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Forage vs. Grain Legumes: Contrasting Effects on Soil Organic Carbon Stocks–Evidence From 30 European Field Experiments

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ABSTRACT

Sustainable land management can play an important role in climate change mitigation by reducing soil organic carbon (SOC) losses or even by sequestering C in soils. This can be achieved through practices that increase C inputs to the soil and/or improve the quality of these inputs, thereby facilitating the removal of atmospheric carbon dioxide (CO_2) and storing it in the soil as SOC. In this study, we investigated the potential of an increased share of legumes in crop rotations to enhance SOC accrual—defined as the increase in SOC stocks at a given land unit compared to the baseline scenario—using data from 30 mid-term (MTEs, 5–20 years) and long-term (LTEs, 20+ years) field experiments across Europe. Our findings indicate that increasing the proportion of forage legumes in rotations (based on 21 experiments and 39 paired comparisons) led to SOC accrual of up to 13.25 Mg ha^{-1} (0.44 Mg ha^{-1} year^{-1}), while grain legumes (based on nine experiments and 28 paired comparisons) resulted in a decrease in SOC stocks of up to 14.37 Mg ha^{-1} (−0.48 Mg ha^{-1} year^{-1}) compared to the reference treatment. For forage legumes, the largest SOC gains were achieved at sites with the smallest reference SOC stocks and greater share of forage legumes in the rotation. Our observations suggested that the duration of crop growth of the forage legumes (annual vs. perennial) did not exert a significant impact on SOC stock increase, while pedoclimatic zone did. Positive effects on SOC stocks were more pronounced in the Atlantic climatic zone in contrast to the Mediterranean climatic zone. For grain legumes, larger SOC losses were observed with a greater share of grain legumes in the rotation. Overall, integrating forage legumes in cropping systems can enhance their sustainability and present a viable option for climate change mitigation. Finally, we present a regression equation to derive emission factors (EFs) for estimating SOC changes due to the increase of the share of forage legumes in a rotation, and another due to

Abbreviations: fLEGshare, Share of forage legumes in the crop rotation in %; fLEGsharediff, Percentage difference of forage legumes in the treatment compared to the reference (%); gLEGshare, Share of grain legumes in the crop rotation in %; gLEGsharediff, Percentage difference of grain legumes in the treatment compared to the reference (%); LEGshare, Share of legumes in the crop rotation in %; LEGsharediff, Percentage difference of legumes in the treatment compared to the reference (%); SOCref, Soil organic carbon stock of the reference (Mg ha^{-1}); SOCref/cm, Soil organic carbon stock of the reference per cm depth (Mg ha^{-1} cm^{-1}); SOCtreat, Soil organic carbon stock of the treatment (Mg ha^{-1}).

the increase of the share of grain legumes in the rotation. The first can be used to support the assessment of management impacts for the purpose of rewarding carbon farming and the estimation of a national-scale SOC accrual potential, while the second can be used for estimating national-scale SOC losses.

1 | Introduction

About 60%–70% of EU soils (agricultural and non-agricultural) are considered to be affected by one or more soil degradation processes and can be considered unhealthy due to current management practices (European Commission et al. 2020; Panagos et al. 2024). Agricultural soils are either eroded, compacted, suffer from salinization, or lose carbon (C) at a rate of 0.5% per year (European Commission et al. 2020; Panagos et al. 2024). Soil organic matter (SOM), of which soil organic carbon (SOC) is the main component, is crucial for maintaining soil health due to its numerous beneficial effects on soil properties and functions (Jensen et al. 2019; Kopittke et al. 2022; Lehmann et al. 2020). Through improved management, more organic matter and thus more C can be returned to the soil resulting in SOC accrual (i.e., "an increase in SOC stock for a given unit of land, starting from an initial SOC stock or compared to a business-as-usual value") which can lead to SOC loss mitigation (defined as the reduction of SOC losses compared to a business-as-usual scenario due to an anthropogenic intervention (Don et al. 2024)) and potentially to soil C sequestration (Freibauer et al. 2004), which is defined as the "process of transferring C from the atmosphere into the soil through plants or other organisms, which is retained as SOC resulting in a global C stock increase of the soil" (Olson et al. 2014). Enhanced C sequestration in soils through improved agricultural management means that agricultural soils serve as C sinks (negative CO_2 emissions) and thus can contribute to climate change mitigation (Don et al. 2024). Several agricultural practices have been proposed in the literature as potential tools for C accrual and in turn, improved soil fertility. These, among others, include the addition of organic amendments, the incorporation of crop residues into soils, site adapted reduced tillage, or the diversification and improvement of the crop rotations with cover crops and crops that increase C inputs, such as by increasing the share of perennial crops (Paustian et al. 2016; Smith et al. 2008). These practices control and alter SOC stocks by either increasing the inputs of C into the soil by plant roots, root exudates, crop residues or more amendments of exogenous organic matter, or by reducing C turnover (mineralization) (Chenu et al. 2019; Smith et al. 2008).

One practice that contributes to crop diversification and crop rotation improvement is the cultivation of legumes. Legumes, which are the major protein crops and play a significant role in human and animal nutrition, can increase environmental sustainability thanks to their ability to fix atmospheric nitrogen (N) (Jensen and Hauggaard-Nielsen 2003; Stagnari et al. 2017). Legumes are a broad plant family that can be classified into two categories: forage and grain legumes. Common forage legumes include alfalfa, clover, and vetch. Forage legumes are typically used in grazing systems, cut for hay or silage, and are often mixed with grasses (e.g., grass-clover mixtures) to enhance livestock nutrition. On the other hand, grain legumes (also known as pulses) are cultivated for their seeds, which are harvested for human and animal consumption. This group includes peas, beans (e.g., soybeans), and lentils (Ditzler et al. 2021; Reckling et al. 2016a; Sellami et al. 2019).

However, in the last decades, the cultivation of legumes has decreased in Europe, due to low gross margins and unstable yields (Cernay et al. 2015), unfavourable market and policy incentives (Zander et al. 2016), as well as the inability to recognise or evaluate their long-term effects in cropping systems (Preissel et al. 2015). The EU self-sufficiency in protein crops (legumes, pseudocereals, grains and other minor crops) averages around 70% (EUROSTAT 2023), occupying about 3% of its arable land. Nevertheless, EU protein crop production only meets 28% of the demand for high-protein animal feed crops (FEFAC 2024). While the EU is self-sufficient in grass, silage maize, and fodder legumes, it relies heavily on imports for high-protein crops such as soy, essential for poultry and pig diets. These imports can contribute to environmental issues in exporting regions, including deforestation, concomitant with increased greenhouse gas emissions (Nemecek et al. 2008).

The EU faces a longstanding shortage of high-protein crops for human and animal consumption, compounded by environmental concerns and rising demand. This has made protein self-sufficiency a priority, leading to several initiatives (COM (2018) 757 2018; O.J. (C 199 E/07) 2012; O.J. (C 390/01) 2019) and the development of the European protein strategy (European Parliament 2023). Supported by sustainability-based initiatives such as the "Biodiversity Strategy" and the "Farm to Fork Strategy" of the "EU Green Deal" (European Commission, COM (2020) 381, 2020), these efforts aim to reduce import dependency, to promote crop diversification and to reintroduce domestic protein production.

Legume-based crop rotations are included in the list of practices that could be supported by the Eco-schemes under the CAP 2023–2027 and the EU Green Deal for their potential benefits, including climate change mitigation and adaptation, soil degradation prevention, and reduction of pesticide use (OJ L 435, 2021; European Commission 2021). As there is still a large potential to increase legumes in crop rotations (Notz et al. 2023; Reckling et al. 2016a, 2016b; Zander et al. 2016; Zhao et al. 2022), even when livestock numbers would not increase (Nemecek et al. 2008), growing legumes in crop rotations in diverse leys or as intercropping is considered a sustainable agricultural practice. It provides numerous agronomic and environmental benefits, such as reducing the need for mineral N fertilisers, which are energy-intensive to produce, costly, and contribute to greenhouse gas emissions (Harris and Ratnieks 2022; Murphy-Bokern et al. 2017). The main advantages are the reduction of weed pressure, improving soil N content by N fixation from the atmosphere, and reducing the import of legumes when locally produced. However, legumes also increase the complexity of cropping systems, thus creating a potential risk of species

Summary

- Data analysis across 30 mid-term and long-term field experiments with durations between 5 and 55 years
- Including forage legumes in crop rotations can lead to SOC accrual in the European croplands.
- Regression equations for estimating SOC Tier-2 EFs for increased forage and grain legume shares in rotation.

competition when used for intercropping, and require more labour, specific skills, knowledge, and technical equipment, as well as market access (Voisin et al. 2014; Wezel et al. 2013).

