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Hot regions of labile and stable soil organic carbon in Germany - Spatial variability and driving factors

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Abstract

Atmospheric carbon dioxide levels can be mitigated by sequestering carbon in the soil. Sequestration can be facilitated by agricultural management, but its influence is not the same on all soil carbon pools, as labile pools with high turnover may be accumulated much faster, but are also more vulnerable to losses. The aims of this study were to 1) assess how soil organic carbon (SOC) is distributed among SOC fractions on national scale in Germany, 2) identify factors influencing this distribution and 3) identify regions with high vulnerability to SOC losses. The SOC content and proportion of two different SOC fractions were estimated for more than 2500 mineral topsoils (<87 g kg^{-1} SOC) covering Germany, using near-infrared reflectance spectroscopy. Drivers of the spatial variability in SOC fractions were determined using the machine learning algorithm cforest. The SOC content and proportions of fractions were predicted with good accuracy (SOC content: R^2=0.87-0.90, SOC proportions R^2=0.83, ratio of performance to deviation (RPD) 2.4-3.2). Main explanatory variables for distribution of SOC among the fractions were soil texture, bulk soil C/N ratio, total SOC content and pH. For some regions, the drivers were linked to the land-use history of the sites.

Arable topsoils in central and southern Germany were found to contain the highest proportions and contents of stable SOC fractions, and therefore have the lowest vulnerability to SOC losses. North-western Germany contains an area of sandy soils with unusually high SOC contents and high proportions of light SOC fractions, which are commonly regarded as representing a labile carbon pool. This is true for the former peat soils in this area, which have already lost and are at high risk of losing high proportions of their SOC stocks. Those “black sands” can, however, also contain high amounts of stable SOC due to former heathland vegetation, and need to be treated and discussed separately from ‘normal’ agricultural soils. Overall, it was estimated that, in large areas all over Germany, over 30% of is stored in easily mineralisable forms. Thus, SOC-conserving management of arable soils in these regions is of great importance.

1 Introduction

There is increasing interest in soil organic carbon (SOC) in agricultural soils, as it contributes to soil fertility and also to mitigation of climate change when organic carbon (OC) sequestration is enhanced (Post and Kwon, 2000). The pathway of atmospheric carbon to SOC is controlled by land use and agronomic management. However, SOC comprises a large range of compounds, ranging from recently added organic matter, such as root litter and exudates, to highly condensed and transformed organic matter that may even be derived from the geogenic parent material. These different compound classes are stabilised in different ways and therefore have different turnover times (Lehmann and Kleber, 2015). Although SOC is now considered as having a continuum of turnover times, it is mostly described and modelled as consisting of different pools that vary in their turnover time (e.g. labile pool, intermediate pool and stabilised pool). The effects of land use and management are not the same for all soil organic matter compounds, however, but differ between SOC pools (Cardinael et al., 2015; Chimento et al., 2016). This is why the different SOC pools need to be assessed separately from the bulk SOC when discussing the influence of land use and management on stabilisation and storage of SOC.

One method for experimental quantification of the distribution of SOC among different SOC pools is fractionation. Various fractionation procedures for quantifying SOC fractions have been developed, mostly aiming at isolating fractions with differing turnover times (Poeplau *et al.*, submitted). Determining the distribution of SOC among fractions with assumedly different carbon turnover times is one step towards understanding the factors influencing SOC stabilisation.

Some impact factors are consistently reported as being important at site scale for the distribution of SOC among different fractions or pools, one of which is land use. In croplands and grasslands, a similarly large share of bulk SOC is attributed to fractions regarded as stable, while in forest soils a larger proportion of SOC is attributed to more labile SOC fractions (John *et al.*, 2005; Helfrich *et al.*, 2006; Wiesmeier *et al.*, 2014). Tillage can also have an impact on SOC pools, as several studies report higher levels of bulk SOC under no-till conditions compared with conventional tillage, with the

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majority of this increase occurring in the more labile carbon pools (Chan et al., 2002; Devine et al., 2014; Liu et al., 2014).

Fewer studies have examined the SOC distribution into fractions at regional scale and even fewer have examined factors affecting the proportions of SOC distributed among different fractions or pools. Wiesmeier *et al.* (2014) determined the distribution of SOC fractions among 99 Bavarian soils under different land uses using the method of Zimmermann *et al.* (2007a). They found that approximately 90% of the bulk SOC in cropland and grassland soils was distributed in intermediate or stabilised SOC pools, while this was only true for 60% of the SOC found in forest soils. Therefore, those authors suggested that Bavarian soils under cropland and grassland are more suitable for long-term sequestration of additional SOC than soils under forest. They also examined controlling factors for the SOC distribution among fractions in the different land uses (Wiesmeier *et al.,* 2014). Correlation analyses suggested that the intermediate SOC pools in croplands and grasslands were significantly correlated to soil moisture, but none of the functional SOC pools was influenced by temperature or precipitation. The particulate organic matter (POM) fraction of soils under grasslands and croplands was not significantly related to any environmental factor in that study (Wiesmeier *et al.*, 2014). Poeplau & Don (2013a) conducted a study on 24 sites in Europe and found that SOC fractions differed in their degree of sensitivity to land-use change (LUC), with the sensitivity declining with increasing stability in the SOC fractions. Their results indicated that afforestation of cropland shifts SOC from the more stable to the more labile fractions, while on conversion from cropland to grassland the newly sequestered SOC is stored in the intermediate to stable pools. Rabbi *et al.* (2014) examined the relationships between land use, management, climate and soil properties and the stock of three SOC fractions for soils in south-eastern Australia, and observed a high impact of climate and site-specific factors (rainfall, silicon content, soil pH, latitude) and only a minor influence of land use. The dominance of site and climate variables as impact factors in that region may primarily be due to the wide range of site conditions in the area studied.

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There are several regions in north-western Europe and also in northern Germany where the soils exhibit unusually high SOC content while having a high sand and low clay content (Sleutel *et al.*, 2011). These so called 'black sands' have a poor capacity to stabilise SOC by binding onto mineral surfaces, and therefore most SOC is present in the form of POM. A great part of this land surface in northern Germany was covered by heathland and peatland until the end of the 18th century and those soils may behave different than other soils in terms of SOC storage

All methods for carbon fractionation are quite laborious, time-consuming and therefore expensive, and not feasible for large datasets. Therefore, few studies exist on SOC fractions at regional scale, indicating a need for development of more efficient methods to predict carbon fractions in assessment of large datasets. Near-infrared reflectance spectroscopy (NIRS) and mid-infrared spectroscopy (MIRS), in combination with chemometric methods, have been applied successfully to predict carbon fractions (Zimmermann *et al.*, 2007b; Baldock *et al.*, 2013; Cozzolino & Moro, 2006; Reeves *et al.*, 2006). Thus, since prediction of SOC fractions has been demonstrated to be possible using spectroscopic methods, it should also be possible to go beyond small datasets at field scale in order to examine how SOC fractions are distributed regionally and the factors that drive this distribution.

The present study is part of the German Agricultural Soil Inventory. A set of 145 topsoil samples, representative of German agricultural soils, was fractionated and used to calibrate NIRS predictions of the constituent fractions for >2500 sites with mineral soils all over Germany. Additional climate, management and geographical data were gathered for all sites and a machine learning algorithm was employed to answer the following research questions:

1) How is SOC distributed among the fractions at national scale?
2) Which drivers are relevant for this distribution?
3) Can regions of high vulnerability to carbon losses be identified by this predictive approach?

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2 Material & Methods

2.1 Study area, sampling and sample selection

Germany has a total surface area of 357 000 km² and its climate is temperate, marine and continental. Mean annual precipitation (MAP) ranges between 490 and 2090 mm and mean annual temperature (MAT) between 5.7 and 11.2 °C. Around half the country's surface area is used for agriculture, with cropland accounting for 71% of this area, grassland for 28% and other crops (e.g. vines) for 1%.

