

Estimating organic carbon stocks of mineral soils in Denmark: Impact of bulk density and content of rock fragments

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ABSTRACT

Management measures to reduce atmospheric carbon dioxide concentrations by increasing soil organic carbon (SOC) storage need verification, e.g., by periodic sampling of soils to estimate resulting changes in SOC stock. Estimates of SOC stocks are affected by content of rock fragments (systematic bias) and soil bulk density (random but significant effect), both of which may vary significantly between soils. We investigated the importance of using site-specific bulk density and correcting for rock fragment content on estimates of SOC stock in 0–50 cm depth of agricultural mineral soils, collected in 2019 in the Danish National Square Grid. We found that use of an average bulk density value for a given soil type category produced valid estimates of SOC stocks for regional/national inventories. However, large variations in bulk density were found within a given soil type category, which can result in over- or under-estimation at local sites. This calls for measurement of site-specific bulk density and rock fragment content to produce valid estimates of field-scale SOC stock, e.g., to be used in farm carbon credit schemes.

1. Introduction

Sequestration of atmospheric carbon dioxide (CO₂) in soil organic carbon (SOC) pools of agricultural land remains intensively debated as a means of mitigating global warming (Powlson et al., 2011; Minasny et al., 2017; Rumpel et al., 2020; Smith et al., 2020). Increased incorporation and stabilization in soils of carbon (C) extracted from the atmosphere by photosynthesis could, at least temporarily, counteract the increase in the atmospheric CO₂ concentration. At the same time, SOC has beneficial effects on a range of soil ecosystem functions, such as aggregate stability, water holding capacity, microbial activity and nutrient cycling (Schjønning et al., 2004; Adhikari and Hartemink, 2016). The SOC pool in the upper one meter of soils contains about twice the amount of C in the atmosphere, wherefore even small changes in global SOC storage have a significant impact on atmospheric CO₂ concentrations (Jobbagy and Jackson, 2000; Rumpel, 2014). The quantity of SOC in soils reflects mainly the balance between C inputs and the release of gaseous C associated with soil microbial activity. The capacity

of agricultural soils to accumulate additional inputs of plant-derived C depends on initial SOC contents and management regimes (Peltre et al., 2016; Jensen et al., 2022), and thereby the time it takes for a given soil to reach a new steady-state level of SOC. Consequently, the potential of soils to accommodate organic C is finite (Powlson et al., 2011). Moreover, management options implemented to store greater amounts of atmospheric CO₂-C in soil are reversible and may affect the release of other greenhouse gases (GHG), such as nitrous oxide (Powlson et al., 2011).

The effect of any SOC-increasing initiative on SOC stocks needs verification either by resampling of previously sampled locations after varying periods of time or by baseline determination of SOC stocks combined with simulation models that have been thoroughly validated against empirical data sets preferably from long-term field experiments (Taghizadeh-Toosi and Olesen, 2016). Initiatives to document and monitor changes of SOC stocks in mineral agricultural soils are pursued, e.g., in Germany, France and Denmark, with changes in SOC stocks being included in national inventories of GHG emissions (Smith et al.,

Abbreviations: BD_{db}, database average total soil bulk density; BD_{fine}, site-specific fine-soil bulk density; BD_{total}, site-specific total soil bulk density; JB category, Jordbunds-kategori (from Danish) soil type category; SOC, soil organic carbon.

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2020). A high degree of accuracy in SOC stock estimates is essential to validate the impact of national policies for offsetting GHG emissions through enhanced SOC storage. Site-specific information on rock fragment content and soil bulk density at different soil depths have previously been reported to be essential for the accuracy of SOC stock determinations, especially for soil types rich in rock fragments (Schrumpp et al., 2011; Rytter, 2012; Poeplau et al., 2017). However, this information may not always be available.

In Denmark, approximately 60% of the land area is in agricultural use, with mineral soils (<6% SOC) accounting for 2.5×10^6 ha. Danish national stocktaking of SOC in arable soils relies on a system of sites (also termed grid areas; each $50 \text{ m} \times 50 \text{ m}$), established in 1986 at farmer's fields and designed as a grid of sites at a mutual distance of 7 km (the National Square Grid, NSG). These sites have been used for determination of changes in SOC by decadal resampling. Taghizadeh-Toosi et al. (2014) reported data for samplings in 1986, 1997 and 2009, with calculations of SOC stocks guided by soil bulk densities retrieved from a national soil profile database (Krogh et al., 2003).

A fourth SOC sampling campaign was performed in 2019. This campaign also included measurements of rock fragment content and site-specific soil bulk density in the 0–50 cm soil layers. These parameters were not included in previous samplings. The objective of the current study was to evaluate the importance of site-specific information on rock fragment content and soil bulk density for inventories of SOC stocks in Danish agricultural soil types, using statistical analyses. We compared the more precise calculation method applying site-specific information (Poeplau et al., 2017) with the previous method applying national average values of bulk density for the Danish soil categories (Taghizadeh-Toosi et al., 2014).