Even if there are some (meta)review studies (Kumar et al. 2018; Virk et al. 2022) that investigate the role of legumes in SOC storage (i.e., SOC pool size measured as SOC stocks or SOC concentration (Don et al. 2024)), most research on leguminous crops focuses on N-fixation and cycling (Costa et al. 2020). In particular, there is a need to test, identify, and document the effects of including legumes in crop rotations on SOC storage (Haddaway et al. 2015) considering the entire rotation, a cropping system assessment (Plaza-Bonilla et al. 2018) and the role of the different legume species (Costa et al. 2020).

In this study, we aimed to assess how increasing the proportion of legumes in crop rotations affects SOC stocks and to evaluate their SOC accrual potential. To quantify this, we estimated SOC emission factors (EFs) for European croplands using data from mid-term (5–20 years MTEs) and long-term (20+ years, LTEs) field experiments conducted across Europe. Relevant experiments with varying legume shares were selected, and SOC stocks and EFs were estimated for each treatment pair. After evaluating the effects of management-related factors such as the use of perennial versus annual legumes, and forage versus grain legumes on SOC EFS, we applied linear mixed models (LMMs) to analyse the impact of legume share differences on SOC EFs. The models included soil, climate, and management-related variables to account for potential influencing factors on SOC EFs. Using backward stepwise elimination and considering different data limitations, we identified the optimum model, which can be used for deriving EFs and subsequently predict SOC stock changes associated with increase share of legumes in crop rotations.

2 | Materials and Methods

2.1 | Data Sources and Pairs Selection

The CarboSeq crop and soil management database (Ruysschaert et al. 2024) contains metadata and data of MTEs and LTEs in Europe. It was established to support the estimation of EFs for several crop and soil management practices in arable land, including zero and non-inversion tillage, cover crops, crop choice, residue management, agroforestry, and irrigation (Panagea et al. 2023). The classification used in the database is followed in this research and defines experiments lasting 5–20 years as Mid-Term Experiments (MTEs) and those exceeding 20 years as Long-Term Experiments (LTEs).

For this study, data was extracted from version: 2022_10_24 of the database, using the export module developed especially for this database (Blanchy 2022). For the extraction, one main prerequisite was formulated, that is, the only difference between the reference and the alternative management option (hereafter, referred to as 'treatment') is the crop rotation system applied, whereas any other factor (e.g., mineral, and organic fertilisation, tillage, etc.) remains the same. In the CarboSeq crop and soil management database, 128 entries were identified as potentially relevant, that is, with different rotations among the different treatments. After manually checking each entry and removal of duplicates, 35 experiments that included different crop rotations which include leguminous crops were further selected. These experiments included treatments with different crop rotations, minimum necessary data for analysis (measured SOC stocks or SOC concentration) and had a duration of more than 5 years. Among the 35 experiments, 28 were found to be relevant for the category "increased LEGshare" (i.e., treatments with different shares of legumes in the rotation or conversion to rotation only with legumes).

Apart from data derived from the CarboSeq crop and soil management database, a structured query in the Web of Science in October 2022 (shown in Appendix S2) was used to search for additional published literature testing the completeness of the database. The final dataset related to increased LEGshare included 32 experiments with 72 pairs of reference and treatment.

In the selected experiments, treatments with different crop management were identified: (i) treatments with increased LEGshare, (ii) treatments where the cropping system converted to rotation only with legumes, and (iii) treatments where the legumes were used as intercrops in orchards. The latter crop management was not included in the analysis due to sparse data. To compare rotations with varying proportions of legumes, we calculated the percentage difference in legume presence between the treatment and the reference rotations, referred to as LEGsharediff. This represents the difference in the percentage of years legumes are included in the rotation. For example, if the reference rotation was 4 years with legumes present only in 1 year out of the four (1/4 = 25%), and the treatment rotation also spanneds 4 years but includes legumes for 3 years (3/4 = 75%), the LEGsharediff would be 50%.

Then, the type of legumes and the growth cycle duration, was evaluated. The leguminous crops were separated into grain legumes, that is, those that are cultivated for their grains (e.g., faba bean, soybean, chickpea), and forage legumes, that is, those that are cultivated for their entire aboveground plant parts (e.g., clover, alfalfa, vetch etc.). The duration of the crop growth was distinguished as annual and perennial. In treatments where the leguminous crop was planted for 1 year, the duration of growth was considered as annual, while in treatments where the crop remained in the field for more than 1 year the duration of growth was considered as perennial. Treatments where the crop remained on the field for more than 5 years were excluded as according to the EUROSTAT glossary, they are considered grasslands (EUROSTAT 2018).

After removing all the data and experiments that fall in the abovementioned categories, the final dataset related to the increased share of forage and grain legumes in the rotation was analysed for estimating the SOC EFs included 30 experiments with 67 pairs of reference and treatment. The information for these experiments and pairs were extracted from the CarboSeq crop and soil management database version: 2022_11_16.

2.2 | Soil Organic Carbon Emission Factor and Stock Changes Estimation

SOC EFs refer to the quantifiable effects at which C is either stored in or emitted from soil as a result of a given management activity compared to a baseline scenario. Depending on their detail and estimation methods, the EFs are used in the different tiers to support greenhouse gas inventories (IPCC 2019), enabling better climate policy formulation and effective land management practices to reduce CO_2 emissions and potentially mitigate climate change.

In line with the IPCC (2006, 2022), the SOC EF is defined as the ratio of the SOC stocks up to 30 cm soil depth of the treatment to the SOC stocks of the reference (Equation 1). In this study, a reference was a crop rotation with no legumes or a lower LEGshare than in the treatment. In the literature, the EF is also referred to as the C response ratio (RR) or management factor (Ogle et al. 2005). A relative change greater than 1 corresponds to SOC stock increase in the treatment in comparison to the reference, while relative change less than 1 is a SOC stock decrease.

$$\mathrm{EF} = \frac{\text{SOC Stock}_{\text{treatment}}}{\text{SOC Stock}_{\text{reference}}} \tag{1}$$

where EF stands for the SOC emission factor of a practice, SOC $\text{Stock}_{\text{treatment}}$ is the SOC stock in the treatment, and SOC $\text{Stock}_{\text{reference}}$ is the SOC stock in the reference, both measured at the same time and in the same soil sampling depth.

The EFs can be used to estimate changes in the SOC stocks due to the different practices.

Considering that $\Delta\ \text{SOC Stock} = \text{SOC Stock}_{\text{treatment}} - \text{SOC Stock}_{\text{reference}}$ and based on the Equation (1) the formula to estimate the SOC stock changes is:

$$\Delta\ \text{SOC Stock} = (\mathrm{EF} - 1) * \text{SOC Stock}_{\text{reference}} \tag{2}$$

2.3 | Carbon Stocks Estimation Based on Data Availability

When SOC stocks were not reported, they were estimated using measured data of SOC concentration, bulk density (BD) and sampling depth. In cases where BD values were not available, the pedotransfer function (PTF) (Equation 3) proposed by Hollis et al. (2012) was used to estimate BD. In the case that the textural properties were not known, the clay and sand percentages required to estimate the BD were derived from maps that are based on the LUCAS topsoil physical properties database for Europe (Ballabio et al. 2016).

$$\begin{aligned} \mathrm{BD} = {} & 0.80806 + \left(0.823844 * e^{-0.27993 * OC\%}\right) \\ & + (0.0014065 \times \text{Sand}\%) - (0.0010299 \times \text{Clay}\%) \end{aligned} \tag{3}$$

where BD expressed in Mg m^{-3} stands for bulk density, OC represents the organic C content in % in the layer of which the BD is estimated, Sand and Clay represent the sand and clay content in mass percentage in this soil layer.

2.4 | Statistical Analysis

2.4.1 | Data Inspection

To assess whether the type of legumes used in a crop rotation (forage versus grain legumes) had a significant impact on SOC EFs, we initially performed simple *t*-tests. We compared the EFs of forage legumes and grain legumes with each other and with a reference value of 1, which indicates no change in SOC stocks. The *t*-tests were used to determine if the EFs significantly deviated from 1, which would indicate either an increase or decrease in SOC due to the legume type. In addition to *t*-tests, variance analysis was performed to evaluate the overall variability in the data across the different groups.

To evaluate the robustness of the results and account for the influence of individual experiments, we conducted a Leave-One-Out (LOO) sensitivity analysis for both forage and grain legumes. In this analysis, one experiment was excluded at a time from the dataset, and the mean EFs and 95% Confidence Intervals (CIs) were recalculated. The purpose of this analysis was to determine if any single experiment had an undue influence on the overall results. The log-transformed values of the EFs were used in the LOO analysis to ensure the data conformed better to a normal distribution and to reduce the effect of extreme values on the estimates. The mean and CIs were then back transformed from the log scale to the original scale.