Soil samples were taken in the course of the ongoing German Agricultural Soil Inventory. By May 2017, 2900 agricultural sites had been sampled based on an 8 km x 8 km sampling grid. At each site, a soil profile was characterised by a soil scientist and soil samples were taken from five fixed depth increments. All soils were classified in the field according to the German Soil Classification System (Sponagel et al., 2005).

The topsoils (0-10 cm) of 145 calibration sites, representative for the whole dataset, were chosen according to the following criteria: 1) Maximum difference in NIR spectra, according to the Kennard-Stone algorithm (Daszykowski et al., 2002), 2) consistent spatial distribution within Germany, 3) exclusion of sites with SOC content >87 g kg^{-1} in any horizon, as such soils may be organic (> 30% organic substance) or in transition between organic and mineral soils and it was assumed that the processes governing the variability of SOC in organic soils differ from those in mineral soils, and 4) representative mapping of land use, soil type and carbon stock.

2.2 Laboratory analyses

All samples were analysed for gravimetric water content, electrical conductivity (EC), pH, SOC content (g kg^{-1}, by dry combustion), soil inorganic carbon content (g kg^{-1}) after removing organic carbon in a muffle kiln, texture (by the pipette method), rock content, root content and bulk density (with repeated soil rings). The SOC stocks were calculated as suggested by Poeplau *et al.* (2017), taking into account the stone and root content of the soil.

2.3 Fractionation of calibration samples

The topsoil samples (0-10 cm depth) of the selected calibration sites were dried at 40°C to constant weight and sieved to a size <2 mm. Three different fractions were prepared:

1) To obtain the fraction that contains free particulate organic matter (fPOM), 20 g of soil sample were placed in a falcon tube, which was then filled to 40 mL with sodium polytungstate (SPT) solution (density=1.8 g mL^{-1}). The sample was dispersed ultrasonically at 65 J mL^{-1}, with the probe energy supply calibrated using the procedure explained in Puget *et al.* (2000). The tube was centrifuged at 4000 rpm until there was a clear distinction between the fPOM fraction and the remaining soil pellet. The supernatant was then filtered through a 45 µm filter paper and a ceramic filter using vacuum filtration. The fPOM fraction remained on the filter and was rinsed with distilled water until the electrical conductivity of the filtered water was below 10 µS m^{-1}. The fPOM fraction was then dried at 40°C, weighed and milled.

2) To obtain the particulate organic matter occluded in aggregates (oPOM) fraction, the falcon tube containing the pellet was again filled to 40 mL with SPT solution. The pellet was mixed with the solution using a vortex shaker and then ultrasonic dispersion was applied, again at 450 J mL^{-1}, in order to destroy soil aggregates. The sample was centrifuged and the oPOM fraction was processed as described above for the fPOM fraction.

3) The remaining soil pellet was assumed to contain the mineral-associated organic matter (MOM or heavy) fraction. The pellet was washed three times with 40 mL of distilled water, dried, weighed and milled in the same way as the fPOM and oPOM fractions. The organic carbon (C) and total nitrogen (N) content of the three fractions were determined through thermal oxidation by dry combustion using an elemental analyser (LECO Corp.). Basic descriptive statistics were calculated for the data on the fractionated calibration sites, including mean absolute and relative proportions of the SOC fractions divided between different land uses and soil texture classes. An ANOVA was conducted to

determine whether the differences between cropland and grassland land uses were significant and to test for significant differences between soil texture classes. The Games Howell post-hoc test was used for this purpose.

2.4 Near-infrared spectroscopy and chemometrics

As the oPOM fraction generally contained a small proportion of total SOC, it was combined with the fPOM fraction to give a 'light fraction' for the purpose of prediction. Soil samples were dried at 40°C, sieved through a 2 mm sieve and finely milled in a rotary mill. Before analysis, the samples were dried again at 40°C and equilibrated to room temperature for a few minutes in a desiccator. The soil samples were scanned with spot size 4 cm diameter in a Fourier-Transform near-infrared spectrophotometer (FT-NIRS, MPA - Bruker Optik GmbH, Germany). Spectral data were measured as absorbance spectra (A) according to A = log (1/R), where R is the reflectance expressed in wave number from 11000 to 3000 cm^{-1} (NIR region) with 8 cm^{-1} resolution and 72 scans. The final spectrum was obtained by averaging two replicates.

To improve the model accuracy a spectral pre-treatment was applied, using Savitzky-Golay first derivative smoothing (3 points) and wavelength selection from 1330 to 3300 nm, since these regions contain the main absorbance information. The calibration set consisted of the 145 calibration site samples, and the remaining samples were used for prediction. Partial least squares regression (PLSR) was performed in the pls package (Mevik et al., 2015), based on near-infrared (NIR) spectra and reference laboratory data. A cross-validation was applied using leave-one-out to avoid over- and under-fitting. To obtain the carbon fractions and ensure that the sum of light and heavy fractions was equal to total SOC content, the log ratio of the light and heavy fraction was predicted (Jaconi et al., in prep.). Model performance was evaluated using the root mean square error of cross-validation (RMSECV), Lin's concordance correlation coefficient (ρc) and the coefficient of determination (R^2) between predicted and measured carbon content in the fractions. In addition, residual prediction deviation (RPD) was calculated, using the classification system devised by Viscarra Rossel *et al*. (2006).

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NIRS in combination with chemometric methods was found to give accurate prediction of the carbon content in light and heavy fractions of the soil. For prediction of carbon content in the fractions (g kg^{-1}), the coefficient of determination (R^2) between predicted and measured carbon content in the fractions was found to be 0.87-0.90 and RMSECV was 4.37 g kg^{-1}. The RPD was 2.9 for the prediction of carbon content in the light fraction and 3.2 for the prediction in the heavy fraction. For prediction of carbon proportions in the fractions (%), R^2 was 0.83, RMSECV 11.45% and RPD 2.4 (Fig. S1; for more details see Jaconi *et al.*, submitted). The accuracy of prediction of both SOC content and proportions of the light and heavy SOC fractions was very good and was comparable with that in other studies that have used NIRS to predict SOC fractions (Cozzolino and Moro, 2006; Reeves et al., 2006). It can thus be concluded that prediction of SOC fractions with NIRS is a fast, inexpensive and accurate method.

2.5 Drivers of soil organic carbon distribution in fractions

A total of 75 potential drivers of differences in carbon proportions in different fractions was compiled from the soil analysis data, complemented with data from a farm survey and geographical data (for a complete list of predictors, see Table S2). The farm survey related to management practices implemented over the 10 years prior to sampling. Using this information, carbon and nitrogen inputs and outputs were calculated for the sites. When data were missing in the survey responses, yields were calculated using regional yield estimates. Carbon and nitrogen inputs through mineral or organic fertiliser were also calculated based upon the survey data. Climate and site data acquired from GIS data layers completed the set of predictor variables (climate data from Deutscher Wetterdienst, normalised difference vegetation index (NDVI) data from ESA, elevation data from Bundesamt für Kartographie und Geodäsie). For the sites in the federal states of Lower-Saxony (north-western Germany) and Mecklenburg-Western Pomerania (north-eastern Germany), the land-use history was researched using historical maps (dating back to 1873-1909), as many regions in these states are known to have a heathland or peatland legacy.

The conditional inference forest algorithm (cforest; Hothorn *et al.*, 2006), was used to identify the most influential drivers of SOC distribution among the different fractions. Cforest is an ensemble model and uses tree models as base learners that can handle many predictor variables of different types and can also deal with missing values in the dataset (Elith et al., 2008). The cforest algorithm is similar to the better known random forest algorithm, a non-parametric data mining algorithm that uses recursive partitioning of the dataset to find the relationships between predictor and response variables (Breiman, 2001).

