terms of soil physical and hydrological function, since they do not typically burrow in soil (Bottinelli et al., 2020). Anecic and endogeic earthworms both contribute directly to soil hydrological functionality through their burrowing behaviour by creating macro- and mesopores (Blouin et al., 2013). In particular, the deep burrows created by anecic earthworms are known to create large macropores which are important to the soil's hydrological system (Blouin et al., 2013; Hallam and Hodson, 2020). It is therefore important to consider the response of different ecotypes within earthworm communities, to fully understand how land disturbances might impact the biological functionality of the soil.

The grazing density (LU ha^{-1}) at organic sites in this project is slightly lower than at conventional sites, due mainly to the larger land area needed for 'clean grazing' systems (see Table 1.2, Chapter 1), whereby fields are left ungrazed for periods of >12 months to prevent helminth infection (Stubbings et al., 2020; Soil Association, 2021). In conventional farming, antiparasitic veterinary medicines such as anthelmintics are administered regularly throughout the year to prevent infections of helminths. Anthelmintics are commonly used to prevent and treat infections of parasitic worms (helminths) in livestock (Stubbings et al., 2020). Organic grazing management requires a treatment-based approach to the use of anthelmintics, rather than treating whole flocks to prevent infection. This treatment-based approach can result in: i) lower grazing densities as farms tend to require more land area to reduce cross-infection, and so that fields can be left ungrazed for long periods of time, thereby breaking the lifecycle of the helminth (Stubbings et al., 2020; Soil Association, 2021); ii) fewer anthelmintic residues being deposited via dung and urine, as individual sheep are treated once infection in the individual is evident, rather than treating the whole flock prior to/to prevent infection (Stubbings et al., 2020; Soil Association, 2021). Evidence suggests that anthelmintic residues in dung may cause organisms to avoid the contaminated dung deposits (Yeates et al., 2007; Goodenough et al., 2019), which could therefore potentially be altering earthworm communities and biomass due to disturbances in their food supply.

There is, therefore, potential for earthworm species distribution and biomass to differ between organically and conventionally managed pastures, due to differences in managing helminth infections, which can result in lower grazing densities and the possibility of fewer anthelmintic residues being deposited through animal waste. Hence, this study investigates and compares earthworm density, species distribution and biomass under both organically and conventionally managed field sites. It also considers the distribution of the populations between earthworm functional groups due to their different contributions to the soil biological function.

3.2 Methodology

3.2.1 Earthworm collection, identification & biomass

Earthworm collection took place in 2019 on 27 April (NYM), 1-2 May (YD), and 3-4 May (FoB) (Figure 1.8;Figure 1.9 &Figure 1.10), during the most active period for earthworms in temperate climates (Edwards and Bohlen, 1996). Four sample points were selected at each of the 6 sites (3 pairs of sites) (see Table 1.6) by dividing the site into quarters and selecting one sample point at random in each quarter. Soil blocks of 20 x 20 x 20 cm were dug and removed at each sample point, and live earthworms removed by hand-sorting (Butt and Grigoropoulou, 2010; Sherlock, 2018). A vermifuge solution of 10 g mustard powder suspended in 2 L water was then poured into each pit and left to infiltrate through the soil to expel any deeper burrowing anecic species (Butt & Grigoropoulou, 2010). The pit was monitored over a 30-minute period and any emerging earthworms collected. Earthworms were then returned to the laboratory where individual earthworms were placed in 20% *v/v* industrial methylated spirit to anaesthetise and straighten them before being placed in 80% *v/v* industrial methylated spirit to preserve for later identification and biomass measurements (Sherlock, 2018). Adult earthworms (with visible clitellum) were identified following Sherlock (2018), and the number of juveniles recorded. The earthworms were then dried at 60 ºC for 48 hours and their total dry biomass recorded.

The diversity of adult earthworms (8 species) was calculated using the Shannon-Weiner diversity index ($H' = -\Sigma_i\, p_i \ln(p_i)$, where p_i is the proportion of the total count from the ith species) (Gotelli, 2008). This index value was then used to calculate overall species evenness under each management system using Pielou's evenness index: $J' = H' / \log S$, where S is the total number of species.

3.2.2 Statistical analysis

Data distributions of earthworm density and biomass failed to meet the requirements of ANOVA even after transformation. Therefore, non-parametric analyses were used. Kruskal-Wallis tests were undertaken to test for differences ($p < 0.05$) in earthworm biomass and density between organic and conventional sites. Regression analyses were conducted to test for relationships between earthworm density and pH, and earthworm biomass and pH. All statistical analyses were completed using Minitab 19.

3.3 Results

3.3.1 Earthworm density and biomass

Overall, grazing management did not impact total earthworm density across all sites (p = 0.156). When examining each location separately, there was no significant difference in total earthworm density between organic and conventional sites at NYM (p = 0.659) and YD (p = 0.885). However, management had a significant impact on population density at FoB, where the organic site had a much larger earthworm population (median: 225 individuals m^{-2}; 75 individuals m^{-2} for conventional, p = 0.019).

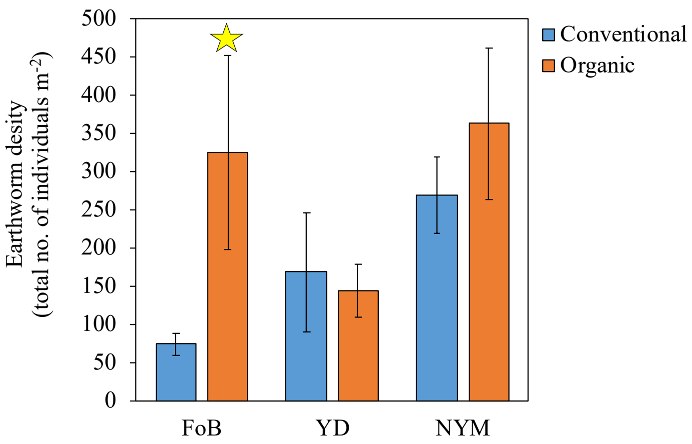

Figure 3.1: Mean earthworm density for all sites. FoB = Forest of Bowland, YD = Yorkshire Dales, NYM = North York Moors. Star indicates significant difference at the organic site at FoB (p = 0.019). Error bars are one standard error, n = 4 (sample points) for each site.

Grazing management had a significant impact on the density of *Allolobophora chlorotica,* with a much higher population density of this species under organic management (medians: 88 individuals m^{-2} under organic; 13 individuals m^{-2} under conventional, p = 0.003), with this species accounting for 33% total abundance under organic sites, compared to 16% under conventional (Figure 3.2a). The difference was particularly high at FoB, where *A. chlorotica* accounted for 44 % of the population under the organic treatment, and 8% under conventional grazing. Management was not a significant factor in variations of earthworm biomass across all sites (p = 0.299). Species evenness (J) was similar for both management systems (J = 0.63 for conventional and 0.55 for organic sites), with

