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Hot-water extractable C and N as indicators for 4p1000 goals in a temperate-climate long-term field experiment: A case study from Hungary

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ABSTRACT

Soil organic matter (SOM) consists of various labile and stable fractions, which are differently influenced by agricultural activities and land-use change. This study, aimed at investigating the feasibility of achieving 4p1000 goals on conventionally tilled plough-land, was carried out in a 42 years-old long-term field experiment (LTE), in which the effect of three farmyard manure (FYM) doses and the FYM equivalent NPK mineral fertiliser rates can be examined, together with that of mineral fertiliser with and without ploughed-in plant residues. The soil total organic carbon (SOC) and nitrogen (SN) and the labile hot-water soluble C (HWC) and N (HWN) fractions were determined (0–0.3 m) and used as indicators. The parameters of the tilled area were compared with those of a grassland area with similar characteristics. The suitability of these parameters for use as soil quality indicators (SQI) was also examined in terms of soil fertility. The results showed that the most sensitive fraction for the detection of treatment effects were HWC and HWN. Increases in these labile organic fractions were significantly related to the gains of SOC stocks. Based on the close correlation between these factors and both SOC and crop yield it is recommended that they should be used as indicators for the prediction of changes in SOC and in studies on soil fertility or soil quality. The initial C stock of 40.46 Mg ha^{-1} became 40.27–47.05 Mg ha^{-1} on the tilled soil after four decades, while on the grassland it rose to 69.31 Mg ha^{-1}. The carbon sequestration rate (CSR) in the various fertilisation systems exhibited the following order: plant residue incorporation >FYM addition > mineral fertilisation (0.147–0.156, 0.101–148 and –0.021–0.065 Mg ha^{-1} yr^{-1}, respectively), while an outstandingly high value of 0.687 Mg ha^{-1} yr^{-1} was recorded for the grassland. Despite the fact that the carbon sequestration potential (CSP) remained high (55.36–62.48%), the achievement of 4 per 1000 aims can only be ensured in treatments involving high rates of organic matter and only in the short term.

1. Introduction

Soil organic matter (SOM) is the largest reservoir of terrestrial organic carbon (C), is quantitatively balanced by equivalent respiration and C inputs. Since soil C quantity exceeds that of the atmosphere and vegetation, even a tiny shift in global SOM stocks can have a marked impact on the atmospheric CO_2 level (Lefèvre et al., 2017). The Kyoto Protocol made it possible for biospheric C sinks and sources to be ratified to comply with emission reduction targets. Numerous anthropogenic activities that may increase soil organic carbon (SOC) were listed by the Intergovernmental Panel on Climate Change, and the management of cultivated land and grasslands was proposed as a viable way of decarbonization (Soussana et al., 2004). The agricultural sector is responsible for a third of greenhouse gases (GHG) emissions (Vermeulen et al., 2012). Since ill-advised LULUCF (land-use, land-cover changes and forestry) sector changes are the 2nd highest anthropogenic C sources, EU directives prescribing reduction in CO_2 emissions by 2050, and changing the earlier 80% target value (Eurostat, 2019) to 100%, could partially be achieved by agriculture via an increase in the C-sequestration of the soils. By the adapted, new, stricter regulations, the objectives of the

Abbreviations: BMP, best mamagement practice; C_f, fine fraction, silt and clay particle-sized bound carbon; C_L, labile carbon; C_{NL}, non-labile carbon; CSP, carbon sequestration potential; CSR, carbon sequestration rate; FYM, farmyarm manure; GHG, greenhouse gas; HWC, hot-water extractable carbon; HWN, hot-water extractable nitrogen; LTE, long-term field experiment; SN, total soil nitrogen; SOC, total soil organic carbon.

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European Green Deal would make Europe the first climate-neutral continent (Clima, 2020).

Changes in SOC stocks are a slow process and may vary greatly as a function of soil type, texture, clay plus silt fraction, vegetation and land-use history; they are positively correlated with precipitation and negatively with temperature (Powlson et al., 2011; Wang et al., 2016). In addition to the indigenous fertility of the soil, both the initial SOC content and its composition are important, as a significant gain in the labile fractions may occur during crop production (Janzen et al., 1998; Soussana et al., 2004).

One of the most frequently used indicators of agronomic sustainability and soil quality (SQI) is SOM (or SOC). Together with pH, texture and clay content, and available P and K, these provide a picture of the physical, chemical and biological properties of the soil (Bongiorno et al., 2019; Costantini et al., 2016; Juhos et al., 2019; Reeves, 1997; Tóth et al., 2008). SOM is extremely complex, representing a sort of continuum ranging from labile to recalcitrant substances, but from the agricultural point of view it can be divided into a stable and a labile portion. The size of the readily decomposable fraction is greatly influenced by the intensity of soil cultivation, the crop grown and the standard of the cultivation technology. This pool changes dynamically, predicting changes in SOM, and provides energy for soil microorganisms and nutrients to plants, so it is of fundamental importance for the understanding of soil fertility, acting as a 'fine' indicator of soil quality (Körschens et al., 1998; Loveland and Webb, 2003; Trimble et al., 2018).

The "4 per Mille" (4p1000, 4 per 1000) Initiative proposed by the French Minister of Agriculture at the Paris Climate Conference in 2015 suggested that an increase of 0.4% yr^{-1} in SOC (calculated as the quotient of the 2400 Gt SOC stock in the 0–2 m soil layer and the 8.9 Gt fossil C $year^{-1}$ emission) could largely compensate for the increase in atmospheric C (4p1000 Initiative, 2018). The scientific response to this initiative was contradictory. According to Chambers et al. (2016) the 4p1000 goals can only be achieved with radical land-use change coupled with enhanced C-sequestration, with the help of best management practices (BMP) and by adapting cultivation practices to the given growing site. This was confirmed by Minasty et al. (2017), who analysed shallow soil horizons of varying depth (0–0.6 m), where management practices made C sequestration possible. In a temperate climate the majority of SOC was found in the topsoil and, although a significant amount of SOC is also present in deeper soil layers, the study stated, listing several references, that there is 'very little evidence of whether subsoil stocks are changing'. De Vries (2017) corroborated the fact that the impact of land management mainly affected the topsoil over a timescale of decades. For example, in a conventionally tilled long-term experiment (LTE), relatively minor deviations in SOC content below the topsoil were detected between the different treatments (Collier et al., 2017), though after a longer period, depending on the site, the SOC increment could be quite considerable (Poulton et al., 2018). Yu et al. (2017) proved evidence that the increase in the subsoil was only significant in the short term when cropland was converted to grassland. Decomposition slows in the subsoil. This is a complex issue, and it is not taking place because a larger portion of SOM is mineral-associated, but due to the limited microbial activity caused by the lack of deeply-rooted plant species, fine roots, a lower rate of exudation and leaching dissolved organic C from surface litter, resulting in the non-availability of labile C. Thus, a higher residence time favours long-term C preservation (Pries et al., 2018).

On degraded soils, which make up almost half of all agricultural soils, it is hoped to achieve a carbon sequestration rate (CSR) of 0.60 Mg ha^{-1} yr^{-1} using agricultural techniques adapted to the local economic and social conditions. The C saturation of arable soils is low worldwide, so the SOC quantity could be substantially increased (Chen et al., 2018; Di et al., 2017; Wiesmeier et al., 2018). However, changes in SOC only occur during the transition from one steady-state to another, upon the adoption of new practices. Since SOC accumulation follows a non-linear, asymptotic curve, it is of finite duration and magnitude (Soussana et al., 2004). In a Rothamsted LTE, for example, the 4p1000 targets were only reached during the early stages of the experiments even when high manure doses were applied (Poulton et al., 2018).

Many studies deal with grasslands in the context of global warming. These ecosystems have an outstanding role in C storage: their soils have high nutrient and SOM content, thus their restoration has the potential to facilitate the storage of large quantities of C (Soussana et al, 2004). Temperate grasslands account for around 20% of the European land area, and although they are found mainly on nutrient-rich soils, their soil properties can be described by considerable regional variabilities (Tóth et al., 2013). In overall, however, the warmer environment results in a positive response in their productivity (Reeves et al., 2014; Yiang et al., 2019; SOCCR2, 2018). It is important to note, that compared with ploughland, which may easily become C sources in response to global warming (Kay et al., 2019), grasslands suffer less damage and are larger, more flexible C sinks. Their C-absorbing ability and the preservation of their C stocks, though declining even in the case of European grasslands, according to modelling, will be retained (Chang et al., 2016). Model-based estimation of the C cycle of agricultural systems, however, can lead to contrasting results and discrepancies, uncertainties in future C data (SOCCR2, 2018). Through refinement of geochemical models and large-scale, multiple model medianes, more accurate prediction of future C cycles and C fluxes can be made possible (Sándor et al., 2020).

The current study investigated quantitative changes occurring in the C and N fractions following long-term organic and inorganic treatments. The main research objective was to assess the suitability of robust soil biochemical parameters as indicators of BMP with special reference to achieving 4p1000 targets. Other questions were how sensitive individual C and N fractions were as indicators, how suitable they were for the prediction of changes in treatments and in SOM, and which C and N fractions were most closely correlated with the crop yield, the best indicator of soil fertility? This last topic is also of vital importance for ecosystem services and food security, C input and net primary production (NPP).

2. Materials and methods

2.1. Study location and area description

The organic and mineral fertilisation LTE was set up in 1963 on the experimental area of the University of Pannonia, near Keszthely, at 120 m a.s.l. (17°14′23.12″E–46°44′38.06″N) on Ramann's brown forest soil (Eutric Cambisol), representing the characteristic soil type of the lower, drier slopes of moderate and low hills (Fig. 1a), in a random block design with four replications, two crop rotations, each with 15 treatments, on 7 × 14 m (98 m^2) plots, with conventional cultivation based on autumn ploughing (Fig. 1d, e, f). The soil is a loam formed on glacial sandy loess deposits. The initial soil analysis revealed that the ploughed layer was poor in humus (1.5–1.7 H% = 0.87–0.99% total SOC), with low N supplies (0.12–0.13%), 22% clay, 33% silt, poor supplies of available phosphorus (27–60 mg kg^{-1}), moderate supplies of potassium (135–160 mg kg^{-1}) and a neutral pH_{KCl} of 7.1–7.3. The area has probably been cultivated since Roman times (Kenéz, 2014), but certainly for many centuries. The map of Lake Balaton published by Samuel Krieger in 1776 proves that the whole of the Keszthely peninsula was under arable cultivation (Zlinszky and Molnár, 2009), as it is today (Fig. 1b, c). The climate of the region is classified as cold temperate continental with some sub-Mediterranean influences. The 30-year mean annual values (1985–2015) are 10.8 °C (9.1–12.1 °C) for temperature, and 629 mm (331–914 mm) for precipitation. July is the warmest (avg. 25 °C) and sunniest month, followed by August and June. The rainfall distribution is uneven, but it is favourable that most of it falls between May and July and September and November.