Bootstrap sampling without replacement was carried out in order to prevent biased variable importance (Strobl et al., 2007). Ten cforest models were created, each containing 1000 trees and using different random subset generators. From these models, the variable importance of predictors was extracted and the relative variable importance was calculated and averaged over all 10 models. Variables were considered important when their relative variable importance was higher than 100/n, where n is the number of predictors in the model. This is the variable importance that each variable would have in a model where all variables are equally important (Hobley et al., 2015). It should be noted that the relative variable importance value obtained from the cforest algorithm does not necessarily imply direct relationships between the proportion of SOC in the light fraction and the main drivers, as the algorithm also takes into account interaction effects between the variables. Model performance was assessed by the coefficient of determination (R^2), as defined by the explained variance of out-of-bag estimates, which represent a validation dataset:

$$R^2 = 1 - \frac{MSE_{OOB}}{Var_z} \qquad (1)$$

where MSE_{OOB} is the mean squared error of out-of-bag estimates and Var_z is the total variance in the response variable.

A range of soils in northern Germany, called 'black sands', behaved quite differently from other soils in the country in terms of the driving factors for SOC distribution among the fractions. Therefore the

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dataset was split into two parts for the cforest analysis and the cforest algorithm was used on: 1) the dataset containing only the black sands from northern Germany (n=264). Those were extracted using the NIR spectra, which were classified into black sands and normal soils using the simca function in the "mdatools" package (Kucheryavskiy, 2017); and 2) on all other soils considered not to be black sands (n=2406). All statistical analyses were conducted using the software R (R, 2013). Maps were generated with the software QGIS.

3 Results

3.1 Carbon distribution among measured fractions (calibration sites)

The fPOM fraction contributed an average of 23% to bulk SOC (23% ±2.36 (mean ± standard error (SE)) in croplands and 25% ±3.79 in grasslands (Fig. 1). The oPOM fraction accounted for an average of 4% of SOC (3% ± 0.49 in croplands, 8% ±1.26 in grasslands) across all calibration sites (Fig. 1). The heavy fraction contributed the largest proportion to bulk SOC (73% in all soils, 73% ± 2.46 in croplands and 68% ± 4.43 in grasslands). The differences between land uses were not significant. There was great variation in the carbon distribution between the fractions, with the fPOM fraction contributing between 3 and 99% to bulk SOC. The absolute carbon content (g kg^{-1}) of the fractions was significantly different for the heavy fraction, with grasslands having significantly higher heavy fraction carbon content than croplands (31 g kg^{-1} compared with 13 g kg^{-1}).

There were significant differences in the contribution of the different fractions to bulk SOC depending on the main soil texture class (Fig. 2). In sandy soils, the fPOM fraction contributed significantly more and the heavy fraction contributed significantly less to bulk SOC than in other soils. For the oPOM fraction, the difference between sandy soils and clayey, silty and loamy soils was not significant. The absolute SOC content (g kg^{-1} soil) was significantly higher in the heavy fraction of clayey soils than in the heavy fraction of all other soil textures and it was significantly higher in the oPOM fraction of sandy soils than in the fPOM fraction of all other soils.

3.2 Influences on soil organic carbon distribution among fractions (calibration and prediction sites)

With the machine-learning algorithm cforest, 75 variables that may act as drivers for the regional distribution of SOC fractions were evaluated (Fig. 3a). For the ‘normal’ soils (non-black sands) dataset, soil texture had the highest explanatory power in predicting the contribution of the light fraction to bulk SOC (Fig. 4), with clay content being negatively and sand content positively correlated with percentage of SOC in the light fractions. The SOC content, bulk soil C/N ratio, land use, soil type, pH and $CaCO_3$ content were also identified as important explanatory variables. The SOC content showed a positive relationship with light-fraction SOC proportion and with bulk soil C/N ratio. The grassland soils showed a higher proportion of bulk SOC in the light fraction than the cropland soils and pH was negatively related to the light-fraction SOC proportion.

The analysis of historical land use data of northern Germany confirmed that the former peatland, heathland and grassland sites had significantly higher (($p<0.01$) proportions of bulk SOC in the light fraction than sites used as cropland in the same period (Fig. 5a). These historical peatland, heathland and forest sites also had significantly higher ($p<0.05$) C/N ratio than the historical cropland and grassland sites (Fig. 5b). Regarding the total SOC content, historical peatland and grassland sites had significantly higher ($p<0.001$) values than historical croplands (Fig. 5c).

For the black sands dataset, bulk soil SOC content was the most important driver of SOC distribution in the fractions (Fig. 3b), followed by C/N ratio, soil temperature in summer and soil bulk density. The SOC content had a positive relationship with percentage of SOC in the light fraction, and with C/N ratio (Fig. 4). For soil temperature there was no clear relationship. There was a negative relationship between SOC proportion in the light fraction and soil bulk density.

3.3 Distribution of soil organic carbon into fractions across Germany

Regions featuring high proportions of SOC in the light fraction (over 40%) nearly all lie in northern Germany (Fig. 7). Medium proportions of SOC in the light fraction (40-60%) were found in Mecklenburg-Western Pomerania and in parts of Brandenburg (north-east Germany). Low proportions (< 40 %) of SOC in the light fraction were found in central and southern Germany.

4 Discussion

4.1 Contribution of soil organic carbon fractions to bulk soil organic carbon

The distribution of carbon among different fractions did not differ significantly between croplands and grasslands, which is in agreement with previous findings for south-east Germany (Wiesmeier et al., 2014). This may partly be due to historical conversion of cropland to grassland still affecting carbon distribution in the fractions. Grasslands and croplands are often located on different soil types, and thus it is not possible to draw direct conclusions on land-use change effects on carbon fractions from such regional inventories. In a previous study using paired land-use change sites, the POM proportion was found to be twice as high in grasslands as in croplands (Poeplau and Don, 2013b). Even though the fraction distribution did not differ significantly between croplands and grasslands in the present study, there was a trend for slightly higher fPOM content in grasslands than in croplands. The significant differences observed in the SOC content of fractions between different land uses were to be expected, as grassland soils in Germany contain on average more than twice as much SOC in the upper 10 cm as cropland soils (42±16 g kg^{-1} compared with 17±9 g kg^{-1}).

4.2 Black sands in Germany

All samples with medium or high proportions of SOC in the light fraction were found to originate from northern Germany. This is the area in which the black sands are present, which store large parts of their SOC in the light fraction. Springob & Kirchmann (2002a) examined the presence of black sands in Lower Saxony in Germany and linked it to the land-use history. In Ap-horizons of soils formerly used as heathland or plaggen, they found a high fraction of SOC resistant to oxidation with HCl. This HCl-resistant fraction was positively correlated with the total SOC content, but soil microbial biomass carbon content showed a negative relationship with total SOC and, when incubated, the specific respiration rates were lowest for the soils with the highest SOC content (Springob & Kirchmann, 2002a). Those authors concluded that a large proportion of the organic matter in the former heathland soils is resistant to decomposition and suggested that low solubility of the SOC could be responsible for its high stability. A recent study (Alcántara et al., 2016) reported similar results for sandy soils under former heathland, which had lower respiration rates per unit SOC and a wider range of C/N ratios than control soils without a heathland history. Certini *et al.* (2015) showed that SOC under heathlands is rich in alkyl C and contains high contents of lipids, waxes, resins and suberin, all of which hinder microbial degradation. This confirms the claim that sandy soils under former heathland and contain high contents of stable SOC even though they also contain a large amount of POM. In such soils, the POM fractions may not be directly linked to higher turnover rates and lower stability.