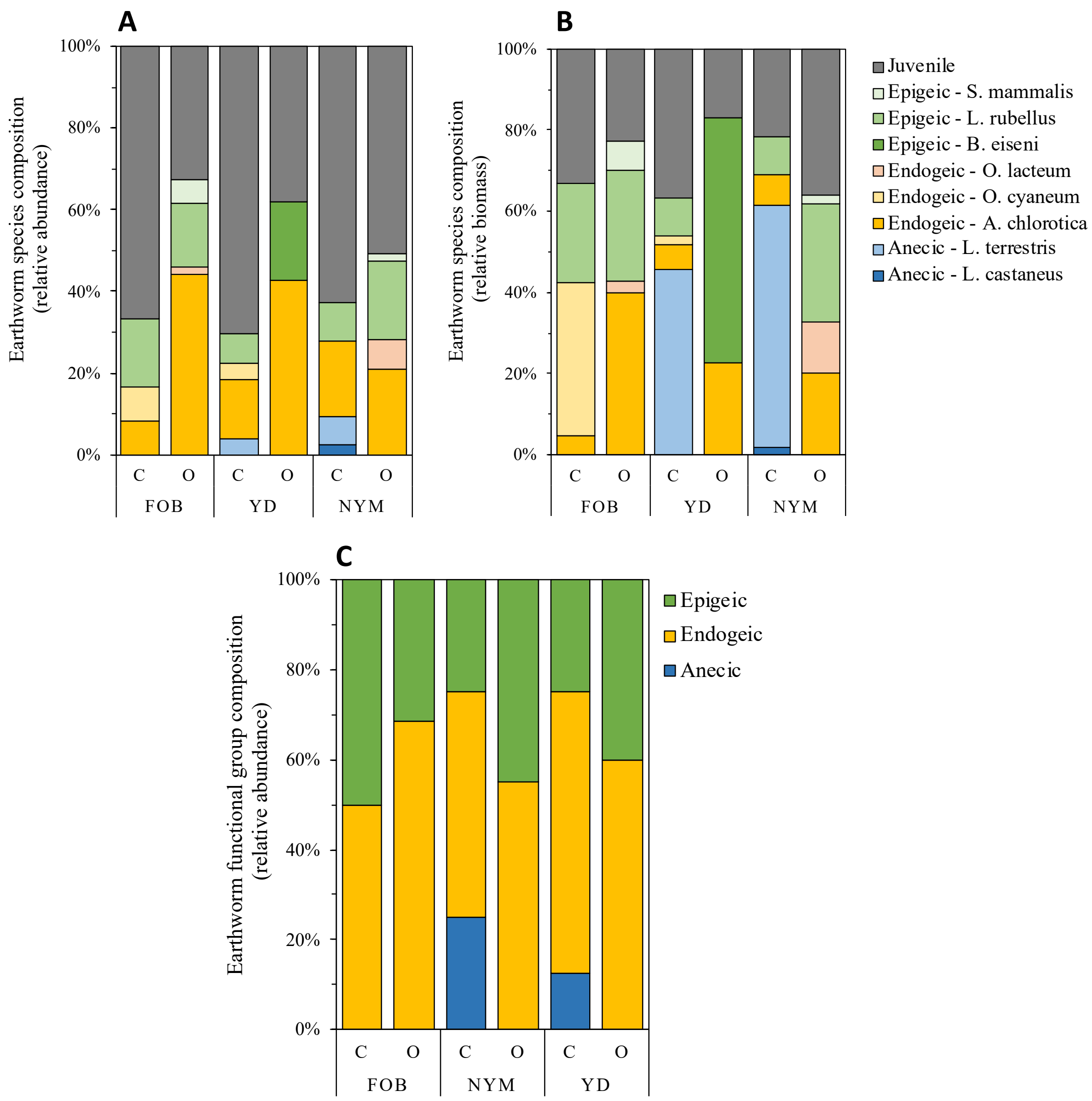

Figure 3.2: Adult earthworm species composition by abundance (A), biomass (B) and earthworm functional group composition by abundance (C).

slightly lower evenness value for organic sites, most likely due to the dominance of *A. chlorotica*.

Organic sites had a significantly higher population density of individuals from the endogeic functional group (Figure 3.2c) (medians of 100 individuals m^{-2} under organic; 25 individuals m^{-2} under conventional; p = 0.002), which is

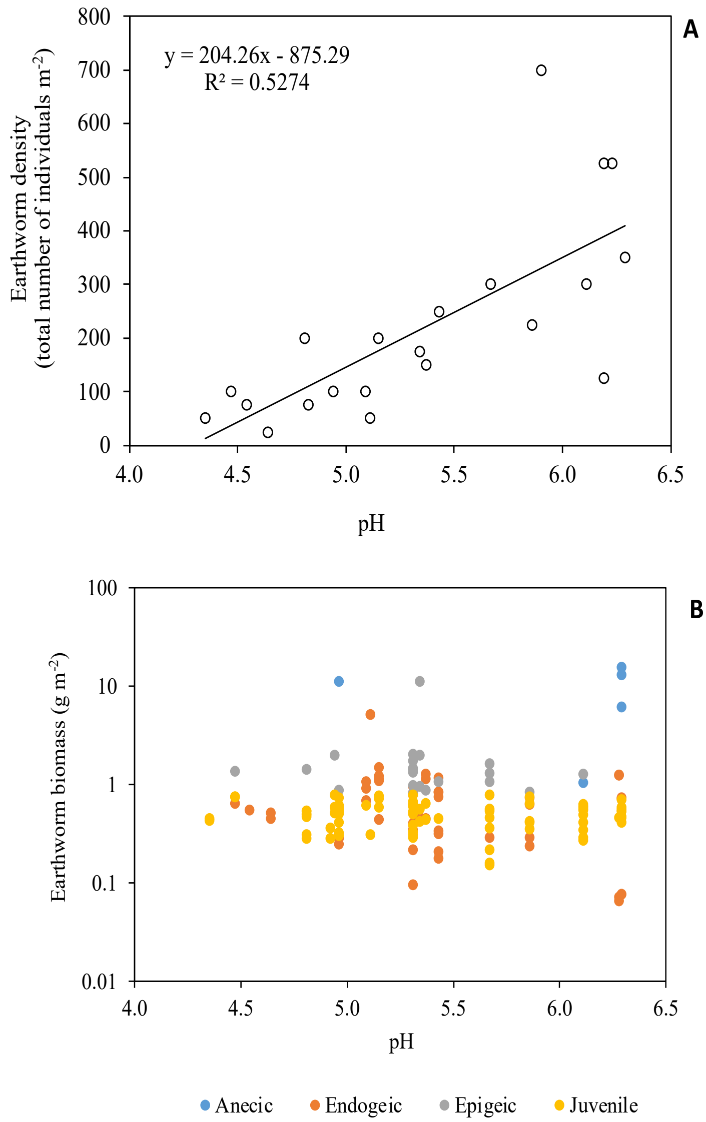

Figure 3.3: Relationship between (A) earthworm density and soil pH across all sites, and (B) earthworm biomass and soil pH across all sites.

unsurprising given that the endogeic population was dominated by *A. chlorotica* (89% overall; 90% under organic sites; 87% under conventional sites). Epigeic species were not impacted by grazing management (p = 0.134). Although anecic species appear to account for a large proportion of relative biomass, this is due to their large size rather than their dominance in the population. Only five anecic adult individuals were found in the entire study (< 5 % of the whole population), and so did not warrant statistical analysis.

Earthworm density across all sites correlated was positively correlated with soil pH (R=0.53, *p*=0.033) (Figure 3.3a). No relationship was observed between earthworm biomass and soil pH within any functional group, across all sites (Figure 3.3b).

3.4 Discussion

3.4.1 Earthworm populations

Total earthworm density and biomass did not differ between organically and conventionally managed sites. However, the population density of *Allolobophora chlorotica* was significantly lower under conventional management compared to organic sites (p = 0.003), suggesting that organic grazing management may be beneficial to this endogeic species, compared to conventional management.

A. chlorotica accounted for over 33% of total abundance at organic sites and 16% under conventional (Figure 3.2a). Median density of *A. chlorotica* under conventional sites (13 individuals m^{-2}) was comparable to other upland grassland studies in the UK & Ireland, e.g.: 2-11 individuals m^{-2} in peaty soil (Cotton and Curry, 1982); 15-40 individuals m^{-2} in ungrazed brown earth (D. A. Davidson and Grieve, 2006; Bishop et al., 2008); and 25 individuals m^{-2} in peaty soil (McCallum et al., 2016). Interestingly, the median density of *A. chlorotica* under organic sites was much higher (88 individuals m^{-2}), and comparable to upland grassland with more neutral pH, particularly upland grassland which had been treated with lime (Bishop et al., 2008; McCallum et al., 2016). (McCallum et al., 2016) found *A. chlorotica* exhibited a strong relationship with soil pH and that abundance peaked in soils at around pH 5.2, whereas the species wasn't found in their sites below pH 4.5. Mean pH at organic sites here was pH 5.4 (Table 2.2), whereas conventional sites FoB-C and YD-C were both pH 4.7 (Table 2.2), and had the lowest abundance of *A. chlorotica*. This indicates that pH might also be an important controlling factor behind the abundance of *A. chlorotica* at these field sites. Indeed, there was a positive correlation between earthworm abundance and pH across all sites.

Historical liming activities may be an important factor here, however the historical liming practices of each site is unknown.