2.4.2 | Model Development

To derive the most significant pedoclimatic and management predictors for the EFs, linear mixed effects modelling (LMM) was used. First, experimental duration and sampling depth—two variables that are expected to play an important role in SOC stock changes—were assessed for their statistical significance on SOC EFs with an a priori LMM. By evaluating their statistical significance beforehand, we aimed to understand their impact on the outcomes more accurately and to avoid bias. Then, all the other available variables were explored for collinearity ($p > 0.05$) and data limitations such as limited/unbalanced data across different climatic zones or soil types. Only variables not strongly correlated ($r^2 < 0.6$) were included in the same model. The predictor variables that were evaluated included climate-related, soil-related, and management-related variables. Climate variables were climatic zone (EEA 2016), aridity index (Trabucco and Robert 2018), aridity class (UNEP 1997), annual precipitation, and mean annual temperature (Fick and Hijmans 2017). Soil-related variables were clay content, USDA textural class, and SOCref/cm, with SOCref/cm being the SOC stock of the reference expressed per cm (by dividing the SOC stocks with the corresponding sampling depth) to account for the impacts of the different sampling depths. Variables related to the management of the crops were crop duration, which includes two levels (i.e., annual and perennial), and LEGsharediff. After this data inspection, in the generic mixed model, climate, soil, and management variables constituted the fixed effects, while the experiment was introduced as a random effect to account for the between-experiment variability.

The initial model (Equation 4) was created to include non-collinear variables from the soil, climate, and management categories.

$$\begin{aligned} \text{EF} \sim & \text{SOCcm} + \text{clay content} + \text{climatic zone} \\ & + \text{duration of crop growth} + \text{LEGsharediff} \\ & + (1 \mid \text{Experiment ID}) \end{aligned} \tag{4}$$

The explanatory variable LEGsharediff was kept in all models and was not considered for elimination as it was used to differentiate different experimental treatments and create the selected pairs. Backward stepwise elimination was used to find the simplest model that explains the data. The final model was fitted using restricted maximum likelihood estimation.

2.4.3 | Software

The analysis was conducted in R (R Core Team 2021), and mixed-effect models were fitted using the package lme4 (Bates et al. 2015). The backward elimination was performed using the step function of the lmerTest package (Kuznetsova et al. 2017) based on the lowest Akaikes Information Criterion (AIC). Graphics and representations of the fitted models were produced with the packages ggplot2 (Wickham 2016) and ggeffects (Lüdecke 2018).

3 | Results

3.1 | Dataset Inspection

Including grain or forage legumes in the crop rotation had contrasting effects on the SOC storage (Figure 1). When evaluated with simple *t*-tests, it was found that the EFs of forage legumes and grain legumes were significantly different from each other ($p < 0.001$) and significantly different from 1, which refers to no change ($p = 0.03$ for forage legumes and $p < 0.001$ for grain legumes). Increasing the share of both annual and perennial forage legumes in the crop rotation led to an increase in SOC and thus EFs > 1.

Increasing the percentage share of forage legumes in the crop rotation (fLEGshare) in crop rotations led to an overall increase in SOC storage, with a mean EF of 1.076 (95% CI: 1.009–1.143) on the original scale. When estimated using a log-transformed approach, the back-transformed mean EF was 1.059 (95% CI: 1.001–1.119), confirming a statistically significant, though marginal, positive effect. Conversely, increasing the percentage share of grain legumes in the crop rotation (gLEGshare) resulted in a decrease in SOC stocks. The mean EF on the original scale was 0.907 (95% CI: 0.868–0.946), indicating a statistically significant negative effect. The log-transformed approach yielded a back-transformed mean EF of 0.901 (95% CI: 0.861–0.943), showing a consistent and statistically significant reduction in SOC storage. These findings confirm that both effects are statistically significant, aligning with our initial conclusions. However, the

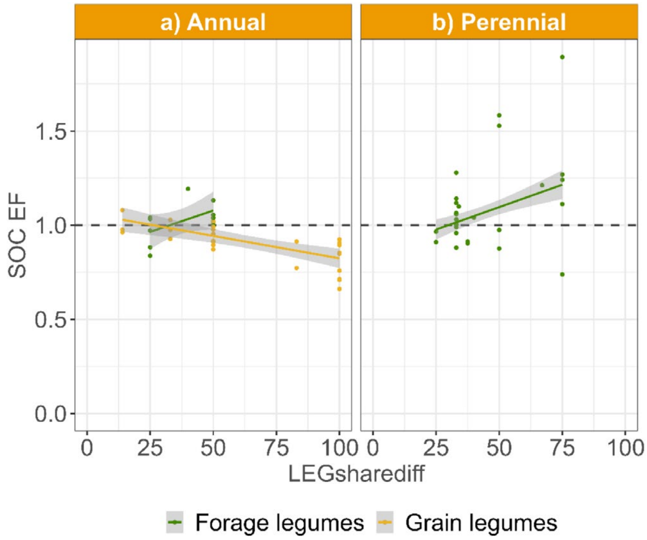

FIGURE 1 | Soil organic carbon emission factors (SOC EF) in relationship with the percentage difference of legumes in the treatment compared to the reference (LEGsharediff). The left panel shows the treatments where the legumes were planted annually. The right panel shows the treatments in which the legumes were planted once and remained in the field for more than one year and up to five years. Within each panel the yellow colour represents the grain legumes, and the green colour the forage legumes.

magnitude of the positive effect for forage legumes was modest, as indicated by the narrow margin above 1. Both the original and log-transformed approaches were used to ensure robustness in the analysis. The original scale provides a direct interpretation of the mean effect, while the log-transformed approach accounts for the skewed distribution of response ratios, reducing the influence of extreme values and improving the accuracy of confidence interval estimation.

The LOO sensitivity analysis confirmed the robustness of the effects of both grain and forage legumes. The negative effect of grain legumes remained consistent, with no single experiment disproportionately influencing the results, while the positive effect of forage legumes also remained statistically significant despite slight variations (see Appendix S2 for details).

As a next step in this study, we further analysed the annual and perennial forage legumes (shown in green in Figure 1) separately from the grain legumes (shown in yellow in Figure 1) to identify which variables led to the potential changes in the SOC stocks and derived EFs for quantifying these changes.

3.2 | SOC EFs for Increasing fLEGshare

The final dataset with only forage legumes used for further analysis included 21 experiments located in 9 different countries (Figure 2) with 39 pairs of treatments and references (i.e., 9 in Italy, 7 in Spain, 5 in Czech Republic, 5 in Denmark, 4 in Germany, 3 in Norway, 3 in Sweden, 2 in Belgium, and 1 in Lithuania). In 29 treatments, the legumes were planted once and stayed in the field for more than 1 year and up to 4 years (perennial), while in 10 treatments, the legumes were planted annually.

