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Significant structural development of a long-term fallow soil in response to agricultural management practices requires at least 10 years after conversion

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Significant structural development of a long-term fallow soil in response to agricultural management practices requires at least 10 years after conversion

Running title: Structural development of long-term fallow soils

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Abstract (250 words, currently 232)

Agricultural practices can have significant effects on the physical and biological properties of soil. The aim of this study was to understand how the physical structure of a compromised soil, arising from long-term bare-fallow management, was modified by adopting different field management practices. We hypothesised that changing agricultural practice from bare-fallow to arable or grassland would influence the modification of pore structure *via* an increase in porosity, pore connectivity, and a more homogenous distribution of pore sizes; and that this change exerts a rapid development of soil structure following conversion. Soil aggregates (< 2 mm) collected in successive years from field plots subjected to three contrasting managements were studied; *viz.* bare-fallow, bare-fallow converted to arable, and bare-fallow converted to grassland. Soil structure was assessed by X-ray Computed Tomography on the aggregates at 1.5 μm resolution, capturing detail relevant to soil biophysical processes. The grassland system increased porosity, diversity of pore sizes, pore-connectivity and pore-surface density significantly over the decade following conversion. However, measured at this resolution, the development of most of these metrics of soil structure required approximately 10 years post-conversion to show a significant effect. The arable system did not influence soil structural development significantly. Only the pore size distribution was modified in grassland in a shorter time frame (2 years post-conversion). Hence development of the soil structural characteristics appears to require at least a decadal timescale following conversion to grassland.

Key words:

Soil structure, 3D pore characteristics, agricultural management practices, X-ray Computed Tomography, porosity

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Highlights:

- The physical structure of a compromised soil was modified by adopting plant-based field management practices.
- Conversion to grassland increased pore size diversity after 2 years.
- Porosity, pore connectivity and pore surface density showed a significant modification between 7 to 10 years after conversion.
- Bare fallow soil management for this extreme period (> 50 years) is detrimental to physical soil properties and the regeneration of the soil structure requires more than 10 years after being reconverted to arable and grassland.

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Introduction

Agricultural practices can have beneficial or detrimental effects on soil functions when applied for decades, depending on the nature of such practices (Ashworth et al., 2017; Bronick and Lal, 2005; Denef et al., 2009; Pagliai et al., 2004). Agricultural management generally aims to increase - or at least stabilise - crop yield, but intensive farming can lead to soil degradation, erosion, compaction and pollution (Bronick and Lal, 2005). Conventional tillage can lead to a decline in soil aggregation and soil structure (Watts et al., 2001), as well as depletion of nutrients and organic carbon within soil (Coleman et al., 1997). Addition of organic matter or crop rotations can prevent soil disruption from tillage by improving soil porosity and aggregation (Abdollahi et al., 2014; Pagliai et al., 2004). In some cases, modification of crop management can have beneficial impacts on soil functions. For example, after 50 years of continuous cultivation, a desert aeolian sandy soil was managed into a sustainable agricultural soil by increasing silt and clay content (a determinant for aggregate formation), soil organic matter and nutrient retention (Su et al., 2010). Moreover, soil aggregate stability is a key factor for soil fertility and physical resilience from external forces, e.g. wind and water (Abivent et al. 2009).

Soil structure plays a fundamental role in the distribution of carbon, soil microorganisms, water and nutrient accessibility (Rabot et al. 2018; Smith et al. 2017). Analysis of soil structure indicates that pore size distribution, assessed by X-ray Computed Tomography (CT), plays an important role in aggregate stability (Menon et al. 2020). Increased diversity of pore sizes (i.e. a more homogenous distribution of pores) is associated with a more complex pore network. This leads to an increase in the number of storage and transmission pores resulting in greater water and nutrient flux (Kravchenko et al. 2014; 2019). The modification of pore size distributions appears to also play a key role in the decomposition of organic matter (Quigley et al. 2018; Smith et al. 2017; Toosi et al. 2017a; 2017b). For example, the presence of small (13-32 µm) and large (136-260 µm) pores decreases organic matter decomposition within macro-aggregates (Toosi et al. 2017b). Pore connectivity is also one of the most important factors, alongside porosity and pore size distribution, to understand soil functions (Rabot et al. 2018). Modification of pore connectivity can have a significant effect upon the distribution and the transport of gas and water (Lucas et al. 2020; Müller et al. 2019; Pires et al. 2019). Pires et al. (2017) demonstrated that pore connectivity was enhanced in zero tillage systems over conventional tillage systems, especially in the upper 10 cm.

In the field, long-term management practices can have substantial impacts on soil structural dynamics (Bacq-Labreuil et al. 2018; Müller et al. 2019; Pires et al. 2019). For example, 50-years of management of a typical silty clay loam soil as bare fallow resulted in significant reductions of carbon and nitrogen, and in the abundance of biological communities (Hirsch et al. 2009) with soil structure also severely compromised (Bacq-Labreuil et al. 2018).

Conversion from bare fallow to arable or grassland increased soil organic carbon, soil

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nitrogen and the population of meso-fauna and fungi within 3 to 5 years following conversion (Hirsch et al. 2017), although, soil structure modification was not assessed in this experiment. The aim of this study was to establish how the micro-structure of a compromised silt-clay loam soil is modified by altered field management over time using soil aggregates (< 2 mm diameter). Three treatments were studied from the converted field of the long-term experiment: continuous bare-fallow, bare-fallow converted to arable, and bare-fallow converted to grassland. We hypothesised that: (1) plants are an active factor in increasing soil porosity, diversity of pore sizes and pore connectivity, and (2) structural development is more rapid in grassland than arable converted systems due to the greater and more persistent presence of vegetation. The precise time for soil structural development is unclear *a priori*, and we aimed to determine this by measuring structural properties on several successive years after conversion.

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Materials and Methods

Soil aggregate sampling

Samples were obtained from conversion plots of the long-term Highfield Ley-Arable experiment at Rothamsted Research, Harpenden, UK (LATLONG 51.8103 N, -0.3748 E). The soil is a silty-clay loam (clay: 27%, silt: 58,4, sand: 14.6%; Jensen et al. 2020b) developed on clay-with-flints over Eocene London Clay (Batcombe series), classified as Chromic Luvisol by FAO criteria (Avery and Catt, 1995; FAO, 2006; Watt and Dexter, 1997). In October 2008, plots of soil managed as bare-fallow by regular tillage to remove any plants since 1952, were converted to arable and grassland managements. The conversion plots had similar soil characteristics. The conversion of the plots from bare-fallow to arable and grassland is explained in details in Hirsh et al. (2017). Arable soil was placed under continuous wheat rotation (winter wheat, *Triticum aestivum* L., *c.v.* ‘Hereward’ seed coated with Redigo® Deter® combination insecticide/fungicide treatment, Bayer CropScience) receiving ammonium nitrate fertilization to provide approximately 220 kg-N ha^{-1} y^{-1}. For the arable and grassland plots additional fertilizers 250 kg-K ha^{-1} and 65 kg-P ha^{-1} was added every three years. Grassland plots were maintained as a managed sward of mixed fescue (*Festuca pratensis* L.), Timothy grass (*Phleum pratense* L.) and white clover (*Trifolium repens* L.) (30 kg ha^{-1}). To remove weeds, bare-fallowed plots were maintained with regular tillage or rotavation at least four times *per* year. Arable and bare-fallowed plots were tilled to a standard depth of 23 cm. Plots have been sampled annually using cores (10 cm height and 3 cm diameter) in October, except in 2018 where the plots were sampled in June. Following sampling, soil was air-dried and sieved at 2 mm before being archived at room temperature. Aggregates (< 2 mm diameter) from continuous bare fallow (bare-fallow), bare fallow converted into arable (arable) and bare fallow converted to grassland (grassland) were randomly selected from samples collected in 2008, 2010, 2012, 2015 and 2018, representing 0-, 2-, 4-, 7- and 10- years *post* conversion. The replication of treatments was a total of 9 scanned aggregates (< 2 mm) *per* year and *per* treatment where randomly selected from 3 independent plots *per* treatment and 3 aggregates (< 2 mm) *per* plot (i.e. 3 replicates *per* plot) to be X-ray CT scanned.

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X-ray Computed Tomography

Aggregates (< 2 mm diameter) were scanned using a Phoenix Nanotom® (GE Measurement and Control solution, Wunstorf, Germany) set at a voltage of 90 kV, a current of 65 µA and at a resolution of 1.50 µm (thus pores below this size were not considered) at the Hounsfield Facility at The University of Nottingham. A total of 1,440 projection images were taken at a 500 ms period using an averaging of 3 images and skip of 2. The total scan time per sample was 60 minutes. Scanned images were reconstructed using Phoenix datos|x2 rec reconstruction software. They were optimised to correct for any movement of the sample during the scan and subjected to noise reduction using the beam hardening correction algorithm, set at 8.

Image analysis

Image analysis was performed using two software packages, ImageJ (Schneider et al., 2012) and QuantIm (Vogel et al., 2010) following the method from Bacq-Labreuil et al. (2018). Briefly, all the images were thresholded using the bin bi-level threshold developed by Vogel and Kretschmar (1996). QuantIm was used to output the 3D characteristics of the pore network calculated from the Minkowski functions where the total porosity (referred to as porosity from here) is the percentage of all the pores >1.5 μm; pore size distribution is the proportion of each size class in the volume normalised to the total pore volume, expressed here as a cumulative value; pore connectivity was calculated from the Euler number and normalised to the total volume (the more negative the Euler number, the greater the pore-connectivity); the pore surface density represents the roughness of the surface of pores: a lower surface density means a lower roughness, i.e. less surface to be colonised by living organisms (Vogel et al., 2010). The Gini-coefficient (G), a statistical measure of distribution, was also determined. It is commonly applied in economics research to estimate the statistical dispersion of income or wealth, and commonly used as a measurement of inequality (Bellù and Liberati, 2006). Here, G was applied to measure the distribution of pore size classes as an indicator of the equality of the pore size distribution. $G \approx 0$ represents an equitable distribution of pores amongst all pore size classes meaning that the soil pores have a homogenous distribution of the pore sizes. $G \approx 1$ represents a heterogeneous distribution of pores which means that a majority of pores have the same sizes.

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Statistical analysis

A standard analysis of variance (ANOVA) was performed using Genstat v 17.1 (VSN International Ltd., 2014) on the porosity. A two-factor ANOVA was conducted on each Minkowski function divided by year using a split plot design with the treatment and the diameter of pores as factors. For total porosity, *G*, the connected porosity and pore surface area, an analysis of co-variance (ANCOVA), was also performed between the arable and grassland with years' post-conversion as a co-variate using SigmaPlot for Windows ver. 14.0 (Systat Software Inc., San Jose, CA). In the case of pore surface area, pore diameter was employed as a second covariate. Parameters were tested following either square root or $\log_{10}$ transformation where necessary to conform to model assumptions of normality (tested using the Shapiro-Wilk test) and homogeneous variances (tested using Levene's test). In each case, ANCOVA was used to test for homogeneity of slopes associated with the change of total and connected porosities and *G* with years' post-conversion of bare fallow to either arable or grassland management. Soil managed as bare fallow throughout the experiment was used to account for temporal changes in soil parameters under continuous management. *Post hoc* pair-wise comparisons were performed employing the Copenhaver & Holland multiple comparisons procedure (Holland and Copenhaver, 1987).