The number of individuals at conventional sites varied widely from 6.25 to 50 individuals m^{-2}, and varied even more so under organic management, ranging from 56 to 144 individuals m^{-2}, similar to the spatial variability of the soil physio-hydrological results in Chapter 2. Earthworms are often found grouped into separate communities within the soil, due to various environmental factors and the complex interactions between them, such as availability of food, soil moisture, avoidance of physical and chemical disturbances, interactions between species, and surface vegetation (Decaëns and Rossi, 2001). Although conventional sites had a slightly higher grazing density than organic sites (Table 1.6), high surfaces pressures from hoof action are unlikely to be the main driving factor in the observed differences in earthworm population density as conventional sites were shown to have lower BD and higher OM than organic sites (Figure 2.1 and Figure 2.2). Instead, it could be that under organic grazing, the spatial pattern of surface disturbances is more variable compared to conventional management, perhaps due to lower grazing densities, which may in turn impact the horizontal distribution of soil fauna communities within the soil.

Endogeic species dominated earthworm communities under both management types, owing to the large number *A. chlorotica,* particularly at organic sites. Possible implications of a higher density of this species might include higher infiltration rates and an increase in the number of soil macropores: endogeic species have been shown to increase soil infiltration in soils where bulk density has been low (Capowiez et al., 2021), and *A. chlorotica* has been shown to have a mean burrowing diameter of 3 mm (Capowiez et al., 2011). Furthermore, *A. chlorotica* has been found to increase the percentage of water stable aggregates and increase the water holding capacity of soil (Hallam et al., 2020). The physio-hydrological properties at all sites determined in Chapter 2 may therefore be related to the high number of *A. chlorotica* found in this study, as all sites had relatively low bulk density, high infiltration rates and high proportion of flow through macropores. However, this interpretation must be approached with some caution, as endogeic species do not create permanent burrows, and connectivity of their burrows to other macropores is generally poor (Capowiez et al., 2011; Capowiez et al., 2014).

However, the effects of different earthworm functional groups on pore size distribution or effective porosity in upland organo-mineral soils is yet to be fully understood and therefore warrants further study.

3.4.2 Implications for soil function and recommendations for future studies

These findings suggest that organic grazing management may be beneficial to *A. chlorotica* populations in upland soils, and that this could possibly be an indication of improved biological functioning at organic sites, as endogeic earthworms are an important species in maintaining good soil structure. The burrowing and feeding behaviour of endogeic species can also result in good soil mixing and accumulation of organic matter within the soil profile through the creation of stable aggregates (Hallam and Hodson, 2020), and so could have important implications for maintaining and securing soil carbon in upland areas, particularly organo-mineral soils. However, it is important to note that juvenile earthworms were not identified to functional group level in this study. Given that they accounted for up to 60% of the total earthworm population in this study, it is recommended that future studies determine at least the functional group of the juvenile worms to further investigate the community structure of earthworm populations under organic grazing management, and how it might impact their life histories.

3.4.3 Conclusions

In this chapter, earthworm communities at organic sites were shown to have a higher density of *A chlorotica*, an important endogeic species. The range of population density values was high under both management systems, but more so under organic management. As with physio-hydrological properties in the Chapter 2, there was a broad range of abundance values, indicating strong spatial heterogeneity in earthworm communities in upland areas. Further studies on the physical properties of burrows of different earthworm functional groups, especially in organo-mineral soil, is recommended, in order to further our understanding of how earthworm communities may influence soil hydrological function.

Chapter 4: Avoidance behaviour of *Allolobophora chlorotica* under exposure to the anthelmintic drugs albendazole, moxidectin, ivermectin and levamisole

4.1 Introduction

Studies have found residues of anthelmintics in the environment (Boxall et al., 2002). Given that I found the population of *Allolobophora chlorotica* to be significantly lower at conventional sites (Figure 3.2), this chapter aims to answer research question 3, by looking at the avoidance behaviour of *A. chlorotica* under anthelmintics known to have been used at the sites in this study (Table 1.6).

Anthelmintics are a group of anti-parasitic medicines commonly used to prevent and treat infections of parasitic worms (helminths) in animals (commercial and companion) and humans, and are widely used in livestock farming (Stubbings et al., 2020). The majority of anthelmintics used in livestock farming belong to one of three groups, based mainly on their chemical structure and mode of action: Benzimidazoles (BZ), Levamisole (LV) and Macrocyclic lactones (ML). Anthelmintics in these groups are all classed as 'broad-spectrum' (except for Triclabendazole, a BZ used to treat liver fluke), meaning they can treat a broad range of helminth infections (Stubbings et al., 2020). Although they can have different modes of action, they all essentially work to induce paralysis in the parasite. Careful guidance is given on the administration of anthelmintics, encouraging targeted use (Stubbings et al., 2020), not only due to rising concern of anthelmintic resistance in helminths (Besier and Love, 2003; Wolstenholme et al., 2004; Pedreira et al., 2006; Pedreira et al., 2006; Matthews, 2014), but also the increasing number of studies suggesting anthelmintics may be harmful to a range of non-target organisms, particularly soil and dung fauna, which could be detrimental to the functionality of grazed soils (Beynon, 2012a; de Souza and Guimarães, 2022).

Much of the research in the past few decades has focused on the impacts of anthelmintics on dung fauna such as dung beetles and dung flies (Hempel et al., 2006; Iwasa et al., 2008; Römbke et al., 2010; Blanckenhorn et al., 2013; Adler et al., 2016; Ambrožová et al., 2021). Where studies have investigated the impacts on earthworms, they are often focused on lethal end points, using concentrations of anthelmintics much higher than those found in the environment (Lowe and Butt, 2007; Beynon, 2012a; de Souza and Guimarães, 2022). In more recent years, sub-lethal endpoints such as avoidance behaviour and motility, have been recommended as more sensitive endpoints for earthworm ecotoxicology tests, especially when looking at low environmental concentrations (Lowe and Butt, 2005; Lowe and Butt, 2007; Goodenough et al., 2019). Furthermore, many studies have used *E. fetida*, a surface-dwelling epigeic species, which is not naturally found in grasslands, and does not feed on soil (Gao et al., 2007; Torkhani et al., 2011; Gao et al., 2015; Bai and Ogbourne, 2016; Lowe et al., 2016; de Souza and Guimarães, 2022). Endogeic species are an ecologically important species due to their role in soil aggregate formation and the maintenance of the soil pore structure through burrowing behaviour. Investigating the impact of anthelmintics on endogeic species such as *A. chlorotica*, a common grassland species, may therefore have more ecological relevance when assessing soil quality. Indeed, *A chlorotica* was the most common endogeic species found at the field sites surveyed in Chapter 3, with conventionally managed sites having significantly smaller populations of this species.

Given the ecological importance *A. chlorotica*, the earthworm population results of the previous chapter, and the recommendations of using sub-lethal end points to investigate the toxicological effects of anthelmintics, this study aims to look at the avoidance behaviour and change in biomass of *A. chlorotica* when exposed to environmentally realistic concentrations of anthelmintics known to have been used at the field sites in this project: Albendazole (ABZ), levamisole (LEV), ivermectin (IVM) and moxidectin (MOX), using a linear concentration gradient within soil (following Lowe et al., 2016).

4.2 Methodology

4.2.1 Earthworm collection and husbandry

Adult *Allolobophora chlorotica* earthworms (clitellum visible) and sub-adult worms were collected from rough grassland. To avoid unwanted effects from previous drug exposure, earthworms were collected from a field in Tadcaster, UK, that has remained ungrazed and without any manure or slurry applications or other organic amendments for at least 10 years. Soil pits approximately 40 cm x 40 cm wide and

30 cm deep were dug, and earthworms were identified and collected by hand-sorting. After collection, worms were kept in their original soil from the field and housed in an environment room at 15ºC for no more than 7 days prior to the start of each experiment.