"Historical" peatlands may have lost much of their former carbon stocks due to a number of reasons: Drained peatlands emit huge amounts of CO_2 (German grasslands on average 27.7 to CO_2 ha^{-1} yr^{-1}, (Tiemeyer et al., 2016)) until the peat has virtually vanished. There might have also been peat extraction, and the remaining peat layer might have been mixed with underlying sand. Finally, former peatland soils were often mixed with large amounts of sand in order to make them usable for arable cultivation, but still often contain substantial proportions of (degraded) peat and therefore have relatively high SOC content, with a large part of the SOC in the light fraction. It has been found elsewhere (Bambalov, 1999; Ross and Malcolm, 1988; Zaidelman and Shvarov, 2000) that the SOC content in sand-mix cultures declines rapidly after mixing with sand and that the decline increases with increasing intensity of mixing. In a 15-year long-term trial, Bambalov (1999) found that the SOC content of a sand-mix culture could only be stabilised (at much lower SOC content than the original peat) by adding organic and mineral fertilisers to the soil. In contrast, Leiber-Sauheitl et al. (2014) found that a peat-sand mixture with a SOC content of 93 g kg^{-1} emitted as much CO_2 as an adjacent shallow "true" peat. Similarly, Frank *et al.* (2017) determined a higher contribution of soil-derived dissolved organic carbon at a peat-sand mixture compared to the peat, which points to a low stability of the SOC in this kind of soils. This means that, for the light fraction of the former peatlands in northern Germany, enhanced stability of the POM cannot be assumed. Thus, for more accurate interpretation of results, the black sands had to be divided into a former heathland group, containing a relatively stable light fraction, and a former peatland group, containing a relatively labile light fraction, although there are transitional vegetation types with heath on peatlands.

Land-use history clearly continues to influence soil SOC dynamics, since the light-fraction SOC proportion and the bulk soil C/N ratio were higher in soils with a heathland or peatland history in the present study. This supports findings by Sleutel *et al.* (2008) that the chemical composition of pairs of relict heathland and cultivated former heathland soils is very similar. Unfortunately former peatlands and heathlands are not necessarily distinguishable due to their SOC content and C/N ratio, so that knowledge on the land use history is necessary. In some cases, however, even the distinction on site can be difficult, e.g. on dry peatlands with heath vegetation (*Calluna, Erica*). In future studies it would therefore be interesting to incubate pairs of former heathland and peatland in order to be able to make accurate claims on the vulnerability of the light fraction SOC in these soils.

The presence of black sands poses a problem for interpretation of the SOC fractions. In most cases, the SOC in the light fraction (fPOM + oPOM fractions) is seen as representing a labile carbon pool with short turnover times. Therefore sites with high proportions of bulk SOC in the light fraction would be seen as being at risk of losing this substantial part of their SOC stock quite rapidly and easily. For the black sands, however, their former heathland land use history has led to quite stable and not easily degradable POM (Overesch, 2007; Sleutel et al., 2008; Springob and Kirchmann, 2002), while for former peatland that was drained and possibly mixed with sand the classification of the light fraction into a labile SOC pool may well be justified (Leiber-Sauheitl et al., 2014). This implies that the results need to be interpreted in a different way for black sands than for other soils.

4.3 Driving factors for carbon distribution into fractions

4.3.1 'Normal' agricultural soils (non-black sands)

The most important driver for the SOC distribution among the fractions in 'normal' soils was the soil texture (Fig. 3a). This is well in line with the frequently reported relationship between clay content and mineral-associated (heavy fraction) SOC, whereby clayey soils can stabilise SOC through mechanisms that protect it against microbial decay by absorption or occlusion (v. Lützow et al., 2006). The SOC that is bound to the mineral phase is mostly assigned to a conceptual stable SOC pool. The negative relationship between SOC content and percentage of SOC in the heavy fraction (Fig. 4) may indicate SOC saturation of the mineral fraction at rising SOC content, so that excess SOC can only be stored as particulate organic carbon.

The positive correlation between C/N ratio and C proportion in the light fraction (Fig. 4) is related to the inherent higher C/N ratio of the light fraction compared with the heavy fraction, so that a higher share of light-fraction C leads to a higher C/N ratio of the total soil. Thus C/N ratio may be useful as an indicator of SOC stability in 'normal' agricultural soils in Germany.

The fact that land use is an important driver for the distribution of SOC among the fractions is mainly due to the fact that topsoils under grassland store a significantly higher share of SOC in the light fraction than topsoils under cropland. This is in line with higher inputs of roots, which make up part of the light fraction, into grassland topsoils. The higher proportion of SOC in the light fraction was also noted in the calibration dataset, but the difference was not significant in that case.

Most arable topsoils in Germany do not contain carbonate. The 9% of arable soils that contained over 5% carbonate in this study consistently had a high proportion of heavy-fraction carbon and were therefore classified as containing mainly stabilised SOC (Fig. 4). Calcium bridges may foster absorption of SOC onto mineral surfaces and, via an active soil fauna, high pH enhances the turnover and transformation of SOC from recently added biomass to mineral-associated SOC that can be stabilised via absorption (Oades, 1984). In general, there was a trend for a higher proportion of SOC in the light fraction with lower pH (Fig. 4), which is well in line with the finding by Rousk *et al.* (2009) that SOC mineralisation is slower in soils with lower pH due to a higher ratio of fungal to bacterial biomass.

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The influence of soil type is mainly due to the Podzol soils storing a much higher proportion of bulk SOC in the light fraction than all other soil type classes (Fig. 6). Podzols often develop on sandy soils and therefore do not have a high capacity for SOC stabilisation in the heavy fraction.

4.3.2 Black sands

In the dataset containing only the black sands, soil total SOC content was the most important driver for the SOC distribution among the fractions, with increasing light fraction with increasing SOC content (Fig. 4). On the one hand, this could indicate saturation of the heavy fraction at high SOC contents, which would lead to further storage in the light fraction only, as already mentioned above for 'normal' soils. Another possible explanation is that those soils with the highest SOC content in the dataset are degraded peatlands, in which a high percentage of the SOC ends up in the light fraction. On former heathlands, the soil total SOC content is also quite high compared with that in other sandy soils and the light fraction is mainly built up from *Calluna vulgaris* litter, since *Calluna* vegetation dominates on many heathlands. *Calluna* litter contains very stable SOC due to high contents of lipids, long-chain aliphatics and sterols, and may persist in the light fraction of soil for decades or even centuries (Sleutel et al., 2008).
There is a close link between land-use history as peatland and heathland and soil C/N ratio, with high C/N ratio in former heathland soils (Alcántara et al., 2016; Certini et al., 2015; Rowe et al., 2006) and also often in former peatlands (Aitkenhead and Mcdowell, 2000). Therefore it is evident that land-use history is a main driver for the high proportions of bulk SOC found in the light fraction in these soils. This is well in line with the significantly higher C/N ratios reported for soils in Lower-Saxony and Mecklenburg-Western Pomerania, which were under heathland or peatland more than 100 years ago (Fig. 5). The influence of land-use history reinforces the relationship between C/N ratio and the light fraction.

In black sands, there was a significant negative relationship between soil temperature and the light-fraction SOC proportion, but this was not found for the other soils (Fig. 4). A negative relationship

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was observed between soil bulk density and proportion of SOC in the light fraction, which was evidently due to the low density of the light fraction affecting overall soil bulk density (Fig. 4).

Even though the land use history was part of the dataset and we could link several of the important driving factors to a history as peatland or heathland, the cforest algorithm did not identify the land use history as important driver for the SOC distribution into fractions. This was the case because we did not have the detailed land-use history data for all sites. But even when running the cforest algorithm only for those sites with known land-use history, it was not selected as important driver. This is probably due to the fact that at the time of the land survey in 1873-1909 some of the former heathland and peatland sites had already been cultivated. Therefore the land-use history would not prove as a reliable indicator. We could confirm this by referring to an older land survey, dating back to 1764-1785. For sites that exhibited typical black sand features (e.g. high SOC proportions in light fractions, high sand content, and high C/N ratio) but were not a heathland and peatland in the 19th century, we often found a heathland or peatland signature on the maps from the 18th century. Unfortunately this land survey from the 18th century is incomplete and we could therefore not rely on it for all sites.