2. Materials and methods

The central and eastern parts of Denmark are classified as warm, humid continental climate (Köppen: Dfb), while the western part is classified as temperate, marine west coast climate (Köppen: Cfb). Mean annual precipitation is 759 mm and mean annual air temperature is 8.7 °C for the period from 1991 to 2020. Recent maps of Danish landscape types and soil properties are presented by Greve et al. (2022).

2.1. Soil sampling

This study relies on measurements in arable mineral soils classified as JB1 to JB7 (Table 1) in the Danish soil type classification system based on the texture of the 0–25 cm soil layer (Madsen et al., 1992). These soils (JB1–JB7) represent 93% of the Danish agricultural area. Soils with >6%

Table 1

Classification of Danish mineral soils (<6% SOC). Soil JB numbers refer to the Danish soil classification system and are based on the texture of topsoil (Madsen et al., 1992).

Soil type	JB Category	Clay	Silt	Fine sand	Total Sand
		<2 μm	2–20 μm	20–200 μm	20–2000 μm
		Weight %	Weight %	Weight %	Weight %
Coarse sand	1	0–5	0–20	0–50	75–100
Fine sand	2	0–5	0–20	50–100	75–100
Loamy sand	3	5–10	0–25	0–40	65–95
Loamy sand	4	5–10	0–25	40–95	65–95
Sandy loam	5	10–15	0–30	0–40	55–90
Sandy loam	6	10–15	0–30	40–90	55–90
Loam	7	15–25	0–35	0–85	40–85

SOC (organic soils) and heavy clay soils were excluded because the number of available NSG sites with these soil types was too low for statistical analysis. For the 2019 sampling campaign, performed during November 2018 to February 2019, precise GPS coordinates were used to locate the NSG sites sampled in 2009 of which 395 were available for sampling (Fig. 1). Each site (grid area) was divided in 100 cells of $5 \text{ m} \times 5 \text{ m}$ (Supplementary Fig. S1). Soil was sampled in 16 cells (randomly chosen in 2009 and revisited in 2019) using a 1-cm inner diameter gouge auger manually hammered into the soil. In few cases with hard subsoils, a percussion auger (5 cm diameter) mounted on a Gator Utility Vehicle (John Deere) was used. The extracted soil cores were divided into 0–25 and 25–50 cm depth avoiding carry-over between soil layers. The 16 soil cores from each depth layer were pooled to a composite sample for C analyses. At 24 out of the 395 sites, an additional soil sample was pooled from 16 other $5 \text{ m} \times 5 \text{ m}$ cells (selected randomly a priori) not included in the regular sampling campaign. These supplementary samples were used to evaluate within-field variation in SOC contents.

The 2019 campaign further included sampling of intact soil cores (100 cm^3) from four $5 \text{ m} \times 5 \text{ m}$ cells at each site for measurement of bulk density and rock fragment content. At each of the four cells, a small pit was excavated to expose a horizontal soil surface at ca. 10 and 35 cm depth, i.e., for collecting 100-cm^3 samples representing the 0–25 cm and 25–50 cm soil layers. The samples were collected using metal rings (6.1 cm diameter, 3.4 cm height) that were hammered into the soil using a special flange to ensure horizontal orientation. The surfaces of the soil cores were trimmed using a knife and the 100-cm^3 samples were transferred quantitatively to individual plastic bags for transport to the laboratory. During excavation and sampling, a visual assessment was made of the volumetric rock fragment content according to broad categories of <1%, 1–5%, 5–10% and >10%. These field observations were recorded and subsequently compared with the laboratory analyses of rock fragment content in the 100-cm^3 soil rings.

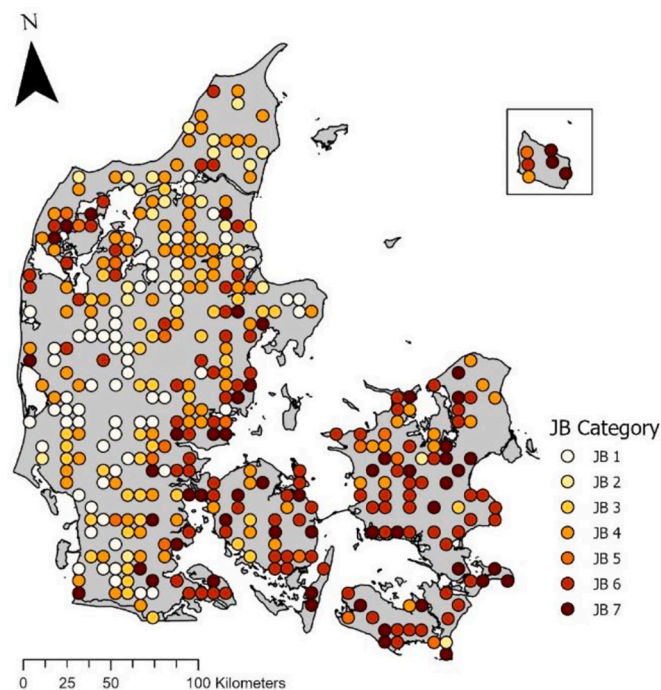


Fig. 1. Map of Denmark showing the grid-points in the National Square Grid used in the current sampling campaign. The color of circles indicates the soil category, from the sandiest JB1 soils (light) to more clayey JB7 soils (dark). See Table 1 for definition of the Danish JB soil classification.

