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Integrating upscaling simulation methods for predicting soil organic Carbon changes in Spain

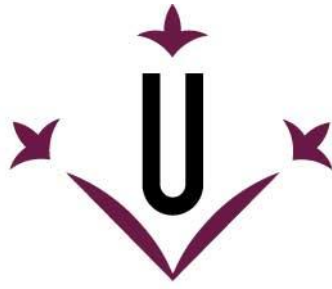
Asma Jebari

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Universitat de Lleida

TESI DOCTORAL

Integrating upscaling simulation methods for predicting soil organic Carbon changes in Spain

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Memòria presentada per optar al grau de Doctor per la Universitat de Lleida
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SUMMARY

Soil organic carbon (SOC) stocks and greenhouse gas (GHG) emissions assessment at the regional scale under climate change scenarios are of paramount importance in implementing management practices to mitigate climate change effect. The main objective of this Thesis was to assess SOC changes and GHG emissions under different agricultural systems (croplands and grasslands) and climatic conditions (Mediterranean and moist temperate) in Spain under different climate scenarios. Furthermore, different alternative management practices to mitigate climate change effects for the considered agroecosystems were also evaluated.

A calibrated version of the SOC model RothC was constructed to estimate the changes in SOC under climate change conditions for croplands of Mediterranean Spain across a total surface area of 23 300km² during the 2010 to 2100 period. It was also simulated current and future (2010–2100) net GHG emissions in more than 4050 km² of moist temperate Spanish grasslands associated to dairy production under different climate scenarios. For SOC dynamics estimation, the RothC model was modified to fit to managed moist temperate grasslands considering: (1) the incorporation of distinction for plant residues components (i.e., above- and below-ground residues and rhizodeposition) in terms of quantity and quality, (2) ruminant excreta quality, and (3) the extension of soil moisture up to saturation conditions. For GHG estimation, it was used mainly Tier 2 IPCC methodologies to estimate the CH₄ and N₂O emissions from enteric fermentation, manure storage and handling, and grassland soils.

According to my findings, among both agroecosystems (i.e., croplands and grasslands), climate change generally led to a decline in SOC content compared with baseline scenarios. Furthermore, C input was the key factor of SOC storage across Mediterranean croplands and moist temperate Spanish grasslands. Additionally, it was found that air temperature rather than precipitation was the climatic factor contributing to most of variation in SOC changes values. Moreover, livestock density was the

main factor affecting net GHG emissions in the grasslands associated to dairy production of Northern Spain.

It was concluded that changes in management could enhance the amount of SOC sequestered and reduce GHG emissions under climate change conditions. Under Mediterranean croplands, no-tillage, in the case of rainfed crops, and vegetation cover, for olive groves and other woody crops, were the alternative management strategies to alleviate climate change effects and SOC loss. In addition, under moist temperate grassland-based dairy livestock systems, alternative manure management practices (particularly, anaerobic digestion) were efficient to mitigate the climate change effects and to reduce the net GHG emissions, while more mitigation could be achieved by optimising the livestock density management.

RESUMEN

La evaluación de stocks de C orgánico del suelo (COS) y emisiones de efecto invernadero (GEI) a escala regional bajo escenarios de cambio climático es de fundamental importancia a la hora de implementar estrategias de manejo para mitigar el cambio climático. El principal objetivo de esta Tesis es evaluar los cambios del COS y GEI en diferentes sistemas agrícolas (e.g., tierras de cultivo y pastos) y diferentes condiciones climáticas (Mediterráneo y templado húmedo) de España bajo diferentes escenarios climáticos. Además, evalué las estrategias de manejo con el objetivo de mitigar los efectos del cambio climático.