4.4 Hot regions of labile and stable carbon in Germany

Taking together all the important explanatory variables discussed above, regions in which the SOC can be classified as mostly labile were identified. These were soils with a high proportion of bulk SOC in the light fraction and without a heathland history. Such soils are mainly located in northern Germany and some have a peatland history (Fig. 7). These soils can be seen as vulnerable to losses of a large proportion of their SOC in the topsoil easily and rapidly. Loss of SOC could occur e.g. through a change in management that reduces carbon inputs to the soil and therefore fails to maintain the light fraction, for example a land use change from grassland to cropland (Poeplau et al., 2011) or reduced input of organic fertilisers or crop residues (Dalal et al., 2011; Srinivasarao et al., 2014). Losses of SOC could also occur due to higher temperatures, which could lead to enhanced microbial activity and therefore enhanced mineralisation of SOC in the light fraction (e.g. Knorr *et al.*, 2005). In the case of former peatlands many soils may already be losing significant parts of their SOC (Leiber-Sauheitl et al., 2014; Tiemeyer et al., 2016).

For a soil to be definitively identified as being vulnerable to SOC losses, it not only needs to have a high proportion of bulk SOC in the light fraction, but also a high absolute SOC content in this fraction. The map in Fig. 8 shows the absolute SOC content of the light fraction at sites of the German Agricultural Soil Inventory. Comparing Fig. 7 and Fig. 8, it is evident that sites which store a high proportion of their SOC in the light fraction generally also have high absolute SOC content in the light fraction. This implies that those sites are really the most vulnerable to SOC losses, as they not only have high proportions of SOC in the light fraction, but also the highest absolute SOC content in the light fractions to lose. As the SOC in former peatland soils has been shown to be easily mineralised (Bambalov, 1999), management of such sites should be aimed at stabilising the SOC stocks and preventing further degradation of the peat. When there is a heathland history, it can be assumed that the SOC in the light fraction is quite stable, but that does not imply that freshly added litter will also be stable. In fact, it is quite likely that it will not be stable if no heathland vegetation is planted. This implies that the SOC stocks on these sites will decline when the resistant litter is not replenished.

Regions with soils with a high proportion of stable SOC are located mainly in central and southern Germany (Fig 7). In these regions, soils consistently store over 60% of their SOC in the heavy fraction, in which the SOC is bound mostly to the mineral surfaces of clay minerals. Thus, these soils have the lowest vulnerability to losing their SOC, as losses mostly occur from the light fraction. However, even in these regions up to 40% of bulk SOC is stored in the light fraction and this may be lost. Therefore apparent lower vulnerability does not mean that SOC-conserving soil management is not needed in these regions. It should be noted that the quality of the SOC in the light fraction is probably not the same in all soils, land-use (history) and climate regions. Therefore, the vulnerability and turnover time of the light fraction may also vary considerably within different regions. This can be seen in the light fraction C/N ratio for example, which ranged between 11 and 43 for the 143 calibration sites studied here.

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5 Conclusions

Identification of the distribution of SOC fractions in German soils allowed clear identification of regions where the SOC in agricultural soils is most vulnerable to being lost. The cforest analysis provided indications of the factors driving the distribution of SOC into the different fractions. It was found that soil texture, bulk soil SOC content, bulk soil C/N ratio, land-use history and pH were the main drivers for this distribution in 'normal' soils. In 'black sand' soils in northern Germany, the SOC distribution into the fractions mainly depended on total SOC content and soil C/N ratio and was directly linked to the land-use history. Former peatland or heathland still has a great influence on the composition of soil SOC decades or even centuries after cultivation of the soil. In some regions of Germany the majority of bulk SOC is stored in the light fraction, but this does not always imply that this SOC is labile. Use of SOC fractionation techniques coupled with NIR spectroscopy to extrapolate to a national soil inventory dataset was successful in predicting POM factions. However, additional knowledge on land-use history was required to determine whether this POM is vulnerable to losses or not. This study focused on the topsoil only, as it has comparatively high SOC stocks and is most vulnerable to changes in management. Future studies should also examine the SOC distribution in the subsoil, as this would enable exploitation of all possibilities for sequestering additional SOC in the soil, in order to mitigate the CO_2 content in the atmosphere. Regarding soil management measures, this study provided indications on where the most prudent and SOC-conserving management techniques are advisable for different regions of Germany.

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Figures

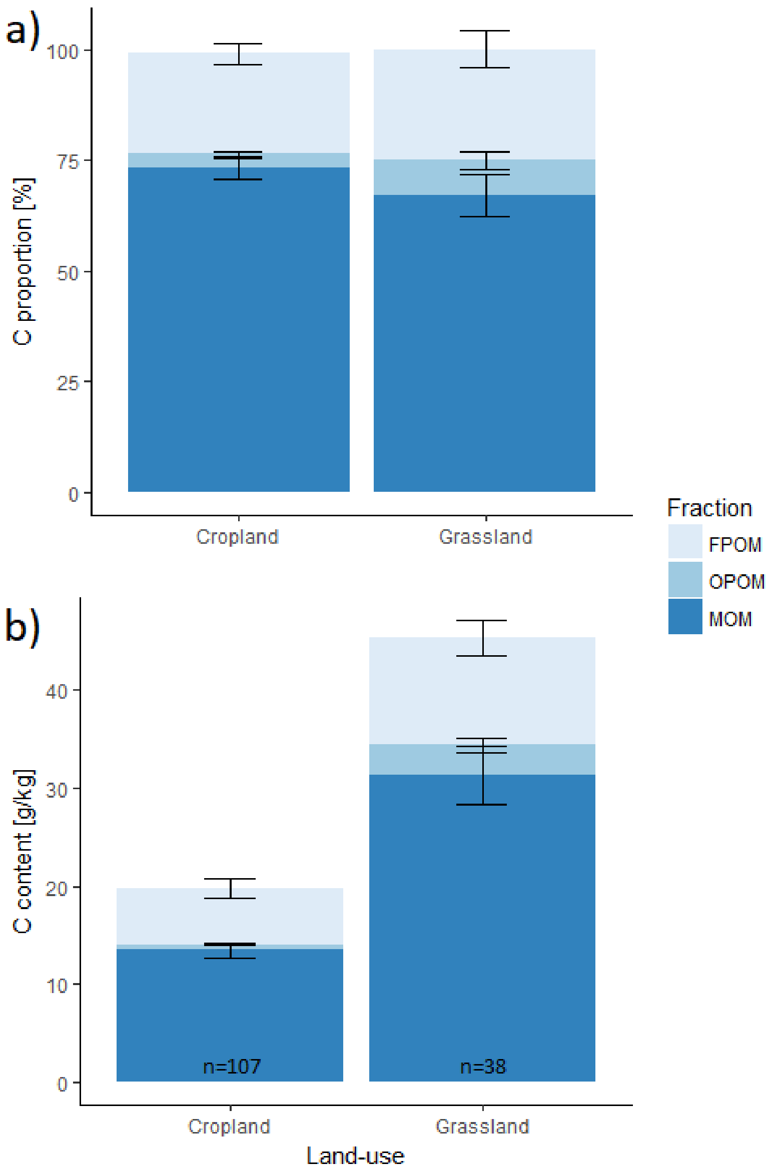

Fig. 1: a) Proportion (%) and b) absolute content (g kg^{-1}) of soil organic carbon (SOC) in the free particulate organic matter (fPOM), occluded particulate organic matter (oPOM) and mineral-associated organic matter (MOM) fraction in soils under cropland and grassland for the 145 calibration sites that were fractionated. Error bars denote standard error of the mean.

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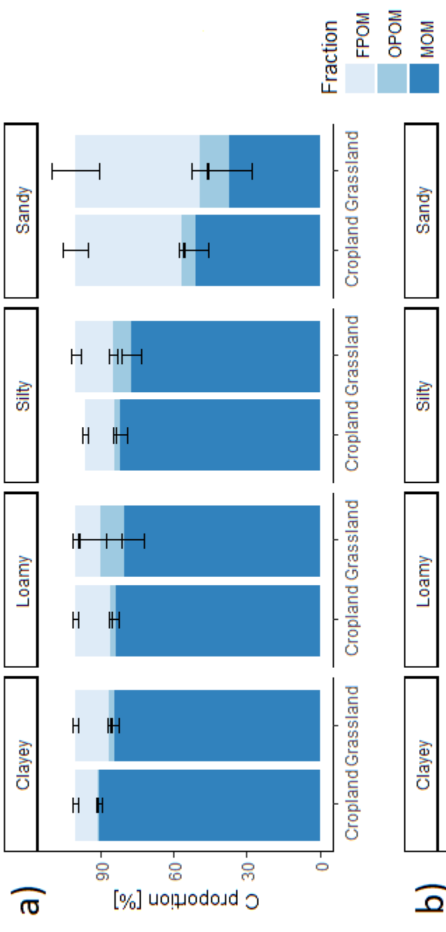

Fig. 2: a) Proportion (%) and b) absolute content (g kg^{-1}) of soil organic carbon (SOC) in the free particulate organic matter (fPOM), occluded particulate organic matter (oPOM) and mineral-associated organic matter (MOM) fraction in different soil texture classes for the 145 calibration sites that were fractionated. Error bars denote the standard error of the mean.