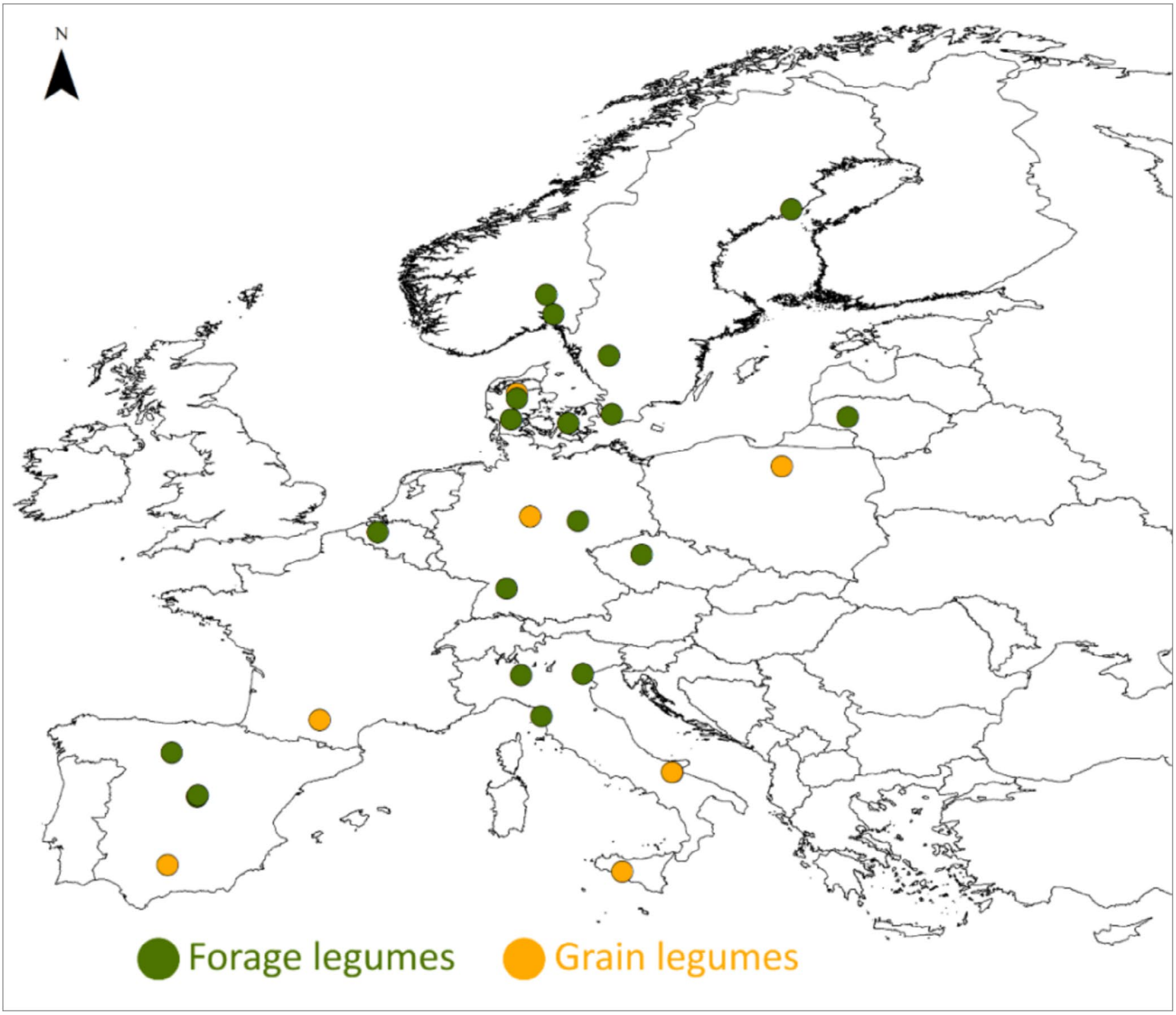

FIGURE 2 | Spatial distribution of the mid- and long-term field experiments on legumes in crop rotations in Europe. Green dots present the experiments which included forage legumes and yellow dots those with grain legumes.

The different forage legumes used were alfalfa (15 treatments), vetches (7 treatments), and different clover species or mixtures of clovers with other legumes or grasses (15 treatments) (Figure 3). These crops were planted either as main annual crops in the rotation, or as intercrops (mixed culture), or as main crops in temporary leys (duration 1–4 years), or as mixtures in the grass grown in the temporary leys. The forage legumes either replaced other crop types such as cereals (in 16 treatments) or root/tuber crops (in 4 treatments) or were used as extra main crops in the rotation, without replacing an existing crop and hence leading to an expansion of the rotation (14 treatments) (Figure 3). Most of the observed pairs were in areas with a Continental climate ($n=20$) followed by those in Mediterranean ($n=8$), Boreal ($n=6$) and Atlantic ($n=5$) climate zones. An overview of the basic information per experiment is shown in Table 1 and detailed information of the pairs and experiments is included in Appendix S1. The statistical parameters (minimum, maximum, mean and median) of the dataset are presented in Table 2.

The a priori LMM that was performed to evaluate the possible effects of experimental duration and sampling depth revealed that neither experimental duration ($p=0.81$) nor the sampling depth ($p=0.17$) had a significant impact on the obtained EF (Appendix S2 Table S1). To further assess the impact of experiment duration, we conducted separate analyses for MTEs (≤ 20 years) and LTEs (> 20 years). Results showed that the positive effect of forage legumes was more pronounced in LTEs, while neither duration nor sampling depth had a significant influence in these datasets (see Appendix S2 for details). As the effect of experiment duration and sampling depth were not statistically significant, these variables were not included in the initial model (Equation 4) to keep the model as simple as possible.

After performing backwards elimination on the initial model (Equation 4) based on the AIC criterion the best fitting model (Equation 5) used the predictor variables $SOC_{cm/ref}$, ($p=0.003$) and climatic zone ($p=0.025$).

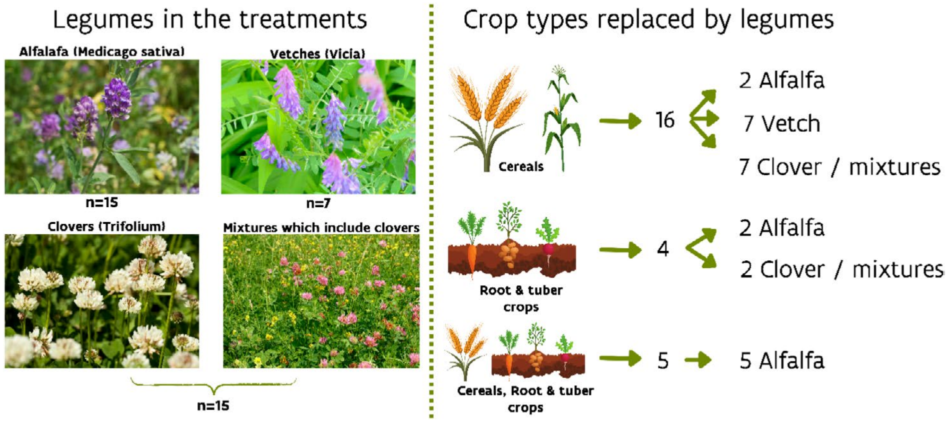

FIGURE 3 | Left: Legume types prevalent in the analysed studies. The number n indicates the considered pairs. Right: number of treatments that the existing crops were replaced by legumes within one crop rotation.

$$\mathrm{EF} \sim \mathrm{fLEG}_{\mathrm{sharediff}} + \mathrm{SOC}_{\mathrm{ref/cm}} + \text{Climatic zone} + (1 \mid \text{Experiment ID}) \quad (5)$$

The summarised results of the best-fitting model are presented in Table S4.

The mathematical expressions of the estimated predictive model take the following forms for each climatic zone:

$$\text{Atlantic: } \mathrm{EF} = 1.636388 + 0.002432 \times \mathrm{fLEG}_{\mathrm{sharediff}} - 0.208378 \times \mathrm{SOC}_{\mathrm{ref/cm}} \quad (5a)$$

$$\text{Boreal: } \mathrm{EF} = 1.582028 + 0.002432 \times \mathrm{fLEG}_{\mathrm{sharediff}} - 0.208378 \times \mathrm{SOC}_{\mathrm{ref/cm}} \quad (5b)$$

$$\text{Continental: } \mathrm{EF} = 1.317307 + 0.002432 \times \mathrm{fLEG}_{\mathrm{sharediff}} - 0.208378 \times \mathrm{SOC}_{\mathrm{ref/cm}} \quad (5c)$$

$$\text{Mediterranean: } \mathrm{EF} = 1.183496 + 0.002432 \times \mathrm{fLEG}_{\mathrm{sharediff}} - 0.208378 \times \mathrm{SOC}_{\mathrm{ref/cm}} \quad (5d)$$

where $\mathrm{fLEG}_{\mathrm{sharediff}}$ refers to the additional fraction of forage legumes in the rotation (%) compared to the reference treatment and $\mathrm{SOC}_{\mathrm{ref/cm}}$ to the SOC stock of the reference treatment per cm depth.

The range of the predicted EFs relative to the fLEGsharediff for each climatic zone is shown in Figure 4A, while Figure 4B presents the predicted EFs relative to the SOCref/cm. The higher the SOCref/cm, the lower the potential to increase the SOC stock by increasing the fLEGshare.

In Figure 4A, the predicted EFs increase with increasing fLEGsharediff. However, the response differs between zones. The Atlantic zone shows the highest predicted EF values, indicating a greater potential for SOC accrual when forage legumes are more frequently included in crop rotations. The Boreal and Continental zones show a similar trend, though the response is slightly lower than in the Atlantic zone. In contrast, the Mediterranean zone shows the lowest EF values, suggesting less potential to increase SOC stocks. Additionally, the effect of SOCref/cm is evident, as higher SOCref/cm values (e.g., 3.5 Mg ha^{-1} cm^{-1}, orange line) lead to lower EF values, meaning that soils with initially higher SOC levels have less potential for further SOC accrual. Figure 4B highlights the inverse relationship between EF and SOCref/cm across climatic zones, where higher SOCref/cm results in lower EF values.

It is important to note that the distribution of available data was not balanced among climatic zones. For example, the Mediterranean zone mainly includes observations with relatively low SOCref/cm, whereas the Boreal zone includes mostly high SOCref/cm values. Furthermore, there is a significant disparity in data availability between zones, with only five observations in the Atlantic zone compared to 20 in the Continental zone. Thus, a simpler prediction model that does not consider the different climatic zones was created and tested, inferring that the trend that is presented in the Atlantic and Continental climatic zones which have a wider spread of data, will follow also in the Mediterranean and Boreal climatic zones.

It had the following structure:

$$\mathrm{EF} \sim \mathrm{fLEGsharediff} + \mathrm{SOCref/cm} + (1 \mid \text{Experiment ID}) \quad (6)$$

The AIC for the best-fitting model (Equation 5) is −38.95, compared to −32.49 for the simpler model (Equation 6). Additionally, the conditional R^2 values are 0.93 for Equation (5) and 0.92 for Equation (6), demonstrating that both models explain a high proportion of the variance.