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Results

Visual appraisal of soil structures

Representative 2D images showed that after 1 and 3 years, all three treatments had similar pore architectures in terms of size and shape (Fig. 1a-f). After 5 years, arable and grassland started to display different pore configurations clearly manifest by a greater proportion of larger pores (>40 µm; Fig. 1g-i). The evolution of the pore characteristics over time was apparent, after 7- and 10-years *post* conversion for the arable and grassland treatments especially for vugh (i.e. irregular) and crack shaped pores (Fig. 1j-o). In contrast the size and distribution of pores was relatively consistent over time.

Total Porosity

Before the conversion (in 2008) and after 2 and 4 years, there were no significant treatment effects on porosity ($P>0.05$; Fig. 2a-c) compared to 7- and 10-years post conversion (respectively $P=0.029$ and $P=0.002$; Fig. 2d, e). After 7 years, the porosity in the grassland soil was greater than in bare-fallow or arable soils which were similar (Fig. 2d). However, after 10 years, porosity increased in the presence of plants according to the ranking; bare-fallow < arable < grassland (Fig. 2e). No significant change in $\log_{10}$ total porosity was observed in the continuous bare-fallow soil over the 10 years (slope = 0.026, $t = 0.104$, $p = 0.917$) (Supplementary Fig. 1). However, total porosity in soils converted to arable (slope = 0.713, $t = 3.4$, $p = 0.0014$) and grassland (slope = 0.41, $t = 3.7$, $p < 0.001$) managements increased over the same period (Fig. 1f). ANCOVA comparing the arable and grassland soils identified a significant time response in $\log_{10}$ porosity ($F_{1,86} = 31.0$, $p < 0.001$) but no significant difference in the rates of change (slope) in total porosity ($F_{1,86} = 0.8$, $p = 0.36$). The resulting equal slopes model identified a significant difference in the adjusted mean $\log_{10}$ total porosity of each treatment ($F_{1,87} = 23.5$, $p < 0.001$) with grassland accumulating significantly greater $\log_{10}$ porosity (0.985 ± 0.025, equivalent to 9.66 ± 1.05%, adjusted mean ± standard error of the mean) than arable soil (0.848 ± 0.021, equivalent to 7.05 ± 1.05%)

Pore size distribution

Before the conversion (in 2008), there was no significant treatment effect on the cumulative pore size distribution ($P>0.05$; Fig. 3a). Between 2- to 10-years post conversion, there was a significant diameter by treatment interaction with respect to the cumulative pore size

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distribution (2 and 7 years: *P<0.001*; 4 and 10 years: *P<0.05*; Fig. 3b-e). After 2 years post conversion, there was a greater proportion of smaller pores under bare-fallow and arable treatments than grassland: for bare-fallow and arable, approximately 50% of pores were smaller than 3.56 µm and 70% of pores smaller than 5.97 µm compared to grassland where 50% of pores were smaller than 5.97 µm and 70% smaller than 14.9 µm. Moreover, the proportion of pores larger than 42 µm was greater under grassland (13% of pores) than bare-fallow and arable (respectively 1% and 2% of pores; Fig. 3b). After 4 years, this trend was not apparent: the difference between grassland compared to bare-fallow and arable was less significant than after 2 years. The proportion of pores smaller the 9.26 µm was greater under bare-fallow and arable compared to grassland but the proportion of pores larger than 42 µm was not significant between all treatments (Fig. 3c). After 7 years, the trend observed after 2 years was more apparent: the proportion of pore sizes smaller than 14.9 µm was greater ranking from bare-fallow > arable > grassland and the proportion of pore sizes over 42 µm was greater under arable and grassland (respectively 7% and 10% of pores) than bare-fallow (2% of pores; Fig. 3d). After 10 years post conversion, this trend was also observed, but only for pores smaller than 9.26 µm, where the proportion of pores followed the ranking; bare-fallow > arable > grassland (Fig. 3e). Beyond this pore size, the proportion of pore sizes was not significantly different between bare-fallow and arable. The proportion of pore sizes greater than 42 µm was highest under grassland (15% of pores) than bare-fallow and arable (respectively 4% and 2% of pores; Fig. 3e).

The general trend in all three treatments was a shift to a more even distributions of pore sizes, manifest as a decrease in G over time (Fig. 3f). ANCOVA indicated equal rates of change of G between treatments ($F_{2,129} = 0.18$, $p = 0.834$). Using an equal slopes model, there was a significant effect of land management upon G ($F_{2,131} = 9.1$, $p < 0.001$) with grassland having a significantly lower adjusted mean G (0.420 ± 0.033) than either bare-fallow (adjusted mean

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$G = 0.566 \pm 0.028$; $t = 3.6$, $p < 0.001$) or arable (adjusted mean $G = 0.571 \pm 0.024$; $t = 3.7$, $p < 0.001$). There was no significant difference in adjusted mean G between arable and bare-fallow ($t = 0.13$, $p = 0.900$).

Pore connectivity

Before the conversion (in 2008) and after 7-years post conversion, there was no significant pore diameter by treatment interaction with regards to pore connectivity ($P= 0.05$; Fig. 4a, d). However, there was a significant pore diameter by treatment interaction after 2-, 4- and 10-years (with 2- and 10-years: $P<0.001$; 4-years: $P<0.05$; Fig. 4b, c, e). After 2- and 4- years, the difference was significant only for the pore sizes smaller than 3.56 µm. After 2-years, pore connectivity was greater ranking from bare-fallow > grassland > arable (Fig. 4b) and after 4-years, pore connectivity was greater under arable and grassland than bare-fallow (Fig. 4c). After 10-years post-conversion, the same trend as after 4-years was shown with a greater difference in the values (Fig. 4e). There was no significant trend in square root transformed connected porosity (Supplementary Fig. 2) in bare-fallow (slope = -0.00023, $t = 0.051$, $p = 0.959$). However, both arable (slope = 0.017, $t = 4.0$, $p = 0.0002$) and grassland (slope = 0.022, $t = 5.3$, $p < 0.0001$) showed increases in connected porosity with time. ANCOVA comparing arable and grassland indicated a significant influence of time *post* conversion upon square root transformed connected porosity ($F_{1,85} = 43.9$, $p < 0.001$) but no significant heterogeneity of slopes ($F_{1,85} = 0.98$, $p = 0.326$). Using an equal slopes model, a significant effect of management was detected ($F_{1,85} = 4.4$, $p = 0.039$): grassland was associated with greater connected porosity (0.048 ± 0.0002%, adjusted mean ± standard error) than arable (0.011 ± 0.0002%).

Pore surface density

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Before the conversion (in 2008) and at 4- and 7-years *post* conversion, there was no significant pore diameter by treatment interaction with regards to pore surface density ($P>0.05$; Fig. 5a, c, d). There was a significant diameter by treatment interaction after 2- and 10-years (respectively $P<0.05$ and $P<0.001$; Fig. 5b, e). After 2-years, the difference in pore surface density was greater ranking from bare-fallow > arable > grassland for the pore sizes equal to 1.86 µm, and the difference between arable and grassland was not significant for the pore sizes equal to 3.56 µm. Beyond this pore size, there was no significant difference between treatments (Fig. 5b). Ten years *post* conversion, pore surface density was greater ranking from grassland > arable > bare-fallow, for all pore sizes smaller than 14.9 µm, there was no significant difference beyond this pore size (Fig. 5e). Years post-conversion and pore diameter were both used as covariates in ANCOVA analysis of pore surface area. Accounting for these two covariates, an equal slopes model identified a significant effect of land management upon pore surface area ($F_{2,2020} = 9.1$, $p < 0.001$). *Post hoc* pair-wise comparison of adjusted means indicated that grassland supported a greater pore surface area (0.00784 ± 0.000493 $\mu m^2\ \mu m^{-3}$) than either arable (0.00617 ± 0.000452 $\mu m^2\ \mu m^{-3}$; difference = 0.00167, $t = 3.4$, $p = 0.001$) or bare-fallow (0.00593 ± 0.000443 $\mu m^2\ \mu m^{-3}$; difference = 0.00191, $t = 3.9$, $p < 0.001$). There was no significant difference in pore surface area between arable and bare-fallow (difference = 0.000240, $t = 0.495$, $p = 0.621$).

Discussion

The plots studied here were derived from long-term bare-fallow management converted to arable and grassland. A lack of a significant treatment effect on porosity until 7-years post conversion suggests that modification of micro-porosity takes several years (Fig. 2). Another study on the same soil 2 and 4 years post conversion found some recovery of meso-faunal populations after 2-years of conversion and an increase in soil organic matter and microbial

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abundance after 4- and 2-years respectively (Hirsch et al., 2017). However, our study showed that development of micro-scale porosity apparently takes longer. This might be related to carbon cycling processes which are modified by the microbial communities and plants (*via* decomposition of organic matter and rhizodeposition). This is likely to affect soil structure at the micro-scale, but not instantaneously. Increased pore formation under grassland compared to arable is consistent with a previous study, which showed greater resistance to, and development from physical stresses of soil structure from grassland (Gregory et al., 2009). They posited that the greater proportion of organic matter enhanced the elastic recovery of soil structure (Gregory et al., 2009).

Pore size distributions showed a more rapid response to altered management than porosity for the grassland treatment: after only 2-years of conversion (in 2010), a greater diversity of pore sizes was observed, and this trend was also recorded in the data after 7- and 10-years (Fig. 3). The Gini-coefficient indicated that soil converted to grassland established a more even distribution of pore sizes than the other treatments, meaning that grassland treatment had a greater diversity of pores after 2-, 7- and 10-years post conversion (Supplementary Fig. 2) leading to enhanced functionality. This increase in pore size diversity might be due to the increase of presence of plants, active organisms and organic matter (Hirsch et al., 2017) as well as the absence of tillage. A study focused on the soil organic carbon on the same experiment found that the conversion of the bare-fallow soil to grassland led to an increase of soil organic carbon (+46 %) 7-years post conversion (Jensen et al. 2020a). There is no data regarding the conversion from bare-fallow to arable. Thus, the increase of organic matter in the converted soil may play a role in the more homogenous distribution of the pore sizes. Indeed, in a silty clay soil, the greater organic matter content increases the proportion of pores between 0.5 to 500 µm (Metzger and Yaron, 1987; Watts and Dexter, 1997) leading to a more equitable distribution of pore sizes, i.e. a greater diversity of pore sizes. The greater

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diversity of pore sizes under soil converted to grassland was consistent with a previous study describing the long-term effect of grassland management on the same field experiment (Bacq-Labreuil et al., 2018). After 4-years the pore size distribution did not follow this trend (Fig. 3c), which could be due to weather conditions prior sampling in that year. Indeed, 2008 and 2012 (at the start and 4-years post conversion respectively) were the wettest years during the experimental period (Supplementary Fig. 3). In the presence of water, clay particles can swell, and the compression of entrapped air in capillary pores can disrupt the pore architecture, and affect the pore size distribution (Denef et al., 2001; Grant and Dexter, 1990). Pore networks are re-structured upon re-wetting due to the nature of soil particles. Changes in pore size between 2-, 4- and 7-years post-conversion raises the question of the dynamics of this mechanism. The pore size distribution may have had a heterogeneous response over time due to the impact of the wet year 4-years post-conversion, which shows the rapid development of the pore size distribution after a sustained wet period compared to the impact of agricultural practices (Supplementary Fig. 3). For the arable treatment, this trend was not observed even 10-years post conversion, which might be due to the associated tillage practices.