Rotation 'A' of the LTE allows the effect of three farmyard manure (FYM) doses and the FYM-equivalent NPK mineral fertiliser rates to be examined, together with FYM + NPK combinations, while in rotation 'B'

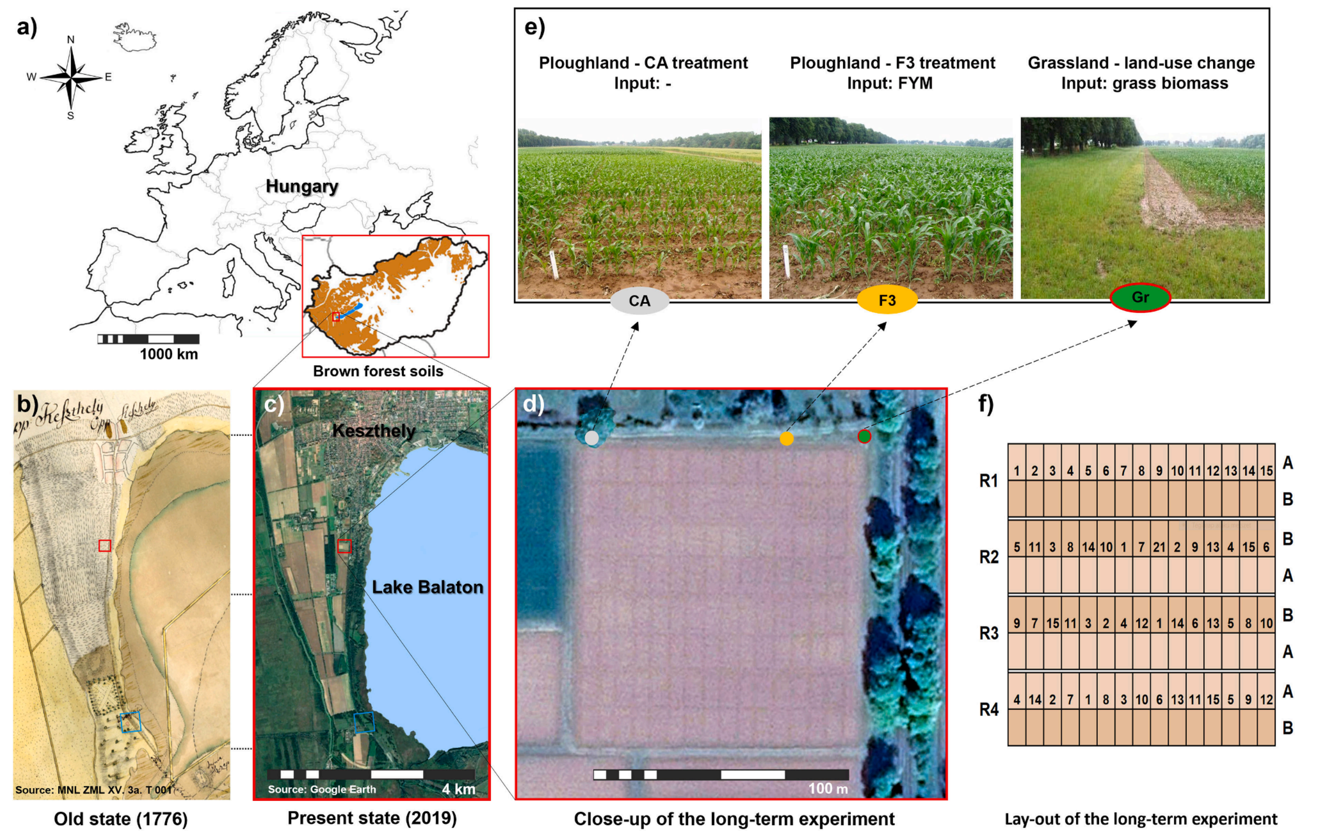

Fig. 1. a) Location of the long-term field experiment (LTE), b, c) its land use history, and d, e, f) presentation.

the effect of mineral fertiliser with and without ploughed-in plant residues can be compared (Table 1). FYM and mineral PK were applied twice in each 5-year period, while N fertiliser was supplied annually. The crop sequence and composition in rotation 'A' was changed three times, while in rotation 'B' they remained the same. Since 2003, crop compositions have been the same, but their sequences differ (Table 1). Rotation 'A' started two years later. The coordination of the rotations, economic considerations (reduction in transportation and FYM

Table 1
Treatments, applied fertilizer doses and crop sequences in rotation 'A' és a 'B' of the long-term field experiment, with the abbreviations of selected treatments (highlighted in bold). CA: rotation 'A' control; F1, F2 and F3: three doses of FYM; M1, M2 and M3: FYM-equivalent mineral fertiliser treatments; CB: rotation 'B' control; M: approx. 4 equ.; Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw. FYM: farmyard manure. 1x – applied once in 5 years, 2x – applied 2 times in 5 years. The mineral fertilizers were applied in the form of ammonium nitrate (27% N), superphosphate (17% P_2O_5) and KCl (60% K_2O).

Rotation 'A' No.		Treatments	Active ingredients kg ha^{-1} year^{-1}	Rotation 'B' No.		Treatments	Active ingredients kg ha^{-1} year^{-1}
1	**CA**	**Control**	$N_0P_0K_0$	1	**CB**	**Control**	$N_0P_0K_0$
2		½ FYM + ½ equivalent NPK	$N_{44}P_{38}K_{49}$	2		½ FYM + ½ equivalent NPK	$N_{44}P_{38}K_{49}$
3		1 FYM (35 t ha^{-1} 5 year^{-1}, 1x)	$N_{44}P_{38}K_{49}$	3		1 FYM (35 t ha^{-1} 5 year^{-1}, 1x)	$N_{44}P_{38}K_{49}$
4	**M1**	**1 equivalent NPK**	$N_{44}P_{38}K_{49}$	4		1 equivalent NPK	$N_{44}P_{38}K_{49}$
5		1 FYM + NPK	$N_{160}P_{154}K_{118}$	5		1 FYM + NPK	$N_{160}P_{154}K_{118}$
6		1 FYM + NP	$N_{160}P_{154}$	6		1 FYM + NP	$N_{160}P_{154}$
7		1 equivalent NPK + NPK	$N_{160}P_{154}K_{118}$	7	**M**	**1 equivalent NPK + NPK**	$N_{172}P_{118}K_{181}$
8	**F1**	**1 FYM** (35 t ha^{-1} 5 year^{-1}, 2x)	$N_{44}P_{38}K_{49}$	8	**Mm**	**1 equivalent NPK + NPK + m**	$N_{172}P_{118}K_{181}$
9		2 equivalent NP	$N_{88}P_{76}$	9		1 equivalent NPK + NP + m	$N_{172}P_{118}$
10	**F2**	**2 FYM** (70 t ha^{-1} 5 year^{-1}, 2x)	$N_{88}P_{76}K_{98}$	10		1 equivalent NPK + NPK + w	$N_{172}P_{118}K_{181}$
11	**M2**	**2 equivalent NPK**	$N_{88}P_{76}K_{98}$	11		1 equivalent NPK + NP + w	$N_{172}P_{118}$
12	**F3**	**3 FYM** (105 t ha^{-1} 5 year^{-1}, 2x)	$N_{132}P_{117}K_{147}$	12	**Mmw**	**1 equivalent NPK + NPK + m + w**	$N_{172}P_{118}K_{181}$
13	**M3**	**3 equivalent NPK**	$N_{132}P_{117}K_{147}$	13		1 equivalent NPK + NP + m + w	$N_{172}P_{118}$
14		1 FYM + 3 equivalent NPK	$N_{176}P_{152}K_{196}$	14		4 equivalent NPK	$N_{176}P_{152}K_{196}$
15		4 equivalent NPK	$N_{176}P_{152}K_{196}$	15		4 equivalent NPK	$N_{176}P_{152}K_{196}$

Crop sequence
1963–1985: sugar beet – maize – maize – winter wheat – red clover1963– potato – winter wheat – winter wheat – maize – maize
1985–2003: sugar beet – maize – maize – winter wheat – winter wheat
2003– potato – maize – maize – winter wheat – winter wheat

application costs) led to a time shift in the rotations as related to each other. Eleven treatments were chosen for this study. CA: the control plot for rotation 'A'; F1, F2 and F3: three doses of FYM; M1, M2 and M3: the FYM-equivalent mineral fertiliser treatments; CB: the control plot for rotation 'B'; M (approx. 4 N equ.); Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw. The detailed description and the applied NPK doses are shown in Table 1.

Since it was set up the experiment has been surrounded by a semi-natural, agriculturally not managed, regularly (two to three times a year) mowed grassy margin. The lack of cultivation allows the regeneration of the ex-arable land to be investigated. The grassland is characterized by the predominance of perennial ryegrass (*Lolium perenne*), while six other monocotyledonous species: orchard grass (*Dactylis glomerata*), yellow foxtail (*Setaria pumila*), green foxtail (*Setaria viridis*), couch grass (*Agropyron repens*), smooth meadow grass (*Poa pratensis*) and cockspur (*Echinochloa crus-galli*), also occur. The number of dicotyledonous species was larger (27), but these were only found sporadically, though they became more frequent at a greater distance from the LTE. Apart from N-fixing clover (*Trifolium*) species, broadleaf and narrowleaf plantain (*Plantago major, P. lanceolata*), common knotgrass (*Polygonum aviculare*) and common yarrow (*Achillea millefolium*) were also characteristically found.

2.2. Soil sampling and laboratory analysis

In the present analysis, representing approximately 40 years of treatments, SOC data were analysed in three years (2002, 2005 and 2007) during the 2000–2010 sampling period, while SN was measured in 2004 and pH, available P and available K in 2004 and 2007. The HWC measurement period covered three successive years, 2004–2005–2006, while parallel HWC and HWN analyses were carried out in June and October 2005. A new sampling period began in 2019. After harvest in October soil samples were taken from the ploughed horizon (0–0.3 m) of the first three replications at 15 points diagonally across the plots and from the grassy margin using an auger. The soil samples were prepared for analysis by removing visible plant residues and rootlets and passing the material through a 2 mm sieve.

The hot-water extractable C and N was determined by the method described by Körschens and Schultz (1999). Briefly, 20 g of soil subsamples from a couple of day old field-moist samples stored in plastic bags in cool place, were weighted into a digestion flask and then boiled in 100 mL distilled water (DW), connected to a reflux condenser, in order to digest the organic matter. After 60 min the reaction was stopped by placing the soil suspensions in cold water. After cooling and centrifuging (for 10 min at 3500 rpm) 3 × 10 mL aliquots were pipetted from the supernatant, 10 mL chromosulphuric acid ($Cr_2K_2O_7{\cdot}H_2O_4S$) was added and the samples were kept at 125 °C for 20 min. The cooled liquids were diluted with 20 mL DW, after which the solutions were titrated with 0.2 M ammonium iron(II) sulphate (Mohr's salt). The amount of acid consumed was used to calculate the amount of oxidised labile organic C.

The Kjeldahl method was used to determine total nitrogen (SN) and, with minor modifications, HWN (Buzás et al., 1988), except that the SN were determined from air-dried, finely-ground soil subsamples, and HWN from the same solution as HWC. Due to the toxic gases, the determination of HWN was done under fume hood. A spatula of reduced iron (Fe(II)), 2 mL sulfuric acid and 4 mL salicylic sulfuric acid were added to 10 mL solution to enhance the violent reaction, after which titanium oxide (TiO_2) catalyst was added to release N during the 380 °C, 60 min digestion. The addition of cc. NaOH led to the formation of ammonium that was transferred to 2% boric acid. The solutions were titrated with 0.01 M hydrochloric acid (HCl).

Total organic C (SOC) was estimated from finely-ground subsamples with Tyurin's method: 10 mL 0.2 M potassium dichromate ($Cr_2K_2O_7$) was added to 0.2 g dry soil. The organic matter was oxidised by 5 min heating over a flame. The cooled solution was diluted to 100 mL with DW. The excess dichromate was determined by titration against 0.2 *N* ammonium iron(II) sulfate (Mohr's salt). The determination of SOC, available phosphorus and potassium (P_a, K_a) using ammonium acetate-lactate extractant, and that of soil pH in neutral salt (KCl) solution were all performed according to Buzás et al. (1988).

2.3. Calculations and statistical analysis

The factor required to convert HWC into the labile C (C_L) fraction can be derived from the slope of the HWC-SOC regression equation (Körschens and Schultz, 1999; Schulz, 1997). This is influenced by the soil texture and the climate (Weigel et al., 2011). Freytag (1987) reported a conversion factor value of 13–16 (avg. 15.02) on the basis of LTEs for clayey loam soils, which gave the strongest correlation ($R^2 = 0.91$), Eq. (1):

$$C_L = HWC \times 15 \tag{1}$$

The non-labile C fraction can be expressed as the difference of SOC and C_L, Eq. (2):

$$C_{NL} = SOC - C_L \tag{2}$$

The SOC and SN stocks [Mg ha^{-1}] can be calculated according to the next, Eq. (3):

$$SOC\ or\ SN\ Stock = C \times BD \times T \tag{3}$$

where C is the SOC or SN concentration [g kg^{-1}], BD is the bulk density [g cm^{-3}], and T is the thickness of the soil layer [dm].

Soil bulk density (BD) was determined according to Blake (1965). Undisturbed samples were collected with calibrated sample cylinders (5 × 5 cm) from the 0.10–0.20 m soil layers and oven-dried at 105 °C for 72 h. As the BD values were not consequent across the treatments, it proved more reliable to apply SOC-BD regression on treatments of pivotal importance (CA, F1, F3, M1, M3, CB, Mmw, rep. 1–3) by substituting SOC values in Eq. (4) if BD values were missing:

$$BD = 2.0893 \times e^{-0.33 \times SOC}\ \left(R^2 = 0.81\right) \tag{4}$$

The saturation (maximum) C storage value ($C_{f\text{-}max}$) of the clay + silt fine particle-sized (C_f, <20 μm) fraction can be calculated from the clay–silt % content using databases. Wiesmeier et al. (2019) collected regression equations between the % clay-silt fraction and the associated C content from various studies, differentiated according to climate, soil type, land use, soil depth and clay minerals. By substituting the clay-silt value (55%) based on similar soils in the temperate zone, the $C_{f\text{-}max}$ ranges for the arable and grassland soils in the LTE were calculated as 18.12–19.06 and 19.46–33.91 g kg^{-1} (average: 18.60 and 27.15 g kg^{-1}), respectively (Fig. S1). Based on the equation given by Hassink (1997) for grassland soils in the temperate and tropical zones this value would be 24.17 g kg^{-1}.