The summarised results of the model are presented in Table S5. The mathematical expressions of the final estimated predictive model were:

$$\mathrm{EF} = 1.182198 + 0.003957 \times \mathrm{fLEGsharediff} - 0.129105 \times \mathrm{SOCref/cm} \quad (6a)$$

As in the different climatic zones, it was observed that the predicted EFs are increasing when the fLEGsharediff is increasing, that is, when forage legumes are included more years in the crop rotation (Figure 5A), and when the SOC stock of the reference is

TABLE 1 | Overview of experiments considered in the analysis. Experiment ID as is in the CarboSeq crop and soil management database v 2022_11_16, Country (superscript: Climatic zone); Sampling depth of the SOC data used in the analysis; Duration of the experiment at the moment that used SOC data sampled; Soil type according to the USDA classification; Main reference.

		Experiment ID	Country	Sampling depth	Duration	Soil type	Reference
Forage legumes	Perennial	Apelsvoll	Norway[a]	30	15	L	(Riley et al. 2008)
		Crop rotation experiment (CR)	Italy[b]	20	38	L	(Lugato et al. 2007)
		CROPSYS_Flakkebjerg	Denmark[b]	25	11	SL	(Schjønning et al. 2012)
		CROPSYS_Foulum	Denmark[b]	25	22	SL	(De De Notaris et al. 2021)
		Experimental farm near As Rotation & Fertilisation	Norway[a]	20	48	CL	(Cuvardic et al. 2004)
		Ihinger Hof	Germany[b]	25	14	SiL	(Friedel et al. 1996)
		Kaltinenai	Lithuania[a]	20	18	SL	(Feiza et al. 2008)
		KUNZO13V59.1	Czech Republic[b]	20	55	CL	(Kunzová 2013)
		La	Sweden[a]	20	16	SL	(Röing et al. 2005)
		Lo	Sweden[b]	20	16	SL	(Röing et al. 2005)
		Lodi - POC	Italy[b]	30	11	SL	(Tomasoni et al. 2011)
		Melle2	Belgium[c]	10	38	SL	(van van Eekeren et al. 2008)
		O_Ler	Denmark[c]	20	30	SL	(Christensen 1988)
		Ro	Sweden[a]	20	17	SiL	(Röing et al. 2005)
		U_Sand	Denmark[c]	20	30	LS	(Christensen 1988)
	Annual	CHRIS90V94.1	Denmark[c]	20	29	SL	(Christensen 1990)
		El Encin	Spain[d]	30	11	LS	(Hernanz et al. 2002)
		INIA-LTE-ROT	Spain[d]	30	18	SL	(Martín-Lammerding et al. 2015)
		MASCOT	Italy[d]	25	5	L	(Mazzoncini et al. 2010)
		rot_Etzdorf	Germany[b]	30	36	SiL	(Deumelandt et al. 2010)
		Torrepadierne	Spain[d]	30	6	CL	(Sombrero and de Benito 2010)
Grain legumes		10_CREA experimental farm Manfredini	Italy[d]	30	24	SiL	(Vanino et al. 2022)
		3_CREA experimental farm Manfredini	Italy[d]	30	16	CL	(Farina et al. 2013)
		CENTS Flakkebjerg	Denmark[b]	25	7	SL	(Hansen et al. 2015)
		CENTS Foulum	Denmark[b]	25	7	SL	(Hansen et al. 2015)
		GLBR	France[c]	30	6	L	(Plaza-Bonilla et al. 2016)
		Harste	Germany[b]	30	14	SiL	(Grunwald et al. 2021)
		Malagon	Spain[d]	30	20	C	(López-Bellido et al. 2020)
		Pietranera	Italy[d]	15	18	C	(Laudicina et al. 2014)
		RYCHC06V52.1	Poland[b]	25	37	SiL	(Rychcik et al. 2000 C.E.)

Abbreviations: C, clay; CL, clay loam; L, loam; LS, Loamy sand; SiL, Silt loam; SL, Sandy loam.
[a]Boreal.
[b]Continental.
[c]Atlantic.
[d]Mediterranean.

lower (Figure 5B). So, the higher the SOC stock of the reference, the lower the potential to increase the SOC stock by increasing the fLEGshare. It was also observed that when the SOC stock of the reference was higher than 2 Mg ha^{-1} cm^{-1} (which corresponds to about 60 Mg ha^{-1} for the 0–30 cm depth) the predicted EFs can be less than 1, especially when the fLEGshare is low.

TABLE 2 | Summary of the key variables in the dataset of forage legumes in the crop rotation which were used for the estimation of emission factors (EFs) and analysis. SOC stands for soil organic carbon. ($n = 39$ pairs of treatments and reference). The individual values for each pair of each experiment are shown in Appendix S1.

	Variable	Minimum	Mean	Median	Maximum
Experiment	Sampling depth (cm)	10.0	23.2	20.0	30.0
	Experimental duration (years)	5.0	28.2	29.0	55.0
Soil	SOCref (Mg ha^{-1})	11.4	42.5	39.2	88.5
	SOCtreat (Mg ha^{-1})	20.7	44.2	39.8	88.7
	SOCref/cm (Mg ha^{-1} cm^{-1})	0.57	1.86	1.64	3.69
	Clay content (%)	3.1	16.0	15.0	24.6
Climate	Annual precipitation (mm)	428	668	775	880
	Annual mean temperature (°C)	3.1	10.0	9.2	14.2
	Average aridity index	0.28	0.75	0.74	1.23
	EF	0.74	1.08	1.03	1.89

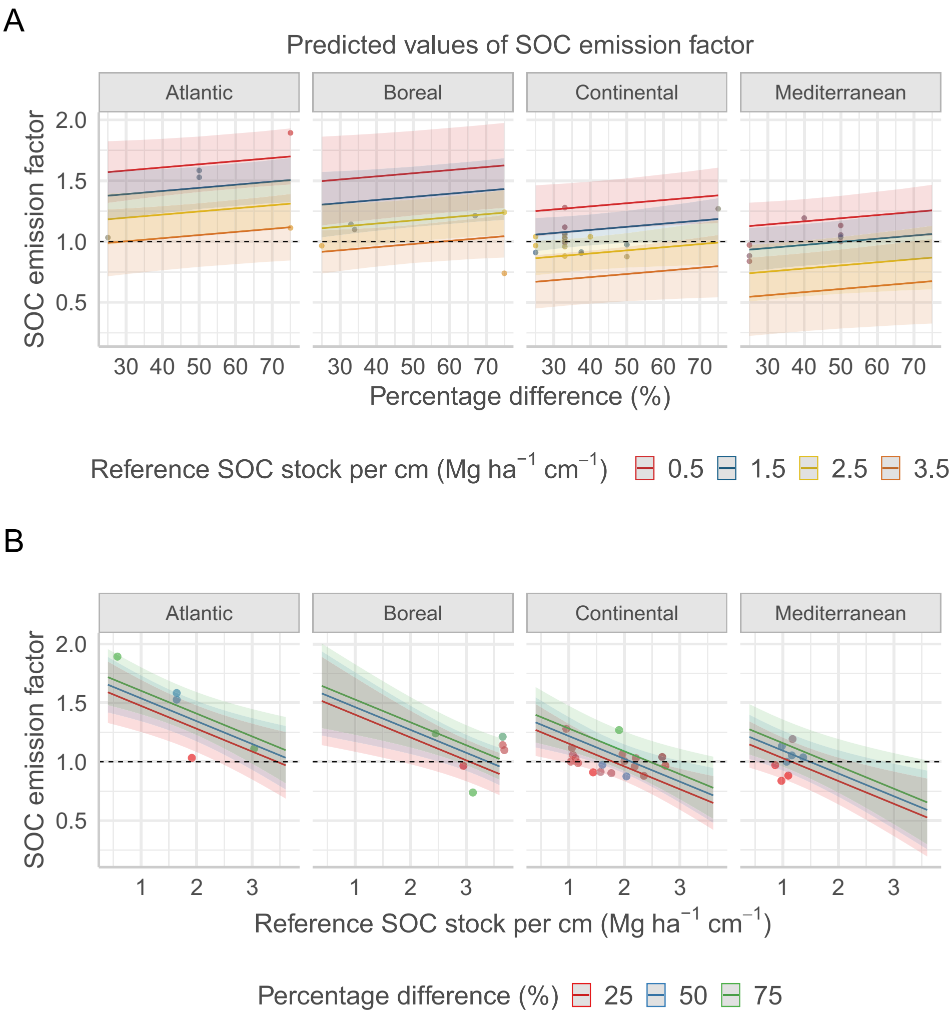

FIGURE 4 | (A) Estimated marginal means (predicted values) for the emission factor (EF) relatively to the percentage difference of forage legumes between the treatments and the reference (fLEGsharediff) for each climatic zone, when the soil organic carbon of the reference per cm (SOCref/cm) is 0.5 Mg ha^{-1} cm^{-1} (red line), 1.5 Mg ha^{-1} cm^{-1} (blue line), 2.5 Mg ha^{-1} cm^{-1} (yellow line) and 3.5 Mg ha^{-1} cm^{-1} (orange line) and confidence intervals at 95% level. (B) Estimated marginal means (predicted values) for the EF relatively to the SOCref/cm for each climatic zone, when the fLEGsharediff is 25% (red line), 50% (blue line) and 75% (green line) and confidence intervals at 95% level.