4.2.2 Study set-up

A. chlorotica were exposed to each compound separately at linear gradients of six concentrations of each anthelmintic in plastic containers (0.6 × 0.13 × 0.1 m) (n = 4 replicate containers for each treatment). This linear gradient avoidance test was designed following Brami et al. (2017) and Lowe et al. (2016), which were based on ISO 17512-1 (2008). Concentration gradients were created by spiking 1 kg moist soil (per dose) with the required concentration and placing each section sequentially from lowest to highest concentration within the container. The highest concentration was in section F for each treatment, and was derived from the environmentally realistic concentrations previously found in the literature (Paulson and Feil, 1996; Hempel et al., 2006; Iwasa et al., 2008; Römbke et al., 2010; Porto et al., 2021) (Table 4.1). No chemical analyses of the spiked soil were performed to determine the soil concentration of anthelmintics, therefore chemical concentrations are nominal and not actual.

Kettering loam, obtained from Boughton Loam Ltd., UK (Clay 24%, Silt 18%, Sand 58%, Organic content 6.72%, pH 6.8) was chosen as the soil substrate, which is recommended as a standard medium for use in toxicology tests and has been widely used in earthworm research (Lowe and Butt, 2005). Six soil preparations were made by mixing the desired concentration of anthelmintic with 50 mL deionized water, then mixing this into 100 g soil. The soil mixture was then left

Table 4.1: Concentrations of anthelmintics used in this study. Section B = 20%, section C = 40%, section D = 60%, section E = 80%, section F = 100% of the environmentally realistic concentrations derived from the literature.

Anthelmintic	Concentration in each section of the gradient (mg kg^{-1})						Environmentally realistic concentration derived from
	A	B	C	D	E	F	
Albendazole	0.0	2.6	5.2	7.8	10.4	13.0	Porto et al. (2021), residues found in ewe feces
Levamisole	0.0	4.0	8.0	12.0	16.0	20.0	Paulson and Feil (1996), based on 5% excretion of unmetabolised product in urine after 7.5 mg kg^{-1} oral given to a 55 kg ewe
Ivermectin	0.0	0.2	0.4	0.6	0.8	1.0	Römbke et al. (2010), Hempel et al. (2006), residues found in cattle dung
Moxidectin	0.0	0.2	0.4	0.6	0.8	1.0	Iwasa et al. (2008), residues found in cattle dung

for 24 hours at 15ºC to allow sorption of the anthelmintic thereby reducing bioavailability to a more realistic level. Spiked soil was then mixed with the remaining 900 g of soil. Spacers created from laminated card were placed at even intervals into each container, and the spiked soil placed into the sections. Control tubs (*n* = 4 replicates) were set up in the same way, whereby 50 mL deionized water was mixed into 1 kg soil sections placed in each container.

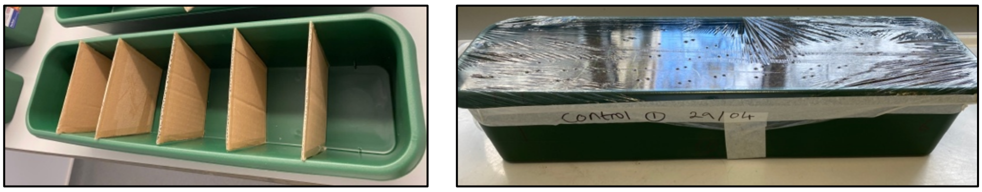

Figure 4.1: Photograph of container before adding soil and earthworms (left) and photograph of one of the control replicates after the dividers had been removed, prior to being placed in the environment chamber. The dividers in the left image are before lamination.

Two earthworms of similar size were placed into each section after having their biomass recorded. Once the earthworms had burrowed into the soil, the dividers were carefully removed, and the container covered in clingfilm to prevent earthworms escaping and excessive moisture loss. The clingfilm was then pierced with a needle to allow ventilation. The four containers were then kept in darkness in a temperature-controlled incubator at 15º C for 7 days. After 7 days, the dividers were inserted into the soil at the same position as at the start of the experiment and the soil removed. The number of earthworms in each section and the live biomass of each worm was then recorded.

4.2.3 Statistical analysis:

Biomass distribution met the assumption of a general linear model. ANOVA general linear models were therefore used followed by post hoc Tukey tests to analyse the differences in biomass at days 0 and 7 under each compound and at each concentration level at day 7. Distribution of worm numbers under all treatments did not meet the assumptions required for ANOVA general linear models and so instead Kruskal Wallis tests were used to analyse differences in earthworm locations along the concentration gradient at day 7 compared to day 0 in each treatment group, to test for differences in earthworm numbers between the sections along the gradient at day 7. All statistical analysis was completed using Minitab 19.

4.3 Results

4.3.1 Distribution of earthworms

At the end of the experiment, all earthworms were recovered with 100% survival for all treatments. There was no significant change in earthworm distribution at day 7 compared to day 0 in the Control experiment, apart from in section C where there

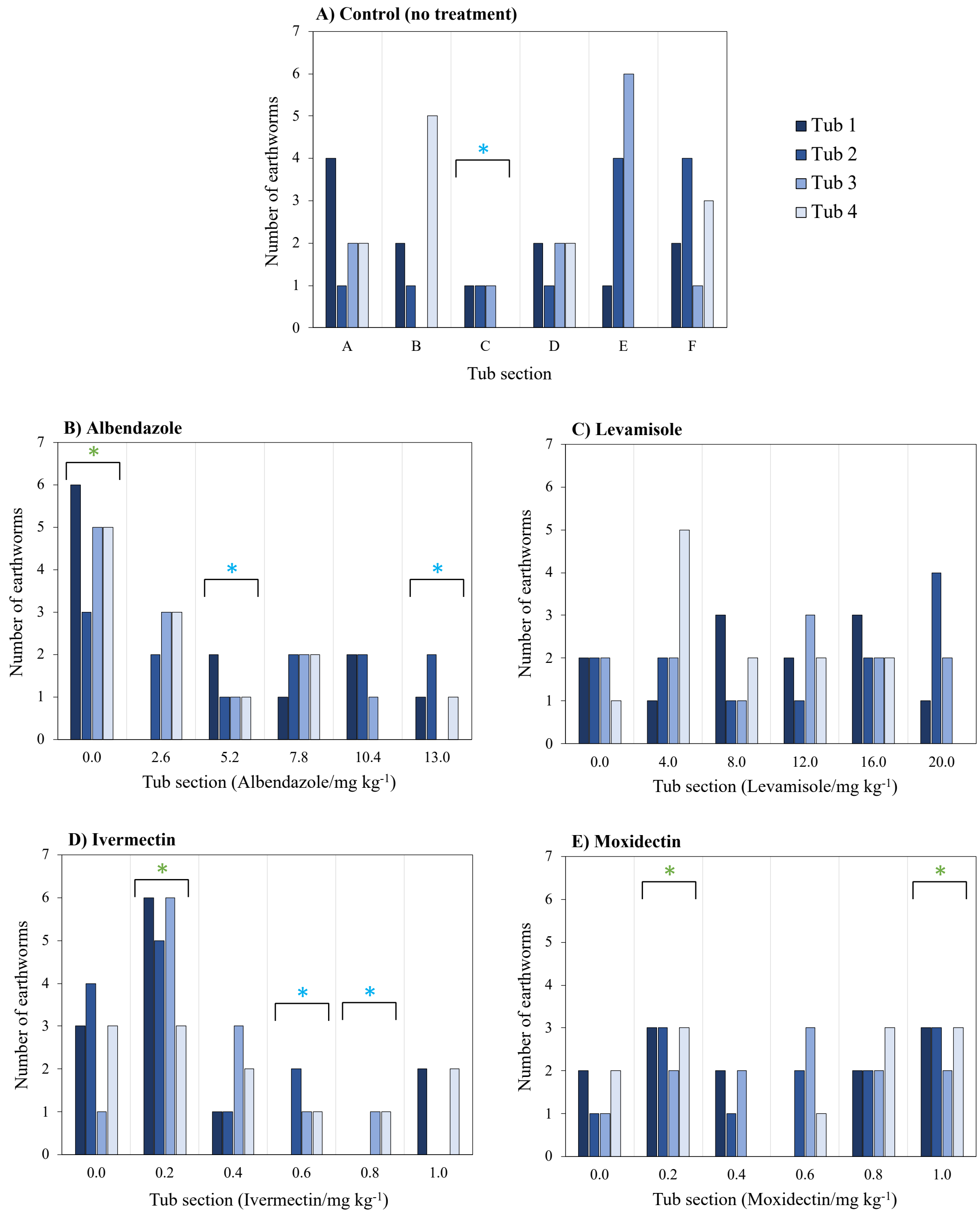

Figure 4.2: Number of earthworms in all tubs (containers) for each treatment at day 7. Asterisks indicate a significant difference in worm numbers in that section at day 7 compared to day 0, green asterisk = increase in numbers, blue asterisk = decrease in numbers ($p < 0.05$).

was a decrease in earthworms across all tubs ($p = 0.011$) (Figure 4.2a). However, there was no significant difference in earthworm numbers between all sections along the gradient at day 7 ($p > 0.05$).