The C saturation deficit can be calculated as the difference between C saturation and C_f. The value of C_f was reported to be 77–85% of the SOC of cultivated topsoil by Wiesmeier et al. (2014) and Di et al. (2017) and 85% by Angers et al. (2011), with a value of 69% for grassland soil (Chen et al., 2018). The average of the 77–85% range, 81%, was used in the present calculations.

The carbon sequestration potential (Mg ha^{-1}) was calculated as follows, Eq. (5):

$$C_{spd} = T \times C_{sd} \times BD \times (100 - ce) \times 10^{-2} \tag{5}$$

where T is the thickness of soil layer (m), C_{sd} is the C saturation deficit (g kg^{-1}), BD is the bulk density (kg m^{-3}), ce is the proportion of rough components (%).

The carbon sequestration potential as a percentage was calculated as follows, Eq. (6):

$$C_{spd}\% = C_{spd} / C_{sat-den} \times 100 \tag{6}$$

where $C_{sat-den}$ is the difference of the measured and the C saturation values.

The Sensitivity Index (SI) indicates those C and/or N fractions that are the most responsive to the treatments and that can be used as the most sensitive indicators, Eq. (7):

$$SI = \left(C\ or\ N_{fraction\ in\ treatment} - C\ or\ N_{fraction\ in\ control}\right) / \left(C\ or\ N_{fraction\ in\ control} \times 100\right) \quad (7)$$

One-way ANOVA was used to determine the effect of the treatments on the C and N fractions and to compare the values of the parameters. Statistical analysis was performed with the Microsoft Office Excel XLSTAT program. Correlations between the parameters were evaluated using Pearson's linear correlation analysis, with LSD and Duncan's test as post hoc tests. A total of 33 samples were analysed in the first three replications, or 36 including the grassland (n = 33, 36). Data for the 1998–2013 period were used for the analysis of crop yields. The potato yield was converted into cereal units (GE t ha^{-1} = Z × 0.25).

3. Results

3.1. Effects of treatments and land-use change on the C and N fractions, yield differences between treatments

The fertilization modes significantly affected the concentration of soil organic carbon (SOC). SOC values statistically differed between control 'A' and the higher organic treatments; FYM (F2, F3), between control 'B' and Mm, Mmw treatments. Total soil-N (SN) content changed in the opposite way to the SOC values and the inputs, gradually declining for the three rates of FYM, while the values only exhibited slight differences in the case of mineral fertilisation. The F1 and M2 treatments gave significantly higher and slightly lower values, respectively. HWC and HWN changed proportionately to the inputs in the treatments. Considerably higher HWC values (1.97 and 1.90 times the treatment mean) were recorded on the grassland soil. Statistically verified clear proof was obtained of the beneficial effect of FYM, mineral fertilisation and the ploughing in of plant residues. HWN proved to be more sensitive, rising to a greater extent. The treatment effects were significant with the exception of F1 for HWC and F1 and M for HWN (Tables 2 and 3). The slope of the linear regression, used as a conversion factor, was 15.35 (R^2 = 0.842), averaged over the two land-use systems, and 11.82 (R^2 = 0.544) in case of the LTE (Fig. S2). The C_{NL} fraction were more advantageously influenced by the organic treatments, with statistical difference only between F3 and M3 treatments. Except for the stable C_{NL} fraction, the deviations between the two land use systems were significant ($p < 0.05$) (Table 2). The C_L ratio was smallest on non-fertilised soils, improving proportionately with the inputs. Values of F3, M1, M2 and Mmw that of the grassland were also significantly higher as compared to the control, and to the tilled soil of LTE, respectively (Fig. 2). The relationship between the N fertiliser rates and C_L was very close in the case of treatments with FYM and plant residues, while only a moderate correlation was observed for the mineral fertiliser treatments. The N rates explained 90.0, 86.0 and 31.7% of the C_L values (Fig. S3).

The SOC density changed proportionately to the inputs in the treatments, ranging between 40.27 and 47.05 Mg ha^{-1} (Table 3). Compared to the initial mean value of 40.46 (37.93–42.99) Mg SOC ha^{-1}, there was a slight reduction in SOC on the unfertilised plots and in the M1 treatment, while no change was observed in the M2 treatment and a considerable increase was recorded for the M3 treatment. The ploughing in of plant residues had a positive effect, similar to that of FYM. The SOC stock of the grassland soil was considerably higher than the mean of fertilised treatments, and exhibited the greatest C gain as a consequence of land-use change. The difference was significant ($p < 0.05$) compared with both FYM, which resulted in a substantial C increment, and mineral fertiliser, which was only sufficient to maintain the initial SOC level. Compared with mineral fertilisation, substantial increases in C-sequestration were caused by both the FYM doses and by the incorporation of plant residues combined with mineral fertiliser application (Fig. 3a). As regards N gain it can be stated that in many treatments, despite continual nutrient supplies, the total N stock fluctuated around the low initial value of 5.73 Mg SN ha^{-1} (Fig. 3b). A turning point was observed at higher doses: the N stock began to rise from an N dose of 132 kg ha^{-1} (M3) and reached the initial level at 176 kg N ha^{-1} (M). The values only shifted in a positive direction for lower FYM rates and in the M and Mm treatments, and were 16–18% lower in the control and in plots given mineral fertiliser.

According to the crop yield analysis three main treatment-groups can be separated: 1) the controls, 2) FYM and M1, 3) M2, M3 and M combined with plant residues. All the treatments had a significant effect on the results at all three dose levels compared to the control, and this was also true of mineral fertiliser compared to farmyard manure. Depending on the crop grown, M2 gave the highest values in 40% of the years.

Table 2

Soil organic carbon (SOC), hot-water extractable carbon (HWC), labile carbon (C_L), non-labile carbon (C_{NL}), total soil nitrogen (SN) and hot-water extractable nitrogen (HWN) fractions in the 0–0.3 m soil layer of treatments and grassland. Different lower and upper case superscripts indicate significant differences between treatments and between cropland and grassland, respectively, at $p < 0.05$. Mean ± standard deviation (SD) values. ns: non-significant. CA: rotation 'A' control; F1, F2 and F3: three doses of FYM; M1, M2 and M3: FYM-equivalent mineral fertiliser treatments; CB: rotation 'B' control; M: approx. 4 equ.; Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw.

Treatment	C fractions SOC	HWC	C_L	C_{NL}	N fractions SN	HWN
			mg kg^{-1}			
Cropland			Rotation 'A'			
CA	8321.3 ± 5.4^{cd}	246.46 ± 19.25^{c}	3696.9 ± 327.3^{c}	4624.1 ± 332.7^{ab}	1052.5 ± 31.3^{bc}	18.93 ± 0.02^{b}
F1	9632.2 ± 198.1^{bc}	287.40 ± 3.95^{bc}	4311.0 ± 77.7^{bc}	5321.2 ± 261.9^{a}	1275.0 ± 130.7^{a}	28.87 ± 0.01^{ab}
F2	10142.6 ± 217.6^{b}	341.08 ± 11.60^{b}	5116.1 ± 243.2^{b}	5026.5 ± 400.4^{a}	1215.8 ± 152.9^{ab}	31.80 ± 0.01^{a}
F3	10561.6 ± 415.7^{a}	411.76 ± 14.53^{a}	6176.5 ± 562.8^{a}	4385.2 ± 611.9^{b}	1103.3 ± 74.9^{bc}	37.09 ± 0.02^{a}
M1	8559.6 ± 578.9^{d}	320.43 ± 42.66^{b}	4806.5 ± 639.9^{b}	3752.8 ± 278.3^{ab}	1038.3 ± 82.2^{bc}	30.88 ± 0.01^{a}
M2	8811.6 ± 109.8^{bcd}	353.60 ± 40.50^{ab}	5304.0 ± 1 012.1^{ab}	3507.7 ± 1104.7^{ab}	1021.7 ± 71.4^{c}	33.00 ± 0.02^{a}
M3	9639.1 ± 273.4^{bc}	353.64 ± 30.48^{ab}	5304.7 ± 533.9^{ab}	4334.5 ± 503.7^{a}	1085.0 ± 31.2^{bc}	37.47 ± 0.03^{a}
			Rotation 'B'			
CB	8496.9 ± 548.7^{b}	244.73 ± 8.59^{c}	3670.9 ± 279.6^{c}	4826.0 ± 373.8^{a}	1165.0 ± 49.5^{a}	28.26 ± 0.01^{b}
M	9351.9 ± 386.1^{b}	312.80 ± 12.33^{b}	4692.0 ± 517.0^{b}	4659.9 ± 681.5^{a}	1260.0 ± 98.9^{a}	35.75 ± 0.02^{ab}
Mm	10463.2 ± 585.0^{a}	357.34 ± 12.13^{ab}	5360.0 ± 363.8^{ab}	5103.1 ± 817.6^{a}	1230.0 ± 98.9^{a}	40.59 ± 0.03^{a}
Mmw	10722.9 ± 246.5^{a}	384.58 ± 16.43^{a}	5768.7 ± 493.0^{a}	5059.0 ± 728.1^{a}	1240.1 ± 14.1^{a}	44.36 ± 0.02^{a}
Grassland						
Gr	15100 ± 200.0^{A}	647.20 ± 44.25^{A}	9708.0 ± 1149.6^{A}	5392.0 ± 1349.6 ^{ns}	1829.7 ± 24.2^{A}	63.58 ± 0.09^{A}

Table 3
Soil organic carbon (SOC), hot-water extractable carbon (HWC), labile carbon (C_L), non-labile carbon (C_{NL}), total soil nitrogen (SN) and hot-water extractable nitrogen (HWN) stocks in the 0–0.3 m soil layer of treatments and grassland. Different lower and upper case superscripts indicate significant differences between treatments and between cropland and grassland, respectively, at $p < 0.05$. Mean ± standard deviation (SD) values. ns: non-significant. CA: rotation 'A' control; F1, F2 and F3: three doses of FYM; M1, M2 and M3: FYM-equivalent mineral fertiliser treatments; CB: rotation 'B' control; M: approx. 4 equ.; Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw.

	C fractions					N fractions
Treatment	SOC	HWC	C_L	C_{NL}	SN	HWN
			Mg ha^{-1}			
Cropland			Rotation 'A'			
CA	40.27 ± 0.03^{d}	1.19 ± 0.11^{c}	17.89 ± 1.58^{c}	22.38 ± 1.61^{ab}	5.09 ± 0.15^{b}	0.092 ± 0.02^{b}
F1	44.96 ± 0.92^{ab}	1.34 ± 0.02^{bc}	20.12 ± 0.36^{bc}	24.84 ± 1.22^{a}	5.95 ± 0.61^{a}	0.135 ± 0.01^{a}
F2	45.49 ± 0.98^{ab}	1.53 ± 0.07^{ab}	22.95 ± 1.09^{ab}	22.54 ± 1.80^{ab}	5.45 ± 0.69^{ab}	0.143 ± 0.01^{a}
F3	47.05 ± 1.85^{a}	1.83 ± 0.17^{a}	27.52 ± 2.51^{a}	19.54 ± 2.73^{bc}	4.92 ± 0.33^{b}	0.165 ± 0.02^{a}
M1	39.55 ± 2.67^{d}	1.48 ± 0.20^{bc}	22.21 ± 2.96^{bc}	17.34 ± 1.29^{c}	4.80 ± 0.38^{b}	0.143 ± 0.01^{a}
M2	41.29 ± 0.51^{cd}	1.66 ± 0.32^{ab}	24.86 ± 4.74^{ab}	16.44 ± 5.18^{c}	4.79 ± 0.33^{b}	0.155 ± 0.02^{a}
M3	43.35 ± 1.23^{bc}	1.59 ± 0.16^{ab}	23.86 ± 2.40^{ab}	19.49 ± 2.26^{bc}	4.88 ± 0.14^{b}	0.169 ± 0.03^{a}
			Rotation 'B'			
CB	40.51 ± 2.62^{b}	1.17 ± 0.09^{b}	17.50 ± 1.23^{b}	23.01 ± 1.78^{a}	5.55 ± 0.24^{a}	0.135 ± 0.01^{b}
M	43.05 ± 1.78^{ab}	1.44 ± 0.16^{a}	21.60 ± 2.38^{a}	21.45 ± 3.14^{a}	5.80 ± 0.46^{a}	0.165 ± 0.02^{ab}
Mm	46.43 ± 2.60^{a}	1.59 ± 0.11^{a}	23.79 ± 1.61^{a}	22.65 ± 3.63^{a}	5.46 ± 0.44^{a}	0.180 ± 0.03^{a}
Mmw	46.80 ± 1.08^{a}	1.64 ± 0.12^{a}	24.65 ± 1.77^{a}	22.08 ± 3.18^{a}	5.41 ± 0.06^{a}	0.194 ± 0.02^{a}
Grassland						
Gr	69.31 ± 0.94^{A}	2.97 ± 0.36^{A}	44.56 ± 5.35^{A}	24.75 ± 6.16 ^{ns}	8.32 ± 0.15^{A}	0.292 ± 0.09^{A}

Compared to M, deviations of plant residue treatments were non-significant (Table 4).