3.3 | SOC EFs for Increasing gLEGshare

The final dataset with only grain legumes used for further analysis included eight experiments located in five different countries (Figure 2) with 28 pairs of treatments and references (i.e., 12 in Poland, 6 in Italy, 4 in Spain, 3 in Denmark, 2 in France and 1 in Germany). The different grain legumes used were peas, chickpeas, faba bean, and field beans. The grain legumes either replaced other crop types such as cereals or were used as extra main crops in the rotation, without replacing an existing crop and hence leading to an expansion of the rotation. Most of the observed pairs were in areas with a Continental climate ($n = 16$) followed by those in Mediterranean ($n = 10$) and Atlantic ($n = 2$) climate zones. The statistical parameters (minimum, maximum, mean and median) of the dataset are presented in Table 3.

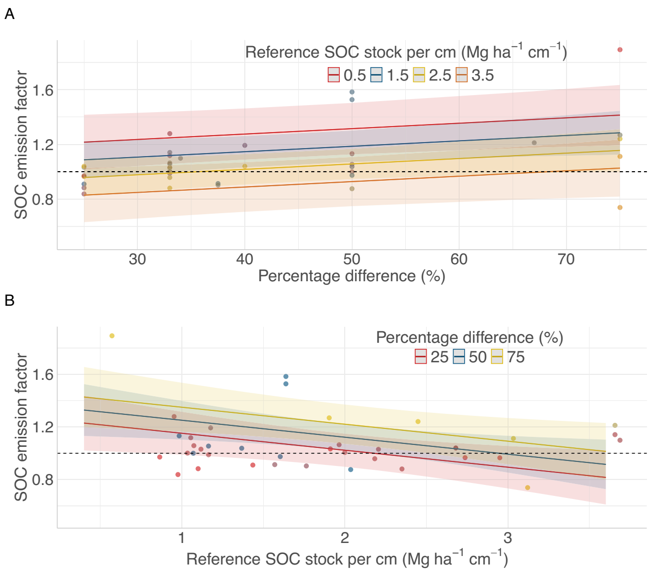

FIGURE 5 | (A) Estimated marginal means (predicted values) for the emission factor (EF) relatively to the percentage difference of forage legumes between the treatments and the reference (fLEGsharediff) when the soil organic carbon of the reference per cm (SOCref/cm) is 0.5 Mg ha^{-1} cm^{-1} (red line), 1.5 Mg ha^{-1} cm^{-1} (blue line), 2.5 Mg ha^{-1} cm^{-1} (yellow line) and 3.5 Mg ha^{-1} cm^{-1} (orange line) and confidence intervals at 95% level. (B) Estimated marginal means (predicted values) for the EF relatively to the SOCref/cm, when the fLEGsharediff is 25% (red line), 50% (blue line) and 75% (yellow line) and confidence intervals at 95% level.

The a priori LMM that was performed to evaluate the possible effects of experimental duration and sampling depth revealed that neither experimental duration ($p = 0.80$) nor the sampling depth ($p = 0.19$) had a significant impact on the obtained EF (Appendix S2 Table S2). Similarly with the forage legumes, we conducted a separate analysis for medium-term (≤ 20 years) and long-term (> 20 years) experiments to assess the impact of duration on the effect of grain legumes. The negative effect was more pronounced in LTEs but remained statistically significant in both cases, with no significant influence of duration or sampling depth (see Appendix S2 Table S3). Therefore, these variables were not included in the initial model (Equation 4).

After performing backwards elimination on the initial model (Equation 4 without the 'crop growth duration' variable as all grain legumes are annual) based on the AIC criterion, the best fitting model (Equation 7) used the predictor LEGsharediff ($p < 0.001$). The summarised results of the model are presented in Appendix S2 Table S6. The mathematical expression of the estimated predictive model takes the following form

$$EF \sim 1.0643179 - 0.0024389 \times \text{gLEGsharediff} \quad (7)$$

where gLEGsharediff refers to the additional fraction of grain legumes in the rotation (%) compared to the reference treatment.

The range of the predicted EFs relative to the gLEGsharediff is shown in Figure 6. The predicted EFs are lower when the gLEGsharediff is increasing, that is, when grain legumes are

TABLE 3 | Summary of the key variables in the dataset of grain legumes in the crop rotation which was used for the estimation of emission factors (EFs) and analysis. ($n = 28$ pairs of treatments and reference).

	Variable	Minimum	Mean	Median	Maximum
Experiment	Sampling depth (cm)	15	25.71	25	30
	Experimental duration (years)	6	24.61	22	37
Soil	SOCref (Mg ha^{-1})	18.47	38.90	31.28	74.88
	SOCtreat (Mg ha^{-1})	17.60	35.99	27.13	69.99
	SOCref/cm (Mg ha^{-1} cm^{-1})	0.62	1.54	1.28	2.59
	Clay content (%)	9.20	26.60	13.23	69.40
Climate	Annual precipitation (mm)	456	594	637	745
	Annual mean temperature (°C)	7.29	11.31	8.56	17.66
	Average aridity index	0.29	0.63	0.80	1.07
	EF	0.66	0.91	0.91	1.08

Note: SOCref stands for Soil organic carbon stock of the reference, SOCtreat for Soil organic carbon stock of the treatment, and SOCref/cm for Soil organic carbon stock of the reference per cm depth. The individual values for each pair of each experiment are shown in the Appendix S1.

A

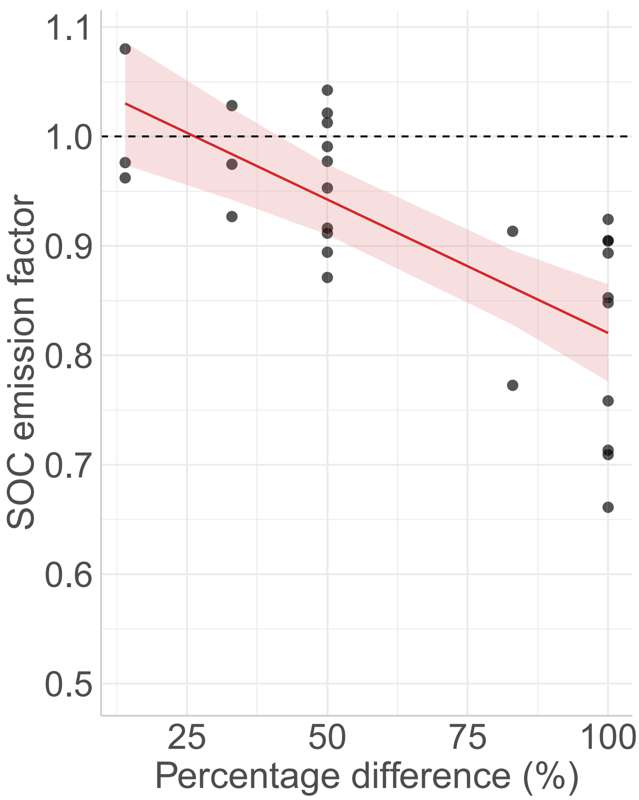

FIGURE 6 | Estimated marginal means (predicted values) for the EF relative to the percentage difference of grain legumes between the treatments and the reference (gLEGsharediff) and confidence intervals at the 95% level.

included for more years in the crop rotation, or the rotation is converted to a monoculture of legumes (gLEGsharediff = 100%).

3.4 | The Use of the Regression Model for Scenarios and Stock Estimations

In this study, the EF proposed is not a numerical response ratio but rather regression equations to include the variables that affect it.

Considering different scenarios for increased fLEGshare (Table 4) and gLEGshare (Table 5), (i) the EFs, (ii) the change of SOC stocks from the reference, and (iii) the annual change of SOC stocks are presented. The average duration of the alternative rotation required for these changes is considered to be 30 years, which is approximately the average duration of experiments included in the dataset for grain and forage legumes together (27.8).

For example, if a 4-year rotation only including cereal crops is considered and forage legumes are added to the system for 1 year, the percentage of more forage legumes in the treatment than in the reference will be 25%, and for 3 years it will be 75%. In a soil with reference SOC stock of 60 Mg ha^{-1} in the 0–30 cm depth layer, there is a potential for increasing the SOC stocks by about 1.37 Mg ha^{-1} for 25% more forage legumes, up to about 13.25 Mg ha^{-1} when including 75% more forage legumes, respectively. This corresponds to about 0.05 Mg ha^{-1} year^{-1} up to about 0.44 Mg ha^{-1} year^{-1} when the specific rotation is applied on average for 30 years.

It is observed that, if for example a 4-year rotation without legumes is considered and grain legumes added in the system for 1 year (i.e., the percentage of more grain legumes in the treatment than in the reference will be 25%) there will be no meaningful change in the SOC stocks, while if all crops in the rotation are replaced with grain legumes then the SOC stock can present a decrease up to 14.37 Mg ha^{-1} when the specific rotation is applied on average for 30 years.