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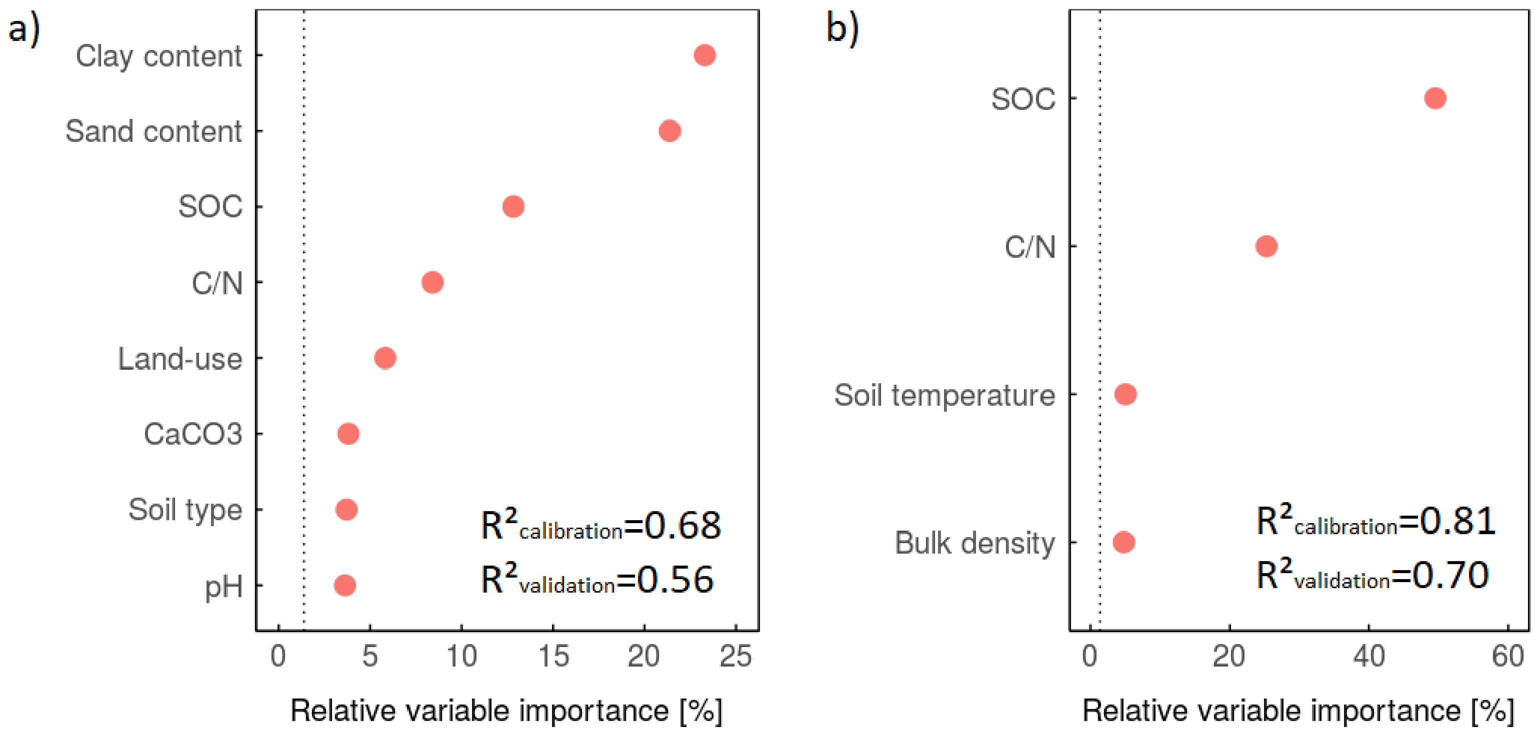

Fig. 3: Mean relative variable importance according to conditional inference forest (cforest) algorithm for predicted proportion of soil organic carbon (SOC) in the light fraction. The vertical line indicates the threshold value of relative variable importance above which a variable was regarded as important. a) Variable importance for all soils that are not black sands and b) variable importance for only black sands.

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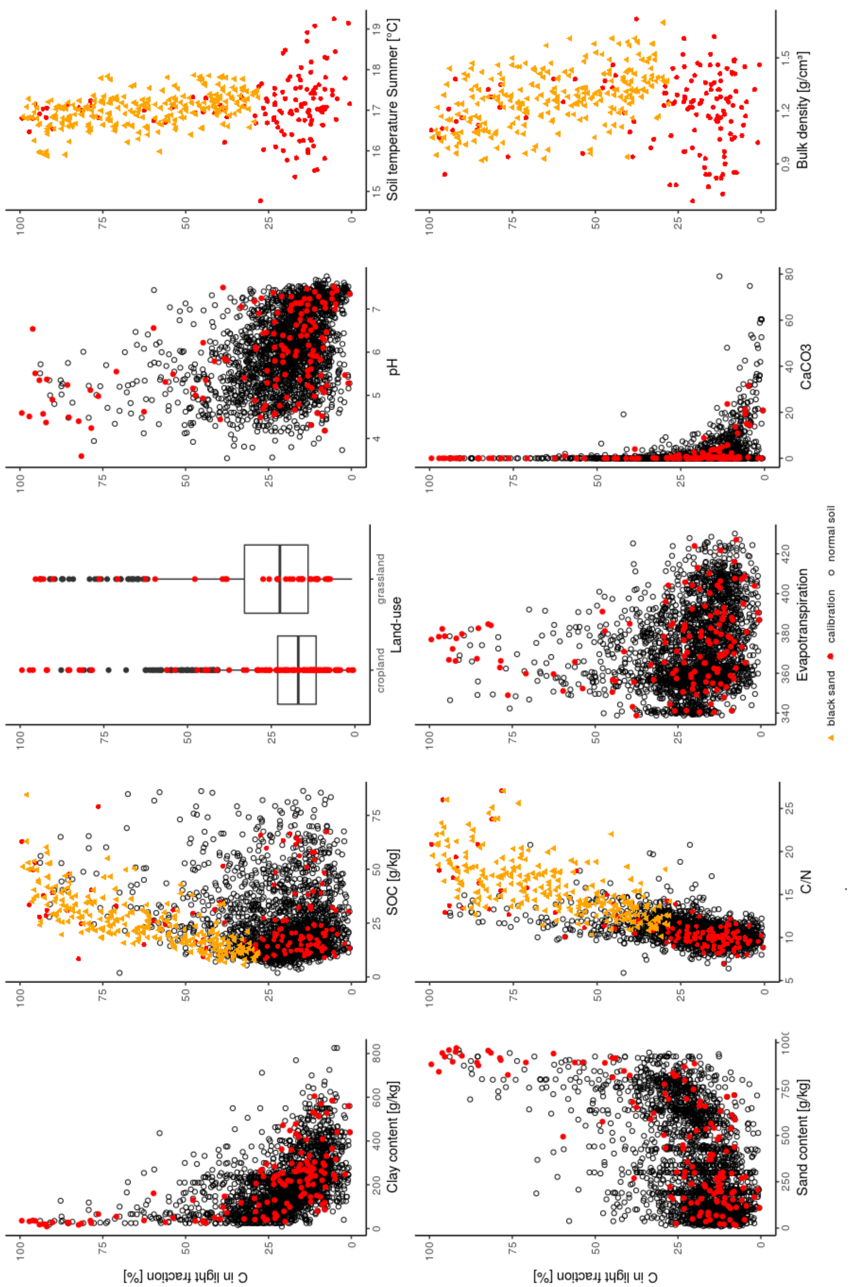

Fig. 4: Relationship between soil organic carbon (SOC) proportion in the light fraction and influential variables. Calibration sites are shown as red dots, normal soils as black dots and black sands as orange triangles.