En el estudio de modelización espacial, se adoptó una versión calibrada del modelo de COS RothC para estimar los cambios en los stocks de COS en condiciones de cambio climático en las tierras de cultivo de la España mediterránea en una superficie total de 23 300 km² durante el período 2010 a 2100. También simulé las presentes y futuras (2010-2100) emisiones netas para unos 4050 km² de pastos asociados a la producción lechera de la zona templada húmeda de España. Para la estimación del COS, se modificó el modelo RothC para adaptarlo a los pastos templados húmedos considerando: (1) la incorporación de los diferentes componentes de los residuos vegetales (parte aérea, parte subterránea y rizodeposición) diferenciando su calidad y cantidad, la diferenciación de la calidad de la excreta de los rumiantes, y la extensión de la función de humedad del suelo considerando condiciones de saturación. Para la estimación de los GEI, se usó la metodología refinada del IPCC (Tier 2) considerando emisiones de CH₄ and N₂O provenientes de la fermentación entérica, del manejo de la excreta y del suelo de los pastos.

Según los resultados encontrados en ambos agroecosistemas (es decir, tierras de cultivo y pastos), el cambio climático generalmente condujo a una disminución en el contenido de COS en comparación con los escenarios *baseline* de referencia. Concluimos que el aporte de C es el factor clave del almacenamiento de COS en las tierras de cultivo mediterráneas y los pastos templados

húmedos y que la temperatura del aire es el factor climático que contribuyó más a las variaciones en el COS. Además, la densidad ganadera fue el factor que más afectó a las emisiones netas en los pastos asociados a la producción lechera en el Norte de España.

En conclusión, las alternativas de manejo mejoraron la cantidad de COS almacenado y eran estrategias efectivas para reducir las emisiones GEI bajo las condiciones futuras del cambio climático. La siembra directa, en el caso de los cultivos de secano, y la cubierta vegetal, para los olivares y otros cultivos leñosos, fueron las alternativas de manejo eficaces para reducir los efectos del cambio climático y la pérdida de COS. Además, en el caso de pastos templados y húmedos asociados a la producción lechera, las prácticas alternativas de manejo del estiércol (en particular, la digestión anaeróbica) ayudaron a mitigar los efectos del cambio climático y a reducir los GEI netos, mientras que se podría lograr una mayor mitigación mediante la optimización de la densidad ganadera.

RESUM

L'avaluació d'estocs de C orgànic del sòl (COS) i emissions d'efecte hivernacle (GEH) a escala regional sota escenaris de canvi climàtic és fonamental a l'hora d'implementar estratègies de maneig per mitigar el canvi climàtic. El principal objectiu d'aquesta Tesi és avaluar els canvis del COS i GEH en diferents sistemes agrícoles (en terres de cultiu i de pastura) i en diferents condicions climàtiques (clima mediterrani i temperat humit) d'Espanya sota diferents escenaris climàtics. A més, s'avaluen les estratègies de maneig de la terra amb l'objectiu de mitigar els efectes del canvi climàtic.

A l'estudi de modelització espacial, es va adoptar una versió calibrada del model de COS RothC per estimar els canvis en els estocs de COS en condicions de canvi climàtic a les terres de cultiu de l'Espanya mediterrània en una superfície total de 23 300 km² durant el període 2010-2100. També es van simular emissions presents i futures (2010-2100) netes per a uns 4050 km² de terres de pastura associades a la producció lletera de la zona temperada humida d'Espanya. Per a l'estimació del COS, es va modificar el model RothC per adaptar-lo a les terres de pastura temperades humides, considerant: (1) la incorporació dels diferents components dels residus vegetals (part aèria, part subterrània i rizodeposició) diferenciant-ne la qualitat i la quantitat, (2) la diferenciació de la qualitat de l'excreta dels remugants, i (3) l'extensió de la funció d'humitat del sòl considerant condicions de saturació. Per a l'estimació dels GEH, es va fer servir la metodologia refinada de l'IPCC (Tier 2) considerant emissions de CH₄ i N₂O provinents de la fermentació entèrica, del maneig de l'excreta i del sòl de les pastures.