In the Albendazole treatment gradient, Kruskal-Wallis tests showed a significant increase in earthworm numbers at day 7 in the untreated section ($p = 0.013$), and that this differed significantly from all other sections ($p < 0.05$). 42-50% of worms were recovered from the untreated sections in three of the four tubs (Figure 4.1). There was a significant decrease in worms in the section containing the highest concentration (13.0 mg kg^{-1}) (p=0.046) and the second lowest concentration (5.2 mg kg^{-1}) (p=0.040). However, there was no significant difference in earthworm numbers between all the treated sections of the gradient at day 7 ($p > 0.05$).

There was a significant change in earthworm distribution along the Ivermectin treatment gradient at day 7, with 42-50% of earthworms recovered from the section containing 0.2 mg kg^{-1} ($p = 0.013$) (Figure 4.2). Comparisons between all sections at day 7 showed the number of earthworms in sections containing 0.0 and 0.2 mg kg^{-1} to be significantly different to the other sections. Although the numbers of worms in sections containing 0.6 and 0.8 mg kg^{-1} were significantly lower at day 7 (p=0.046 and 0.013 respectively), there were no significant differences between sections at day 7 in sections containing 0.4 mg kg^{-1} or above ($p > 0.05$).

Under the Moxidectin treatment gradient there was a statistically significant increase in earthworm numbers at 0.2 and 1.0 mg kg^{-1} ($p = 0.040$). However, the increase was relatively small, changing from two earthworms to three in three out of four tubs, and there was no significant decrease in any other section along the gradient. There was no significant change in earthworm distribution at day 7 across the Levamisole treatment gradient, with the number of earthworms along the gradient varying across the different tubs. For example, at 4.0 mg kg^{-1} the number of earthworms ranged from 1-5, and at 20.0 mg kg^{-1} the number of earthworms ranged from 1-4 (Figure 4.2c).

4.3.2 Biomass

There was a significant reduction in mean biomass at day 7 for earthworms exposed to Albendazole when compared to the control (mean loss of 26 %, $p = 0.001$) (Figure 4.3 & Figure 4.4). Figure 4.3 is presented to show in which section the change in biomass occurred. There was no significant difference in mean earthworm biomass between sections along the concentration gradients at day 7. No other treatment showed any significant changes in biomass.

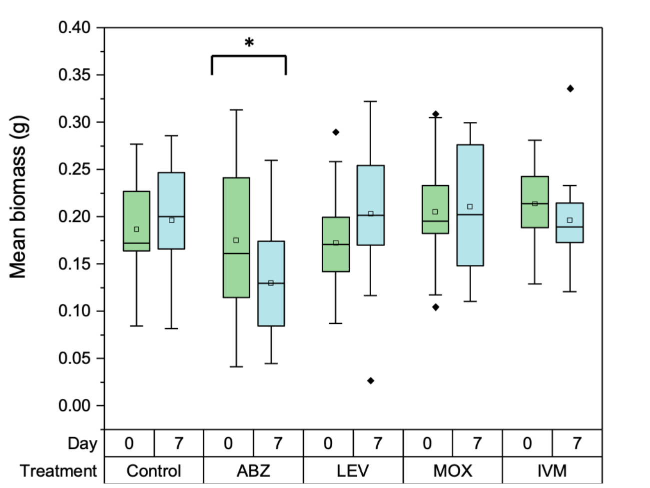

Figure 4.3. Mean biomass of earthworms at day 0 and day 7 for each treatment. Asterisk indicates a significant difference in mean biomass for that treatment at day 7 compared to day 0 ($p = < 0.05$). ABZ = albendazole, LEV = levamisole, IVM = ivermectin, MOX = moxidectin. Horizontal lines within boxes indicate the median, box ends indicate the 25th and 75th percentile, square markers indicate the mean, and whiskers show the minimum and maximum values (excluding outliers, indicated by black dots, which are more than 1.5 interquartile ranges).

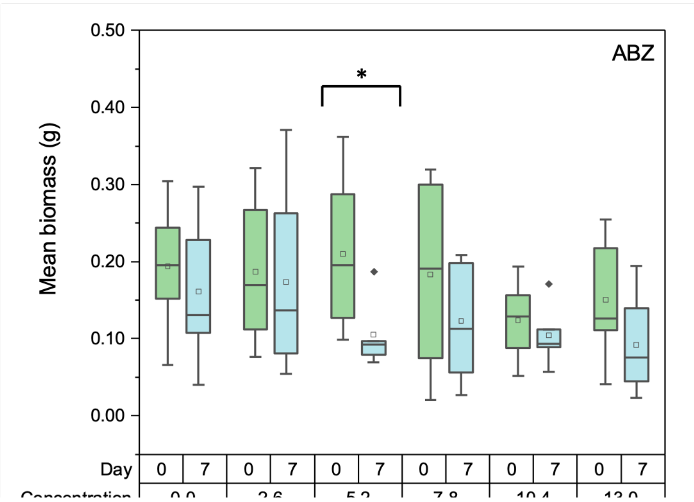

Figure 4.4. Mean biomass of earthworms at day 0 and day 7 for each section of the concentration gradient of albendazole. Asterisk indicates a significant difference in mean biomass for that treatment at day 7 compared to day 0 ($p = < 0.05$). ABZ = albendazole. Horizontal lines within boxes indicate the median, box ends indicate the 25th and 75th percentile, square markers indicate the mean, and whiskers show the minimum and maximum values (excluding outliers, indicated by black dots, which are more than 1.5 interquartile ranges).

4.4 Discussion

4.4.1 Albendazole and ivermectin

Avoidance behaviour was observed in *A. chlorotica* after 7 days of exposure to environmentally realistic concentrations of Albendazole (ABZ) and Ivermectin (IVM), with a significantly higher number of earthworms recovered from the untreated section of both experiments (Figure 4.2). These findings suggests that *A. chlorotica* may be able to detect the presence of, and actively avoid, these compounds at the realistic concentrations applied to the soil environment.

Although, to date, no literature has been found on the avoidance behaviour of *A. chlorotica* under exposure to the contaminants I used, the results in this study do support findings of the few papers which investigate other similar sub-lethal end points in earthworm species under exposure to anthelmintics. Rates of faecal disappearance over 28 days has been observed to be significantly lower when sheep have been treated with ABZ and IVM bolus, which was partially attributed to decreased earthworm activity (Yeates et al., 2007). During a field experiment, Svendsen et al. (2003) found *A. chlorotica* to be disinterested in dung treated with IVM bolus and fenbendazole bolus (a BZ compound, similar to ABZ). More recently, Goodenough et al. (2019) found that that motility (defined as the time taken for an individual earthworm to burrow between two points) of *Lumbricus terrestris* was 28% longer after 12 weeks exposure to soil spiked with < 2.5 mg kg^{-1} IVM. Contrastingly, Torkhani et al. (2011) found no avoidance response of *L. terrestris* and *E. fetida* species when exposed to IVM, and interestingly, found more *E. fetida* worms in the sections containing higher concentrations (64 and 256 mg kg^{-1} dry soil), suggesting that this species may actually be attracted to the compound. However, *E. fetida* is an epigeic species which does not feed on the soil. It is possible that the geophagous endogeic species, such as *A chlorotica*, are exposed to more of the contaminant, which could explain the effect on avoidance behaviour observed in this study.