For pore connectivity, the conversion to grassland had a small effect after 2- and 4-years compared to after 10-years post conversion (Fig. 4 b, c, e). However, pore connectivity data after 10-years post conversion, for both arable and grassland converted soils (Fig. 4e), suggested the pore network was less connected compared to Bacq-Labreuil et al., (2018). This indicated that a longer time may be required to develop the connectivity of a pore network than the overall porosity. Increased connectivity of pores promotes water, gasses and nutrient flows within the pore structure (Dexter, 1988; Tisdall and Oades, 1982). Therefore, subtle increases in pore connectivity might increase water, gas and nutrient flux within the soil. As well as the pore connectivity, the pore surface density was significantly increased in

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the grassland and arable 10-years post-conversion (Fig. 5e). Our results are congruent with Bacq-Labreuil et al. (2018), which showed grassland managed consistently for over 200 years has an increased pore surface density compared to arable and bare-fallow soils *i.e.* the pore-solid interface which led to a greater surface of the pore where micro-organisms and plant roots can colonise and water films can develop. A greater pore surface density in the converted plot means that the grassland and the arable have a more complex structure of pores than the bare-fallow soil (Müller et al. 2019). The greater surface density for the grassland compared to the arable might be induced by the greater SOC content and the absence of tillage for this treatment (Hirsch et al. 2017; Jensen et al. 2020a). This can lead to the formation of new habitats and niches which can be beneficial for microbial community diversity (Holden, 2011). The greater pore surface area might increase water and nutrient uptake by the microbial community and plants.

This study suggests that conversion of degraded bare-fallow soil to grassland requires at least 10-years after conversion before being effective in terms of significant development of soil structure at aggregate scale, as assessed by the overall and connected porosity. Moreover, the conversion from bare-fallow to arable had no significant effect on soil structural properties after a decade. In general, the recovery of meso-fauna and organic matter (Griffiths et al., 2000; Hirsch et al., 2017) were more rapid than the recovery of soil structure (Gregory et al., 2009). The pore size distribution was the only characteristic which was more sensitive to changes induced by wetting and drying cycles and living organisms.

Our first hypothesis was supported since porosity, pore size diversity, pore connectivity, and pore surface area density were all enhanced in grassland soil. Moreover, the conversion to arable management did not affect soil structural development significantly. The conversion to grassland increased the range of pore sizes after 2-years, consistent with our second

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hypothesis. However, all other Minkowski functions (porosity, pore connectivity and pore surface area density) responded to change more slowly. The mechanisms behind the development of pore sizes appeared to be dynamic and possibly dependent upon weather conditions before sampling. Apart from the pore size distribution, the magnitude of the grassland effects on all other Minkowski functions was lower than the difference observed after a minimum of 50 years of management (Bacq-Labreuil et al., 2018). In this study, the effect of grassland upon porosity and pore connectivity were two-fold greater than bare-fallow management. Here, the difference was significant but not as major. Bare-fallow soil management for this long period (> 50 years) is detrimental to both physical and biological soil properties and the development of the soil structure after this requires more than 10 years after the conversion the grassland

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Conclusions

The soil structural development of a degraded silt-clay loam soil, as quantified by micro-scale topological metrics, requires at least 10-years of a grassland management before showing any significant effects.. These observations raise the question on the application to certain managements in agricultural practices. For example, instead of applying a bare-fallow treatment in a crop rotation, it would be beneficial for the soil characteristics to apply a vegetation cover *i.e.* cover crops which increase organic matter inputs and influence soil structure, leading to a ‘conditioning’ of soil physical and biological characteristics for the next crop. This would prevent further degradation of soil and help its development if the soil characteristics were compromised. Moreover, the development of soil structure is apparently a long process in the context of current agricultural practices and perceived imperatives. Thus, a modification of cropping managements should be anticipated to require some time before the observation of beneficial impacts on soil structural dynamics.

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Figure 1: Representative 2D X-ray attenuation images of soils subjected to different forms of management over 10-years following conversion to each treatment. Base resolution is 40 µm; (P) pores are the darker shades and (S) soil matrix are the lighter shades which relate to the attenuation of the X-ray (a sharpening algorithm has been passed over these images to increase contrast of features); (a, d, g, j, m) bare-fallow; (b, e, h, k, n) arable; and (c, f, i, l, o) grassland.

Figure 2: Porosity (based on resolution of 1.5 µm) in bare fallow, arable and grassland soils in the years following: (a) 0-year; (b) 2-years; (c) 4-years; (d) 7-years; (e) 10-years. Bars are means (n = 9) expressed as the percentage of pores relative to the total volume, whiskers denote pooled standard errors. (f) Porosity evolution 0- to 10-years *post* conversion, data points represent means (n = 9), whiskers denote pooled standard errors for clarity and trend lines are linear regressions.

Figure 3: Cumulative pore size normalized to the total volume in relation to bare-fallow, arable and grassland in the years following conversion: (a) 0-year; (b) 2-years; (c) 4-years; (d) 7-years; (e) 10-years. Data points indicate means (n=9), whiskers denote pooled standard errors. (f) Gini coefficient development 0- to 10-years post conversion, data points represent means (n = 9), whiskers denote pooled standard errors for clarity and trend lines are linear regressions.

Figure 4: Pore connectivity normalized to total volume in continuous bare fallow, arable and grassland soils following conversion: (a) 0-year; (b) 2-years; (c) 4-years; (d) 7-years; (e) 10-years. Data points indicate means (n=9), whiskers denote pooled standard errors.

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Figure 5: Pore surface density in continuous bare fallow, arable and grassland soils following conversion: (a) 0-year; (b) 2-years; (c) 4-years; (d) 7-years; (e) 10-years. Data points indicate means (*n* = 9), whiskers denote pooled standard errors.

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Appendix

Significant structural development of a long-term fallow soil in response to agricultural management practices requires at least 10 years after conversion

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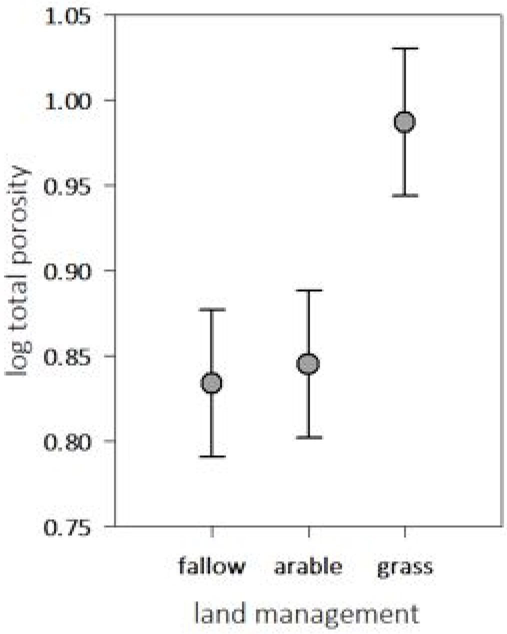
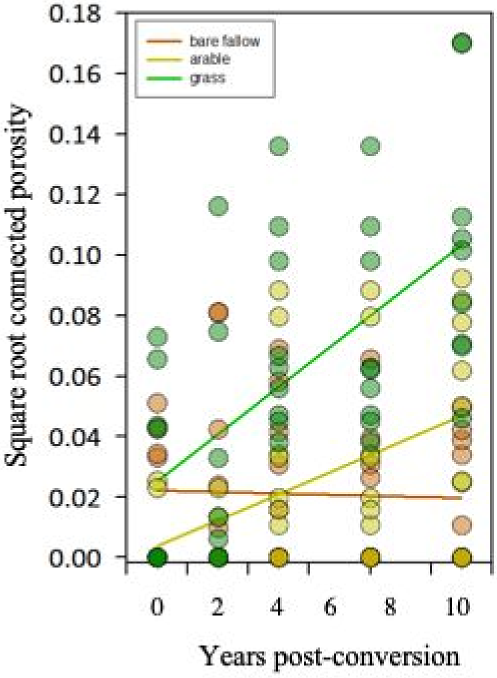


Supplementary Figure 1: Log_{10} total porosity, points represent the adjusted means generated from an analysis of covariance, employing time *post* conversion (years) as the covariate. Errors represent the 95% confidence intervals associated with the adjusted means



Supplementary Figure 2: Square root transformed connected porosity: points represent the replicates and the lines are a linear regression for each treatment.