Segons els resultats trobats en els dos agroecosistemes (és a dir, terres de cultiu i de pastura), el canvi climàtic condueix a una disminució en el contingut de COS en comparació amb els escenaris de base de referència. Concloem que l'aportació de C és el factor clau de l'emmagatzematge de COS a les terres de cultiu mediterrànies i les terres de pastura temperades humides i que la temperatura de l'aire és el factor climàtic que contribueix més a les variacions del COS. A més, la densitat ramadera és el factor que més afecta les emissions netes a les terres de pastura associades a la producció lletera al Nord d'Espanya.

En conclusió, les alternatives de maneig milloren la quantitat de COS emmagatzemat i es mostren com a estratègies efectives per reduir les emissions GEH sota les condicions futures de canvi climàtic. La sembra directa, en el cas dels cultius de secà, i la coberta vegetal, per als camps d'oliveres i altres cultius llenyosos, són les alternatives de maneig que resulten més eficaces per a reduir els efectes del canvi climàtic i la pèrdua de COS. A més, en el cas de les terres de pastura temperades i humides associades a la producció lletera, les pràctiques alternatives de maneig dels fems (en particular, la digestió anaeròbica) afavoreixen a mitigar els efectes del canvi climàtic i a reduir els GEH nets, alhora que contribueixen a la mitigació mitjançant l'optimització de la densitat ramadera.

INTRODUCTION

Soil organic carbon storage and its potential to mitigate greenhouse gas emissions from agriculture

Agriculture is practiced on 49% of the global ice-free land surface, 12% as cropland, and 37% as pasture, and is responsible for 12% of the total direct anthropogenic greenhouse gas (GHG) emissions (IPCC 2019). In this context, some cultivated soils have lost from one-half to two-thirds of the original SOC pool with a cumulative loss of 30-40 Mg C ha⁻¹. Additional C losses in managed grasslands occur through methane (CH₄) emissions most of it from enteric fermentation (Martin et al. 2010). However, it is estimated that agricultural lands have the potential to re-plenish up to 66% of historical carbon (C) loss, if managed properly (Lal 2004). Globally, the proportion of organic C stored in soils is approximately twice as large as that in the atmosphere and three times that in vegetation (Batjes 1996). As a consequence, a small change to this soil C has the potential to significantly modify the C dioxide (CO₂) concentration of the atmosphere (Schlesinger and Andrews 2000). Thus, one of the most effective ways of mitigating GHG emissions is to increase rates of soil organic C (SOC) sequestration in agricultural lands (Smith et al. 2008), with an additional benefit of improving soil structure and conditions (Lal 2017). In this context, the 4 per 1,000 initiative (<http://www.4p1000.org/>) launched by France at the COP 21 in 2015 stressed the fact that C sequestration in agricultural soils should be considered as one of the key GHG mitigation strategies for reaching the global temperature goals of the Paris Agreement. The maintenance of SOC and soil structure together with protection of soils against erosion were the two pillars to protect and enhance the soil quality and functions in the Common Agricultural Policies (CAP) (Borrelli et al. 2016).

To address global needs for food security and climate change mitigation, priority actions to increase SOC stocks need to focus on improved management practices having high potential for increasing C storage (Whitehead 2020). For instance, Zomer et al. (2017) estimated that global cropland soils could sequester 26–53% of the target C storage of the 4 per 1,000 Initiative (Soussana et al. 2019). However, predicting the impacts of management on grassland soil stocks is problematic because of the complex interactions among climate and soil types (Conant et al. 2017), and management practices including grazing intensity (Zhou et al. 2017), the removal of biomass and the return of C in dung (Soussana and Lemaire 2014). Also because of the interaction with the N cycle and the potential nitrous oxide (N₂O) trade-offs by some SOC sequestration strategies (Guenet et al. 2021).