2.2. Soil analyses

Soil samples for C analyses were air-dried for 2 days at room temperature (20 °C) and crushed manually. A representative subsample was further dried for 2 days at 40 °C, before being passed through a 2-mm sieve. Subsamples of sieved and homogenized soil (ca. 1 g) were analyzed for total C (TC) content by dry combustion using a Vario Max cube CN analyzer (Elementar Analysensysteme GmbH, Germany). Inorganic C (IC) was quantified when an effervescence test with drops of 10% HCl indicated the presence of carbonates in the soils. Measurements of IC were performed using a Scheibler apparatus where the volumetric CO₂ production was quantified after acidifying ball milled soil samples of 0.1 to 2 g (Sørensen and Bülow-Olsen, 1994). SOC was calculated as the difference between TC and IC.

The soil samples from the metal rings ($V_{\text{sample}} = 100 \text{ cm}^3$) for determination of bulk density and rock fragment content were dried at 105 °C (24 h) for measurement of the total dry weight (W_{total} , g). The soils were then washed on a 2-mm sieve to recover the rock fragments, which were dried and weighed (W_{rock} , g). The weight of the rock fragments was converted to volume (V_{rock} , cm^3) using a standard rock fragment density of 2.65 g cm^{-3} . The bulk density of the total soil sample (BD_{total} , g cm^{-3}) and that of the rock fragment-free fine soil fraction (BD_{fine} , g cm^{-3}) was calculated as:

$$BD_{\text{total}} = \frac{W_{\text{total}}}{V_{\text{sample}}} \quad (1)$$

$$BD_{\text{fine}} = \frac{W_{\text{total}} - W_{\text{rock}}}{V_{\text{sample}} - V_{\text{rock}}} \quad (2)$$

2.3. Calculations of SOC stocks

As in the previous inventory (Taghizadeh-Toosi et al., 2014), SOC contents were first converted to SOC stocks (Mg C ha^{-1}) using average estimates of total bulk density from a national Danish soil database (BD_{db}) for each soil type and depth interval (see Table 2):

$$SOC \text{ stock} = C_{\text{di}} * BD_{\text{db},i} * d \quad (3)$$

where C_{di} is the organic C content (%) in soil from the i^{th} soil layer, $BD_{\text{db},i}$ is the average database total bulk density (g cm^{-3}) of the i^{th} soil layer (Table 2), and d is the depth of the i^{th} soil layer (cm).

Next, using the site-specific estimates of rock fragment contents and bulk density of rock-free fine soils obtained in the 2019 sampling campaign, SOC stocks were also estimated as suggested by Poeplau et al.

(2017):

$$SOC \text{ stock} = C_{\text{di}} * BD_{\text{fine},i} * d * (1 - RF_i) \quad (4)$$

where C_{di} and d are defined as above, $BD_{\text{fine},i}$ is the site-specific bulk density (g cm^{-3}) of the rock fragment-free fine soil fraction of the i^{th} soil layer, and RF_i (unitless) is the volumetric rock fragment content of the i^{th} soil layer.

The importance of including site-specific bulk density and rock fragment content when estimating SOC stocks was analyzed based on the entire JB1-JB7 dataset from 2019 ($n = 395$), i.e., based on comparative calculations using Eqs. 3 and 4. The effects of improving Eq. 3 by including only site-specific bulk density or only rock fragment content was also evaluated through comparison of the resulting difference to the SOC stock estimate of Eq. 4.

2.4. Data analyses

Student's and Welch's t -tests were used to determine significance of differences between average and site-specific total soil bulk densities for the JB soil categories. The same tests were used for analysis of the within-field difference in SOC between soil from regular and supplementary samplings. We applied classical random sampling estimators of variance, even though it has in other studies been documented that spatial autocorrelation will affect the variance (Brus and Saby, 2016). We argue that the spatial autocorrelation is low in the Danish landscape due to the large spatial variation in soils and management (Adhikari et al., 2013), and we applied the analyses specifically for separate soil texture classes, which thus reduces effects of spatial autocorrelation. Analysis of variance (ANOVA) was applied to determine the difference in effect of the change in method between the JB categories, and paired Sign tests were applied to determine the significance of the differences between Eq. 4 and the other three methods. A non-linear exponential model was fitted to evaluate the relationship between bulk density and organic C content. The statistical analyses were performed in R 4.0.4 (R Core Team, 2021).