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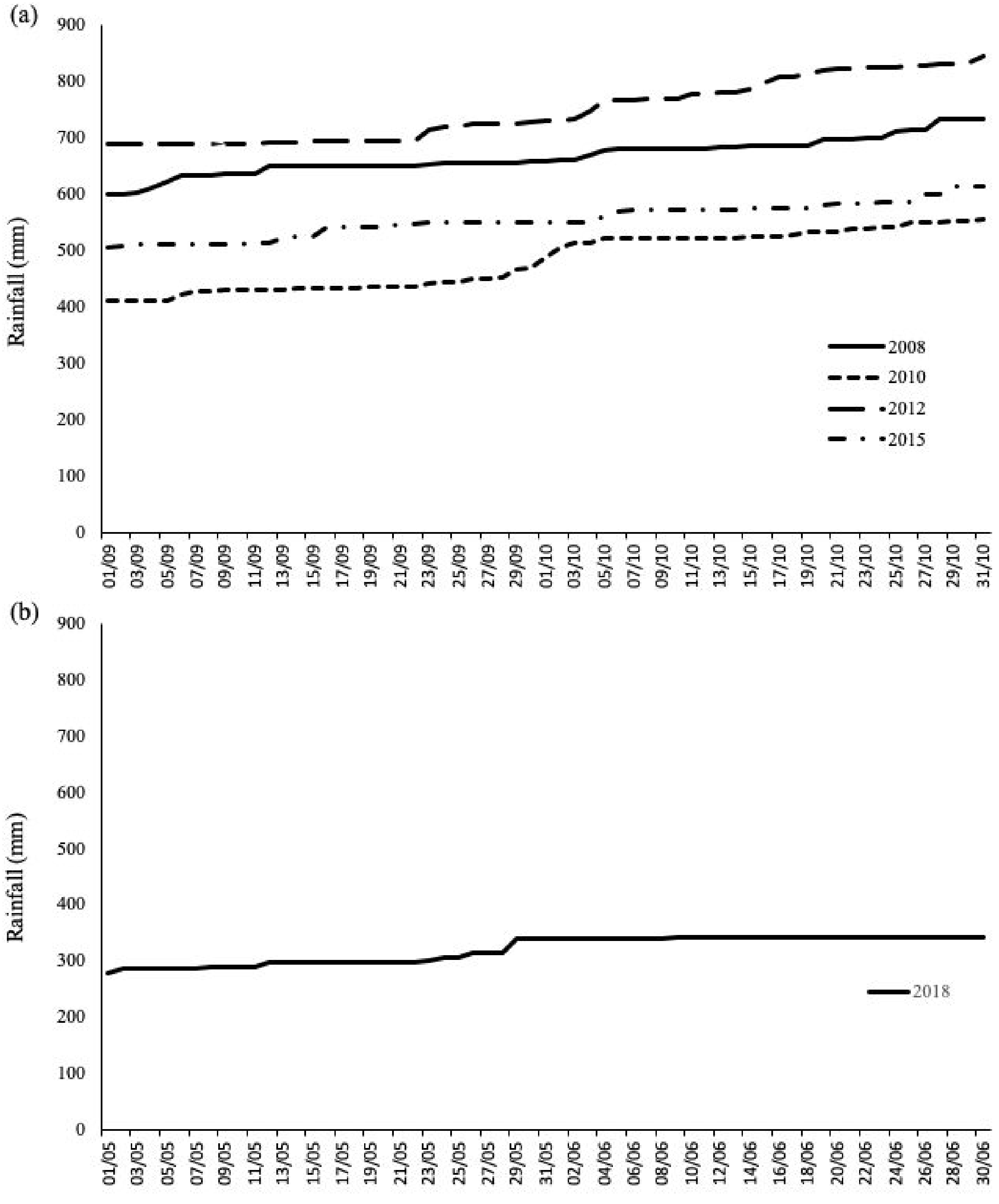
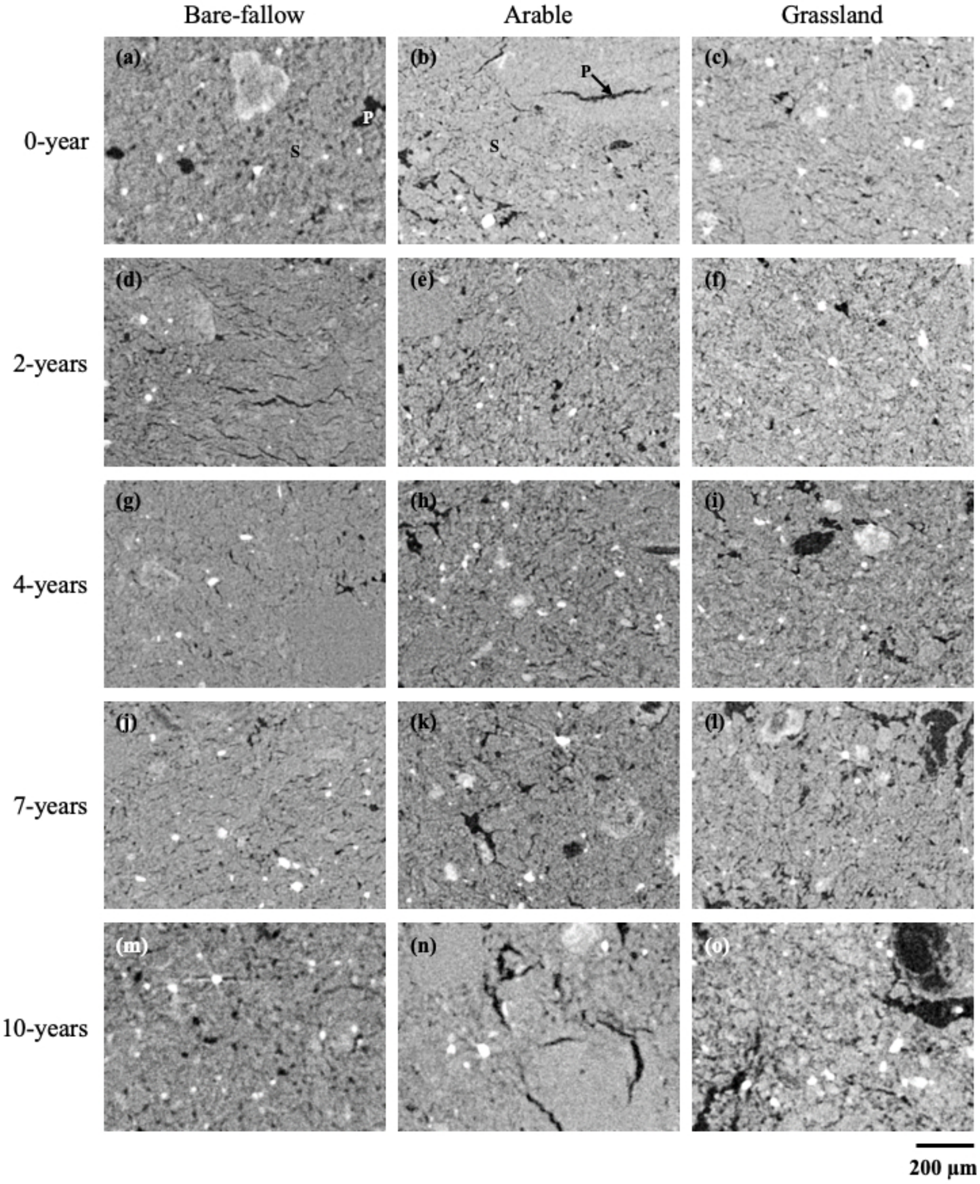


Supplementary Figure 3: Cumulative rainfall (mm) on the Highfield from (a) September to October for 2008 to 2015; and (b) May to June for 2018, as the sampling time were different.



Representative 2D X-ray attenuation images of soils subjected to different forms of management over 10-years following conversion to each treatment. Base resolution is 40 μm; (P) pores are the darker shades and (S) soil matrix are the lighter shades which relate to the attenuation of the X-ray (a sharpening algorithm has been passed over these images to increase contrast of features); (a, d, g, j, m) bare-fallow; (b, e, h, k, n) arable; and (c, f, i, l, o) grassland.

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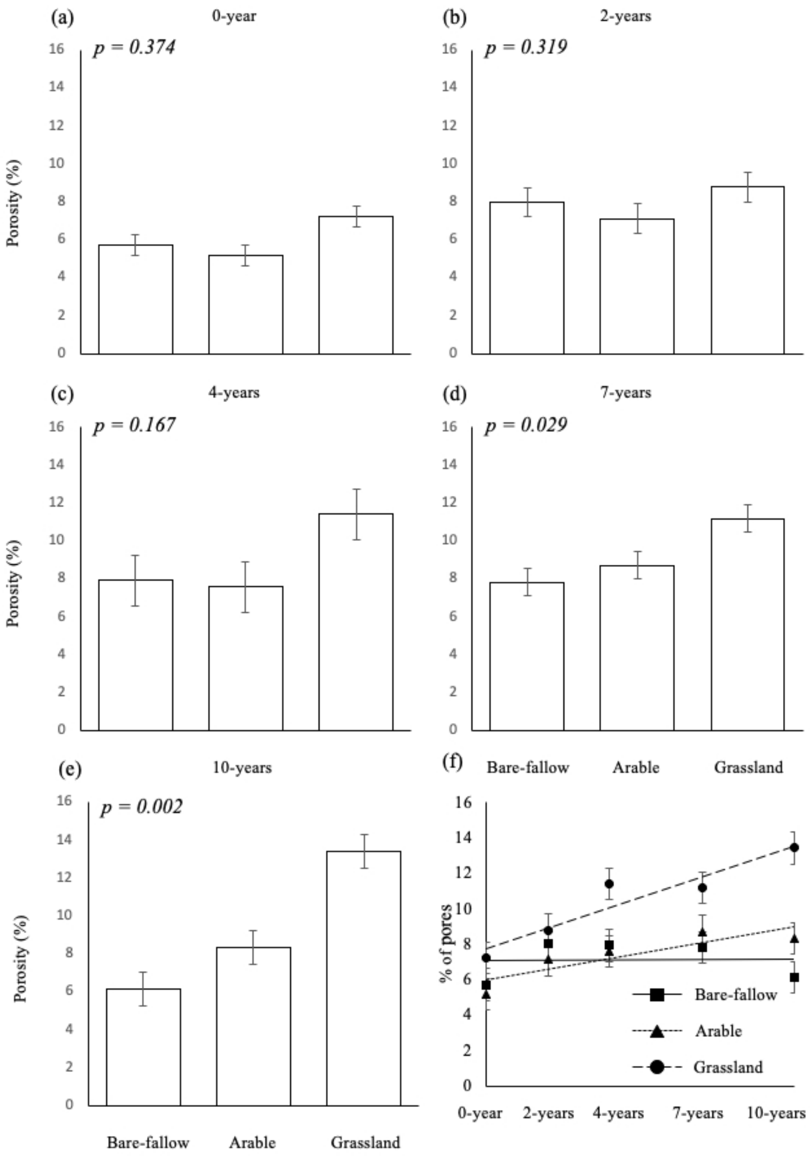

Porosity (based on resolution of 1.5 μm) in bare fallow, arable and grassland soils in the years following: (a) 0-year; (b) 2-years; (c) 4-years; (d) 7-years; (e) 10-years. Bars are means (n = 9) expressed as the percentage of pores relative to the total volume, whiskers denote pooled standard errors. (f) Porosity evolution 0- to 10-years post conversion, data points represent means (n = 9), whiskers denote pooled standard errors for clarity and trend lines are linear regressions.

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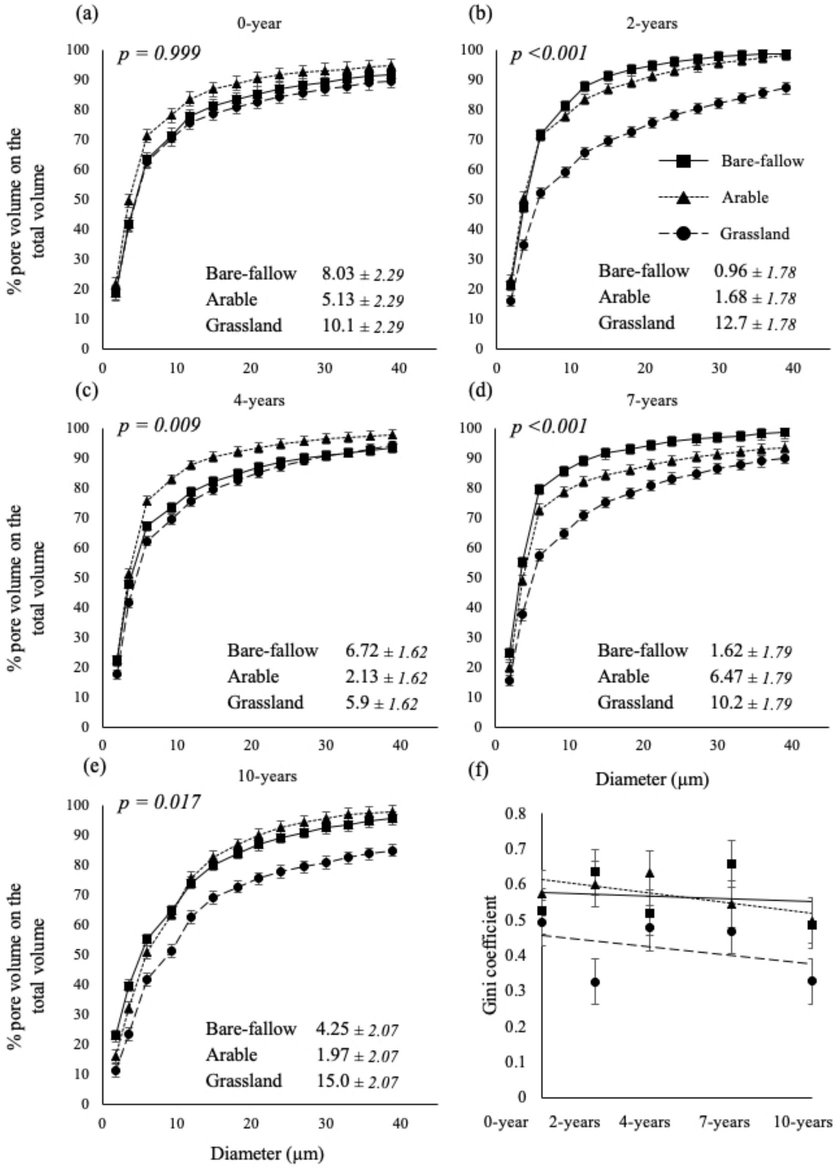

Cumulative pore size normalized to the total volume in relation to bare-fallow, arable and grassland in the years following conversion: (a) 0-year; (b) 2-years; (c) 4-years; (d) 7-years; (e) 10-years. Data points indicate means (n=9), whiskers denote pooled standard errors. (f) Gini coefficient development 0- to 10-years post conversion, data points represent means (n = 9), whiskers denote pooled standard errors for clarity and trend lines are linear regressions.