Agriculture in Spain: Contribution to climate change and mitigation potential

At a National spatial level, Spain has the fourth highest land area assigned to agriculture of the European Union (EU) countries. Thus, agricultural activities in Spain generate significant GHG emissions to the atmosphere. In 2019, the agricultural sector in Spain emitted 37,794 kt of CO₂-e (UNFCCC 2020). This value represented 12% of the total GHGs emitted in the country. Enteric fermentation represented the main GHG emitter (about 42.4% of the total GHGs emitted by the Spanish agricultural sector) followed by agricultural soils (32.5%) and manure management (22.7%) (MITECO 2021). These GHGs are weighted by their 100-year Global Warming Potentials (GWPs), using values consistent with the IPCC fourth assessment report: GWP (N₂O) = 298 and GWP (CH₄) = 25. Spain's diversity in climate and geography circumstances greatly influences the large heterogeneity in agriculture and livestock production systems and thus their associated environmental challenges. As study cases, we opted for different regions of Spain characterised by different climatic conditions (Mediterranean vs moist temperate) and ecosystems (croplands vs grasslands).

Croplands under Mediterranean region as a contributor to climate change and an opportunity for SOC storage: Aragon as an example

Aragon has an irregular orography and large climate heterogeneity, where the Mediterranean climate prevails except in the Pyrenees. It is located at the centre of the Ebro Depression, presenting one of the severest climates in Spain as it experiences a dry summer climate (Mediterranean or semiarid) with scarce rainfall and high potential evaporation, as well as a total annual rainfall ranging from 400 to 600 mm.

Most of the land is cultivated by rainfed crops (74%). The irrigated land, however, is far more productive and accounts for the better part of Aragon's agricultural output. Woody crops in Aragon represent 30% of the sales of vegetative products of the study area, with almonds (*Prunus dulcis* L.), olives (*Olea europaea* L.), and grapes (*Vitis vinifera* L.) as the main crops (DGA 2012).

Irrigated crops (mainly corn (*Zea mays* L., 47%) and barley (*Hordeum vulgare* L., 22%)) are commonly grown as part of a rotation while rain-fed crops, mainly barley (65%) and durum wheat (*Triticum aestivum* L., 31%) are commonly managed under either cereal–fallow rotations or monocropping together with intensive tillage (56% of rainfed surface is intensively tilled). Historically, practices such as intensive tillage and long fallowing has exacerbated SOC loss resulting in an excellent opportunity to sequester atmospheric C in soils through the adoption of alternative management practices (Moreno et al. 2010).

Dairy production under moist temperate conditions and their contribution to climate change

The livestock sector contributes nearly half the total agricultural emissions in Spain according to the Spanish Ministry for the Ecological Transition. According to data from the European Commission (EUROSTAT 2019), Spain is one of the seven major producers of cow milk in the EU. Of the 156 million tonnes that the EU is estimated to produce per year, Spain provides 5.1%, having provided 7.2 million tonnes in 2019. The dairy farming activity is mainly located in regions of the Northern moist temperate zone, as grasslands of these areas are very productive as a consequence of adequate climatic conditions of frequent rainfall and cool temperature (Smit et al. 2008). The climate provides also favourable conditions for microbial soil processes such as denitrification (Estavillo et al. 1994) potentially resulting in large N₂O emissions (Merino et al. 2001). The dairy farming activity is characterised by a gradient of productive intensification with farm size, explained by an increase of the per animal and per hectare productivity (Flores-Calvete et al. 2016).