3. Results and discussion

3.1. Bulk density

According to Walter et al. (2016), site-specific bulk density remains a most neglected soil parameter when estimating changes in SOC stocks. We found that mean total soil bulk densities measured in the 2019 sampling campaign corresponded closely to the database averages of

Table 2

Mean, standard deviation (SD) and number of observations (n) of average total soil bulk density (BD_{db}) from the national Danish soil database, adopted by Taghizadeh-Toosi et al. (2014), and from site-specific total soil bulk density measurements in the current study (BD_{total}). Soils are grouped according to the Danish JB soil classification system (Table 1) and depth interval (0–25 cm and 25–50 cm). P -values are shown for Students t -tests comparing BD_{db} and BD_{total} for each combination of JB soil category and depth. Percentages of sites where BD_{total} deviates more than 5% and 10% from the corresponding BD_{db} value are given.

Soil depth interval	JB category	BD_{db} (g cm^{-3})			BD_{total} (g cm^{-3})			P value	Percentage of sites deviating more than	
		Mean	SD	n	Mean	SD	n		5%	10%
0–25 cm	1	1.44	0.11	185	1.43	0.11	64	0.53	47	24
	2	1.40	0.08	38	1.38	0.11	27	0.44	37	19
	3	1.43	0.14	96	1.42	0.11	40	0.66	44	24
	4	1.39	0.14	143	1.36	0.13	97	0.09	56	22
	5	1.51	0.16	23	1.44	0.17	17	0.20	65	29
	6	1.46	0.18	87	1.43	0.13	95	0.20	63	27
	7	1.49	0.18	75	1.45	0.18	55	0.21	60	40
25–50 cm	1	1.50	0.12	118	1.49	0.12	64	0.59	52	20
	2	1.47	0.10	25	1.42	0.13	27	0.13	52	19
	3	1.43	0.14	58	1.48	0.13	40	0.07	73	32
	4	1.45	0.15	83	1.47	0.13	97	0.35	63	26
	5	1.55	0.20	14	1.55	0.18	17	1.00	41	29
	6	1.52	0.24	37	1.57	0.15	95	0.24	72	43
	7	1.60	0.12	71	1.59	0.18	55	0.72	51	27

total soil bulk density values of individual soil types presented by Taghizadeh-Toosi et al. (2014) for both the 0–25 and 25–50 cm soil depth intervals (Table 2). The two means were not significantly different at $P < 0.05$ for any combination of soil type and depth interval. Thus, the use of JB category average bulk densities appears to be a valid approach to calculate SOC stocks at a national/regional scale, although it is the combination of bulk density, SOC content and rock fragment content that eventually determines the variance of the mean SOC stock.

For a given soil type, however, there were notable differences between the database averages and the individual site-specific total soil bulk densities. For 37–73% of the sites, the site-specific total soil bulk density differed >5% from the mean database value while 19–43% of the sites differed >10% (Table 2). Furthermore, the difference between site-specific and the database mean total soil bulk densities for a given JB category was not distributed evenly around the mean (Fig. 2), and thus the category-based mean total soil bulk density is a poor representation for all sites within a category. This means that the national JB category mean should be applied with caution when calculating stocks at individual sites.

Estimates of SOC stocks link directly to soil bulk density. Therefore, Fig. 2 also illustrates that the use of site-specific rather than mean bulk densities may provide widely different field-scale estimates of SOC stocks. The effect of variation in total soil bulk density on SOC stock estimates is markedly affected when sites with high or low soil C content also show large differences between the site-specific and the mean bulk density. Indeed, this pattern is likely, since we observed an inverse relationship between SOC content and total soil bulk density (Fig. 3). An exponential model to describe this relationship explained 37% of the observed variation in bulk density, similar to the goodness-of-fit reported by Schrupf et al. (2011) for agricultural sites in Europe. Consequently, establishing baseline SOC stocks and verifying field- and farm-scale changes in SOC stocks associated with regulations and credit schemes related to GHG offsets calls for information on site-specific soil bulk densities (Post et al., 2001; Gojdt et al., 2009; Schrupf et al., 2011; Walter et al., 2016). However, our reported effects of shifting from mean to site-specific bulk density of JB categories link specifically to Danish agricultural land, which is typically relatively high in sand content and low in rock fragment content, and thus the extrapolation of the quantitative effects to other settings remains uncertain.

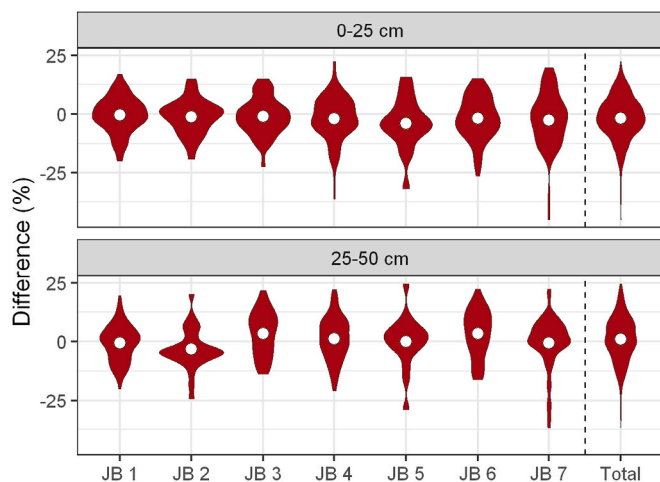


Fig. 2. Violin plot of the differences (%) between the mean total bulk density of a JB category (BD_{db}) according to national soil databases (Taghizadeh-Toosi et al., 2014) and the site-specific total bulk densities measured in the current sampling campaign (BD_{total}) for the seven JB categories as well as across the whole dataset (total). The white dots signify the mean differences.