3.2. Changes of C-sequestration over time and achievement of 4 per 1000 targets

Compared to the initial value of 64.94 Mg ha^{-1}, the CSP in the treatments varied between 58.36 and 69.40 after two decades and decreased slightly to 58.35–65.85 Mg ha^{-1} after four decades. The organic treatments were more effective in reducing CSP. An increase in CSP was originally observed for CA and in the inorganic treatments, and later for both controls and only in the M1 treatments (Table 5). When the experiment was set up CSP was 61.61% and a substantial portion of the SOC was missing, while after four decades this value was 55.36–62.48%, with a seemingly similar value of 61.63% for the grassland, but the C saturation value was considerably higher for the grassland soil (180.61 Mg ha^{-1}) than for the ploughed area (105.40 Mg ha^{-1}). The initial 140.15 Mg ha^{-1} CSP of the grassland indicated that the SOM quantity was greatly exhausted, while the later value of 111.30 Mg ha^{-1} indicated an increase.

With the exception of the control and the M1 treatments, CSR exhibited on average a modest value of 0.081 Mg ha^{-1} yr^{-1}. Based on CSR the fertilisation systems could be ranked in the order: plant residue incorporation > FYM application > mineral fertilisation (0.147–0.156, 0.101–0.148 and –0.021–0.065 Mg ha^{-1} yr^{-1}) (Table 5). A treatment effect was still not perceptible after two decades, and considerable deviations could be observed for the replications, due to the previous history of the fields and the soil patches. Over the longer period SOC contents proportionate to the inputs were detected, though there was a pronounced drop in the initially intensive CSR (Fig. S4).

The 0.04% increase in SOC was only exceeded in the F1, F2, M and Mmw treatments, but positive deviations could not be observed after 40 years; the intensity of C-sequestration fell by a third (F3) or half (F2, M3, Mmw). Only half (F1, M, Mmw) or 1/3 (F2, F3) of the desired global CSR rate of 0.60 Mg ha^{-1} yr^{-1} was achieved after 20 years, while after 40 years this value was at most ¼ (F3, Mm, Mmw). Despite the regular removal of foliage biomass, the exceedingly high value of 0.687 Mg ha^{-1} yr^{-1} recorded for the grassy margin met the 4p1000 requirements. The high organic doses maximised C-sequestration, while the additional C storage promoted by the application of inorganic nutrients was less pronounced (Table 5).

3.3. C and N ratios

The SOC and SN ratios investigated rose proportionately with the inputs. With the exception of HWC:HWN the control plots gave the lowest values, and significant differences were observed between the treatments. In rotation 'B' the Mm and Mmw treatments resulted in higher ratios than for the control and, with the exception of HWC:HWN, for the M treatment. The SOC:SN ratio was 7.03–9.62 (avg. 7.96%). HWC made up 2.88–3.90% of SOC (avg. 3.44%), and was 4.29% for the grassland. HWN made up 1.79–3.75% of SN (avg. 2.89%), and was 3.51% for the grassy margin. The HWC:HWN ratio varied within 8.29–11.75 (avg.10.10), and was 10.56 for the grassland soil (Table 6).

3.4. Sensitivity of the C and N fractions

The greatest sensitivity index (SI) was found for the HWN and HWC fractions, the sensitivity of which rose almost proportionately with the inputs, with diverse values: the HWC value of the control treatments was around 0, while the HWN value deviated slightly from zero (–19 to +19%). The highest HWN values were recorded for F3 and M3 (46 and 49%) and for the Mm and Mmw treatments (59 and 71%). The trend for SOC was very similar to that of HWC, but the SI values were proportionately smaller. C_{NL} and SN responded similarly, representing the two least sensitive fractions, with very low values, which were often inversely proportional to the doses. The SI values of HWC, HWN, SOC and SN of the grassland soil were significantly higher than those of the treatments, suggesting significant changes (Fig. 4, see Table S1 for details).

3.5. Correlation analysis: relationship with soil properties

HWC and HWN were found to be in strong positive correlation with SOC and SN, and SOC with SN. Correlation analysis on the C and N fractions, the chemical soil parameters and the crop yield data, which give the clearest illustration of soil fertility, and on various ratios revealed how suitable these were for expressing the degree of mineralisation, for replacing the SOC:SN ratio and for use as a potential indicator of mineralisation (Table 7). Based on their correlation with yield data, the correlation coefficients established the order SOC < HWC <

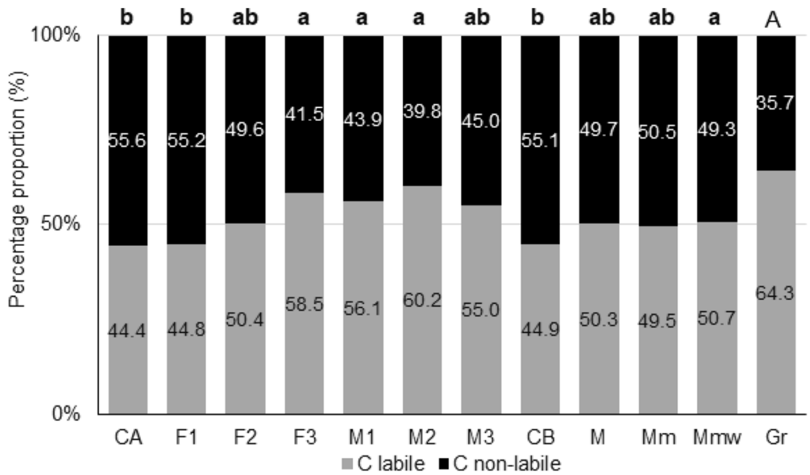

Fig. 2. Percentage proportion of labile and non-labile organic C fractions. Different lower and upper case superscripts indicate significant differences between treatments and between cropland and grassland, respectively, at $p < 0.05$. CA: rotation 'A' control; F1, F2 and F3: three doses of FYM; M1, M2 and M3: FYM-equivalent mineral fertiliser treatments; CB: rotation 'B' control; M: approx. 4 equ.; Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw.

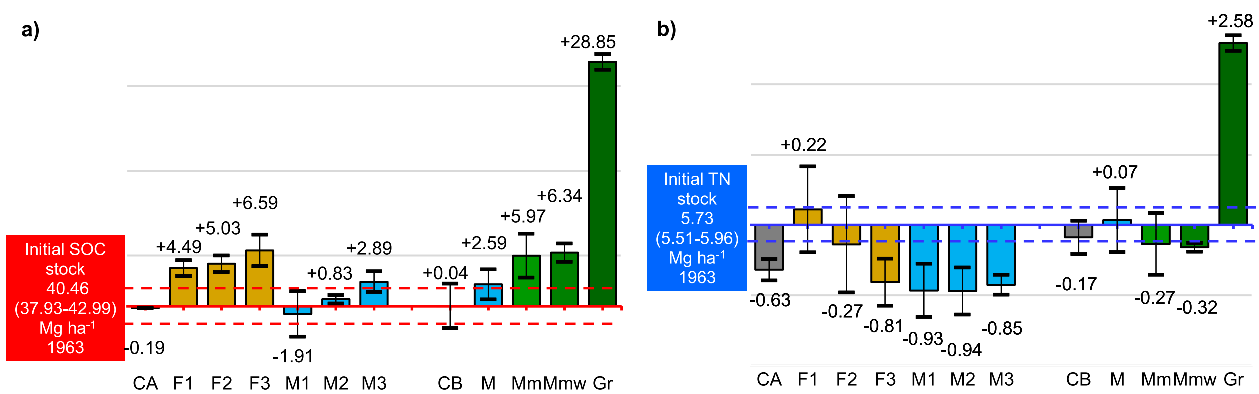

Fig. 3. a) Total soil organic carbon (SOC) and b) total soil nitrogen (SN) gain for the 0–0.3 m soil layer of treatments and grassland. Dashed lines indicate the initial value range. Whiskers represent standard deviation. CA: rotation 'A' control; F1, F2 and F3: three doses of FYM; M1, M2 and M3: FYM-equivalent mineral fertiliser treatments; CB: rotation 'B' control; M: approx. 4 equ.; Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw.

Table 4
Crop yield values in the treatments. Different lower case superscripts indicate significant differences between treatments at $p < 0.05$. Mean ± standard deviation (SD) values are presented. CA: rotation 'A' control; F1, F2 and F3: three doses of FYM; M1, M2 and M3: FYM-equivalent mineral fertiliser treatments; CB: rotation 'B' control; M: approx. 4 equ.; Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw.

Treatment	Yield t ha^{-1}
	Rotation 'A'
CA	3.54 ± 0.34^{d}
F1	4.57 ± 0.38^{c}
F2	4.77 ± 0.18^{c}
F3	5.27 ± 0.07^{b}
M1	5.20 ± 0.21^{b}
M2	6.33 ± 0.44^{a}
M3	6.61 ± 0.42^{a}
	Rotation 'B'
CB	3.61 ± 0.25^{b}
M	7.43 ± 0.32^{a}
Mm	7.07 ± 0.20^{a}
Mmw	7.05 ± 0.33^{a}

HWN, and, for the ratios, SOC:SN < HWC/SOC% < HWN/SN%. Strong correlations were found for available P and for the P_a/SOC% and P_a/HWC% indices and a weaker correlation for the HWN/HWC% ratio.

4. Discussion

4.1. Effect of treatments and land-use change on soil carbon storage

The effects of treatments on SOC were clearly discernible in the LTE after a period of several decades (Hoffmann et al., 2006) (Tables 2 and 3). Significant C gain was recorded in the favourable treatments and the grassland soil, so SOC could be substantially increased. SN, on the other hand, only reached the initial level at higher N rates and in the absence of intensive nutrient uptake (F1, F2) (Fig. 3).

The advantage of FYM over mineral fertiliser and the favourable effect of straw incorporation corroborated the *meta*-analysis of LTEs (Powlson et al., 2011; Reeves, 1997). The grassy margin is regularly mowed, which according to Poeplau and Don (2013) results in the more frequent decay of the leaves and roots and in greater C input from the larger, denser root system. In addition, the lignin, polyphenols and more aromatic compounds under a grassland tend to be recalcitrant to degradation (Dean, 2000), and the roots and their associated microflora stabilize the soil aggregates (Soussana et al., 2004). These effects following the conversion of cropland to grassland also exert a significant influence on SOC accumulation in the subsoil (Yu et al., 2017).

The estimation of C storage requires knowledge of soil type, texture, management history (Janzen et al., 1998) and also of clay minerals (Di et al., 2017). The quantity of C is mainly limited by the rough clay plus fine silt fraction, as the fine fraction bound C (C_f) makes up the bulk of the SOC, and its saturation limits the C-sequestration ability of the soils

Table 5
C sequestration potential (CSP), difference compared to 4p1000 requirements and carbon sequestration rate (CSR) values in the 0–0.3 m soil layer of treatments and grassland after 20 and 40 years. Different lower and upper case superscripts indicate significant differences between treatments and between cropland and grassland, respectively, at $p < 0.05$. ns: non-significant. CA: rotation ‘A’ control; F1, F2 and F3: three doses of FYM; M1, M2 and M3: FYM-equivalent mineral fertiliser treatments; CB: rotation ‘B’ control; M: approx. 4 equ.; Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw.