The main production model is a small familiar farm with an average of cows per herd of 31.6 and an average milk production per cow per year (kg) of 7.323 kg (Flores-Calvete et al. 2016), with a low–medium level of industrialization and a feeding regime based in local forages (pasture, grass and corn silage) supplemented with concentrate feed (Orjales et al. 2018). Grassland ecosystems of dairy production in Northern Spain are commonly based on grass-white clover mixture; mainly ryegrass with around 5% of white clover (*Trifolium repens*. L.). The intensification of dairy production systems favoured manure management which increases the potential for both CH₄ and N₂O emissions (Petersen 2018). Urine and faeces of lactating dairy cows are generally excreted in the stable. This manure is stored as liquid (slurry) in tanks or lagoons. However, excreta from dry dairy cows and heifers are generally mixed with straw and other bedding and handled as solid manure, farmyard manure (FYM). Cow slurry is spread in most farms on their grassland fields, while mineral fertilizer N application is almost negligible.

Soil Organic Carbon and GHG emissions modelling

Direct measurements and long term experiments allowed to get reliable and credible estimations of SOC stocks (Smith et al. 2019) and helped to assess the controlling factors of SOC change (e.g., Kühnel et al. 2019). Simulation models, however, play a prominent role in SOC research because they provide a mathematical framework to integrate, examine and test the understanding of SOC dynamics (Campbell and Paustian 2015). In other words, simulation models provide the capacity for numeric evaluation of SOC at different time and spatial scales and the capability to forecast the impacts of interacting management practices and climatic conditions on SOC stocks (Wang et al.

2020). This has led to an expanding use of soil models specifically to predict SOC dynamics in order to apply policies or to make decisions on management (Campbell and Paustian 2015). Soil organic carbon storage modelling studies have been conducted at widely different scales (i.e., plot level, regional level and national or global level) or as part of C footprint assessment (e.g., Batalla et al. 2015) through process-based SOC models (Farina et al. 2020; Abramoff et al. 2021) or more empirical and simplified approaches (IPCC 2019). However, simulation models could present some uncertainty in SOC estimation, ascribed to the physical and biogeochemical processes incorporated in C cycle (Brilli et al. 2017). In this sense, model calibration improves the accuracy of SOC estimations and reduce model uncertainty especially when supported by measurement or include estimates of uncertainty (Fitton et al. 2017). Therefore, model improvement helps to get better representation of the ecosystem dynamics and the related advantages for stakeholders (Brilli et al. 2017).

In particular, the simultaneous quantification of the main C and GHG flows and of how they are affected by farmer's practices is a very complex task that can be affordable with the support of modelling tools specifically developed for this purpose. Indeed, several modelling approaches have been designed to assess the management practices at the agronomic and environmental levels for the particular case of dairy systems (e.g., Del Prado et al. 2013) in Northern Spain, using a combination of models e.g., grassland model NGAUGE, Brown et al. (2005); Vergé et al. (2012) developed ULICEES model to estimate the carbon footprint of Canadian livestock production including dairy production. Similarly, Rotz et al. (2020) simulated with the Integrated Farm System Model the C footprint for dairy farms of Pennsylvania. Regarding SOC simulation, different models vary considerably in the assumptions and the approach used to represent C processes (e.g., number of C pools, type of decomposition kinetics used and initialisation) (Riggers et al. 2019). The RothC (Coleman and Jenkinson, 1996) and the Century (Parton et al. 1988) models are examples of well-established SOC models that have been applied to numerous field studies worldwide under various types of agricultural management and agroclimatic regions, while C-TOOL (Taghizadeh-Toosi et al. 2014), YASSO07 (Tuomi et al. 2009), ICBM (Andren and Katterer 1997) were used mainly in national GHG inventories. The advantage of RothC over more complex process models like Century is the relatively small number of required input parameters, which most closely reflects the availability of parameters on larger scales. The RothC model simulates the turnover of organic C in no waterlogged topsoil using a monthly time step. It allows for the effects of soil type, temperature, moisture content and plant cover on the turnover process. RothC has been calibrated under agricultural dryland conditions by Farina et al. (2013). However, managed grasslands under moist temperate conditions are of paramount importance in terms of SOC dynamics. They have particularities with respect to grazing animals leading to soil

compaction, changes in vegetation growth and quality and animal dejections that need to be introduced in the RothC model as part of net GHG estimation. In this regard, an integrated modelling framework -consisting in the RothC model to simulate SOC changes and Tier 2 IPCC methodologies to estimate the CH₄ and N₂O emissions from enteric fermentation, manure storage and handling, and pasture soils- would allow us to account for emission variability resulting from pedo-climatic conditions and management practices.