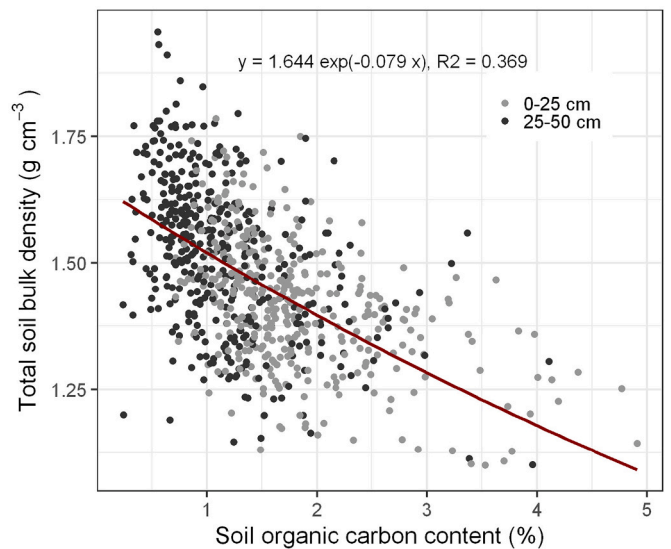


Fig. 3. The relationship between site-specific total soil bulk density (BD_{total}) and the C concentration at 0–25 cm (gray circles) and 25–50 cm soil depth (black circles). An exponential model (line) was fitted to the data.

3.2. Rock fragment content

The volumetric rock fragment contents measured by laboratory analyses was higher than 0.05 for 9% and 16% of the samples from the 0–25 cm and 25–50 cm soil layers, respectively (Table 3). This aligns well with estimates obtained by the visual assessment in the field when soils were sampled, although the visually assessed rock fragment content in subsoils tended to be slightly smaller (Table 3). However, the general concordance between rock fragment content assessed by visual assessment and by laboratory analyses substantiates the validity of the laboratory approach based on relatively small soil samples (100 cm^3). For soils with greater contents of rock fragments than normally found in Denmark (e.g., >15%), it may be challenging to determine the rock fragment contents by soil sampling, and alternative methods such as field-scale estimation by electrical resistivity may be considered (Tetegan et al., 2012).

The distribution of rock fragment content (laboratory data) was right-skewed (Fig. 4), which is expected as the lower limit is zero whereas the upper limit could be relatively high as compared to the mean. The median rock fragment content in the 0–25 cm layer was smallest for the sandy soils (JB1 and JB2) and tended to increase with soil depth for all JB categories.

The presence of rock fragments affects the calculation of SOC stock by decreasing the volume of soil with SOC storing capacity (Eq. 4). Ignoring significant rock fragment contents leads to overestimation of the SOC content, with the decrease in SOC estimates being proportional to the rock fragment content. Poepflau et al. (2017) found that correcting for volumetric rock fragment content below 0.05 had negligible effects on SOC stock estimates; whereas small, but systematic, effects emerged

Table 3

Volumetric rock fragments in 0–25 cm and 25–50 cm depth as measured by analysis of soil samples in the laboratory ($n = 4$ per site and depth, 100 cm^3 each) and estimated by visual assessment in the field during soil sampling.

Volumetric rock fragment	Frequency by soil sampling (%)		Frequency by field observation (%)	
	0–25 cm	25–50 cm	0–25 cm	25–50 cm
<0.05	91	83	93	89
0.05–0.10	8	13	5	8
>0.10	1	3	3	3