4 | Discussion

The calculation of EFs offers a standardised approach to estimate greenhouse gas emissions resulting from specific activities, such as changes in land use or land management practices compared to a reference (IPCC 2019, 2022; Lehtinen et al. 2014). Here the same principle was used to estimate the impact of agricultural practices on CO_2 emissions from soils expressed as changes in the SOC stocks, allowing for the calculation of the SOC accrual potential in support of the development of policies and incentives to promote climate mitigation.

This study presents EFs that can be used to estimate SOC stock changes due to the increase in the LEGshare in a Tier-2 approach and on a large scale in Europe. This was achieved by using a dataset of European MTEs/LTEs distributed in the different European pedoclimatic conditions.

While EFs offer a valuable tool for estimating SOC stock changes resulting from agricultural management practices, their application requires careful consideration of the limitations and uncertainties associated with their estimations and use. EFs are typically based on data from a limited number of sites, which may not represent the full range of environmental and management conditions across different regions. This can result in underestimation or overestimation of the effects of certain management practices, also determining uncertainty in the estimates, particularly when extrapolating EFs to regions with different conditions (GHG Protocol 2014). Here, to increase the robustness of the results and to provide an EF that can be applied and extrapolated in Europe, MTEs and LTEs that represent the full range of environmental and management conditions across different regions were considered. The proposed EFs were complemented with additional information on soil properties, climate conditions, and other relevant management information of the dataset as using EFs outside of these ranges should be approached with caution. For forage legumes, the climatic zone was even found to be a statistically significant explanatory variable. Nevertheless, the distribution of available data was not balanced among and within the different climatic zones, raising concerns if specific EF can be drawn for each climatic zone of Europe (apart from the Continental). Thus, a more generalised EF regression is proposed for the whole of Europe.

In the analysis for the estimation of the EFs, experiments with different durations were considered. Changes in soil properties tend to occur slowly (Grosse et al. 2020) and SOC equilibrium is reached over the long term. However, even shorter-term

TABLE 4 | Emission factors (EFs), change of SOC stocks (Mg ha^{-1}) and annual change of SOC stocks considering different scenarios for reference SOC stock and fLEGshare estimated for 30 cm soil depth and when applied on average for 30 years based on the selected model. Blue colour indicates EF higher than one, SOC stocks of the treatment higher than the reference and ultimately increase in SOC stocks, while red colour indicates EF lower than one, SOC stocks of the treatment lower than the reference and ultimately SOC stock losses.

	Emission factor (Equation 6a)			Change of SOC stocks (Mg ha^{-1}) (Equation 2)			Annual change of SOC stocks (Mg ha^{-1} year^{-1})		
	Percentage of more forage legumes in the treatment rotation than in the reference (%)								
Reference C stock at 0–30 cm depth layer (Mg ha^{-1})	25%	50%	75%	25%	50%	75%	25%	50%	75%
20	1.20	1.29	1.39	3.90	5.88	7.86	0.13	0.20	0.26
40	1.11	1.21	1.31	4.36	8.32	12.27	0.15	0.28	0.41
60	1.02	1.12	1.22	1.37	7.31	13.25	0.05	0.24	0.44
80	0.94	1.04	1.13	−5.05	2.86	10.78	−0.17	0.10	0.36

TABLE 5 | Emission factors (EFs), change of SOC stocks (Mg ha^{-1}) and annual change of SOC stocks considering different scenarios for reference SOC stock and gLEGshare estimated for 30 cm soil depth and when applied on average for 30 years based on the selected model. Blue colour indicates EF higher than one, SOC stocks of the treatment higher than the reference and ultimately increase in SOC stocks, while red colour indicates EF lower than one, SOC stocks of the treatment lower than the reference and ultimately SOC stock losses.

	Change of SOC stocks (Mg ha^{-1}) (Equation 2)			Annual change of SOC stocks (Mg ha^{-1} year^{-1})		
	Percentage of more forage legumes in the treatment rotation than in the reference (%)					
Reference C stock at 0–30 cm depth layer (Mg ha^{-1})	25%	50%	100%	25%	50%	100%
20	0.07	−1.15	−3.59	0.00	−0.04	−0.12
40	0.13	−2.31	−7.18	0.00	−0.08	−0.24
60	0.20	−3.46	−10.77	0.01	−0.12	−0.36
80	0.27	−4.61	−14.37	0.01	−0.15	−0.48
Emission factor (Equation 7)	1.00	0.94	0.82			

experiments can provide valuable insights and highlight important trends (Debreczeni and Körschens 2003). While MTEs may not capture the equilibrium state for SOC stocks, they provide meaningful information complementary with LTEs and therefore were kept in the analysis. To evaluate the effects of experimental duration on EFs, priori models were employed. The analysis revealed that the duration of experiments was not a statistically significant factor either for grain or for forage legumes. Similarly, when focusing exclusively on MTEs (< 20 years) or when considering part of the dataset without the longer-term experiments (< 30 years, < 40 years) the experimental duration remained an insignificant factor. While the importance of experimental duration is well recognised in soil science for understanding and validating the sustained impacts of agricultural practices on SOC changes, considering the model complexity versus the model accuracy, for the purpose of estimating EFs at a Tier 2 level, it was decided to keep only the significant explanatory variables. Also, a more geographically representative dataset—including MTEs—can provide valuable insights about the trends. This is particularly relevant as long-term agricultural interventions are challenging to plan and implement in real-world scenarios. Therefore, EFs derived from shorter-duration experiments can be effectively used to estimate potential SOC changes resulting from agricultural practices. Given these considerations, it was assumed that the obtained EFs, estimated using experiments with a duration of 5–55 years, are valid after 30 years, which is approximately the average duration of experiments included in the dataset.

The priori analysis revealed also that the varying sampling depths among the experiments did not significantly affect the EFs. This can be explained by the fact that all experiments were only sampled in the topsoil (up to 30 cm depth). Topsoils in European croplands are typically ploughed and thus homogenised, so the difference between sampling depths within the topsoil should be negligible. Assuming thus that the SOC is equally distributed and not stratified within the sampled soil layer, the SOC stock of the reference used as an explanatory variable in the models was normalised to depth and expressed per cm to homogenise the SOC stock across the different studies considered, given that the sampling depths varied between 10 and 30 cm.

The individual EFs from each experiment, as calculated here, considered only the latest time point to estimate the difference between the treatment and reference at that point (i.e., comparison with a baseline scenario) and did not consider the initial conditions. Even if EFs can be useful for estimating SOC differences between two management practices, they do not provide direct information on whether an applied management measure leads to C sequestration in soils or SOC loss mitigation, which are two related but distinct concepts (Don et al. 2024) that can both be captured by EFs as the initial conditions are not taken into consideration. Thus, if the target is to evaluate if a practice leads to C sequestration in soils (global net C increase), it is crucial to consider the initial SOC stocks together with the evolution of the reference SOC stock.

4.1 | Effects of Introducing Legumes Into the Crop Rotation

The magnitude of the C accrual potential in soils varies among the different leguminous species depending on their type, whether they are grown on an annual or perennial basis, their decomposition rates, their C/N ratio, and the climatic conditions in which they grow (Mesgar et al. 2024; Whitbread et al. 2000; Young et al. 2009). We demonstrated that the type of legume cultivated plays a significant role in the potential to increase the SOC stocks. Forage legumes such as alfalfa, clover, and vetch were shown to increase SOC stocks, as is also shown by Guan et al. (2016). In contrast, grain legumes did not enhance the SOC stocks, as also found by Oliveira et al. (2019). A similar trend was demonstrated in the global metanalysis of Beillouin et al. (2023). They showed that annual grain legume-based rotations led to a 3.8% relative SOC change, and annual/perennial forage legume-based rotations to a 4.1% relative increase in SOC, compared to exclusive cereal-based rotations. This occurred possibly because forage legumes, even if they are annual, usually remain in the field for a longer period in comparison to grain legumes and therefore provide higher C inputs through their rooting system. Also, various leguminous forage crops, such as alfalfa (Fan et al. 2016) and red clover, have deeper rooting systems and secrete a great proportion of root exudates which can promote deeper and longer-lasting SOC storage (Bolinder et al. 2007; Jensen et al. 2012; van der Pol et al. 2022). Root-derived C contributes more to the stable soil C pools and prolongs the time that assimilated C remains in the soil compared to aboveground crop residue-derived C (Kätterer et al. 2011; Poeplau et al. 2021).