Treatment	CSP				Compared to 4p1000 aims		CSR		
	20 yrs		40 yrs		20 yrs	40 yrs	20 yrs	20–40 yrs	40 yrs
	C (Mg ha^{-1})	%	C (Mg ha^{-1})	%	C (Mg ha^{-1})		C (Mg ha^{-1} year^{-1})		
Cropland				Rotation ’A’					
CA	63.66^{ab}	60.40^{ab}	65.13^{a}	61.79^{a}	–2.61^{ab}	–7.77^{d}	0.064^{ab}	–0.068^{a}	–0.004^{d}
F1	58.36^{b}	55.37^{b}	60.44^{cd}	57.35^{a}	2.69^{a}	–3.08^{ab}	0.329^{a}	–0.228^{a}	0.101^{ab}
F2	60.55^{ab}	58.30^{ab}	59.91^{cd}	56.84^{b}	0.50^{ab}	–2.55^{ab}	0.219^{ab}	–0.106^{a}	0.113^{ab}
F3	61.19^{ab}	58.06^{ab}	58.35^{d}	55.36^{b}	–0.14^{ab}	–0.99^{a}	0.187^{ab}	0.039^{a}	0.148^{a}
M1	69.40^{a}	65.85^{a}	65.85^{a}	62.48^{b}	–8.35^{b}	–8.49^{d}	–0.223^{b}	0.203^{a}	–0.021^{d}
M2	65.71^{ab}	62.34^{ab}	64.11^{ab}	60.82^{a}	–4.66^{ab}	–6.75^{cd}	–0.039^{ab}	0.057^{a}	0.019^{cd}
M3	62.96^{ab}	59.73^{ab}	62.05^{bc}	58.87^{a}	–1.91^{ab}	–4.69^{bc}	0.099^{ab}	–0.034^{a}	0.065^{bc}
				Rotation ’B’					
CB	60.36^{a}	57.27^{a}	64.89^{a}	61.57^{a}	0.69^{a}	–7.53^{b}	0.229^{a}	–0.228^{b}	0.001^{b}
M	58.56^{a}	55.56^{a}	62.35^{ab}	59.15^{a}	2.49^{a}	–4.99^{ab}	0.319^{a}	–0.255^{ab}	0.064^{ab}
Mm	61.97^{a}	58.79^{a}	58.97^{b}	55.95^{ab}	–0.92^{a}	–1.61^{a}	0.148^{a}	–0.001^{a}	0.147^{a}
Mmw	59.72^{a}	56.66^{a}	58.60^{b}	55.60^{b}	1.33^{a}	–1.24^{a}	0.261^{a}	–0.104^{ab}	0.156^{a}
Grassland									
Gr		111.30^{A}		61.63 ^{ns}			21.27^{A}		0.687^{A}

4per1000 requirements: 44.35 and 48.04 Mg SOC ha^{-1} and SOC increases of + 9.61% and + 18.72% after 20 and 40 years, respectively.

Table 6
C and N ratios for treatments and grassland. Soil organic carbon (SOC), hot-water extractable carbon (HWC), total soil nitrogen (SN) and hot-water extractable nitrogen (HWN). Different lower case superscripts indicate significant differences between treatments at $p < 0.05$. Mean ± standard deviation (SD) values. ns: non-significant. CA: rotation ‘A’ control; F1, F2 and F3: three doses of FYM; M1, M2 and M3: FYM-equivalent mineral fertiliser treatments; CB: rotation ‘B’ control; M: approx. 4 equ.; Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw.

Treatment	HWC/SOC (%)	HWN/SN (%)	HWC:HWN	SOC:SN
Cropland		Rotation ’A’		
CA	2.96 ± 1.33^{b}	1.79 ± 0.41^{d}	11.75 ± 0.19^{a}	7.91 ± 0.24^{b}
F1	2.98 ± 0.43^{b}	2.28 ± 0.29^{cd}	9.98 ± 0.48^{b}	7.62 ± 0.92^{b}
F2	3.36 ± 0.55^{ab}	2.65 ± 0.47^{bc}	10.76 ± 0.72^{ab}	8.44 ± 1.12^{ab}
F3	3.90 ± 0.48^{a}	3.36 ± 0.28^{a}	11.23 ± 1.87^{ab}	9.62 ± 0.98^{a}
M1	3.74 ± 0.79^{a}	2.98 ± 0.23^{ab}	10.42 ± 1.76^{ab}	8.31 ± 1.26^{ab}
M2	4.01 ± 0.15^{a}	3.22 ± 0.12^{ab}	10.69 ± 1.46^{ab}	8.65 ± 0.62^{ab}
M3	3.67 ± 0.22^{ab}	3.46 ± 0.52^{a}	9.49 ± 0.65^{b}	8.89 ± 0.49^{ab}
		Rotation ’B’		
CB	2.88 ± 0.65^{b}	2.34 ± 0.01^{b}	8.67 ± 0.67^{a}	7.03 ± 0.32^{b}
M	3.34 ± 0.62^{ab}	2.80 ± 0.22^{ab}	8.75 ± 0.20^{a}	7.30 ± 0.78^{b}
Mm	3.42 ± 0.41^{ab}	3.15 ± 0.80^{ab}	8.90 ± 0.95^{a}	8.29 ± 0.29^{ab}
Mmw	3.59 ± 0.04^{a}	3.75 ± 0.43^{a}	8.29 ± 0.16^{a}	8.73 ± 0.09^{ab}
Grassland				
Gr	4.29 ± 0.64 ^{ns}	3.51 ± 1.11 ^{ns}	10.56 ± 1.08 ^{ns}	8.33 ± 0.10 ^{ns}

(Chen et al., 2018; Hassink, 1997). Due to continual microbial by-product transfer and the C derived from non-decomposed plant residues, organic compounds accumulate on minerals (Samson et al., 2020). The Keszthely soil is dominated (59%) by illite clay minerals of the transitional, nonexpandable 2:1 type (Csathó et al., 2011). Such clay minerals do not possess a C preservation effect (Kennedy et al., 2014), so the C adsorption and aggregate stability of these soils are lower and more sensitive to tillage (Di et al., 2017). The stabilisation of C is determined not only by the chemical properties of SOM, but also by organo-mineral complexes; the organic matter becomes inaccessible to microbes (Pavithra et al., 2018). Physical protection, however, does not necessarily provide strong stability, as the land use may have a destabilising effect. The stable (C_f) CSP deficit was reported to be over 50% globally, which could be robustly reduced by means of C input (Di et al., 2017; Wiesmeier et al., 2014), or by less tillage, without ploughing, as

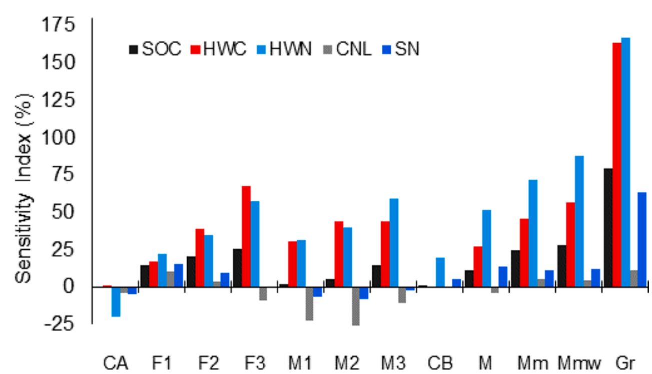

Fig. 4. Sensitivity indices of soil organic carbon (SOC), hot-water extractable carbon (HWC), labile carbon (C_L), non-labile carbon (C_{NL}), total soil nitrogen (SN) and hot-water extractable nitrogen (HWN) fractions for the 0–0.3 m soil layer of treatments and grassland. CA: rotation ‘A’ control; F1, F2 and F3: three doses of FYM; M1, M2 and M3: FYM-equivalent mineral fertiliser treatments; CB: rotation ‘B’ control; M: approx. 4 equ.; Mm: NPK + ploughed-in maize stalks; Mmw: NPK + ploughed-in maize stalks + wheat straw.

was verified in a nearby LTE (Rieder et al., 2018). Conventional tillage is unfavourable for C accumulation due to the disturbance of the soil and the disruption of aggregates (Bongiorno et al., 2019). This was also reported earlier by Reeves (1997) in his *meta*-analysis of numerous LTEs in various parts of the world. The decline in SOC is reversible and can be returned to the original level by means of reclamation (Richard et al., 2017). However, this process is also reversible due to the promotion of macroaggregate formation through the addition of plant residues (Dunai and Tóth, 2015; Rieder et al., 2018), because particulate organic matter (POM) makes up the major part of the increase (Trimble et al., 2018). Unlike the C_f and the inter-microaggregate C, POM respond sensitively to disturbance (Rieder et al., 2018; Schulz et al., 2002), and residue retention, while C_f is sensitive to manuring (Samson et al., 2020).

The high CSP values for the LTE and the grassland soil (Table 5) agree with results of Chen et al. (2018) and Wiesmeier et al. (2018), who found that on a world scale CSP is high, with values of 50–69% for arable land and 30% for grassland. However, the estimated carbon sequestration capacity cannot be fully achieved, because the labile, transitional POM pool of the coarse mineral fraction is also part of the SOC (Di et al.,

Table 7
Pearson's correlation coefficients between soil quality parameters.

Parameters	HWC	SOC	C_{NL}	HWN	SN	K_a	P_a	pH_{KCl}	Crop yield	SOC: SN	HWC: HWN	HWN/HWC%	HWC/SOC%	HWN/SN%	P_a/SOC	P_a/HWC
HWC	1.000	0.894*	−0.386*	0.890*	0.727*	0.666*	0.639*	0.313*	0.586*	0.299	0.164	−0.147	0.795*	0.648*	0.560*	0.308**
SOC		1.000	0.444*	0.797*	0.894*	0.536*	0.607*	0.412*	0.482*	0.252	0.101	−0.084	0.452*	0.419*	0.431*	0.411*
C_{NL}			1.000	−0.006	0.465*	−0.155	−0.036	0.121	−0.110	−0.033	−0.006	0.103	−0.579*	−0.364*	−0.154	0.126
HWN				1.000	0.698*	0.687*	0.642*	0.229	0.761*	0.156	−0.291	0.301	0.684*	0.799*	0.577*	0.426*
SN					1.000	0.020	−0.002	0.282	0.106	−0.292	−0.349**	−0.084	0.274	0.149	−0.079	0.004
K_a						1.000	0.819*	0.064	0.570*	0.421*	−0.109	0.137	0.480*	0.687*	0.777*	0.920*
P_a							1.000	0.129	0.821*	0.503*	−0.084	0.100	0.427*	0.642*	0.976*	0.920*
pH_{KCl}								1.000	0.156	0.094	0.178	−0.228	0.160	0.137	0.054	0.038
Crop yield									1.000	0.264	−0.349**	0.348**	0.455*	0.723*	0.830*	0.775*
SOC:SN										1.000	0.358**	−0.345**	0.323**	0.480*	−0.439*	−0.351**
HWC/HWN											1.000	−0.991	0.237	−0.338**	−0.090	−0.199
HWN/HWC%												1.000	−0.229	0.349**	0.101	0.200
HWC/SOC%													1.000	0.755*	0.447*	0.137
HWN/SN%														1.000	0.619*	0.429*
P_a/SOC															1.000	0.940*
P_a/HWC																1.000

* and ** represent significance at the 0.05 and 0.1 level, respectively.
P_a, K_a – available P and K.

2017).

The present results corroborate previous findings that C storage can be enhanced by increasing the quantity of fertilizer and the 4p1000 targets can be reached during the early stages of the experiments (Table 5). In Rothamsted, for example, when the FYM quantity was doubled to 40 t ha^{-1} yr^{-1}, CSR fell by 75% over the next 10 years, and a significant increase in CSR was only recorded for the double or quadruple straw quantity or by applying high N doses (Poulton et al., 2018). A higher rate of 240 kg N ha^{-1} was no more effective: the gain in SOC dropped to half after 20 years and a third after 40 years (Reiter, 2015). In a recent *meta*-analysis of straw treatments Han et al. (2018) detected an intense decrease in CSR over six decades. Straw has a weaker value than FYM, but the ploughing in of plant residues supplemented by 140 or 210 kg N ha^{-1} had a beneficial impact on humus formation (Dunai and Tóth, 2015), as confirmed in the present work.