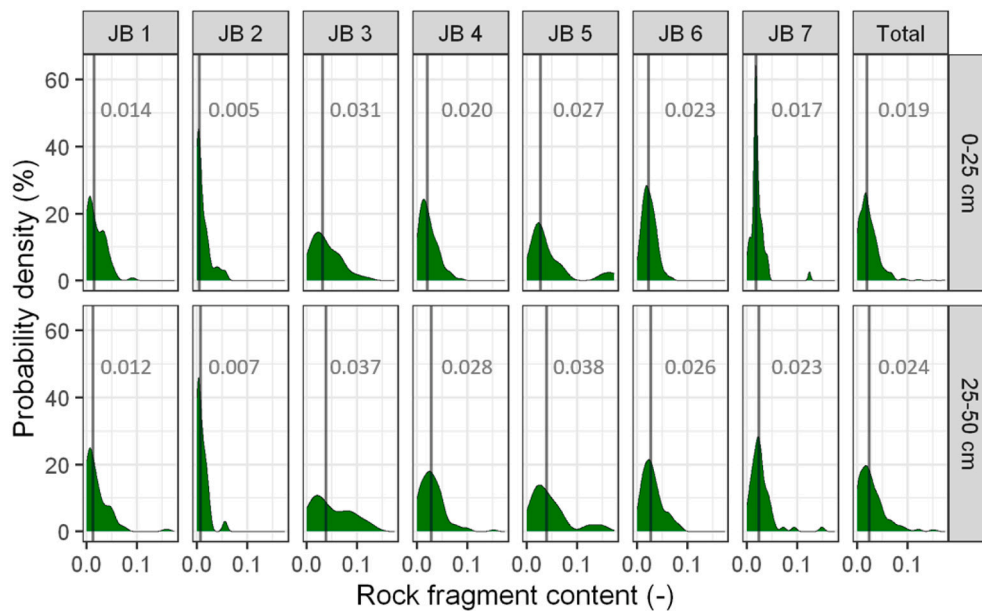


Fig. 4. The distribution of measured volumetric rock fragment content (unitless) for the seven JB categories and across the whole data set (total). Data are shown for the depth intervals of 0–25 cm (upper panels) and 25–50 cm soil depth (lower panels). The vertical lines and gray numbers indicate the median rock fragment content.

when rock fragment content ranged from 0.05 to 0.10. Most soils included in the current study had small volumetric rock fraction contents (Table 3) and little effect on SOC stock estimates was expected. For soils richer in rock fragments and soil with high organic matter contents and (thus low bulk density), correcting for the volumetric rock fragment content becomes essential (Schrumpp et al., 2011; Rytter, 2012; Poeplau et al., 2017).

3.3. SOC stocks calculations: accounting for site-specific bulk density and rock fragment content

We compared SOC stocks based on Eq. 3 (mean total soil bulk density

for JB category, no correction for rock fragment content) with SOC stocks based on Eq. 4 (site-specific fine-soil bulk density and correction for rock fragment content). Here, the use of Eq. 3 generally overestimated SOC stocks (Fig. 5) by a median value of 3.2 and 1.1 Mg C ha⁻¹ for the 0–25 and 25–50 cm soil depth, respectively, corresponding to a relative overestimation of SOC stocks by 5.7% and 4.1% (Table 4). The statistical significance of the differences in SOC stock estimates by Eq. 3 and Eq. 4 was tested using a paired Sign test, and all but two combinations of soil category and depth showed statistically significant differences between the two methods (Table 4). Still, even though statistically significant, the differences in the SOC stock estimate of the two methods were relatively small (i.e., median relative differences around

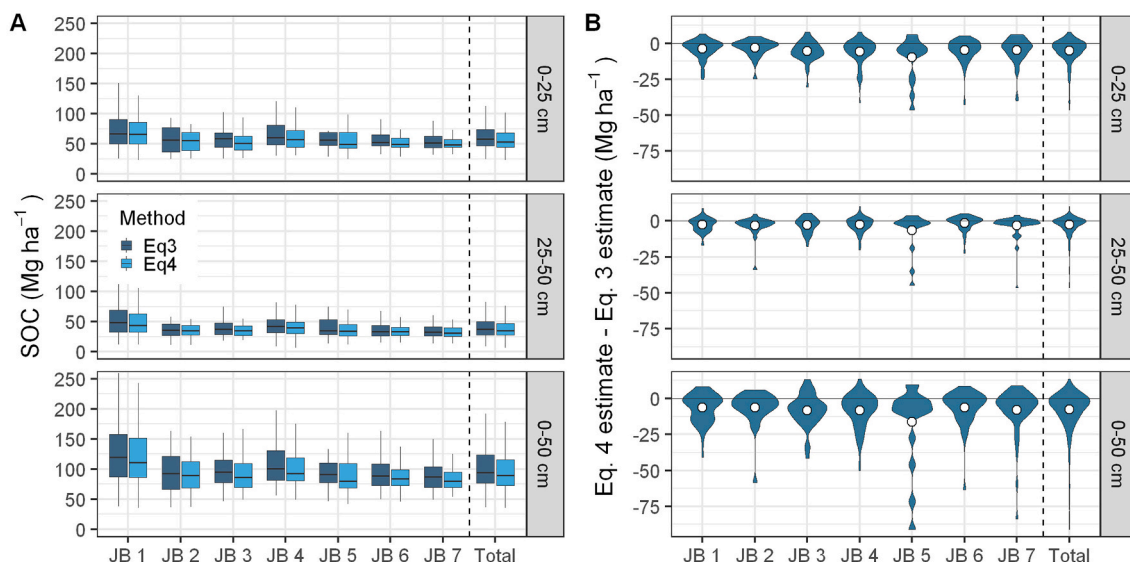


Fig. 5. A) Box plots of the SOC stock estimates (Mg ha⁻¹) for each JB category and for the whole data set (total) in the soil depth intervals of 0–25 cm (upper panels), 25–50 cm (middle panels) and 0–50 cm combined (lower panels). The dark and lighter blue colors show calculations by Eq. 3 and Eq. 4, respectively. The black line in the interquartile boxes shows the median and whiskers extend to the furthest observation within 1.5 times the interquartile range. Outliers are removed for clarity. B) Violin plots of the calculated differences in SOC estimate (Mg ha⁻¹) by Eq. 4 and Eq. 3 for each JB category and depth interval, as well as the total across the data set. The white circle signifies the mean difference.