Importance of regionalization and climate change projection in the Spanish context

In Spain, the number of studies integrating the different components (e.g. soil, plant or animal) into scales larger than the animal or the field level (e.g, regional) is limited (Álvaro-Fuentes et al. 2016). Linking geographic information systems (GIS) with SOC/GHG model like RothC at regional scale, enables us not only to consider the local parameters that control SOC/GHG dynamics (e.g., soil properties, climate, and land use), but also to analyse their spatial variability (Farina et al. 2017).

In terms of climate change, in Europe, the Mediterranean region is a recognized hot spot for the next decades (IPCC 2013), where drought and extreme meteorological events are expected to severely affect many economic sectors of the region, including agriculture and food security (Ventrella et al. 2012). Under climate change scenarios, only few studies have studied the climate change impact in the Mediterranean Spanish croplands (Álvaro-Fuentes et al. 2012; Pardo et al. 2017). Therefore, there is a need for more studies dealing with the spatial analysis of climate change impact on Mediterranean croplands of Spain to better make the assessment relevant for decision making. In addition, scientific research focused mainly on the potential for long term SOC stocks in managed grassland soils rather than net GHG emissions. Consequently, there is also a need for future studies to synthesize the net effect of management activities on the GHG balance (Eze et al. 2018). In this context, White et al. (2012) concluded that climate change effects on temperate grasslands remain poorly understood and this underscores the need for further research.

Currently, climate is changing, with nearly 0.8°C rise in global average temperature since the 19th Century and altered precipitation patterns expected throughout the 21st Century (IPCC 2013; Jenkins et al. 2008). Rising temperatures in the course of climate change are discussed to cause significant declines of SOC in temperate agricultural soils (Wiesmeier et al. 2016). Since emissions of GHG from soil derive from biological processes, leading to production and consumption/storage of CO₂, N₂O, and CH₄, that are sensitive to soil temperature and water content, climate change may impact significantly on future emissions (Baldock et al. 2012). For instance, increasing temperature has been

shown to increase the ratio of N₂O/ nitrate from nitrification (Goodroad and Keeney 1984) and reduce N₂O/N₂ ratios from denitrification (Castaldi 2000). Higher temperature also induces higher CH₄ emissions derived from manure storage (IPCC 2019). The impact of agricultural management practices on emissions of N₂O and CH₄ must then be considered, given their high levels of radiative forcing relative to CO₂ (298 and 25 times that of CO₂ over a 100-year time frame, respectively). Therefore, to better inform soil management policy, research should focus on the impacts of the projected climate change on net GHG exchange for temperate grasslands (Eze et al. 2018). This will provide a clearer picture of the full implication of grassland management to climate change.

Main objectives of the thesis

The main objective of this thesis was to assess soil organic carbon (SOC) changes and greenhouse gas (GHG) emissions at a regional scale for different agroecosystems and climatic conditions in Spain (i.e., croplands under Mediterranean climatic conditions and managed grasslands associated to dairy production under Northern moist temperate conditions). We also aimed to evaluate different alternative management practices to mitigate GHG emissions in both agroecosystems and under climate change conditions.

In chapter 1, the general aim was to evaluate the impact of climate change and different management strategies on SOC changes in Mediterranean Spanish agroecosystems. For this aim it was considered the region of Aragon as a case study to assess SOC dynamics for different crops under different climate scenarios compared to a baseline reference and to assess alternative management practices scenarios to mitigate the climate change effect.