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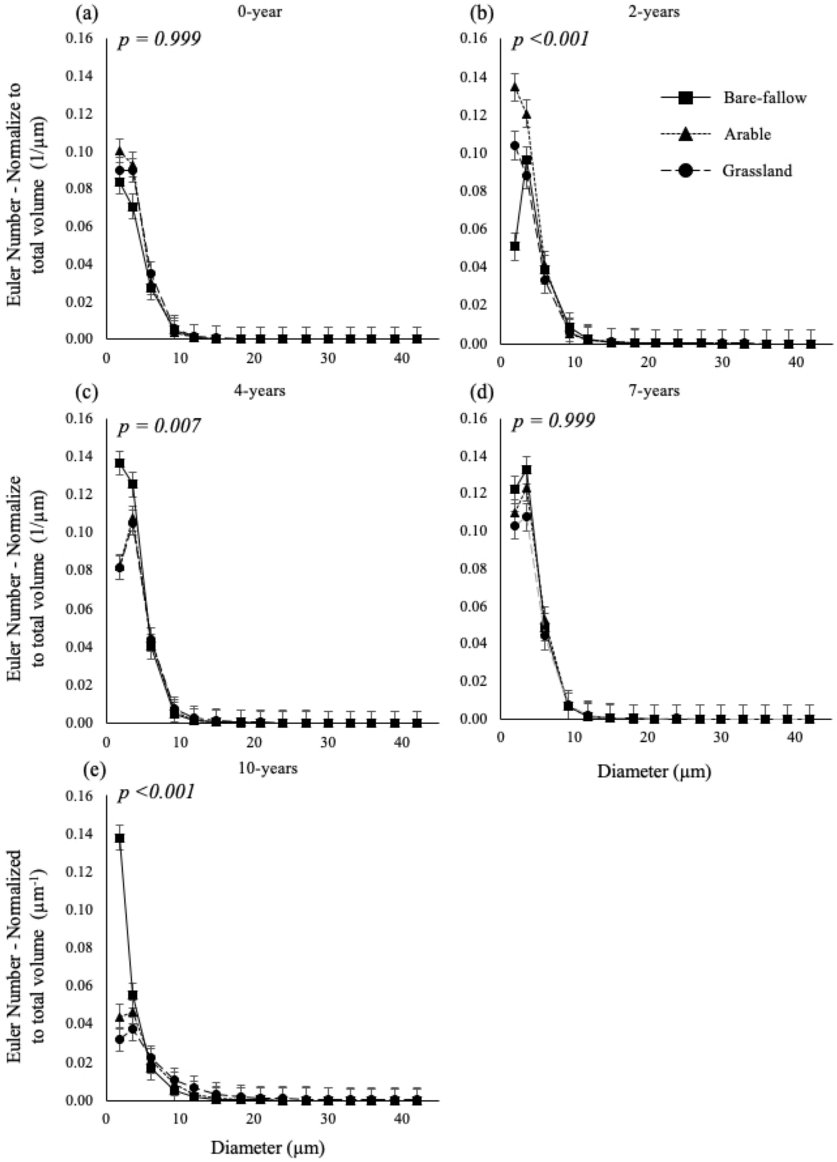

Pore connectivity normalized to total volume in continuous bare fallow, arable and grassland soils following conversion: (a) 0-year; (b) 2-years; (c) 4-years; (d) 7-years; (e) 10-years. Data points indicate means (n=9), whiskers denote pooled standard errors.

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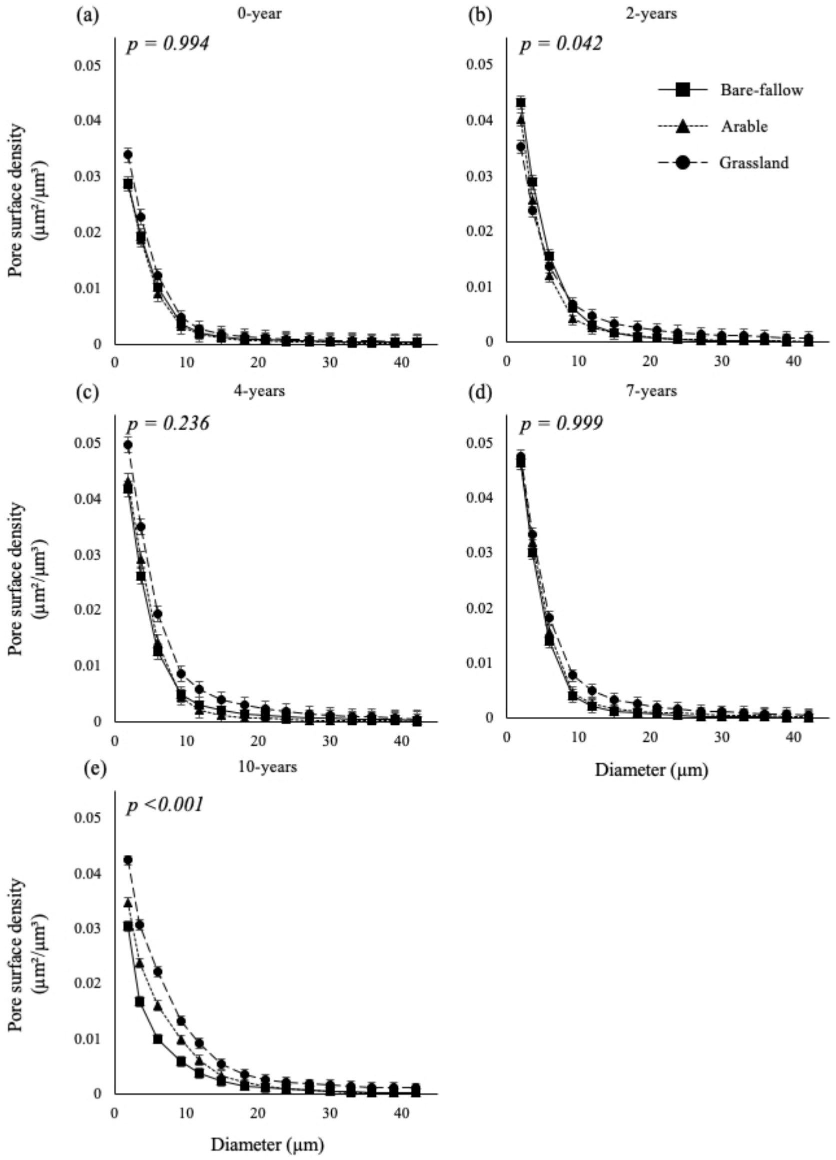

Pore surface density in continuous bare fallow, arable and grassland soils following conversion: (a) 0-year; (b) 2-years; (c) 4-years; (d) 7-years; (e) 10-years. Data points indicate means (n = 9), whiskers denote pooled standard errors.

176x244mm (96 x 96 DPI)

Significant ~~S~~structural ~~recovery~~ development of a long-term fallow soil in response to ~~annual or perennial~~agricultural management practices ~~cropping~~ requires at least 10 years after conversion

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Abstract (250 words, currently ~~230~~23~~3~~12)

Agricultural practices can have significant effects on ~~soil~~ the physical and biological ~~soil~~ properties of soil. ~~Crop rotation and modification of cropping systems can lead to marked effects on these properties and enhance plant growth, sequestration of carbon and reorganise soil structure.~~ The aim of this study was to understand how the physical structure of a compromised soil, arising from ~~a~~ long-term ~~bare~~ bare-fallow ~~period~~management, was modified by adopting different field management practices. ~~Soil aggregates collected on successive years from field plots subjected to three contrasting management regimes were studied, *viz.* bare fallow, bare fallow converted to arable, and bare fallow converted to grassland.~~ We hypothesised that ~~a~~ chang~~e~~ing ~~of plant inputs~~agricultural practice~~s~~ from bare-fallow to arable ~~and~~or grassland would influence the modification of pore structure *via* an increase ~~of~~ in porosity, pore connectivity,~~,~~ and a more homogenous distribution of pore sizes;~~diversity of pore sizes and pore connectivity;~~ and that this~~e~~ ~~effect of plants~~change exerts a rapid ~~recovery~~ development of soil structure ~~after~~ following conversion. Soil aggregates (< 2 mm) collected ~~o~~in successive years from field plots subjected to three contrasting managements were studied~~,~~; *viz.* bare-fallow, bare-fallow converted to arable, and bare-fallow converted to grassland. Soil structure was assessed by X-ray Computed Tomography ~~of 2 mm~~on the aggregates at 1.5 µm resolution, ~~to~~ captur~~e~~ing detail relevant to ~~key~~ soil biophysical processes. ~~The greatest presence of plants, here represented by t~~The grassland system~~,~~ increased ~~significantly~~ porosity, diversity of pore sizes, pore-connectivity and pore-surface density significantly over the decade following conversion. However, measured at this resolution, the ~~recovery~~ development of most of these metrics of soil structure required approximately 10 years post-conversion to show a significant effect ~~of plant presenceafter the conversion to grassland~~. The arable system did not ~~affect~~influence ~~the~~ soil structur~~e~~al development significantly. Only the pore size distribution was modified in grassland in ~~by~~

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~~plants in~~ a shorter time frame (2 years post-conversion). Hence ~~Full~~ d~~D~~evelopment~~recovery~~ of the soil structural characteristics~~, therefore,~~ appears to require at least a decadal ~~time-scale~~timescale ~~after being converted~~following conversion to grassland.

Key words:

Soil structure, ~~soil recovery,~~ 3D pore characteristics, ~~cropping systems~~agricultural management practices, X-ray Computed Tomography, porosity

Highlights:

- ~~How t~~The physical structure of a compromised soil was modified by adopting ~~different~~ plant-based field management practices.
- ~~The presence of plantse~~Conversion to grassland increased ~~the~~ pore size diversity ~~of pore sizes~~ after ~~only~~ 2 years~~s post-conversion~~.
- Porosity, pore connectivity and pore surface ~~area~~ density showed ~~recovery~~ a significant modification between 7 to ~~11~~ 10 years ~~post-~~after conversion.
- Bare fallow soil management for this extreme period (> 50 years) is detrimental to ~~both~~ physical ~~and biological~~ soil properties and the ~~recovery~~ regeneration of the soil structure requires more than 10 years after being reconverted to arable and grassland.

Introduction

~~When applied for decades to soil, a~~Agricultural practices can have ~~both~~ beneficial or detrimental effects on soil function~~propertie~~s~~,~~ when applied for decades, depending on the nature of such practices (Ashworth et al., 2017; Bronick and Lal, 2005; Denef et al., 2009; Pagliai et al., 2004). Agricultural management generally aims to increase - ,~~—~~or at least

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stabilise ~~—~~ crop yield, but intensive farming can lead to soil degradation, erosion, compaction and pollution (Bronick and Lal, 2005). Conventional tillage can lead to a decline ~~of~~ in soil aggregation and soil structure (Watts et al., 2001), as well as ~~a~~ depletion of nutrients and organic carbon within soil (Coleman et al., 1997). ~~The a~~Addition of organic matter or crop rotations can prevent soil disruption from tillage by improving soil porosity and aggregation (Abdollahi et al., 2014; Pagliai et al., 2004). In some cases, m~~M~~odification of crop~~ping~~ management can have beneficial impacts on soil ~~properties~~functions. For example, after 50 years of continuous cultivation, a desert aeolian sandy soil was managed into a sustainable agricultural soil by increasing silt and clay content ~~, which was~~ (a determinant for aggregate formation), soil organic matter and ~~the content of silt and clay (caused by irrigation) leading to an increase of aggregation~~nutrient retention (Su et al., 2010). Moreover, soil aggregate stability is a key factor for soil fertility and physical resilience from external forces, e.g. wind and water (Abivent et al. 2009).