Table 4

Median difference in absolute (Mg ha^{-1}) and relative (%) SOC stock relative to Eq. 4 when estimated by Eq. 3 and by Eq. 3 improved by including rock fragment content (Eq. 3 with RF) or site-specific fine soil bulk density (Eq. 3 with BD_{fine}). Thus, as compared with Eq. 4, the column 'Eq. 3 with RF' indicates the effect of using BD_{db} rather than BD_{fine} , while the column 'Eq. 3 with BD_{fine} ' indicates the effect of neglecting RF. A positive value indicates that the SOC stock is overestimated as compared to the calculation using Eq. 4. Data are presented for each combination of JB category and soil depth, as well as for each depth across the JB categories. Significance of the absolute differences between (paired sign test) are indicated (* = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$; ns, not significant $P > 0.05$).

		Eq. 3		Eq. 3 with RF		Eq. 3 with BD_{fine}	
		Mg ha^{-1}	%	Mg ha^{-1}	%	Mg ha^{-1}	%
0–25 cm	JB 1	2.3 ***	3.3	1.6 ns	2.2	1.0 ***	1.4
	JB 2	1.9 ^{ns}	3.1	1.0 ns	1.9	0.4 ***	0.5
	JB 3	4.9 ***	9.4	2.4 ***	4.8	1.6 ***	3.1
	JB 4	3.5 ***	5.9	2.5 ***	4.8	1.2 ***	2.0
	JB 5	5.1 *	11.4	2.2 *	7.7	1.5 ***	2.7
	JB 6	3.5 ***	6.1	2.1 **	−3.8	1.1 ***	2.3
	JB 7	2.6 **	5.1	1.5 ns	3.2	1.0 ***	1.7
	Median	3.2***	5.7	2.0***	3.3	1.0***	1.9
25–50 cm	JB 1	1.6 *	5.2	0.8 ns	1.6	0.5 ***	1.2
	JB 2	1.6 **	5.0	1.6 *	4.5	0.3 ***	0.7
	JB 3	1.6 *	5.1	0.0 ns	0.0	1.6 ***	3.7
	JB 4	1.8 **	4.2	0.8 ns	1.9	0.9 ***	2.8
	JB 5	1.6 *	6.3	0.7 ^{ns}	2.5	1.1 ***	3.8
	JB 6	−0.1 ns	−0.2	−0.9 ns	−3.0	0.9 ***	2.6
	JB 7	1.4 ***	3.7	0.4 ns	1.1	0.7 ***	2.3
	Median	1.1***	4.1	0.3***	0.9	0.8***	2.4

5%).

There was no systematic effect of clay content, a proxy for the JB category, on the difference in SOC estimates by Eq. 3 and Eq. 4, while both the relative and absolute difference between calculation methods scaled positively with SOC stocks (Supplementary Fig. S2). This may be linked to the strong inverse relationship between SOC contents and bulk density that exists across soil categories (Fig. 3), meaning that, e.g., soils with high SOC content typically have a site-specific bulk density that is lower than the soil category mean. Hence, the difference in SOC stock estimate between Eq. 4 and Eq. 3 might scale with the difference between the observed site-specific SOC content and the average soil SOC content of the JB category.

Testing the individual effects of not using site-specific bulk density and neglecting rock fragment content indicated a median overestimation by 2.0 and 1.0 Mg C ha^{-1} , respectively, compared to Eq. 4 for the 0–25 cm soil depth (Table 4). Indeed, for all JB categories, the overestimation of SOC stock in the topsoil was more affected by not using site-specific bulk density than by neglecting contents of rock fragments. For the 25–50 cm soil depth, there was a median overestimation of 0.3 Mg C ha^{-1} by not using site-specific bulk density and 0.8 Mg C ha^{-1} by neglecting rock fragment content (Table 4).

Overall, across the two soil depths, there was a consistent and always significant effect of neglecting rock fragment contents (Table 4), which reflects a systematic bias (overestimation) in the calculated SOC stock. This bias scales directly with the rock fragment content, as the soil volume that contains organic carbon decreases proportionally with the rock fragment content.

For soil bulk density, the use of average database values, rather than site-specific values, can result in both over- and underestimation of the SOC stocks (Fig. 2; Table 4), which will depend on the variation within the group (here JB category) that is assigned the same average bulk density. However, in the present study, the median effects for individual JB categories were often non-significant, especially in the subsoil. In general, the effect of using average database bulk density, rather than site-specific bulk density will be random, but depending on the difference between the two estimates. Therefore, it is not possible to

generalize an effect of using average bulk density rather and site-specific values that will apply outside of this study.