In chapter 2, the SOC model RothC was modified and calibrated to improve SOC estimation in managed grasslands under moist temperate climatic conditions of Northern Spain. In this chapter, it is described the basis of the proposed modifications, carried out a simple sensitivity analysis and validate predictions against data from existing field experiments from four different sites in Europe with similar climate and land use systems.

In chapter 3, the main objective was to estimate the net GHG emissions in grassland-based dairy cattle systems under moist temperate conditions of Northern Spain at a subnational scale. In this chapter, the changes in SOC stocks and GHG emissions, derived from enteric fermentation, manure storage and handling, and pasture soil, were estimated over 30-yr period (1981-2010).

In chapter 4, the aim was to assess net GHG emissions under regional scale for moist temperate grasslands associated to dairy production in Northern Spain under different climate scenarios. We also addressed alternative management practices to reduce manure related GHG emissions under climate change conditions.

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CHAPTER 1: MODELING REGIONAL EFFECTS OF CLIMATE CHANGE ON SOIL ORGANIC CARBON IN SPAIN

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Abstract

Soil organic C (SOC) stock assessments at the regional scale under climate change scenarios are of paramount importance in implementing soil management practices to mitigate climate change. In this study, we estimated the changes in SOC sequestration under climate change conditions in agricultural land in Spain using the RothC model at the regional level. Four Intergovernmental Panel on Climate Change (IPCC) climate change scenarios (CGCM2-A2, CGCM2-B2, ECHAM4-A2, and ECHAM4-B2) were used to simulate SOC changes during the 2010 to 2100 period across a total surface area of 23,300 km². Although RothC predicted a general increase in SOC stocks by 2100 under all climate change scenarios, these SOC sequestration rates were smaller than those under baseline conditions. Moreover, this SOC response differed among climate change scenarios, and in some situations, some losses of SOC occurred. The greatest losses of C stocks were found mainly in the ECHAM4 (highest temperature rise and precipitation drop) scenarios and for rainfed and certain woody crops (lower C inputs). Under climate change conditions, management practices including no-tillage for rainfed crops and vegetation cover for woody crops were predicted to double and quadruple C sequestration rates, reaching values of 0.47 and 0.35 Mg C ha⁻¹ yr⁻¹, respectively.

1 Introduction

Agricultural activities significantly contribute to global emissions of the major greenhouse gases (CO₂, CH₄, and N₂O) (Paustian et al., 2004). Nevertheless, in the agricultural sector, there are several options to mitigate climate change effects by either reducing the sources of greenhouse gas emissions, enhancing their sinks (e.g., negative emissions via CO₂ sequestration), or reducing CO₂ emissions by substitution of biological products for fossil fuels or energy-intensive products (Smith et al., 2014). In the recent decades, special attention has been paid to soil organic C (SOC) sequestration and its role in mitigating climate change. Although estimates are subject to a large degree of uncertainty, optimistic studies (e.g., Hansen et al., 2013) suggest that global agricultural soils could sequester at least 10% of the current annual emissions of 8 to 10 Gt yr⁻¹. In particular, SOC mitigation potential for European croplands is estimated between 9 and 38 Mt CO₂ yr⁻¹ by 2050 (Frank et al., 2015). Furthermore, increasing SOC is vital to the ecosystem functioning as a consequence of the positive effects on soil structure, water retention, and cation exchange capacity (van Keulen, 2001), thus contributing to enhanced soil quality and water availability (Johnston et al., 2009). Agricultural intensification has caused growth in the use of management practices that lead to decreasing SOC stocks (e.g., monoculturing, over-tillage). The application of improved agricultural management practices that lead to both enhanced SOC sequestration and