Soil structure plays a fundamental role in the distribution of carbon, soil microorganisms, water and nutrient accessibility (Rabot et al. 2018; Smith et al. 2017). ~~The a~~Analysis of soil structure ~~led to suggest~~indicates that pore size distribution, assessed by X-ray Computed Tomography ~~(X-ray~~ (CT), plays an important role in aggregate stability (Menon et al. 2020). ~~The greater~~Increased diversity of pore sizes ~~diversity~~ (i.e. a more homogenous distribution of pores) ~~creates~~is associated with a more complex pore network. This ~~might~~ leads to an increase ~~of the~~in the number of storage and transmission pores resulting in ~~an increase of~~greater water and nutrient ~~flows~~flux (Kravchenko et al. 2014; 2019). The modification of ~~the~~ pore size distributions appears to also play ~~also~~ a key role in the decomposition of organic matter (Quigley et al. 2018; Smith et al. 2017; Toosi et al. 2017a; 2017b). For example, the presence of small (13-32 µm) and large (136-260 µm) pores decrease~~s the~~ organic matter decomposition within macro-aggregates (Toosi et al. 2017b). ~~The p~~Pore connectivity is also

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one of the most important factors, alongside~~ with~~ porosity and pore size distribution, to understand soil functions (Rabot et al. 2018). ~~The m~~Modification of pore connectivity can have a significant effect upon the distribution and the transport of gas and water (Lucas et al. 2020; Müller et al. 2019; Pires et al. 2019). Pires et al. (2017) demonstrated that pore connectivity was enhanced in zero tillage systems over conventional tillage systems, especially in the upper 10 cm.

In the field, long-term

~~Furthermore, microbial biomass – a fundamental component of soil fertility – is highly sensitive to tillage (Ashworth et al., 2017). Drijber et al. (2000) showed that on bare-fallow plots microbial biomass was reduced by approximately 30% compared to cropped plots after 25 years. The composition of the bacterial communities is more closely related to soil characteristics (such as pH and soil texture) than cropping management, and the fungal community is more associated with nutrients within the soil than cropping management (Lauber et al. 2008). By contrast, a recent study showed that crop management did not have any impact on the microbial community structure, but had an effect on the distribution of genes coding for different functions (Neal et al., 2017). This study focused on phosphatase-coding genes, and showed that bare-fallowed soil contained more genes coding for extracellular and outer-membrane associated enzymes compared to grassland and arable soils, leading to a community with greater foraging potential to access nutrients from a greater distance under the bare-fallow (Neal et al., 2017). Moreover, different~~ ~~m~~~~M~~management practices can have ~~a ~~substantial impacts on soil structural ~~-~~dynamics (Bacq-Labreuil et al. 2018; Müller et al. 2019; Pires et al. 2019). For example, ~~After ~~50~~-~~-years ~~under a~~of management of a typical silty clay loam soil as bare fallow ~~treatment ~~resulted in significant reductions of carbon and nitrogen~~n was markedly reduced~~, and in the~~the~~ abundance of biological communities ~~was reduced ~~(Hirsch et al. 2009). ~~and sS~~with soil

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structure ~~was~~ also severely compromised (Bacq-Labreuil et al. 2018). Conversion from bare fallow to arable or grassland increased soil organic carbon, soil nitrogen and the population of meso-fauna and fungi within 3 to 5 years ~~after~~ following conversion (Hirsch et al. 2017), ~~.~~ ~~However~~although, soil structure modification was not assessed in this experiment. The aim of this study was to establish how the micro-structure of a compromised silt-clay loam soil is modified by altered field management over time using soil aggregates (< 2 mm diameter). Three treatments were studied from the converted field of the long-term experiment: continuous ~~bare~~ bare-fallow, bare-~~-~~fallow converted to arable, and ~~bare~~ bare-fallow converted to grassland. We hypothesised that: (1) plants ~~inputs~~ are an active factor in ~~the modification of soil pore structure *via* an~~ increas~~e~~ing ~~in~~soil porosity, diversity of pore sizes and pore connectivity, and (2) structural ~~recovery~~ development ~~would be~~is more rapid in grassland than arable converted systems due to the greater and- ~~presence of~~more persistent presence of vegetation~~plant populations~~. The precise time for ~~recovery~~ soil structural development is unclear *a priori*, and we aimed to determine this by measuring structural properties on several successive years after conversion.

Materials and Methods

Soil a~~A~~ggregate~~es~~ sampling~~es~~

Samples were obtained from conversion plots of the long-term Highfield Ley-Arable experiment at Rothamsted Research, Harpenden, UK (LATLONG 51.8103 N, -0.3748 E). The soil is a silty-clay loam (clay: 27%, silt: 58,4, sand: 14.6%; Jensen et al. 2020b) developed on clay-with-flints over Eocene London Clay (Batcombe series), classified as Chromic Luvisol by FAO criteria (Avery and Catt, 1995; FAO, 2006; Watt and Dexter, 1997). In October ~~2007~~2008, plots of soil managed as bare-fallow by regular tillage to remove any plants since 1952, were converted to arable and grassland managements. The

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conversion plots had~~ve~~ similar soil characteristics. The conversion of the plots from bare-fallow to arable and grassland i~~wa~~s explained in details in Hirsh et al. (2017). ~~Arable~~ ~~Briefly,~~ ~~a~~Arable soil was placed under continuous wheat rotation (winter wheat, *Triticum aestivum* L., *c.v.* ‘Hereward’ seed coated with Redigo® Deter® combination insecticide/fungicide treatment, Bayer CropScience) receiving ammonium nitrate fertilization to provide approximately 220 kg-N ha^{-1} ~~annumy~~$^{-1}$~~, and~~. For the arable and grassland plots additional fertilizers 250 kg-K ha^{-1} and 65 kg-P ha^{-1} wa~~i~~s added every three years.~~, and~~ ~~grassland~~ Grassland plots we~~a~~re maintained as a managed sward of mixed ~~grasses and forbs~~fescue (*Festuca pratensis* L.), ~~t~~Timothy~~-~~ grass (*Phleum pratense* L.) and white clover (*Trifolium repens* L.) (30 kg ha^{-1}). To remove weeds, bare-fallowed plots we~~a~~re maintained with regular ~~ploughed,~~ tillage ~~and~~or rotavat~~ed~~ion at least four times *per* year. Arable and bare-fallowed plots we~~a~~re tilled ~~with~~to a standard depth of 23 cm. Plots have been sampled annually using cores (10 cm height and 3 cm diameter) in October, except in 2018 where the plots were sampled in June~~e 2018,~~. Following sampling, ~~using cores (10 cm height and 3 cm diameter),~~ ~~the and~~ soil was air-dried and sieved ~~(<~~ at 2 mm~~)~~ before being archived at room temperature. Aggregates (< 2 mm diameter) from continuous bare fallow (bare-fallow), bare fallow converted into arable (arable) and bare fallow converted to grassland (grassland) were randomly selected from ~~the years;~~samples collected in 2008, 2010, 2012, 2015 and 2018, representing ~~1~~0-, ~~3~~2-, ~~5~~4-, ~~8~~ 7- and ~~11~~ 10-years *post*~~-~~ conversion. The replication of treatments was~~:~~ a total of 9 scanned aggregates (< 2 mm) *per* year~~s~~ and *per* treatment~~s~~ where randomly selected from 3 independent plots *per* treatment~~s~~ and 3 aggregates (< 2 mm) *per* plot~~s~~ (i.e. 3 replicates *per* plot) ~~were randomly selected~~ to be X-ray CT scanned~~, therefore a total of 9 scanned aggregates *per* year, *per* treatment were assessed by this approach~~.

X-ray Computed Tomography

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Aggregates (< 2 mm diameter) were scanned using a Phoenix Nanotom® (GE Measurement and Control solution, Wunstorf, Germany) set at a voltage of 90 kV, a current of 65 µA and at a resolution of 1.50 µm (thus pores below this size were not considered~~ in this study~~) at the Hounsfield Facility at ~~the~~ The University ~~Of~~ of Nottingham. A total of 1,440 projection images were taken at a 500 ms period using an averaging of 3 images and skip of 2. The total scan time per sample was 60 minutes. Scanned images were reconstructed using Phoenix datos|x2 rec reconstruction software. They were optimised to correct for any movement of the sample during the scan and subjected to noise reduction using the beam hardening correction algorithm, set at 8.

Image analysis

Image analysis was performed using two software packages, ImageJ (Schneider et al., 2012) and QuantIm (Vogel et al., 2010) following the method from Bacq-Labreuil et al. (2018). Briefly, all the images were thresholded using the bin bi-level threshold developed by Vogel and Kretschmar (1996). QuantIm was used to output the 3D characteristics of the pore network calculated from the Minkowski functions where the total porosity (~~called here~~referred to as porosity from here) is the percentage of all the pores >1.5 µm; pore size distribution is the proportion of each size class in the volume normalised to the total pore volume, expressed here as a cumulative value; pore connectivity was calculated from the Euler number and normalised to the total volume (the more negative the Euler number, the greater the pore-connectivity); the pore surface density represents the roughness of the surface of pores: a lower surface density means a lower roughness, i.e. less surface to be colonised by living organisms (Vogel et al., 2010). The Gini-coefficient (*G*),~~ is~~ a statistical measure of distribution, was also determined. It is commonly applied in economics research to estimate the statistical dispersion of income or wealth, and commonly used as a

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measurement of inequality (Bellù and Liberati, 2006). Here, G was applied to measure the distribution of pore size classes as an indicator of the equality of the pore size distribution.~~:~~ $G \approx 0$ represents an equitable distribution of ~~the~~ pores amongst all pore size classes meaning that the soil pores have a homogenous distribution of the pore sizes~~and~~. $G \approx 1$ represents a heterogeneous distribution of pores which means that a majority of pores have~~s~~ the same sizes.

Statistical analysis

A standard analysis of variance (ANOVA) was performed using Genstat v 17.1 (VSN International Ltd., 2014) on the porosity. A two-factor ANOVA was conducted on each Minkowski function divided by year using a split plot design with the treatment and the diameter of pores as factors. For total porosity, G, the connected porosity and pore surface area, an analysis of co-variance (ANCOVA), was also performed between the arable and grassland with years' post-conversion as a co-variate using SigmaPlot for Windows ver. 14.0 (Systat Software Inc., San Jose, CA). In the case of pore surface area, pore diameter was employed as a second covariate. Parameters were tested following either square root or $\log_{10}$ transformation where necessary to conform to model assumptions of normality (tested using the Shapiro-Wilk test) and homogeneous variances (tested using Levene's test). In each case, ANCOVA was used to test for homogeneity of slopes associated with the change of total and connected porosities and G with years' post-conversion of bare fallow to either arable or grassland management. Soil managed as bare fallow throughout the experiment was used to account for temporal changes in soil parameters under continuous management. *Post hoc* pair-wise comparisons were performed employing the Copenhaver & Holland multiple comparisons procedure (Holland and Copenhaver, 1987).

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Results

Visual appraisal of soil structures

Representative 2D images showed that after 1 and 3 years, all three treatments had similar pore architectures in terms of size and shape (Fig. 1a-f). After 5 years, arable and grassland started to display different pore configurations clearly manifest by a greater proportion of larger pores (>40 µm; Fig. 1g-i). The evolution of the pore characteristics over time was apparent, after ~~8 and 11 years~~~~8~~7- and 10~~1~~-years *post-* conversion for the arable and grassland treatments especially for vugh (i.e. irregular) and crack shaped pores (Fig. 1j-o). In contrast the size and distribution of pores ~~is~~ was relatively ~~stable~~ consistent over time.