Dynamics of SOC stocks are characterized by small annual changes against a large background of existing stock. Thus, field-scale variation in soil C contents remains a challenge for determination of changes in SOC stocks. We accounted for field-scale variation in topsoil (0–25 cm) and subsoil (25–50 cm) by pooling 16 soil samples from randomly selected cells within each grid area. A supplementary sampling of 16 randomly selected cells took place at 24 of sites. Differences between the two sampling events in estimates of total C content were not statistically significant for the topsoil ($P = 0.65$; Fig. 6), while there was small but significant ($P = 0.047$) differences for subsoil samples (Fig. 6). From these results we assume that C contents reported here are representative for the sampling site, which was also concluded by Taghizadeh-Toosi et al. (2014) based on a similar sampling approach.

Across the Danish soil types, the largest SOC stock was seen for JB 1, followed by JB 2 and JB 4, and with smaller and almost similar SOC stocks for JB 3, JB 5, JB 6 and JB 7 (Fig. 5). Thus, the sandiest soils contain the greatest amount of SOC, both in the topsoil and subsoil. The capacity of soils to store SOC generally links to contents of clay and the ability of clay-sized organo-mineral particles to protect SOC against microbial turnover (Christensen, 2001). However, many of the cultivated sandy soils in Denmark originate from old heathland and shrubland that were converted into arable soils some 100 to 150 years ago. At conversion, they most likely contained high levels of SOC and the soils may still have a large reservoir of stable SOC. Many of these soils also have a high C-to-N ratio related to stabilized soil C (Thomsen et al., 2008). Moreover, the sandy soils in the western part of Denmark support dairy production with a high frequency of grass ley in their rotation while the more clayey soils are often under intense cultivation with cereals and other cash crops. These different production systems greatly affect the amount of C returned to soil in animal manure and crop residues.

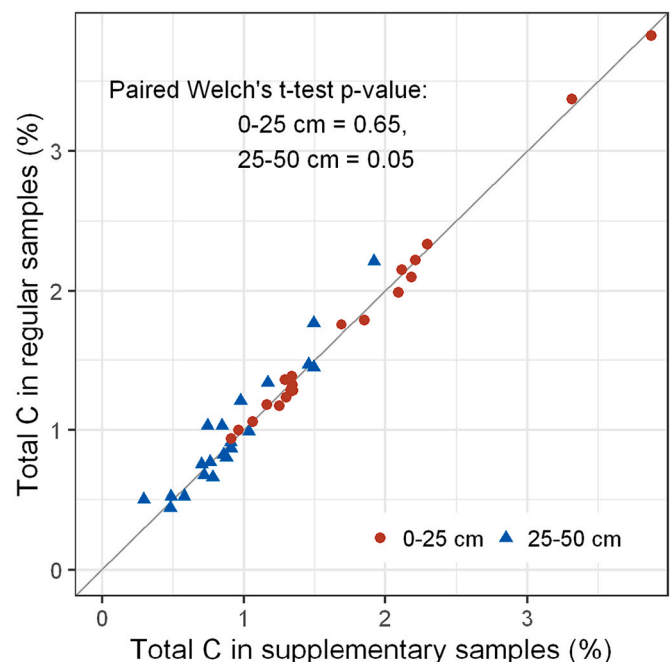


Fig. 6. Correlation between soil organic carbon (SOC) in soil from the regular and supplementary sampling of 16 cells within each of 24 grid-points in 2019 sampling campaign. The red circles are samples from the topsoil (0–25 cm) while the blue triangles are samples from the subsoil (25–50 cm). The figure shows the P -values for Welch's paired t -tests for each layer.

4. Conclusions

An accurate estimate of SOC stock at field scale requires site-specific values of fine-soil bulk density and content of rock fragments. The variances in these parameters within a given JB soil category can be large and when combined may result in estimates of SOC stock for individual sites that differ greatly from estimates based on average values of fine-soil bulk density and rock fragment contents. Thus, the effect of both site-specific fine soil bulk density and rock fragment content are essential for accurate SOC stock estimations at a field-level.

For national inventories, however, information on site-specific fine-soil bulk density and rock fragment content may not be critical for a valid estimate of the SOC stocks in a given JB soil category. This is because the average category-specific fine-soil bulk density differ relatively little from the average of site-specific values and the difference can be both positive and negative, resulting in a non-systematic bias. The effect of the rock fragment content on the estimate of SOC stocks is a systematic bias, but scales with the rock fragment content. As the rock fragment content is relatively low in Denmark, the effect is therefore relatively small.

We found that applying average values of bulk density and neglecting rock fragment contents results in an overestimation of SOC stock by 5.7% for the topsoil and 4.1% for the subsoil in the Danish National Square Grid.

Data availability statement

The data that support the findings in this study are available from the corresponding author upon reasonable request.

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.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.geodrs.2022.e00560>.

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