A reduction in biomass was found in *A chlorotica* after 7 days of exposure to ABZ. Although biomass change of earthworms under exposure to ABZ has not been previously investigated, research has shown that low BZ concentrations in soil can cause mortality in *E. fetida* after exposure to ABZ (Gao et al., 2007), and *L.*

terrestris after exposure to fenbendazole (a similar compound to ABZ) at low concentrations of <1.4 mg kg^{-1} (Goodenough et al., 2019), with mortality being attributed to mitochondrial disruption. In the Goodenough et al. (2019) study, earthworms were exposed fenbendazole for 12 weeks, which is a much longer exposure time than the 7 days used here. It may be that the damage to the mitochondria is one of the mechanisms behind the reduction in biomass found in my study, and that if it were to have been run over a longer period, we might have started to see toxic effects and an increase in mortality in earthworms exposed to ABZ. However, caution must be taken when interpreting the biomass results, as it is also difficult to determine whether the change in biomass was due to a similar change in biomass in all worms, or if the effect is due to a more dramatic change in 1 or 2 individual worms, as individual worms were not labelled or tracked. Other environmental factors can cause changes in earthworm biomass, especially moisture content, which could have impacted the results in this case as live biomass was assessed.

4.4.2 Levamisole and moxidectin

Avoidance behaviour was not observed under environmentally realistic concentrations of either MOX or LEV. Moxidectin (MOX) is part of the ML group of anthelmintics, where the primary mode of action is inducing paralysis in the parasite by binding to gamma-aminobutyric (GABA) and glutamate-gated chloride channels (Cobb and Boeckh, 2009). MOX is a relatively new ‘second-generation’ ML, which is less susceptible to elimination from parasite cells (Cobb and Boeckh, 2009). To date, little is known about its interactions in the environment, nor its impacts on invertebrates (Hempel et al., 2006; Blanckenhorn et al., 2013; Manning et al., 2018). There has only been one study, to the author’s knowledge, which examined the impact of MOX on earthworms (Svendsen and Baker, 2002), and in that study there was no effect on the survival or growth of *Aporrectodea longa* (Ude) (Lumbricidae) over a 10 week period, when fed sheep and cattle dung after MOX treatment. There is a dearth of literature on avoidance behaviour and other sub-lethal effects of these two compounds on earthworms, so it is not known why MOX and LEV produced no avoidance or biomass effect in *A. chlorotica* in my study. It may be that the environmentally realistic concentrations used were below the threshold of any adverse effect, or that earthworms are unable to detect these particular compounds with their chemoreceptors (Sousa et al., 2008).

4.4.3 Implications for soil function & recommendations for future studies

This study supports other findings that anthelmintics such as ABZ and IVM could be negatively impacting the behaviour of ecologically important earthworm species (Yeates et al., 2007; Goodenough et al., 2019). Here, avoidance behaviour has been observed in *A. chlorotica* when exposed to soil containing residues of these compounds. If this avoidance behaviour is representative of field conditions, then it is possible that *A. chlorotica* and other ecologically important endogeic species are avoiding areas of the soil where these compounds have been deposited through urine and faeces (Yeates et al., 2007). It would be beneficial to run this experiment over a longer time period: These compounds have been reported to have high DT_{50} values (Table 1.4; Krogh et al., 2009), so it would be useful to see if the behaviour or tolerance of earthworms to these compounds changes with time. It could be particularly important to repeat these studies under colder more acidic conditions, as it has been shown that these particular conditions can dramatically increase the DT_{50} of macrocyclic lactones. *A. chlorotica* has been found to increase the percentage of water stable aggregates and increase the water holding capacity of soil (Hallam et al., 2020). Changes in the burrowing behaviour of endogeic species could therefore compromise their role in the maintenance of soil aggregates and the soil pore system, and consequently compromise the hydrological functioning of the soil. Therefore, it would be beneficial for future studies to investigate other sub-lethal effects of anthelmintics on endogeic species, such as burrowing behaviour and motility, and to investigate if any observed effects impact the soil pore system or hydrological functioning, so that we can better understand how different management systems impact soil biological function.

4.5 Conclusions

In this chapter, the endogeic earthworm *A. chlorotica* been shown to be sensitive to the anthelmintics ABZ and IVM, exhibiting avoidance behaviour after 7 days exposure to environmentally realistic concentrations of the compounds, with a reduction in mean biomass also observed in the ABZ experiment. No effects were observed in either MOX or LEV experiments. Further studies on the sub-lethal effects of anthelmintics are recommended due to the ecological importance of this earthworm functional group in the maintenance of the soil system.

Chapter 5: General discussion and conclusions

5.1 Section outline

In addition to the discussion and conclusion sections in Chapters 2-4, this section aims to summarise the key findings of the project as a whole, in relation to the overarching research questions:

1. How do soil physical and chemical properties differ under organic upland grazing management compared to conventional management?
2. Do earthworm populations differ under organic upland livestock grazing compared to conventional upland livestock grazing?
3. Do anthelmintic chemicals used in conventional livestock farming impact the avoidance behaviour of earthworms?

The project findings are presented by splitting the three research questions into two categories and discussing the implications for their respective soil functions within these two sections: i) impact of organic grazing on soil physical and hydrological properties, and ii) impact of organic grazing on earthworm populations. This is then followed by two sections which cover the wider implications and limitations of the project's findings, and recommendations for future research.

5.2 Impact of organic grazing on soil physical and hydrological properties

5.2.1 Soil organic matter

The results of this project showed differences in soil physio-hydrological properties under organic grazing systems in upland areas, compared to conventional management systems. However, these differences were likely caused by differences in soil types, with conventional sites at FoB and YD both having a peaty organic horizon of around 20 cm depth. Organic sites contained less organic matter (OM) than conventional sites (Chapter 2), particularly in the top 0-5 cm layer (means of 14% under organic and 39% under conventional), again, likely due to the differences in the depth of the O horizon. Soil OM content was found to negatively correlate well with bulk density at all sites, with sites under organic management having higher bulk density than conventional sites (means of 1.06 g cm^{-3} for organic and

0.85 g cm^{-3} for conventional), again with the biggest difference being observed in the top 0-5 cm layer (means of 0.89 g cm^{-3} under organic compared to 0.63 under conventional). A difference in the vertical distribution of soil physio-hydrological properties between the two management systems was particularly evident (Chapter 2). However, when looking at the horizon depth of the conventional sites compared to organic sites, this difference is to be expected given the much deeper O horizon at conventional sites, due to their soil type (Wilcocks 1 (721c), (Cranfield University, 2023a-f)), whereas organic sites had an O horizon around 5 cm deep. Carbon stocks (Chapter 2) were overall significantly higher under conventional management compared to organic sites (means of 4.14 kg m^{-3} for conventional and 3.12 kg m^{-3}) for organic), however the range of values varied much more widely under organic sites. Carbon balance estimates (Chapter 2), showed that all systems were carbon sinks, sequestering relatively similar amounts of C, ranging 140-179 g/C/m^2/yr across all sites except for FoB-C, which has a gain of 440 g/C/m^2/yr, due to the addition of an additional 270 g/C/m^2/yr through manure applications. The small variations in net C gains across the sites was due predominantly to the differences in their stocking densities, which impacted the consumption of C and removal through animal products, and respiration & dung decomposition outputs. However, the differences in stocking rates were relatively small. The similarity of the net gain of C across all sites suggest that SOC stocks in the conventional systems, although larger, are unlikely to be increasing at a faster rate than organic sites. However, the species composition and productivity of the vegetation community can determine the rate of SOC accumulation (Waters et al., 2017b). Therefore, in future studies it would be useful to conduct vegetation surveys to further our understanding of the carbon balance under these management systems.

Organic sites were found to have more endogeic earthworm species (see section 5.3), particularly *A. chlorotica* (Chapter 3), which could perhaps be an important mechanism behind the more even vertical distribution of OM at organic sites (although this is more likely to be a function of soil type). Endogeic earthworm species typically inhabit depths of 5-15 cm of the soil profile, and can promote good soil mixing through their near-continual burrowing, consumption of soil and OM, and the creation of stable aggregates (Hallam and Hodson, 2020). Earthworms play a key role in the breakdown and transportation of organic matter in upland ecosystems. Other soil fauna also known to also play a key role in the breakdown and transportation of organic matter are dung beetles and nematodes (Bardgett et al., 1999). There are complex interspecies relationships between these soil fauna species, therefore it is important to consider that changes in earthworm populations might also have a knock-on effect on other species.