In the case of low SOC content similar to that of the grassy margin, McLauchlan et al. (2006) recorded a near identical CSR of 0.62 Mg C ha^{-1} yr^{-1} for 40 years after arable areas was transformed back to prairie. Conversion from plowland to grassland may result in as much as 0.4–0.8 Mg ha^{-1} yr^{-1} CSR in some regions in the topsoils of Europe (Lugato et al., 2015). Due to N limitation, however, the recovery of SOC is slow, often incomplete. The plateau of the C sequestration curve suggests the cessation of SOC build-up rather than saturation, which might resume after a longer time span. Furthermore, the increases in the ratio of grasses and in species diversity result in greater phytobiomass (Chang et al., 2017). For example, the establishment of ryegrass-crested dog's-tail *(Lolium perenne-Cynosurus cristatus)*-dominated grassland caused a doubling of C and N sequestation rate, which was further enhanced to a certain extent over the course of 16 years by the presence of N-fixing clover (*Trifolium)* species (De Deyn et al., 2011), which were only found sporadically in the grassy margin of the present experiment.

4.2. Labile C and N fractions are sensitive to soil quality changes

HWC forms an easily decomposable sub-pool of SOC and, due to its rapid dynamics, is a sensitive indicator of anthropogenic effects, such as differences between land-use systems and changes in SOC (Gaublomme et al., 2006; Ghani et al., 2003; Schulz, 1997; Weigel et al., 2011), so its early warning capacity helps to indicate changes in soil quality (Bongiorno et al., 2019).

Quantitative difference in the hot-water extract may be caused by numerous factors, including plant and microbial substances or plant exudates (Yoshikawa et al., 2018), the quality and C:N ratio of the organic residues, the environmental conditions favourable for decomposing agents. Sampling time matters, since at the beginning of the biodegradation the diverse, easily decomposable organic matter content of the plants has a pronounced effect on the water-soluble fraction (Hadas et al., 2004).

HWC can be used to estimate the SOM stocks of soils. In Central European LTEs very low HWC values of < 200, 285 and 550 mg kg^{-1} (2–3 times better than for mineral fertiliser) were reported for unfertilized, NPK + straw and FYM treatments (Schulz et al., 2002; Schulz and Körschens, 2005). The mean HWC contents of 245.5, 342.5, 370.9 and 346.7 mg kg^{-1} detected for the same treatments in the present LTE represented a deterioration in soil structure and fertility, and thus low and medium SOM supplies. HWN increments in the treatments were also reported by Nedvěd et al. (2008). As in the present work, Šeremešić et al. (2013) found a two-fold difference in HWC content between land-use systems. After the cessation of ploughing Landgraf et al. (2003) reported increases of 4.5 and 4 times in HWC and HWN content, but the level characteristic of natural soils has not evolved even after 30 years.

Tilling practices have a decisive effect on SOM composition. The C_L ratio was 44.4–60.2% for the LTE, with higher values in the organic treatments, and 64.3% in the grassland soil (Fig. 2). The decline in the C_L ratio to below 40% in the case of intensive tillage (Yu et al, 2017) or due to the omission of fertilization can be explained by the mineralisation of

SOM (Körschens et al., 1998). Farmyard manuring increased the ratio of high-molecular-weight, good quality components beneficial for soil quality and stability, while high mineral N rates enhanced that of the low-molecular-weight components that improve nutrient-supplying ability. Therefore, their application is not recommended as they result in a depletion of the C stock (Debreczeni and Győri, 1997; Ghani et al., 2003). The N + straw and FYM treatments led to 1.3 and 2.5 times increases in SOC and a more substantial 1.5 and 5 times increase in labile C (Chen et al., 2018). During the grassland period of agro-ecosystem restoration not only the stable but also the labile C content increases; its quantity was reported to be 2–3 times that of the arable soil (Zhao et al., 2015).

4.3. Applicability of labile fractions as soil quality indicators

In the present experiment the SOC:SN ratio varied from 7.03 to 9.62, with a significant difference only for the F3 treatment ($p < 0.05$) (Table 6). In cultivated topsoils the ratio ranges between 8.1 and 15.1 (Brady and Weil, 2010), with lower, 9.3–11.2 values for european temperate croplands and higher, 10.3–16.2 values for grasslands, respectively (Tóth et al., 2013). Lower values were indicative of better substrate quality and degradability (Nedvěd et al., 2008), e.g. due to the presence of leguminous plants (Zhou et al., 2012), while the incorporation of straw increased the ratio (Hazarika et al., 2009).

Many authors used total C and N and their ratio to evaluate soil quality and available N. However, even small changes in SOM may have a significant effect on the ratio, so it is recommended to analyse the closely correlated labile HWC fractions (Schulz and Körschens, 2005; Sparling et al., 1998). These parameters are directly proportional to the plant input (Marty et al., 2017) and all give good correlations with the N-supplying capacity of soils (Thomas et al., 2016).

Various ratios expressive of soil fertility increased during the regeneration of cultivated soils, suggesting better organic inputs and decomposition and favourable mineral N levels, the readily mineralised portion of organic matter (Curtin et al., 2006; Landgraf et al., 2005). The HWC/SOC%, HWN/SN% and HWC:HWN ratios ranged from 2.88 to 3.90, 1.79–3.75% and 8.29–11.75 in the LTE and were 4.26%, 3.51% and 10.56 in the grassland soil. Apart from some variance, the ratios rose proportionately with the inputs, consistently with previous studies. Bongiorno et al. (2019) reported HWC/SOC% values of 1.0–6.0% for European LTEs, Landgraf et al. (2003) found an increasing ratio (1.1–6.9%), in the case of HWC:HWN (3.5–11.0), after the cessation of intensive cultivation. Nedvěd et al. (2008) gave values of 4.27–5.25% HWN/SN%. However, due to the intensive N supplies and uptake in the present LTE, contrary to our results, a lower HWC:HWN ratio of 8 and a wider C:N ratio of 11 than was found by Curtin et al. (2006).

HWC and HWN proved to be the most sensitive indicators, having sensitivity index (SI) values twice as high on grassland soil as in the most favourable organic treatments (Fig. 4). The present results confirmed the findings of earlier studies: the labile C and N fractions gave the most dynamic and most sensitive responses (Liang et al., 2012; Zhong et al., 2015) and could thus be used for the detection of fertiliser-induced changes in SOC. The closest correlation with the yield was found for the labile fractions, due to their nutrient-supplying ability (Das et al., 2017). The ploughing in of organic residues resulted in high biological activity and available nutrient pools, as indicated by the high HWN level, which was similar to that recorded for mineral fertilisation (Martyniuk et al., 2015). This confirmed the findings of Zhou et al. (2012) who reported that 59% of HWN was determined by the plant residue biomass.

Mineralisation indicators are important for the evaluation of soil fertility: the degradation of SOM and organic residues maintains the pools of available nutrients, but they are mostly determined in laboratory incubations. Contradictory results are found in the literature, probably due to the very diverse experimental conditions. To achieve a more reliable relationship, extractable soil C pools and a wider range of soil types and landuses need to be combined. Hot water extraction is a useful method for determining the size of the mineralisable C pool. The extracted C is chemically labile and biologically available, so it can be used as a proxy for decomposable C (Gaublomme et al., 2006; McNally et al., 2018) and as an indicator of SOM and N supplies (Nedvěd et al., 2008; Schulz, 1997). Although Körschens et al. (1990) reported that HWN was less reliable, as it gave fluctuating data, HWN is nevertheless a useful predictor for tillage modes, N dynamics, N_0 (potential N mineralisation) and available N, providing guidelines on the optimum N dose that should be applied (Curtin et al., 2006; Zhou et al., 2012).

In an incubation experiment representing numerous management systems Schomberg et al. (2009) observed that N_0 was strongly correlated not only with SOC and SN, but also with the N fractions obtained with various mineralisation methods, including HWN, since N_0 is partially derived from labile organic N. In another experiment, covering eight landuses, Ahn et al. (2009) found that SOC explained 62% of N_0 and HWC 59%, so they recommended the determination of SOC. By contrast, in a study of 98 arable soils, Ros (2011) reported an opposite correlation order, while a weaker value was obtained for SN (77, 53 and 45%). The quantity of organic matter was found to be decisive, while its quality was less so. Similarly, Zhou et al. (2012) detected a significantly lower net N mineralisation rate in the unfertilized treatment, which was not related to the quality of the cover crops; HWN was in significant correlation with the latter, while HWC was not. In an incubation experiment on manured soils performed by Thomas et al. (2016) a closer correlation with HWN was also demonstrated, with an order of SOC $\geq$ HWN $>$ SN $>$ HWC. Previously, Del Pino Machado (2005) verified that HWN gave the closest correlation with the N mineralised and taken up by plants and SN the second closest, while the N fractions obtained with other extractants proved to be poorer indicators. Recently, in an investigation on major agricultural soil types and land use modes, Lawrence-Smith et al. (2018) found that 87% of the variance in potentially mineralisable N could be explained by HWN, which was thus recommended as a new indicator for the N-supplying ability of the soil.

4.4. Relationships between soil quality parameters

The close correlation found between SOC and SN and between HWC and SOC (Table 7) is in agreement with previous studies (Hazarika et al., 2009; Schomberg et al., 2009; Šeremešić et al., 2013; Zhou et al., 2012). An increase in SOM leads to higher soil fertility and is accompanied by more intense mineralisation and an increase in the labile C fractions (Jensen et al., 2018). Correlation values for labile fractions differ. A study on European LTEs conducted by Bongiorno et al. (2019) demonstrated, for example, that, due to their strong correlations with changes in SOC quality, the permanganate-oxidisable and POM C fractions were better, more sensitive indicators than HWC or SOC, and, according to Filep et al. (2015), than dissolved organic C, a sup-pool of HWC.

The statement by Körschens et al. (1990) that HWC gave the best correlation with SN, was confirmed by Weigel et al. (2011). In the LTE, the weaker correlation between HWC and SN could be explained by the opposing trends observed for the doses and the SN values. In agreement with the results reported by Landgraf et al. (2003) and Nedvěd et al. (2008), a close correlation between HWN and SN was also found in the present work. Landgraf et al. (2003) pointed to close correlations between HWN and available N forms. Namely, a high mineral N level was indicative of intensive mineralisation, and explained why the relationship between the yield and HWN was more significant than that between yield and HWC, as also proven by Thomas et al. (2016). The fact that the yield is closely correlated with P_a > HWN > HWC > K_a > SOC, and with various ratios reflecting soil nutrient supply and fertility, as corroborated by Ros (2011), proves that their functions are manifested through such factors as fertiliser application, the biomass formed, and SOM mineralisation.

5. Conclusions

The 4p1000 targets were achieved in fewer and fewer treatments as time progressed. In contrast to the grassland soil, which exhibited a substantial gain in SOC, the efficiency of the favourable organic treatments (BMP) dropped to half, as indicated by the carbon sequestration (storage) potential (CSP) values. The atmospheric decarbonization targets may be thwarted by the soil, growing site and plant production conditions. Despite the fact that CSP remains high, the achievement of 4p1000 objectives can only be ensured in treatments involving high rates of organic matter and only in the short term.

The present results corroborated the findings in the literature that the most sensitive fraction for the detection of the effects of treatments and land-use change, despite their oscillation, were HWC and HWN. The analysis of sensitivity gave the order: HWN > HWC ≫ SOC. Increases in these labile organic fractions were significantly related to the gains in SOC stocks. Their close correlation with the crop yield and with various soil quality indicators can be attributed to the fact that the organic compounds and nutrients extracted with hot water are sensitive indicators of SOM mineralisation and the nutrient-supplying capacity of soils. The close correlations suggest that the hot water extraction method can be recommended for use in the analysis and monitoring of soil fertility and quality.

CRediT authorship contribution statement

László Bankó: Conceptualization, Software, Visualization, Methodology, Investigation, Data curation, Formal analysis, Writing - original draft. **Gergely Tóth:** Supervision, Validation, Project administration, Writing - review & editing. **Csaba L. Marton:** Supervision, Validation, Writing - review & editing. **Sándor Hoffmann:** Conceptualization, Investigation, Validation, Data curation, Formal analysis, Funding acquisition, Resources, Methodology, Project administration, Supervision, Writing - review & editing.

Declaration of Competing Interest

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

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ecolind.2021.107364.

References

4p1000 Initiative, 2018. Wellcome to the 4 per Mille Initiative. Montpellier, France. https://www.4p1000.org.