Total Porosity

Before the conversion (in 2008) and a~~A~~fter ~~1,~~ ~~3~~ 2 and ~~5~~ 4 years, there were no significant treatment effects on porosity ($P>0.05$; Fig. 2a-c) compared to ~~8 and 11 years~~~~8~~7- and 1~~1~~0-years post- conversion (respectively $P=0.029$ and $P=0.002$; Fig. 2d, e). After ~~8~~ 7 years, the porosity ~~of~~ in the grassland soil was greater than ~~the porosity of~~in bare-fallow ~~and~~ or arable soils which were similar (Fig. 2d). However, after ~~11~~ 10 years, porosity increased in the presence of plants according to the ranking; bare-fallow < arable < grassland (Fig. 2e). No significant change in $\log_{10}$ total porosity was observed in the continuous bare-fallow soil over the ~~11~~ 10 years (slope = 0.026, $t = 0.104$, $p = 0.917$) (Supplementary Fig. 1). However, total porosity in ~~both~~ soils converted to arable (slope = 0.713, $t = 3.4$, $p = 0.0014$) and grassland (slope = 0.41~~07~~, $t = 3.7$, $p < 0.001$) managements increased over the same period (Fig. 1f). ANCOVA comparing the arable and grassland soils identified a significant time response in $\log_{10}$ porosity ($F_{1,86} = 31.0$, $p < 0.001$) but no significant difference in the rates of change (slope) in total porosity ($F_{1,86} = 0.8$, $p = 0.364$). The resulting equal slopes model identified a significant difference in the adjusted mean $\log_{10}$ total porosity of each treatment ($F_{1,87} = 23.5$,

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$p < 0.001$) with grassland accumulating significantly greater $\log_{10}$ porosity (0.985 ± 0.025, equivalent to 9.66 ± 1.05%, adjusted mean ± standard error of the mean) than arable soil (0.848 ± 0.021, equivalent to 7.05 ± 1.05%)

Pore size distribution

~~After 1 year1-year~~ ~~*post*~~Before the~~-~~ conversion (in 2008), there was no significant treatment effect on the cumulative pore size distribution (*P>0.05*; Fig. 3a). Between ~~3~~2- to ~~11~~10-~~-~~years post~~-~~ conversion, there was a significant diameter by treatment interaction with respect to the cumulative pore size distribution (~~3~~ 2 and ~~8~~ 7 years: *P<0.001*; 4~~5~~ and ~~11~~ 10 years: *P<0.05*; Fig. 3b-e). After ~~3~~ 2 years post~~-~~ conversion, there was a greater proportion of smaller pores under bare-fallow and arable treatments than grassland: for bare-fallow and arable, approximately 50% of pores were smaller than 3.56 µm and 70% of pores smaller than 5.97 µm compared to grassland where 50% of pores were smaller than 5.97 µm and 70% smaller than 14.9 µm. Moreover, the proportion of pores larger than 42 µm was greater under grassland (13% of pores) than bare-fallow and arable (respectively 1% and 2% of pores; Fig. 3b). After ~~5~~ 4 years, this trend was not apparent: the difference between grassland compared to bare-fallow and arable was less significant than after ~~3~~ 2 years. The proportion of pores smaller the 9.26 µm was greater under bare-fallow and arable compared to grassland but the proportion of pores larger than 42 µm was not significant between all treatments (Fig. 3c). After ~~8~~ 7 years, the trend observed after ~~3~~ 2 years was more apparent: the proportion of pore sizes smaller than 14.9 µm was greater ranking from bare-fallow > arable > grassland and the proportion of pore sizes over 42 µm was greater under arable and grassland (respectively 7% and 10% of pores) than bare-fallow (2% of pores; Fig. 3d). After ~~11~~ 10 years post-conversion, this trend was also observed, but only for pores smaller than 9.26 µm, where the proportion of pores ~~was~~ followed the ranking~~; from~~ bare-fallow > arable > grassland (Fig.

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3e). Beyond this pore size, the proportion of pore sizes was not significantly different between bare-fallow and arable. The proportion of pore sizes greater than 42 µm was highest under grassland (15% of pores) than bare-fallow and arable (respectively 4% and 2% of pores; Fig. 3e).

The general trend in all three treatments was a shift to a more ~~equitable~~ even distributions of pore sizes, manifest as a decrease in *G* over time (Fig. 3f). ANCOVA indicated equal rates of change of *G* between treatments ($F_{2,129} = 0.18$, $p = 0.834$). Using an equal slopes model, there was a significant effect of land management upon *G* ($F_{2,131} = 9.1$, $p < 0.001$) with grassland having a significantly lower adjusted mean *G* (0.420 ± 0.033) than either bare-fallow (adjusted mean $G = 0.566 \pm 0.028$; $t = 3.6$, $p < 0.001$) or arable (adjusted mean $G = 0.571 \pm 0.024$; $t = 3.7$, $p < 0.001$). There was no significant difference in adjusted mean *G* between arable and bare-fallow ($t = 0.13$, $p = 0.900$).

Pore connectivity

Before the conversion (in 2008) and ~~A~~after ~~1 and 8 years~~~~1- and 8~~7-years post~~-~~ conversion, there was no significant pore diameter by treatment interaction with regards to pore connectivity ($P= 0.05$; Fig. 4a, d). However, there was a significant pore diameter by treatment interaction after ~~3~~2-, ~~5~~ 4- and ~~11~~ 10-years (with ~~3~~ 2- and ~~11~~ 10-years: $P<0.001$; ~~5~~ 4-years: $P<0.05$; Fig. 4b, c, e). After ~~3~~ 2- and ~~5~~ 4- years, the difference was significant only for the pore sizes smaller than 3.56 µm. After ~~3~~ 2-years, pore connectivity was greater ranking from bare-fallow > grassland > arable (Fig. 4b) and after ~~5~~ 4-years, pore connectivity was greater under arable and grassland than bare-fallow (Fig. 4c). After ~~11~~ 10-years post-conversion, the same trend as after ~~5~~ 4-years was shown with a greater difference in the values (Fig. 4e). There was no significant trend in square root transformed connected porosity (Supplementary Fig. 2) in bare-fallow (slope = -0.00023, $t = 0.051$, $p = 0.959$). However,

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both arable (slope = 0.017, $t = 4.0$, $p = 0.0002$) and grassland (slope = 0.022, $t = 5.3$, $p < 0.0001$) showed increases in connected porosity with time. ANCOVA comparing arable and grassland indicated a significant influence of time *post-*conversion upon square root transformed connected porosity ($F_{1,85} = 43.9$, $p < 0.001$) but no significant heterogeneity of slopes ($F_{1,85} = 0.98$, $p = 0.326$). Using an equal slopes model, a significant effect of management was detected ($F_{1,85} = 4.4$, $p = 0.039$): grassland was associated with greater connected porosity (0.048 ± 0.0002%, adjusted mean ± standard error) than arable (0.011 ± 0.0002%).

Pore surface density

Before the conversion (in 2008) and ~~After~~ Aat ~~1~~-~~, 5 and 8 years~~54- and ~~8~~7-years *post-*conversion, there was no significant pore diameter by treatment interaction with ~~respect~~ regards to pore surface density (*$P>0.05$*; Fig. 5a, c, d). There was a significant diameter by treatment interaction after ~~3~~ 2- and ~~11~~ 10-years (respectively *$P<0.05$* and *$P<0.001$*; Fig. 5b, e). After ~~3~~ 2-years, the difference in pore surface density was greater ranking from bare-fallow > arable > grassland for the pore sizes equal to 1.86 µm, and the difference between arable and grassland was not significant for the pore sizes equal to 3.56 µm. Beyond this pore size, there was no significant difference between treatments (Fig. 5b). ~~After 11~~~~Eleven~~Ten years *post-*conversion, pore surface density was greater ranking from grassland > arable > bare-fallow, for all pore sizes smaller than 14.9 µm, there was no significant difference beyond this pore size (Fig. 5e). Years post-conversion and pore diameter were both used as covariates in ANCOVA analysis of pore surface area. Accounting for these two covariates, an equal slopes model identified a significant effect of land management upon pore surface area ($F_{2,2020} = 9.1$, $p < 0.001$). *Post hoc* pair-wise comparison of adjusted means indicated that grassland supported a greater pore surface area (0.00784 ± 0.000493 $\mu m^2\ \mu m^{-3}$) than

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either arable (0.00617 ± 0.000452 μm^2 μm^{-3}; difference = 0.00167, $t = 3.4$, $p = 0.001$) or bare-fallow (0.00593 ± 0.000443 μm^2 μm^{-3}; difference = 0.00191, $t = 3.9$, $p < 0.001$). There was no significant difference in pore surface area between arable and bare-fallow (difference = 0.000240, $t = 0.495$, $p = 0.621$).

Discussion

The ~~conversion~~ plots studied here were derived from long-term bare-fallow management converted to arable and grassland. A ~~L~~lack of a significant treatment effect on porosity until ~~8~~ 7-years post~~-~~ conversion suggests that modification of micro-porosity ~~at this scale~~ takes several years (Fig. 2). ~~Despite~~Another study on the same soil ~~realised after~~ 2 and 4 years post conversion found ~~that the~~some recovery of meso-faunal populations after ~~3~~ 2-years of conversion and an increase ~~of~~ in soil organic matter and microbial abundance after ~~5~~ 4- and ~~3~~ 2-years respectively (Hirsch et al., 2017). However, our study showed that~~,~~ ~~recovery~~ development of micro~~n~~-scale porosity apparently takes longer~~changes more slowly~~. This might be related to carbon cycling processes which are modified by the microbial communities and plants (*via* decomposition of organic matter and rhizodeposition). This is likely to affect soil structure at the micro-scale, but not instantaneously. ~~Greater~~ Increased pore formation under grassland compared to arable is consistent with a previous study, which showed greater resistance to, and ~~recovery~~ development from physical stresses of soil structure from grassland (Gregory et al., 2009). They posited that the greater proportion of organic matter enhanced the elastic recovery of ~~the~~ soil structure (Gregory et al., 2009). Pore size distributions showed a more rapid response to altered management than ~~total~~ porosity for the grassland treatment: after only ~~3~~ 2-years of conversion (in 2010), a greater diversity of pore sizes was observed ~~under the grassland treatment~~, and this trend was also recorded in the data after ~~8~~ 7- and ~~11~~ 10-years (Fig. 3). The Gini-coefficient indicated that