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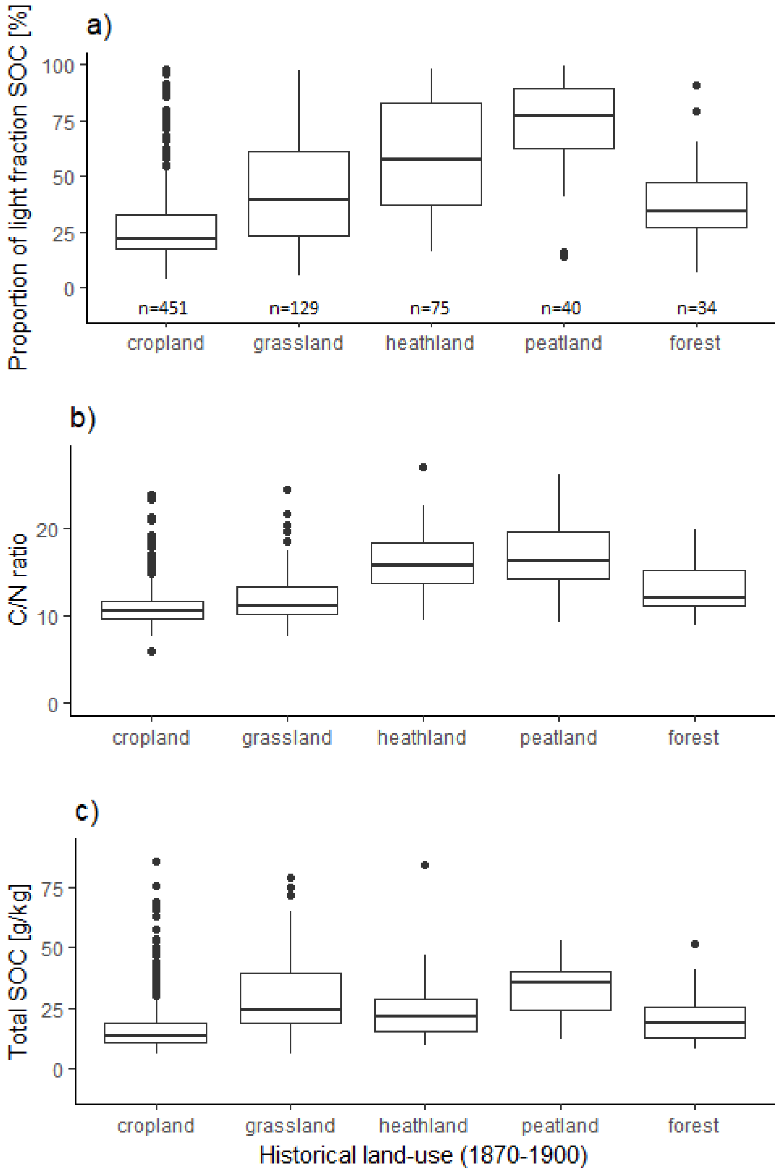

Fig. 5: Relationship between land-use history and a) proportion of light fraction soil organic carbon (SOC), b) carbon/nitrogen (C/N) ratio and c) total SOC content for all sites in the federal states of Lower-Saxony (north-western Germany, n=491) and Mecklenburg-Western Pomerania (north-eastern Germany, n=243).

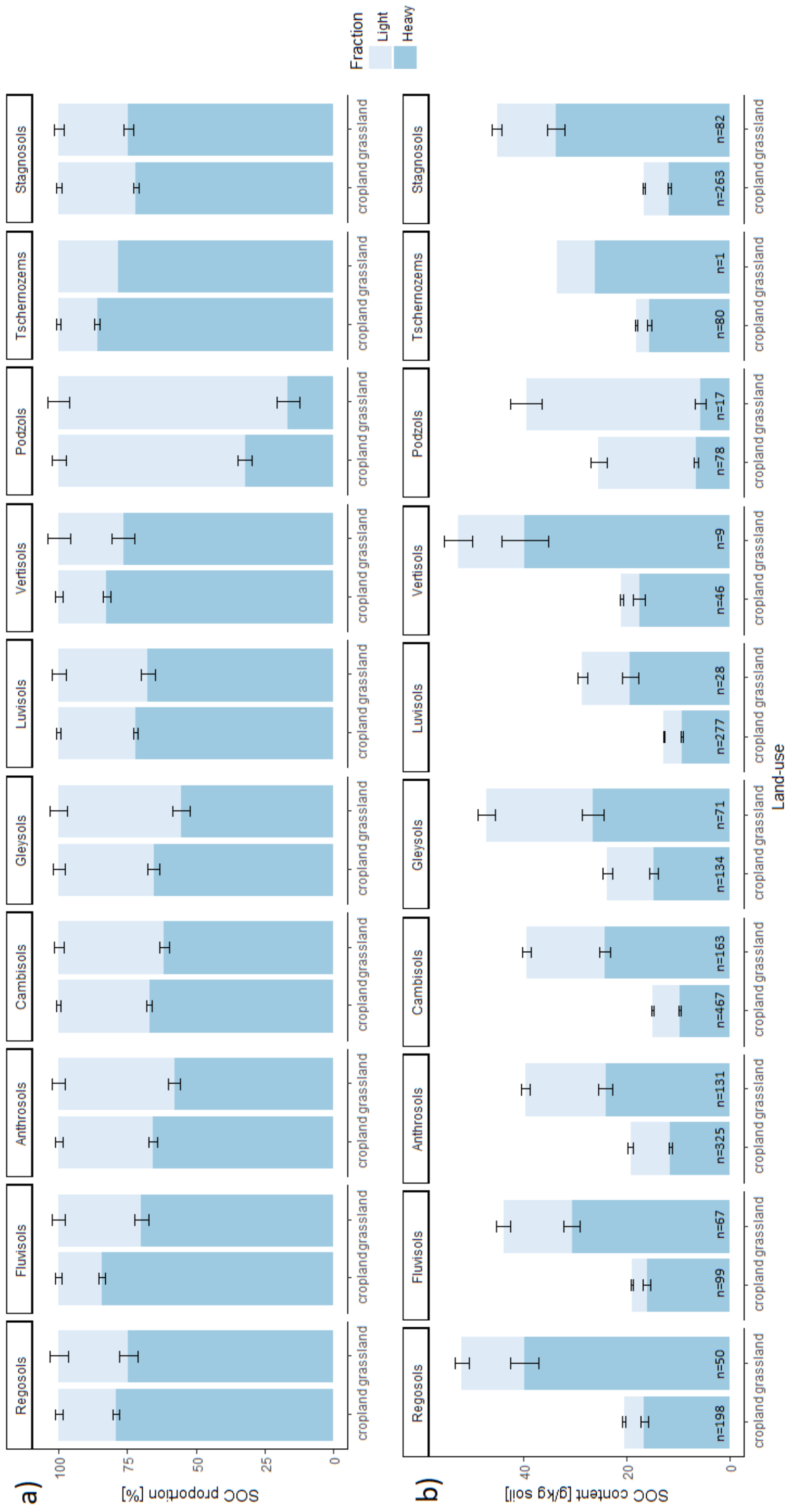

Fig. 6: a) Proportion (%) and b) absolute content (g kg^{-1}) of soil organic carbon (SOC) in the light and heavy fractions in different soil types in the 'normal' soils (non-black sands) dataset. Error bars denote standard error of the mean.