Ahn, M.Y., Zimmerman, A.R., Comerford, N.B., Sickman, J.O., Grunwald, S., 2009. Carbon mineralization and labile organic carbon pools in the sandy soils of a North Florida watershed. Ecosystem. 12, 672–685.

Angers, D.A., Arrouays, D., Saby, N.P.A., Walter, C., 2011. Estimating and mapping the carbon saturation deficit of French agricultural topsoils. Soil Use Manage. 27, 448–452.

Blake, G.R., 1965. Bulk density, in: Black, C.A. (Ed.), Methods of Soil Analysis. Part 1. Am. Soc. Agron. Monograph 9, Madison. WI., pp. 374–390.

Bongiorno, G., Bünemann, E.K., Oguejiofora, C.U., Meier, J., Gort, G., Comans, R., Mäderb, P., Brussaarda, L., de Goedea, Ron, 2019. Sensitivity of labile carbon fractions to tillage and organic matter management and their potential as comprehensive soil quality indicators across pedoclimatic conditions in Europe. Ecol. Indic. 99, 38–50.

Brady, N., Weil, R., 2010. Nutrient Cycles and Soil Fertility. In: Anthony, V.R. (Ed.), Elements of the Nature and Properties of Soils, 3rd Edition. Pearson Education Inc., Upper Saddle River, NJ, pp. 396–420.

Buzás, I., Murányi, A., Rédly, L-né., 1988. Talaj- és agrokémiai vizsgálati módszerkönyv 2. (Szerk. Buzás, I.). A talajok fizikai-kémiai és kémiai vizsgálati módszerei. Mezőgazdasági Kiadó, Budapest.

Chambers, A., Lal, R., Paustian, K., 2016. Soil carbon sequestration potential of US croplands and grasslands: Implementing the 4 per Thousand Initiative. J. Soil Water Conserv. 71, 68A–74A.

Chang, J., Ciais, P., Viovy, N., Vuichard, N., Herrero, M., Havlik, P., 2016. Effect of climate change, CO_2 trends, nitrogen addition, and land-cover and management intensity changes on the carbon balance of European grasslands. Glob. Change. Biol. 22, 338–350.

Chang, X., Chai, Q., Wu, G.L., Zhu, Y., Li, Z., Yang, Y., Wang, G., 2017. Soil organic carbon accumulation in abandoned croplands on the Loess Plateau: Vegetation succession and SOC recovery. Land Degrad. Dev. 28, 1519–1527.

Chen, S., Martin, M.P., Saby, N.P.A., Walter, C., Angers, D.A., Arrouays, D., 2018. Fine resolution map of top- and subsoil carbon sequestration potential in France. Sci. Total Environ. 630, 389–400.

Clima, 2020. https://ec.europa.eu/clima/policies/strategies/2050_en.

Collier, S.M., Ruark, M.D., Naber, M.R., Andraski, T.W., Casler, M.D., 2017. Apparent Stability and Subtle Change in Surface and Subsurface Soil Carbon and Nitrogen under a Long-Term Fertilizer Gradient. SSSAJ. 82, 310–321.

Costantini, E.A.C., Branquinho, C., Nunes, A., Schwilch, G., Stavi, I., Valdecantos, A., Zucca, C., 2016. Soil indicators to assess the effectiveness of restorationstrategies in dryland ecosystems. Solid Earth. 7, 397–414.

Curtin, D., Wright, C.E., Beare, M.H., McCallum, F.M., 2006. Hot water-extractable nitrogen as an indicator of soil nitrogen availability. SSSAJ. 70, 1512–1521.

Csathó, P., Magyar, M., Osztoics, E., Debreczeni, K., Sárdi, K., 2011. Talaj- és diagnosztikai célú növényvizsgálati módszerek kalibrálása az OMTK kísérletekben. II. Környezetvédelmi célú talaj P-teszt módszerek összehasonlítása a tartamkísérletek talajaiban. Agrokémia és Talajtan. 60, 343–358.

Das, D., Dwivedi, B.S., Singh, V.K., Datta, S.P., Meena, M.C., Chakraborty, D., Bandyopadhyay, K.K., Kumar, R., Mishra, R.P., 2017. Long-term effects of fertilisers and organic sources on soil organic carbon fractions under a rice-wheat system in the Indo-Gangetic Plains of north-west India. Soil Res. 55, 296–308.

De Deyn, G.B., Shiel, R.S., Nick, J.O., McNamara, N.P., Oakley, S., Young, I., Freeman, C., Fenner, N., Quirk, H., Bardgett, R.D., 2011. Additional carbon sequestration benefits of grassland diversity restoration. J. Appl. Ecol. 48, 600–608.

De Vries, W., 2017. Soil carbon 4 per mille: a good initiative but let's manage not only the soil but also the expectations. Geoderma. 309, 111–112.

Dean, A.M., 2000. Plant residue biochemistry regulates soil carbon cycling and carbon sequestration. Soil Biol. Biochem. 32, 361–369.

Debreczeni, B-né, Győri, D., 1997. A talajok humuszminőségének és környezetvédelmi kapacításának változása műtrágyázás hatására. Agrokém. Talajtan. 46, 171–184.

del Pino Machado, A.S., 2005. Estimating nitrogen mineralization potential of soils and the effect of water and temperature and crop residues on nitrogen net mineralization. PhD thesis, Georg-August-Universität, Göttingen. https://books.google.hu/books/about/Estimating_Nitrogen_Mineralization_Poten.html?id=vh2ANnDDyCgC&redir_esc=y.

Di, J., Feng, W., Zhang, W., Cai, A., Xu, A., 2017. Soil organic carbon saturation deficit under primary agricultural managements across major croplands in China. Ecosystem Health Sustain. 3, 1364047.

Dunai, A., Tóth, Z., 2015. Szerves- és műtrágyázás tartamhatása a talajaggregátumok stabilitására agyagbemosódásos barna erdőtalajon. Agrokém. Talajtan. 64, 29–52.

Eurostat, 2019. Greenhouse gas emission statistics - emission inventories. https://ec.europa.eu/eurostat/statistics-explained/index.php/Greenhouse_gas_emission_statistics.

Filep, T., Draskovits, E., Szabó, J., Koós, S., László, P., Szalai, Z., 2015. The dissolved organic matter as a potential soil quality indicator in arable soils of Hungary. Environ. Monit. Assess. 187, 479–490.

Freytag, H.E., 1987. Gleichzeitige Ermittlung der Parameter C_{pot} und k für die CMineraliserungsfunktionen aus CO_2-Messungen unter konstanten Bedingungen. Arch. Acker-Pfl. Boden. 31, 23–31.

Gaublomme, E., De Vos, B., Cools, N., 2006. An indicator for microbial biodiversity in forest soils. INBO. R2006.40 Instituut voor Natuur-. en Bosonderzoek, Brussels.

Ghani, A., Dexter, M., Perrott, K.W., 2003. Hot-water extractable carbon in soils: a sensitive measurement for determining impacts of fertilisation, grazing and cultivation. Soil Biol. Biochem. 35, 1231–1243.

Hadas, A., Kautsky, L., Goek, M., Kara, E.E., 2004. Rates of decomposition of plant residues and available nitrogen in soil, related to residue composition through simulation of carbon and nitrogen turnover. Soil Biol. Biochem. 36, 255–266.

Han, X., Jennifer, C.W., Dungait, A.J., Bol, R., Wang, X., Wu, W., Meng, F., 2018. Straw incorporation increases crop yield and soil organic carbon sequestration but varies under different natural conditions and farming practices in China: a system analysis. Biogeosciences. 15, 1933–1946.

Hassink, J., 1997. Preservations of plant residues in soils differing in unsaturated protective capacity. SSSAJ. 61, 131–139.

Hazarika, S., Parkinson, R., Bol, R., Dixon, L., Russell, P., Donovan, S., Allen, D., 2009. Effect of tillage system and straw management on organic matter dynamics. Agron. Sustain. Dev. 29, 525–533.

Hoffmann, S., Csitári, G., Bankó, L., Balázs, J., 2006. Soil fertility characteristics due to different organic and mineral fertilization. Cer. Res. Comm. 34, 203–206.

Janzen, H.H., Campbell, C.A., Izaurralde, R.C., Ellert, B.H., Juma, N., McGill, W.B., Zentner, R.P., 1998. Management effects on soil C storage on the Canadian prairies. Soil. Till Res. 47, 181–193.

Jensen, J.L., Schjønning, P., Christopher, W.W., Christensen, B.T., Clément, P., Munkholm, L.J., 2018. Relating soil C and organic matter fractions to soil structural stability. Geoderma. 337, 834–843.

Juhos, K., Czigány, Sz, Madarász, B., Ladányi, M., 2019. Interpretation of soil quality indicators for land suitability assessment – A multivariate approach for Central European arable soils. Ecol. Indic. 99, 261–272.

Kay, S., Rega, C., Moreno, G., den Herder, M., Palma, J.H.N., Borek, R., Crous-Duran, J., Freese, D., Giannitsopoulos, M., Graves, A., Jäger, M., Lamersdorf, N., Memedemin, D., Mosquera-Losada, R., Pantera, A., Paracchini, M.L., Paris, P., Roces-Díaz, J.V., Rolo, V., Rosati, A., Sandor, M., Smith, J., Szerencsits, E., Varga, A., Viaud, V., Wawer, R., Burgess, P.J., Herzog, F., 2019. Agroforestry creates carbon sinks whilst enhancing the environment in agricultural landscapes in Europe. Land Use Policy. 83, 581–593.

Kenéz, Á., 2014. Keszthely-fenékpuszta római kori régészeti-növénytani leleteinek feldolgozása, különös tekintettel az egykori környezeti állapot jellemzésére. PhD Dissertation. Szent István Egyetem, Gödöllő. http://phd.szie.hu/?docId=14690.

Kennedy, M.J., Löhr, S.C., Fraser, S.S., Baruch, E.T., 2014. Direct evidence for organic carbon preservation as clay-organic nanocomposites in a Devonian black shale; from deposition to diagenesis. Earth Planet. Sci. Lett. 388, 59–70.

Körschens, M., Schultz, E., 1999. Bestimmung die umsetzbaren organiscen Kohlenstoffs im Boden (C_{ums}) und Ableitung von gehaltklassen, in: Die organische Bodensubstanz. UFZ-Umweltfortschunfszentrum Leipzig-Halle GmbH. Nr. 13, 29–39.

Körschens, M., Schulz, E., Behm, R., 1990. Hot water extractable carbon and nitrogen of soils as criteria of their ability for N-release. Zentralbl. Mikrobiol. 145, 305–311.

Körschens, M., Weigel, A., Schulz, E., 1998. Turnover of soil organic matter (SOM) and long-term balances – tools for evaluating productivity and sustainability. Z. Planz. Bodenk. 161, 409–424.

Landgraf, D., Böhm, C., Makeschin, F., 2003. Dynamic of different C and N fractions in a Cambisol under five year succession fallow in Saxony. J. Plant Nutr. Soil Sci. 166, 319–325.

Landgraf, D., Wedig, S., Klose, S., 2005. Medium- and short-term available organic matter, microbial biomass, and enzyme activities in soils under Pinus sylvestris L. and Robinia pseudoacacia L. in a sandy soil in NE Saxony. Germany. J. Plant Nutr. Soil Sci. 168, 193–201.

Lawrence-Smith, E., McNally, S., Beare, M., Curtin, D., Lehto, K., 2018. Updating guidelines for the interpretation of soil organic matter (carbon and nitrogen) indicators of soil quality for state of the environment monitoring. New Zealand Institute for Plant & Food Research Limited. (http://envirolink.govt.nz/assets/Envirolink/Reports/1801-MLDC132-Updating-guidelines-for-the-interpretation-of-soil-organic-matter-carbon-and-nitrogen-indicators-of-soil-quality-for-state-of-the-environment-monitoring.pdf).

Lefèvre, C., Fatma, R., Viridiana, A., Liesl, W., 2017. What is SOC? In: Liesl, W. (Ed.), Soil organic carbon the hidden potential. FAO, Rome, Italy, pp. 1–9. http://www.fao.org/3/I6937EN/i6937en.pdf.

Liang, Q., Chen, H., Gong, Y., Fan, M., Yang, H., Lal, R., Kuzyakov, Y., 2012. Effects of 15 years of manure and inorganic fertilizers on soil organic carbon fractions in a wheat-maize system in the North China Plain. Nutr. Cycl. Agroecosyst. 92, 21–33.