OM and BD findings for both management types were within the ranges expected for good soil functioning. As discussed in Chapter 2, convenrional grazing in upland areas has relatively low stocking rates, and as such the surface pressure from grazing animals may not be high enough to cause significant degradation of the soil surface under either management system (Ward et al., 2016b). In turn, the organic matter may protect the soil surface from hoof action and the breakdown of soil structure.

5.2.2 Soil hydrological function

Chapter 2 showed that organic sites had higher infiltration rates, which changed less with depth than conventional sites (medians of 65 mm hr^{-1} for organic and 33 mm hr^{-1} for conventional). Again, there was a significant difference in the vertical distribution of properties, with conventional sites exhibiting a steady decrease in saturated hydraulic conductivity (*Ks*) with depth. Conventional sites also had higher moisture content at 0-10 cm depth, possibly due to the higher OM matter content, which is known to increase water holding capacity. Interestingly, conventional sites had lower volumetric water content than organic sites (Chapter 2). This could be explained by the higher proportion of macropore space in their deeper peaty O horizons, allowing for more open pore space. The irregular, fibrous structure of peat tends to also have poor connectivity of the smaller pore spaces, therefore resulting in higher moisture content but lower volumetric water content than organic sites.

In contrast, the *Ks* under organic grazing changed little with depth, but the range of values varied by several orders of magnitude. The increased activity from endogeic earthworms under organic sites (Chapter 3) presents a possible mechanism behind the difference in vertical distribution of *Ks* at organic sites. This difference in vertical distribution of *Ks* rates between the two management systems is a particularly important finding in terms of hydrological functioning in upland areas, as saturation-excess overland flow (OLF) tends to dominate organic soils (Joseph Holden et al., 2007; Marshall et al., 2009; Bond et al., 2021). The findings in this study therefore indicate that organo-mineral soils under organic grazing may be less susceptible to saturation-excess OLF than conventionally grazed grassland. Tension infiltrometer measurements presented in Chapter 2 showed proportions of flow through pore size classes under both management systems, with macropore flow dominating both management systems, and conventional sites having slightly higher proportion of flow through the largest macropores. These results are important as little is known about the hydrological functioning of organo-mineral soils. This study presents further evidence that high *Ks* in the upper organic layers is characteristic of this soil type (Bond et al., 2021; Monger et al., 2022), and that macropore systems are functionally important for upland soils (Carey et al., 2007; Holden, 2009; Rezanezhad et al., 2016).

5.3 Impact of organic grazing on earthworm populations

This study is one of the one of the first to look at soil earthworm communities in organo-mineral soils, and found that the endogeic species *A. chlorotica,* was particularly abundant in this soil type (Chapter 3). This study was also the first to look at the avoidance behaviour of *A. chlorotica* under environmentally realistic concentrations of four commonly used anthelmintics (Chapter 4). Chapter 3 showed that the abundance of the earthworm *A. chlorotica* was much higher under organic grazing management, accounting for 33% of the total earthworm population, compared to 16% under conventional management. FoB-O had the highest *A. chlorotica* abundance (33%) and the biggest difference compared to its organic partner site FoB-C (8%). Abundance at YD was also significantly different between management types (YD-C=15%, YD-O=39%), whereas NYM showed no significant difference between sites (NYM-C=19%, NYM-O=21%). *A. chlorotica* is an important endogeic species, commonly found in grassland, so high abundance of this earthworm was generally expected in both management systems. It is rather a surprising outcome that the abundance of this one particular earthworm species was impacted, rather than overall earthworm density. Possible mechanisms behind this difference in abundance could be due to lower pH at two of the conventional sites (Chapter 2). Although *A. chlorotica* survives well in a wide range of pH types, it does have a preference for more neutral soils (>pH 6), and has been shown to have a strong relationship with soil pH (McCallum et al., 2016). In Chapter 4 the relationship between soil pH (taken from pH results in Chapter 2) at field sites and earthworm density was examined, finding a positive correlation between soil pH and earthworm density. Lower grazing densities (Table 1.5) may also result in more spatial variation in surface disturbances, creating more undisturbed areas which *A. chlorotica* may occupy in higher abundance. In addition to this, differences in treatment regimens of anthelmintics between the two livestock management systems (Table 1.1) may be of significance: residues of anthelmintics have been found in dung deposits from treated livestock (Iwasa et al., 2008; Römbke et al., 2010; Porto et al., 2021), which has been shown to negatively impact a variety of dung fauna (Hempel et al., 2006; Iwasa et al., 2008; Römbke et al., 2010; Blanckenhorn et al., 2013; Adler et al., 2016; Ambrožová et al., 2021). Few other studies have looked at the impact of these compounds on endogeic earthworm species, with the majority of earthworm studies investigating epigeic species, and often focusing on lethal end points, using concentrations of anthelmintics much higher than those found in the environment (Lowe and Butt, 2007; de Souza and Guimarães, 2022). Chapter 4 of this study expanded on this information, by investigating the avoidance behaviour of *A. chlorotica* when exposed to four anthelmintic compounds. Avoidance behaviour was then observed in *A. chlorotica* under exposure to albendazole and ivermectin, supporting previous findings that these compounds might have sub-lethal effects on earthworms at environmentally realistic concentrations (Goodenough et al., 2019). The DT_{50} of these compounds have been reported to range widely depending on soil type (Krogh et al., 2009), with evidence showing that when under anaerobic conditions, DT_{50} increased significantly, with the extractable amount remaining unchanged between 14-120 days. Temperature has also been reported as a key driver of the DT_{50} of anthelmintics, and that lower temperatures significantly increased DT_{50} values (Krogh et al., 2009). This is important for upland soils, which are prone to waterlogging and cold temperatures. Furthermore, the high KOC values of anthelmintics outlined in Table 1.4, indicate a strong adherence to soil particles. There is a possibility that transport of these compounds could occur through the movement of POM, however more research is necessary to understand if this is a significant transport mechanism for ivermectin and albendazole. Avoidance behaviour in *A. chlorotica* was not observed under levamisole and moxidectin treatments. There is not much literature on the sub-lethal effects of these two compounds. Consequently, the reasons behind the absence of avoidance or biomass effects in *A. chlorotica* under MOX and LEV treatments in this study remain unknown. It may be that the environmentally realistic concentrations used were below the threshold of any adverse effect, or that earthworms are unable to detect these particular compounds with their chemoreceptors (Sousa et al., 2008).

The work in Chapters 3 and 4, therefore, provide evidence that organic grazing management in upland areas may be more beneficial for soil fauna, particularly *A. chlorotica*, with Chapter 4 indicating that anthelmintics may be a contributing factor towards differences observed in earthworm populations in Chapter 3. However, the findings in Chapter 3 are in contrast to Schon et al. (2011), who found no significant difference in earthworm populations between organic and conventional grazing in New Zealand upland grassland management systems when comparing identical stocking rates. They concluded that organic grazing, through its limited use of antiparaciticides, was no more beneficial to soil invertebrates than conventional grazing management. A key difference between Schon et al. (2011) and this study, is that stocking rates here were not identical. Lower grazing densities in organic livestock farming is a fundamental part of the management practice (Soil Association, 2021; Schon et al., 2011) mention that stocking rates may, therefore, be of more importance when assessing the differences between organic and conventional grazing, through altering the soil physical environment and removal of above-ground biomass. However, although stocking rates in this study were not identical, they were only marginally different (Table 1.6). The key difference between the sites in this study, is that the conventional sites at FoB and YD had very different soil types to that of their organic partner site. The soil pH at these conventionally grazed sites was significantly lower than organic sites (Chapter 2), due to the much deeper and more acidic O horizon (20 cm). Chapter 3 showed a positive correlation between soil pH and earthworm density across all sites, indicating that soil pH is likely the key driver between differences in earthworm density in this study, and not the management system, or grazing density. Thus, the differences in earthworm abundance in this study may not be due the sub-lethal effects of anthelmintics observed in Chapter 4. However, earthworm abundance can increase with plant productivity (Cole et al., 2006), and changes in the plant community can also lead to changes in the earthworm community through changes in nutrient inputs from leaf litter (Cooke and Luxton, 1980). It could be that there are differences in vegetation removal in lower grazing densities at organic sites which may result in changes to the plant communities of the sites, and thus vegetation controls warrant further investigation.