The positive effect of annual and perennial forage legumes on SOC stock accrual seems to be higher with increasing proportion of the forage legumes in the rotation (Jarvis et al. 2017; Zani et al. 2021) and with the use of perennial legume species compared to annual species (Bolinder et al. 2012; Reckling et al. 2016b). In our study, the positive effects of forage legumes on SOC accrual were evident both with annual (Deumelandt et al. 2010; Hernanz et al. 2002; Mazzoncini et al. 2010; Sombrero and de Benito 2010) and perennial forage legumes (Cuvardic et al. 2004; Deumelandt et al. 2010; Kunzová 2013; Röing et al. 2005; van Eekeren et al. 2008) (Figure 1) compared to the reference rotation. Forage legumes, even if grown as annual crops, foster SOC accrual via several mechanisms: First of all, legumes supply a great amount of N to the soil and the subsequent crop thanks to their N fixation capacity (Oliveira et al. 2019). Well-managed legume-based cropping systems, where nitrous oxide (N_2O) emissions and N fertiliser use are reduced, can further contribute to the mitigation of greenhouse gas emissions in agricultural systems (Reckling et al. 2016a).

Additionally, legume cultivation can play an important role in increasing C storage due to its impact on the maintenance of a favourable low soil C/N ratio (Veloso et al. 2019) which allows the microbial community to decompose the added biomass without needing to break down existing SOC to obtain energy (Franke et al. 2008). Consequently, legume cultivation increases N availability in the soil, and especially the readily available N that is added by the roots and root exudates, which substantially favours C storage via improved microbial activity (Mitchell et al. 2017). Changes in soil microbial composition can facilitate root and plant growth that further promotes microbial activity (Bremer and van Kessel 1992; Zak et al. 2003). Then, increased microbial abundance and diversity, root exudates, low weight organic elemental exudation, and residue decomposition stimulated by legume cultivation can together increase aggregate associated C stabilisation (Udom and Omovbude 2019). Enhanced formation of stable aggregates increases SOC accrual due to low C mineralisation and protects SOC against microbial degradation processes for a longer period (Veloso et al. 2019; Virk et al. 2022). Although forage legumes may produce less aboveground residue compared to, for example, cereal crops, they often have a higher proportion of nitrogen allocated to vegetative tissues (stems, leaves, roots), which have lower C:N ratios. This makes their biomass more easily decomposable by soil microbes, promoting microbial activity and facilitating the incorporation of organic matter into the soil, thereby increasing SOC stocks and enhanced aggregate stabilisation (Franke et al. 2008; Gil and Fick 2001). Thus, forage legume cultivation not only increases C and N in the soil but also improves its biological functioning and soil structure. In contrast, grain legumes allocate a significant portion of their nitrogen resources to seed production, as seeds require high N content for protein synthesis (Salon et al. 2001). As a result, the rest of the plant (the vegetative parts) tends to have a higher C:N ratio, which slows down decomposition.

Another benefit of forage legumes is that they can reduce the need for tillage, especially when grown continuously for more than 1 year (Bolinder et al. 2012), which allows for reduced soil disturbance and preserves SOC stocks even more (Virk et al. 2022). Nevertheless, in the case of temporary leys, their termination, whether mechanical or by herbicides, can lead to increased N mineralization and subsequent N_2O emissions depending on the timing of these management activities. As legumes produce N-rich residues, they could lead to increased N leaching to the environment or additional N_2O emissions as the amount of N may not be completely taken up by the next crop (Andrews et al. 2007; Cassman et al. 2002; Jensen et al. 2012). Thus, legumes can contribute to nitrate leaching and N_2O emissions if crop rotation and the synchrony of crop N supply with the demand from subsequent crops are not properly designed and managed. Nevertheless, increasing the fLEGshare in well-managed systems appears to be a promising option for potential SOC accrual and for creating a net reduction in N_2O emissions (Guenet et al. 2021; Jensen et al. 2012; Schmeer et al. 2014).

Our results show that the EFs (response ratio) relative to the reference SOC are influenced also by climatic zone, even if the predicted differences were statistically significantly different only between the Atlantic and the Mediterranean zone. While larger initial SOC stocks generally lead to smaller relative SOC increases, climatic conditions can play a crucial role. Our results indicate that the greatest potential to increase SOC stock by increasing fLEGshare was observed in the Atlantic zone, followed by the Boreal, Continental, and Mediterranean zones. This ranking may seem counterintuitive at first, given that the Boreal and Continental zones exhibit the highest initial SOC stocks. However, this pattern may be explained by differences in climatic conditions and decomposition rates (Wiesmeier et al. 2019). The Atlantic zone exhibited the highest SOC accrual potential, likely due to moderate reference SOC stocks, high biomass inputs and net primary productivity, and a humid climate that favours accumulation. The Boreal zone, despite its high reference SOC stocks (statistically significantly higher than all the other climatic regions), also showed a strong response, likely due to slower decomposition rates in colder conditions, whereas the Mediterranean zone had the lowest SOC increase, likely due to a combination of low initial SOC stocks and climatic constraints. Lower precipitation and water availability limit the net primary productivity, while higher temperatures accelerate organic matter decomposition and limit SOC accumulation potential. However, data distribution across zones was unbalanced, which may have influenced the observed patterns. Since the linear models extrapolate trends beyond observed data points, response ratios in data-sparse regions (e.g., Mediterranean) should be interpreted with caution. Further studies with more balanced datasets are needed to refine these findings, improve model accuracy, and allow for reliable EFs for each climatic zone.

Legume yields in Europe have been found to be unstable, highly dependent on the local conditions and not competitive with maize or cereal crops (Reckling et al. 2018; Sellami et al. 2019). The cultivation of legumes at the expense of existing crops constitutes an extensification that can lead to an increase in cultivated cropland and thus generate higher CO_2 emissions due to land-use change (Beillouin et al. 2023; Houghton et al. 2012; Huang et al. 2023). Nevertheless, this study among others (Jensen et al. 2012) showed that including more forage legumes and temporary leys with legume mixtures in the rotation can be a valuable measure for increasing SOC storage in agricultural systems. Moreover, as forage systems are often combined with increased manure inputs, either external or during grazing, C stocks can be even further increased (Bolinder et al. 2012). Also, legume-based grass mixtures improve both N use efficiency in the cropland as well as the protein quality of the silage (Van den Bossche et al. 2024), and can generate significant agronomic benefits in terms of yield, agronomic quality, cost and feed quality compared with solely grass leys (Malisch et al. 2024; Murphy-Bokern et al. 2017; Peyraud et al. 2009). Further, by reducing the need for synthetic N fertiliser, forage legumes can help to create a more self-sufficient sustainable agricultural management system and contribute to mitigating climate change effects.

5 | Conclusion

We have demonstrated that incorporating forage legumes into a crop rotation leads to SOC accrual, in contrast to grain legumes. Based on 39 paired treatments at 21 MTEs/LTEs with an increasing share of forage legumes in the crop rotation across various pedoclimatic zones of Europe, we derived an average EF of 1.08 (95% CI: 1.001 to 1.119) and we propose a linear regression to calculate EFs for the relative increase of SOC stocks. The controlling factors of this relationship are (1) increased forage legume share in the rotation (positive effect) and (2) the SOC stock per cm of the reference (negative effect). For grain legumes, based on eight experiments and 28 pairs, the SOC stocks decreased with a mean EF of 0.91 and with a 95% CI of 0.861 to 0.943. Respectively, for the proposed linear regression to calculate EFs for the relative increase of SOC stocks due to more grain legumes in the rotation, the controlling factor is the proportion of grain legumes only (negative effect). The linear regressions presented here can be used for estimating the C accrual potential of legumes on a large scale and across Europe. Based on these study results, it is strongly encouraged to reintroduce forage legumes in crop rotations and possibly substitute a fraction of maize-based fodder production, as they can increase SOC stocks in cropland soils, thereby helping to maintain soil health and food security and also to mitigate climate change through CO_2 emissions reduction.

Author Contributions

Ioanna S. Panagea: methodology, software, data curation, investigation, formal analysis, visualization, writing – original draft. **Paul Quataert:** formal analysis, visualization, writing – review and editing, validation, software. **María Alonso-Ayuso:** writing – review and editing, investigation. **Teresa G. Bárcena:** writing – review and editing, investigation. **Maarten De Boever:** writing – review and editing, investigation, methodology. **Mariangela Diacono:** writing – review and editing, investigation. **Anna Jacobs:** writing – review and editing, investigation. **Johannes L. Jensen:** writing – review and editing, investigation. **Felix Seidel:** conceptualization, writing – review and editing, investigation. **Daria Seitz:** writing – review and editing, investigation. **Heide Spiegel:** writing – review and editing, funding acquisition, investigation, conceptualization. **Thijs Vanden Nest:** writing – review and editing. **Axel Don:** conceptualization, methodology, funding acquisition, writing – review and editing. **Greet Ruysschaert:** conceptualization, methodology, validation, supervision, funding acquisition, writing – review and editing.

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Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

The complete EJP SOIL CarboSeq crop and soil management database is available at https://doi.org/10.5281/zenodo.8130195. The export module in the form of a Jupyter Notebook with Voila widgets, to select and extract specific information from the EJP SOIL CarboSeq crop and soil management database, is available at https://doi.org/10.5281/zenodo.7415077.

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Supporting Information

Additional supporting information can be found online in the Supporting Information section.