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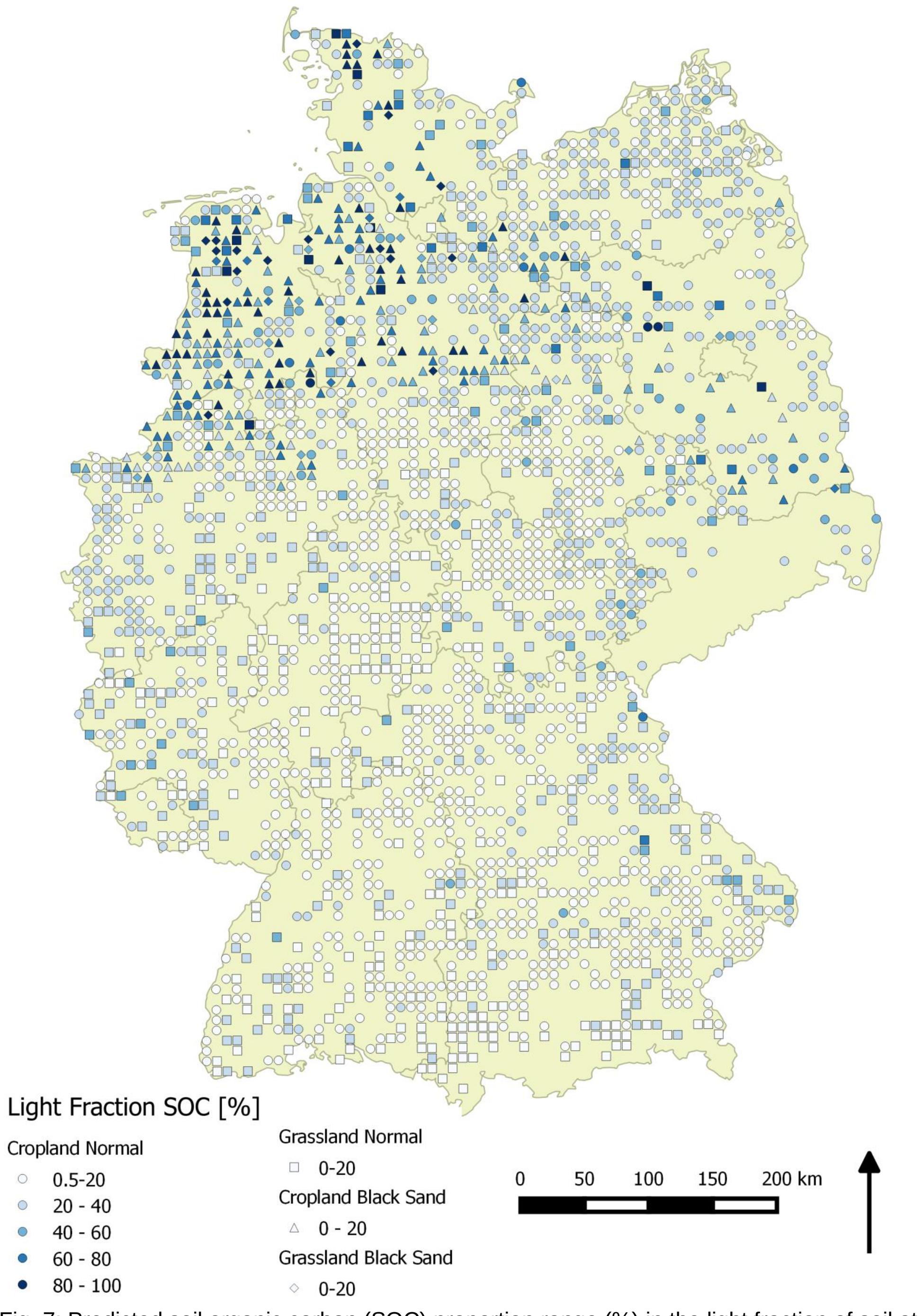

Fig. 7: Predicted soil organic carbon (SOC) proportion range (%) in the light fraction of soil at sites in the German Agricultural Soil Inventory.

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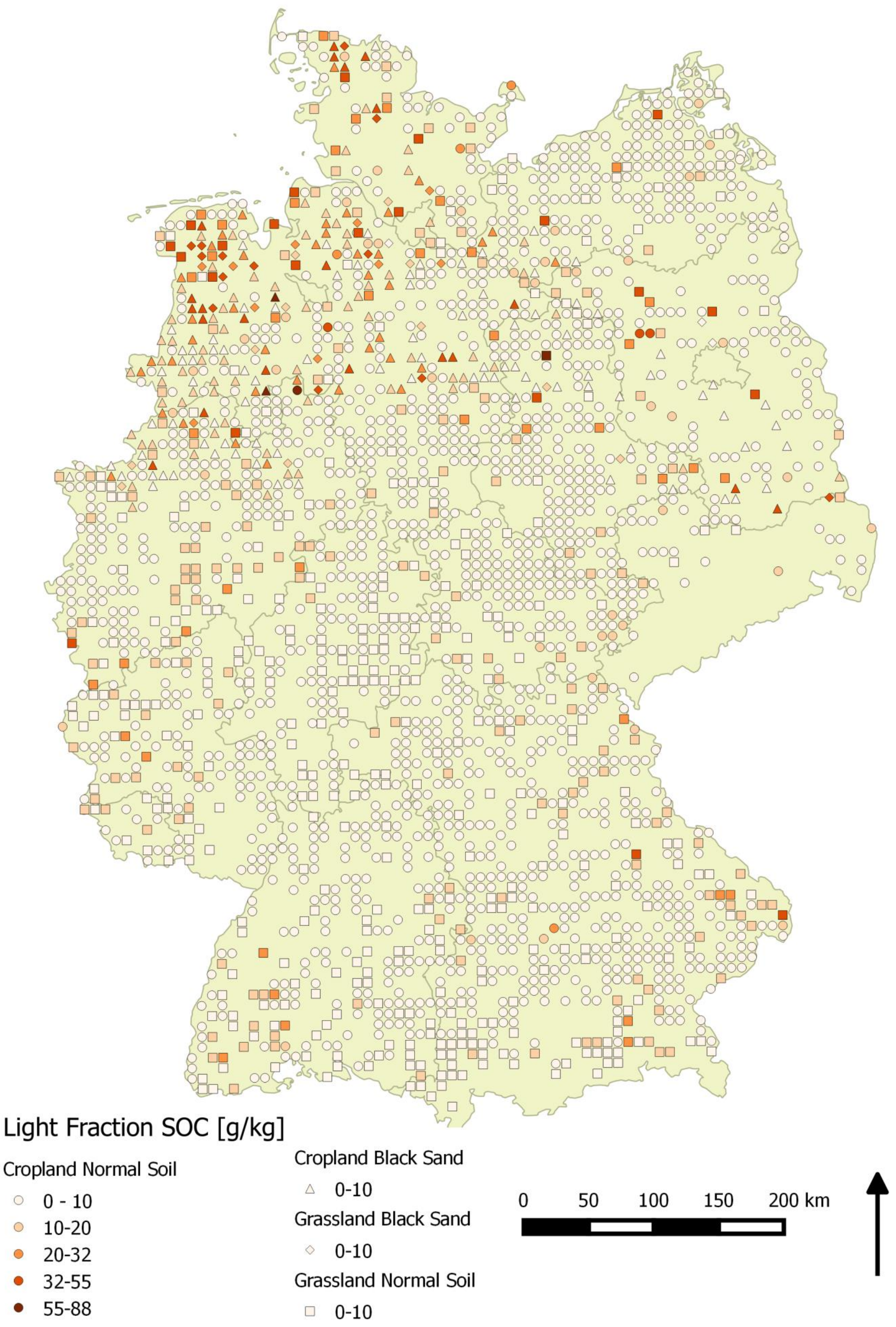

Fig. 8: Predicted absolute soil organic carbon (SOC) content range (g kg^{-1}) in the light fraction at sites in the German Agricultural Soil Inventory.