5.4 Wider implications

Overall, this thesis has investigated the impacts of organic grazing on upland soil functions, and has found no effects which suggest organic livestock management may be helpful towards directly maintaining soil hydrological function, carbon cycling and storage, or nutrient cycling, in upland areas. However, results in Chapter 4 suggested that anthelmintic treatments used in livestock farming might have sub-lethal effects on earthworm populations. If this behaviour reflects environmentally realistic behaviours, then organic sites may be beneficial for protecting soil fauna. There are direct and indirect benefits to soil health through maintaining good soil biological function, such as maintaining the soil pore space and therefore hydrological function, decomposition, nutrient cycling and securing organic carbon. Good maintenance of these soil functions is key to achieving a number of the UN's Sustainable Development Goals (Keesstra et al., 2016), particularly the sustainable management of water through flood mitigation (SDG2); combating climate change though protecting carbon stores (SDG13); and protecting, restoring, and promoting the sustainable use of terrestrial ecosystems (SDG15). However, carbon stocks (Chapter 2) under conventionally managed grasslands in this study were slightly higher, with FoB-C site being the largest carbon sink, likely due to the annual application of farmyard manure. This shows that conventional grazing, especially where manure is being applied, may be better at sequestering carbon than organic management, and therefore be beneficial for meeting climate change mitigation through protecting carbon stores (SDG13).

The findings in Chapters 3 and 4 indicate that earthworms may be negatively impacted by anthelmintic residues, and that organic livestock management, or at least a more 'treatment based' or 'reactionary' approach to the administration of anthelmintics, may be more beneficial to earthworm communities. The selected anthelmintic compounds and the amounts used in this experiment were based on residues of commonly used anthelmintics found in dung from grazed livestock animals. These anthelmintics and concentrations thereof are not unique to upland grazing management and are used globally at all levels of grazing densities on all pasture types. The findings in Chapter 4 add to the growing body of evidence that anthelmintic residues in the environment may be negatively impacting soil biological functionality (Hempel et al., 2006; Blanckenhorn et al., 2013; Adler et al., 2016; Goodenough et al., 2019; Ambrožová et al., 2021; Lagos et al., 2022), which could in turn be damaging soil biological functioning. Soil biological function is crucial not only in maintaining the functionality of the soil system as a whole, but also in maintaining connectivity between ecosystems, by maintaining the flow of resources between them (Bardgett, 2002). Thus, by damaging soil faunal communities, the wider ecosystem could be at risk due to changes in food availability for some species. Upland areas are important habitats for wading birds, particularly in the UK, which has seen a decline in species numbers due to habitat loss and unfavourable land management. Thus, it may be worth considering a more treatment-based approach (rather than preventative) use of anthelmintics in livestock, as a conservation strategy in upland areas.

5.5 Limitations of the project

This study compared organic grazing management only to conventional grazing management, and not to other sustainable grazing management systems. In the UK, sustainable grazing practices are encouraged through the Sustainable Farming Incentive (SFI), and soon to be through the new Environmental Land Management schemes (ELMs) (GOV.UK, 2022). Many of the grant payments available through ELMs that are applicable to upland grazing management include subsidies for many similar practices to those of organic grazing management systems, such as encouraging low stocking rates and grazing exclusion (see grant numbers GS17 and UP6) (GOV.UK, 2022). Therefore, these other sustainable management practices may be just as beneficial for maintaining good soil function, and future studies may consider comparing these to organic grazing systems to further understand the impact of these different management practices. This is particularly important in developing our understanding of the impacts of antiparasitic veterinary medicines on soil biological function under grazing management: although the are many similarities between Sustainable Farming Incentive (SFI) schemes and organic management practices, a key difference is the use of anthelmintic veterinary medicines, due to tightly controlled use of these medicines in organic management standards (Soil Association, 2021).

A key limitation to this study is the site selection and pairing. When choosing field sites, topography was not considered. Slope, altitude, and aspect strongly influence soil development, soil moisture, and decomposition rates. Slower slope gradients facilitate the accumulation of organic matter under higher moisture contents, which can be exacerbated depending on the aspect, which can alter evapotranspiration rates. In this study, conventional sites at FoB and YD had very different topography to their partner sites, and were both in much wetter parts of the catchment, or in the case of YD, not even in the same catchment as the organic site. This also resulted in comparing very different horizon depths at FoB and YD, with both conventional sites having a peaty O horizon of ~20 cm. Future studies should therefore be more careful during the site selection process, to ensure as close a match as possible in terms of topographical setting, both locally and at the catchment scale. Furthermore, this study looked at a very limited number of field sites, only looking at one field per farm, and a limited number of pits were dug in each field (8), to ensure the fieldwork was feasible within the project timeframe. The fieldwork was also limited in that only upland sites in the north of England were used. This may therefore present a narrow view of soil properties under organic grazing management, and a limited view of the functional properties of organo-mineral soils. Larger field studies are therefore recommended to expand on this dataset, particularly on different organo-mineral soil types and different climate zones.

In Chapter 4, the literature was used to derive the environmentally realistic concentrations of anthelmintics used in the avoidance experiment. Caution must be used, therefore, when assuming that these concentrations would be found in upland soil under conventionally grazed livestock. Further sampling and analysis of upland soils for anthelmintic residues is therefore recommended. Similarly, in Chapter 2, calculations for carbon balance were derived from the literature and known characteristics of the field sites. A more detailed analysis of the carbon balance perhaps using in field measurements of respiration rates and trace gas exchange (CH_4, CO_2, N_2O) would also be beneficial.

A further limitation of this project includes the absence of vegetation analysis. Although root biomass was considered in Chapter 2, above ground biomass and vegetation composition was not recorded. Grassland vegetation compositions can have important influences on soil macroporosity and hydrological function, soil organic matter content through leaf litter decomposition, and changes in soil fauna composition (Langlands and Bennett, 1973; Leonard and Andrieux, 1998; Reeder et al., 2004; Cole et al., 2006; van den Berg et al., 2012). It is therefore possible that some of observations in this project were functions of differences in vegetation composition between the two management systems.

5.6 Recommendations for future work

- It is crucial any future field studies consider the topography of the field site and catchment area to ensure better comparisons.
- Larger field studies are recommended to expand the data on infiltration, organic matter and bulk density in organo-mineral soils.
- A more detailed analysis of the carbon balance of each system using in field measurements of respiration rates and trace gas exchange.
- It is recommended that further studies explore the impact of organic grazing on upland vegetation composition and productivity, due to the important interactions between vegetation and soil functioning.
- Further research on the impacts of anthelmintics on other sub-lethal effects in soil fauna.
- Analysis of soils samples under livestock grazing for anthelmintic residues.
- Investigate the environmental fate of anthelmintics, particularly the transfer from dung and urine to the soil.
- Method development for extracting different anthelmintic compounds from organo-mineral soils.
- Analysis of anthelmintic metabolites in soil.
- Examine the sub lethal effects of anthelmintic metabolites on soil fauna.
- Examine soil mesofauna populations under organic farming, particularly springtail and soil mite communities.
- In field measurements of soil infiltration rates under organic grazing management on upland organo-mineral soil.
- It is also recommended that future studies consider comparing organic management to a wider range of grazing management systems so that we may broaden our understanding of the impact of organic management, relative to other sustainable management systems.

5.7 Conclusions

This thesis has shown that the effects of organic grazing systems on soil functions in upland areas are likely to be similar to conventional grazing, and that although significant differences were found in the physio-hydrological properties between the two systems, that these are most likely a function of different soil types, rather than management systems. Carbon balance estimates suggest that both management systems were classed as carbon sinks, with conventional farms sequestering slightly more carbon per year, especially with regular application of manures. Although a higher proportion of endogeic earthworms were found under organic grazing, earthworm populations across all sites were positively correlated with soil pH, and thus most likely be due to differences in soil type between sites. The endogeic earthworm species *A. chlorotica* was found to exhibit avoidance behaviour under exposure to the anthelmintic compounds ivermectin and albendazole, indicating that the regular use of anthelmintics in livestock management may be negatively impacting earthworm behaviour.

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