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soil converted to grassland established a more ~~equitable~~ even distribution of pore sizes than the other treatments, meaning that grassland treatment had a greater diversity of pores after ~~3~~2-, ~~8~~ 7- and ~~11~~ 10-years post- conversion (Supplementary Fig. 2) leading to enhanced functionality. This increase in pore size diversity might be due to the increase of presence of plants, active organisms and organic matter (Hirsch et al., 2017) as well as the ~~cessation~~ absence of tillage. A study ~~looking at~~focused on the soil organic carbon on the same experiment ~~plot~~ found that the conversion of the bare-fallow soil to grassland led to an increase of soil organic carbon (+46 %) ~~after~~ 7-years post conversion (Jensen et al. 2020a). There is no data regarding the conversion from bare-fallow to arable. Thus, the increase of organic matter in the converted soil ~~might have~~may play a role in the more homogenous distribution of the pore sizes. ~~Plants increase aggregation through root action and exudation (Chan and Heenan, 1996; Haynes and Beare, 1997) and they can also break down aggregates (Chan and Heenan, 1996; Materechera et al., 1994) by growing through existing pores in the aggregates and disrupting them. However, plants enmesh soil particles forming large aggregates (Tisdall and Oades, 1982), and release mucilage that binds soil particles with organic matter which in turn stabilises new aggregates and pores (Bronick and Lal, 2005; Chenu et al., 2000).~~ Indeed, in a silty clay soil,~~The addition of organic matter can increase the number of "transmission" (50 – 500 µm) and "storage" (0.5 – 50 µm) pores and decrease the prevalence of macro-pores (> 500 µm) increasing overall functionality. Thus the action of plants and~~ the ~~increase~~ greater ~~of~~ organic matter content increases the proportion of pores between 0.5 to 500 µm (Metzger and Yaron, 1987; Watts and Dexter, 1997) leading to a more equitable distribution of pore sizes, i.e. a greater diversity of pore sizes. The greater diversity of pore sizes under soil converted to grassland was consistent with a previous study describing the long-term effect of grassland management on the same field experiment (Bacq-Labreuil et al., 2018). After ~~5~~ 4-years ~~post-conversion,~~ the pore size distribution did

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not follow this trend (Fig. 3c), which could be due to ~~the~~ weather conditions prior sampling in that year. Indeed, 2008 and 2012 (~~1~~ at the start and ~~5~~ 4-years post~~-~~ conversion respectively) were the wettest years during the experimental period (Supplementary Fig. 3). In the presence of water, clay particles can swell, and the compression of entrapped air in capillary pores can disrupt the pore architecture, ~~and in turn, aggregation which might~~and affect the pore size distribution (Denef et al., 2001; Grant and Dexter, 1990). ~~Therefore, the p~~Pore networks ~~become~~ are re-structured upon re-wetting due to the nature of soil particles. Changes in pore size between ~~3~~2-, ~~5~~ 4- and ~~8~~ 7-years post-conversion raises the question of the dynamics of this mechanism. The pore size distribution may have had a heterogeneous response over time due to the impact of the wet year ~~5~~ 4-years post-conversion, which shows the rapid ~~recovery regeneration~~development of the pore size distribution after a sustained wet period compared to the impact of ~~plant growth~~agricultural practices (Supplementary Fig. 3). For the ~~plot converted to~~ arable treatment, this trend was not observed even ~~after~~ 10-years post-conversion, which might be due to the associated tillage practices ~~on the arable plots~~.

For pore connectivity, the ~~magnitude of the plant~~conversion to grassland ~~effect after 3 and 5 years was~~ had a small effect after 2- and 4-years compared to after ~~11~~ 10-years post-conversion (Fig. 4 b, c, e). However, pore connectivity data after ~~11~~ 10-years post-conversion, for both arable and grassland converted soils (Fig. 4e), suggested the pore network was less connected compared to ~~a previous study (~~Bacq-Labreuil et al., (2018)~~.;~~ This ~~indicating~~ indicated that a longer time may be required to ~~recover~~ develop the connectivity of a pore network than ~~total~~ the overall porosity. Increased connectivity of pores promotes water, gasses and nutrient flows within the pore structure (Dexter, 1988; Tisdall and Oades, 1982). Therefore, ~~the~~ subtle increases in ~~the~~ pore connectivity might increase ~~the~~ water, gas and nutrient ~~movement~~ flux within the soil. As well as the pore connectivity, the pore surface density was significantly i~~nfluenced by the presence of plants~~ncreased in the grassland and

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arable ~~11~~ 10-years post-conversion (Fig. 5e). Our results are congruent with Bacq-Labreuil et al. (2018), which showed ~~plants~~ grassland managed consistently for over 200 years has an increased pore surface density compared to arable and bare-fallow soils~~,~~ *i.e*. the pore-solid interface which led to a greater surface of the pore where micro-organisms and plant roots can colonise and water films can develop. A greater pore surface density in the converted plot means that the grassland and the arable have a more complex~~ed~~ structure of pores than the bare-fallow soil (Müller et al. 2019). The greater surface density for the grassland compared to the arable might be induced by the greater SOC content and the absence of tillage for this treatment (Hirsch et al. 2017; Jensen et al. 2020a). This can lead to the formation of new habitats and niches which can be beneficial for microbial community diversity (Holden, 2011). The greater pore surface area might increase water and nutrient uptake by the microbial community and plants.

This study suggests that ~~the overall impact of the~~conversion ~~from~~of degraded bare-fallow soil to ~~presence of plants~~grassland requires at least ~~10~~ 10-years after conversion before being effective in terms of significant ~~recovery~~ development of soil structure at aggregate scale, as assessed by ~~total~~ the overall and connected porosity. Moreover, the conversion from bare-fallow to arable had no significant effect on soil structural properties after a decade. In general, the recovery of meso-fauna and organic matter (Griffiths et al., 2000; Hirsch et al., 2017) were more rapid than the recovery of soil structure (Gregory et al., 2009). The pore size distribution was the only characteristic which was more sensitive to ~~changes~~ changes induced by~~such as~~ wetting and drying cycles and ~~,~~living organisms.

Our first hypothesis was supported since porosity, pore size diversity, pore connectivity, and pore surface area density were all enhanced in grassland soil. Moreover, the conversion to arable management did not affect soil structural development significantly. The conversion to

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grassland increased the range of pore sizes after 2--years, consistent with our second hypothesis. However, all other Minkowski functions (porosity, pore connectivity and pore surface area density) responded to change more slowly. The mechanisms behind the development of pore sizes appeared to be dynamic and possibly dependent upon weather conditions before sampling. Apart from the pore size distribution, the magnitude of the grassland effects on all other Minkowski functions was lower than the difference observed after a minimum of 50 years of management (Bacq-Labreuil et al., 2018). In this study, the effect of grassland upon porosity and pore connectivity were two-fold greater than bare--fallow management. Here, the difference was significant but not as major. Bare--fallow soil management for this long period (> 50 years) is detrimental to both physical and biological soil properties and the development of the soil structure after this requires more than 10 years after the conversion the grassland

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Conclusions

The ~~S~~soil structural ~~recovery~~ development of ~~the~~ a ~~compromised~~ degraded silt-clay loam soil, as quantified by micro-scale topological metrics ~~related to detail capture at 1.5 µm resolution~~, requires at least ~~10~~ 10-years of a ~~new~~ grassland management before showing any significant effects ~~of the presence of plants~~. ~~Our first hypothesis was supported since porosity, pore sizethe diversity of pore sizes, pore connectivity, and pore surface area density were all enhanced by the presence of plantsin the grassland soil. Moreover, the conversion to arable management did not affect the soil soil structurale development significantly. The presence of plantsThe conversion to grassland increased the range of pore sizes after only 2 years, post-conversion consistent with our second hypothesis. However, all other Minkowski functions (such as porosity, pore connectivity and pore surface area density) showed recovery an increase 7 to 11 years post-conversion to grasslandresponded to change more slowly. The~~

~~mechanisms behind the recovery development of pore sizes appeared to be dynamic and possibly dependent upon weather conditions before sampling. Apart from the pore size distribution, the magnitude of the plant the grassland effects on all other Minkowski functions was lower than the difference observed after a minimum of 50 years of management (Bacq-Labreuil et al., 2018). In this study, the effect of grassland upon porosity and pore connectivity were two-fold greater than bare fallow management. Here, the difference was significant but not as major , which means that soil structure requires more time to completely establish micro-structure after being converted to grassland or arable. Demonstrably, bBare fallow soil management for this extreme long period (> 50 years) is detrimental to both physical and biological soil properties and the recovery development of the soil structure after this requires more than 10 years after the conversion the grassland~~.

These~~is~~ observations raise~~s~~ the question on the application to certain managements in agricultural practices. For example, instead of applying a bare-fallow treatment in a crop rotation, it would be beneficial for the soil characteristics to apply a vegetation cover *i.e.* cover crops which increase~~s~~ ~~the~~ organic matter inputs and ~~impact~~ influence ~~on~~ soil structure, leading to a ‘conditioning’ of soil physical and biological characteristics for the next crop. This would prevent ~~the~~ further degradation of ~~the~~ soil and ~~also~~ help ~~for~~ its ~~recovery~~ development if the soil characteristics were compromised. Moreover, the ~~recovery~~ development of ~~the~~ soil structure is apparently a long ~~process,~~process in the context of current agricultural practices and perceived imperatives. ~~;~~ ~~thus~~T~~t~~hus, a modification of cropping managements ~~might~~ should be anticipated to require some time before the observation of beneficial impacts on soil structural dynamics. ~~Therefore, this should be accounted for the future research and conclusions.~~

Acknowledgements

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Figure 1: Representative 2D X-ray attenuation images of soils subjected to different forms of management over ~~10~~ 10-years ~~after post-~~following conversion to ~~these~~ each treatment~~s~~. Base resolution is 40 µm; (P) pores are the darker shades and (S) soil matrix are the lighter shades which relate to the attenuation of the X-ray (a sharpening algorithm has been passed over these images to increase contrast of features); (a, d, g, j, m) bare-fallow; (b, e, h, k, n) arable; and (c, f, i, l, o) grassland.

Figure 2: Porosity (based on resolution of 1.5 µm) in ~~relation to~~ bare fallow, arable and grassland soils ~~in regards to~~in the years ~~post-conversion~~following: (a) ~~1~~ 0-year; (b) ~~3~~ 2-years; (c) ~~5~~ 4-years; (d) ~~8~~ 7-years; (e) ~~11~~ 10-years. Bar~~-charts~~s ~~were~~ are means ($n = 9$) expressed as the percentage of pores relative to the total volume, whiskers denote pooled standard errors. (f) Porosity evolution ~~from 1~~0- to ~~11~~10--years *post-* conversion, ~~with the points were~~data points represent means ($n = 9$), whiskers denote pooled standard errors for clarity and trend lines ~~were~~ are linear regressions.

Figure 3: Cumulative pore size normalized to the total ~~pore~~ volume in relation to bare-fallow, arable and grassland in ~~regards to~~ the years ~~post-~~ following conversion: (a) ~~1~~ 0-year; (b) ~~3~~ 2-years; (c) ~~5~~ 4-years; (d) ~~8~~ 7-years; (e) ~~11~~ 10-years. Data ~~P~~points indicate means (n=9), whiskers denote pooled standard errors. (f) Gini coefficient ~~evolution~~ development ~~from 1~~0- to ~~11~~10--years post- conversion, ~~with the~~data points ~~were~~ represent means ($n = 9$), whiskers denote pooled standard errors for clarity and trend lines ~~were~~ are linear regressions.

Figure 4: Pore connectivity normalized to total volume in continuous bare fallow, arable and grassland soils ~~in regards to the years post-~~following conversion: (a) ~~1~~ 0-year; (b) ~~3~~ 2-years;

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(c) ~~5~~ 4-years; (d) ~~8~~ 7-years; (e) ~~11~~ 10-years. ~~Points~~ Data points indicate means (n=9), whiskers denote pooled standard errors.

Figure 5: Pore ~~S~~surface density in ~~relation to~~ continuous bare fallow, arable and grassland soils ~~in regards to the years post-~~following conversion: (a) ~~1~~ 0-year; (b) ~~3~~ 2-years; (c) ~~5~~ 4-years; (d) ~~8~~ 7-years; (e) ~~11~~ 10-years. Data ~~P~~points indicate means ($n = 9$), whiskers denote pooled standard errors.

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