Loveland, P., Webb, J., 2003. Is there a critical level of organic matter in the agricultural soils of temperate regions: a review. Soil Till. Res. 70, 1–18.

Lugato, E., Bampa, F., Panagos, P., Montanarella, L., Jones, A., 2015. Potential carbon sequestration of European arable soils estimated by modelling a comprehensive set of management practices. Glob. Chang. Biol. 20, 3557–3567.

Marty, C., Houle, D., Gagnon, C., Courchesne, F., 2017. The relationships of soil total nitrogen concentrations, pools and C: N ratios with climate, vegetation types and nitrate deposition in temperate and boreal forests of eastern Canada. Catena. 152, 163–172.

Martyniuk, S., Koziel, M., Stalenga, J., Jonczyk, K., 2015. Labile factions of soil organic matter and microbial characteristics of soil under organic and conventional crop management systems. Biol. Agric. Hortic. 32, 1–6.

McLauchlan, K.K., Hobbie, S.E., Post, W.M., 2006. Conversion from agriculture to grassland builds soil organic matter on decadal timescales. Ecol. Appl. 16, 143–153.

McNally, S., Beare, M., Curtin, D., Tregurtha, C., Qui, W., Kelliher, F., Baldock, J., 2018. Assessing the vulnerability of organic matter to C mineralisation in pasture and cropping soils of New Zealand. Soil Res. 56, 481–490.

Minasty, B., Malone, B.P., McBratney, A.B., Angers, D.A., Arrouays, D., Chambers, A., Chaplot, V., Chen, Z.S., Cheng, K., Das, B.S., Field, D.J., Gimona, A., Hedley, C.B., Hong, S.Y., Mandal, B., Marchant, B.P., Martin, M., McConkey, B.G., Mulder, V.L., O'Rourke, S., Richer-de-Forges, A.C., Odeh, I., Padarian, J., Paustian, K., Pan, G.X., Poggio, L., Savin, I., Stolbovoy, V., Stockmann, U., Sulaeman, Y., Tsui, C.C., Vagen, T.G., van Wesemael, B., Winowiecki, L., 2017. Soil carbon 4 per mille. Geoderma 292, 59–86.

Nedvěd, V., Balík, J., Černý, J., Kulhánek, M., Balíková, M., 2008. The changes of soil nitrogen and carbon contents in a long-term field experiment under different systems of nitrogen fertilization. Plant Soil Environ. 54, 463–470.

Pavithra, S., Arachchige, P., Hettiarachchi, G.M., Rice, C.W., Dynes, J.J., Maurmann, L., Wang, J., Karunakaran, C., Kilcoyne, A.L.D., Attanayake, C.P., Amado, T.J.C., Fiorin, J.E., 2018. Sub-micron level investigation reveals the inaccessibility of stabilized carbon in soil microaggregates. Sci. Rep. 8, 16810.

Poeplau, C., Don, A., 2013. Sensitivity of soil organic carbon stocks and fractions to different land-use changes across Europe. Geoderma. 192, 189–201.

Poulton, P., Johnston, J., Macdonald, A., White, R., Powlson, D., 2018. Major limitations to achieving "4 per 1000" increases in soil. Glob. Chang. Biol. 24, 2563–2584.

Powlson, D.S., Whitmore, A.P., Goulding, K.W.T., 2011. Soil carbon sequestration to mitigate climate change: a critical re-examination to identify the true and the false. Eur. J. Soil Sci. 62, 42–55.

Pries, C.E.H., Sulman, B.N., West, C., O'Neillf, C., Poppleton, E., Porras, R.C., Castanha, C., Zhu, B., Wiedemeier, D.B., Torn, M.S., 2018. Root litter decomposition slows with soil depth. Soil Biol. Biochem. 125, 103–114.

Reeves, D.W., 1997. The role of soil organic matter in maintaining soil quality in continuous cropping systems. Soil Till. Res. 43, 131–167.

Reeves, M.C., Moreno, A.L., Bagne, K.-E., 2014. Estimating climate change effects on net primary production of rangelands in the United States. Clim. Change. 126, 429–442.

Reiter, L., 2015. Effect of crop residue incorporation on soil organic carbon dynamics: changes in carbon stocks and carbon fractions in an Italian long-term field experiment. Thesis. SLU, Uppsala. https://stud.epsilon.slu.se/8025/.

Richard, T.C., Cerri, C.E.P., Brook, B.O., Paustia, K., 2017. Grassland management impacts on soil carbon stocks: a new synthesis. Ecol. Appl. 27, 662–668.

Rieder, Á., Madarász, B., Szabó, J.A., Zacháry, D., Vancsik, A., Ringer, M., Szalai, Z., Jakab, G., 2018. Soil organic matter alteration velocity due to land-use change: a case study under conservation agriculture. Sustainability. 10, 943.

Ros, G.H., 2011. Predicting soil nitrogen supply. Relevance of extractable soil organic matter fractions. PhD Thesis, Wageningen University Wageningen, The Netherlands. https://www.semanticscholar.org/paper/Predicting-soil-nitrogen-supply-%3A-relevance-of-soil-Ros/771c03cf652fe63daad2aaf900ff4279d0507dac.

Samson, M.-E., Chantigny, M.H., Vanasse, A., Menasseri-Aubry, S., Royer, I., Angers, D. A., 2020. Management practices differently affect particulate and mineral-associated organic matter and their precursors in arable soils. Soil. Bio. Biochem. 148, 107867.

Sándor, R., Ehrhardt, F., Grace, P., Recous, S., Smith, P., Snow, V., Soussana, J.-F., Basso, B., Bhatia, A., Brilli, L., Doltra, J., Dorich, C.D., Doro, L., Fitton, N., Grant, B., Harrison, M.T., Skiba, U., Kirschbaum, M.U.F., Klumpp, K., Laville, P., Léonard, J., Martin, R., Massad, R.S., Moore, A., Myrgiotis, V., Pattey, E., Qing, Z., Rolinski, S., Sharp, J., Smith, W., Wu, L., Bellocchi, G., 2020. Ensemble modelling of carbon fluxes in grasslands and croplands. Field Crop Res. 252, 107791.

Schomberg, H.H., Wietholter, S., Griffin, T.S., Reeves, D.W., Cabrera, M.L., Fisher, D.S., Endale, D.M., Novak, J.M., Balkcom, K.S., Raper, R.L., Kitchen, N.R., Locke, M.A., Potter, K.N., Schwartz, R.C., Truman, C.C., Tyler, D.D., 2009. Assessing indices for predicting potential nitrogen mineralization in soils under different management systems. SSSAJ. 73, 1575–1586.

Schulz, E., 1997. Charakterisierung der organischen Bodensubstanz nach dem Grad ihrer Umsetzbarkeit und ihre Bedeutung für Transformationsprozesse für Nähr- und Schadstoffe. Arch. Agron. Soil Sci. 41, 465–483.

Schulz, E., Körschens, M., 2005. Dauerversuche als Basis zur Ableitung vor Indikatoren der Erfassung bewirtschaftungs bedingter Veränderungen des C-Haushaltes des Ackerböden. Poster.

Schulz, E., Travnikova, L.S., Titova, N.A., Kogut, B.M., Köschens, M., 2002. Influence of soil type and fertilization on accumulation and stabilization of organic carbon in different SOM Fractions. 12th ISCO Conference Beijing.

Šeremešić, S., Dragiša Milošev, D., Sekulić, P., Nešić, L., Ćirić, V., 2013. Total and hot-water extractable carbon relationship in chernozem soil under different cropping systems and land use. J. Cent. Eur. Agric. 14, 1479–1487.

SOCCR2, 2018. Second State of the Carbon Cycle Report. https://carbon2018.globalchange.gov/.

Soussana, J.F., Loiseau, P., Vuichard, N., Ceschia, E., Balesdent, J., Chevallier, T., Arrouays, D., 2004. Carbon cycling and sequestration opportunities in temperate grasslands. Soil Use Manage. 20, 219–230.

Sparling, G., Vojvodic-Vukovic, M., Schipper, L.A., 1998. Hot-water-soluble C as a simple measure of labile soil organic matter: the relationship with microbial biomass C. Soil Biol. Biochem. 30, 1469–1472.

Thomas, B.W., Whalen, J.K., Sharifi, M., Chantigny, M., Zebarth, B.J., 2016. Labile organic matter fractions as early-season nitrogen supply indicators in manure-amended soils. J. Plant Nutr. Soil Sci. 179, 94–103.

Tóth, G., Jones, A., Montanarella, L., 2013. Land-use specific comparative analysis of soil characteristics in the EU. In: Tóth, G., Jones, A., Montanarella, L. (Eds.), LUCAS Topsoil Survey methodology, data and results. JRC-IES, Italy, pp. 35–88. https://esdac.jrc.ec.europa.eu/ESDB_Archive/eusoils_docs/other/EUR26102EN.pdf.

Tóth, G., Montanarella, L., Rusco, E., 2008. Soil quality in the European Union. In: Tóth, G., Montanarella, L., Rusco, E. (Eds.), Threats to Soil Quality in Europe. JRC-IES, Luxembourg, pp. 11–19.

Trimble, B.R., Calderon, F.J., Poulson, S.R., Verburg, P.S.J., 2018. Conversion of a semiarid nevada soil to irrigated agriculture preferentially removes labile carbon. Soil Syst. 2, 38.

Vermeulen, S.J., Campbell, B.M., Ingram, J.S.I., 2012. Climate change and food system. Annu. Rev. Environ. Resour. 37, 195–222.

Wang, G., Luo, Z., Han, P., Chen, H., Xu, J., 2016. Critical carbon input to maintain current soil organic carbon stocks in global wheat systems. Sci. Rep. 6, 19327.

Weigel, A., Eustice, T., Van Antwerpen, R., Naidoo, G., Schulz, E., 2011. Soil organic carbon (SOC) changes indicated by hot water extractable carbon (HWEC). Proc S. Afr. Sug. Technol. Ass. 84, 210–222.

Wiesmeier, M., Burmeister, J., Hübner, R., Kögel-Knabner, I., 2018. Implementation of the 4 per 1000 initiative at the regional scale: A case study for Bavaria. Conference. ISTRO 2018. Paris.

Wiesmeier, M., Hübner, R., Spörlein, P., Geuß, U., Hangen, E., Reischl, A., Schilling, B., von Lützow, M., Kögel-Knabner, I., 2014. Carbon sequestration potential of soils in southeast Germany derived from stable soil organic carbon saturation. Global Carbon Change. 20, 653–665.

Wiesmeier, M., Urbanski, L., Hobley, E., Lang, B., von Lützow, M., Marin-Spiotta, E., van Wesemael, B., Rabot, E., Ließ, M., Garcia-Franco, N., Wollschläger, U., Vogel, H.-J., Kögel-Knabner, I., 2019. Soil organic carbon storage as a key function of soils - A review of drivers and indicators at various scales. Geoderma. 333, 149–162.

Yiang, L., Shao, J., Shi, Z., Zhou, X., Luo, Y., 2019. Responses of grasslands to experimental warming. In: Mohan, J.E. (Ed.), Ecosystem Consequences of Soil Warming: Microbes. Academic Press, Vegetation, Fauna and Soil Biogeochemistry, pp. 347–384.

Yoshikawa, S., Kureda, Y., Uero, H., Kajiura, M., Ae, N., 2018. Effect of phenolic acids on the formation and stabilization of soil aggregates. Soil Sci. Plant. Nutr. 64, 323–334.

Yu, P., Han, K., Li, Q., Zhou, D., 2017. Soil organic carbon fractions are affected by different land uses in an agro-pastoral transitional zone in Northeastern China. Ecol. Indic. 73, 331–337.

Zhao, Y.G., Liu, X.F., Wang, Z.L., Zhao, S.W., 2015. Soil organic carbon fractions and sequestration across a 150-yr secondary forest chronosequence on the Loess Plateau. China. Catena. 133, 303–308.

Zhong, Y., Yan, W., Shangguan, Z., 2015. Soil carbon and nitrogen fractions in the soil profile and their response to long-term nitrogen fertilization in a wheat field. Catena. 135, 38–46.

Zhou, X., Wu, H., Xu, Z., 2012. Dynamics of soil extractable carbon and nitrogen under different cover crop residues. J. Soil Sediment. 12, 844–853.

Zlinszky, A., Molnár, G., 2009. Georeferencing the first bathymetric maps of Lake Balaton. Hungary. Acta Geod. Geophys. Hu. 44, 79–94.