increased soil fertility is therefore required for climate change mitigation (Haddaway et al., 2015). Among these potential mitigating practices, conservation tillage (minimum tillage and no-tillage [NT]) has been recommended for semiarid areas of the Mediterranean basin (López-Garrido et al., 2014), since it (i) increases SOC stocks through developing macroaggregates (Panettieri et al., 2015), (ii) prevents soil disturbance and SOC decomposition (Peterson et al., 1998; Plaza-Bonilla et al., 2010), (iii) enhances water use efficiency (Lampurlanés et al., 2016), and consequently, (iv) may lead to higher yields than conventional tillage systems (De Vita et al., 2007; Morell et al., 2011). The application of organic amendments to agricultural soils and the use of cover crops are also regarded as effective ways of restoring soil C stocks. Experimental and modeling studies have successfully shown an increase in SOC after application of organic amendments such as pruning residues (Sofa et al., 2005) or compost (Mondini et al., 2012), and using cover crops for both woody (Pardo et al., 2017) and arable (Bleuler et al., 2017) cropping systems.

Supplying stakeholders and policymakers with scientifically robust information on soil management practices to restore and increase C stocks is paramount in developing appropriate strategies to mitigate climate change. In this sense, SOC models provide reliable and practical information, as they can estimate SOC potential and assess future trends (Mondini et al., 2012). Using dynamic SOC models at a regional scale, thus linking GIS with soil organic matter (SOM) models (Farina et al., 2017), enables us not only to consider the local parameters that control SOM dynamics (e.g., soil properties, climate, and land use), but also to analyze their spatial variability. In this context, the SOM model RothC (Coleman and Jenkinson, 1996) has been applied to numerous field studies worldwide under various types of agricultural management and agroclimatic regions (Jenkinson et al., 1999; Kaonga and Coleman., 2008; Liu et al., 2009). In Mediterranean Spain, process-based models (e.g., the Century model: Álvaro-Fuentes et al., 2012a) have already been used at the plot (Nieto et al., 2010; Álvaro-Fuentes et al., 2012b; Nieto and Castro, 2013) and regional (Álvaro-Fuentes et al., 2012a; Pardo et al., 2017) scales. Among these studies, Álvaro-Fuentes et al. (2012a) has been the first to investigate the effect of Spanish climate change conditions on SOC changes at the regional level, but they did not use the C model under a spatially explicit environment. Therefore, to our knowledge, there have not been many studies so far investigating SOC dynamics at the regional scale, linking GIS with SOC models and considering climate change conditions. Accordingly, the main general aim of this study was to evaluate the impact of climate change on SOC content at a regional level in Spain using the RothC simulation model. In this context and to make the assessment more relevant for decision making, we included spatial mapping and recommended soil management practices. Furthermore, different alternative management practices to increase SOC sequestration under climate change conditions were also simulated. We hypothesized that (i) climate change

conditions would reduce SOC sequestration capacity, and (ii) alternative management practices could help to enhance soil C sequestration under climate change conditions.

2 Materials and Methods

2.1 Study Area

Our study area (47,719 km²) in Northeastern Spain comprised the entire Aragon autonomous community. This is located at the center of the Ebro River depression, an enclosed basin situated between two high mountain ranges: the Pyrenees in the north and the Iberian mountains in the south. This location has an irregular orography and large climate heterogeneity. Annual precipitation in the central part of the region rarely reaches 400 mm, with mean air temperatures close to 14°C. In contrast, in the northern and southern areas (the Pyrenees and the Iberian mountains, respectively), annual precipitation reaches 1000 mm (Cuadrat, 1999; Peña et al., 2002).

In the Aragon region, agricultural land occupies 49% of the total surface. Field crops occupy almost 80% of the agricultural area, involving rainfed (74%) and irrigated (26%) land. The main crops in the rainfed areas are barley (*Hordeum vulgare* L., 65%) and wheat (*Triticum aestivum* L., 31%). As for irrigated crops, corn (*Zea mays* L., 47%) and barley (22%) are the most abundant crops, with smaller proportions of wheat and alfalfa (*Medicago sativa* L.). Woody crops in Aragon represent 30% of the sales of vegetative products of the study area, with almonds (*Prunus dulcis* L.), olives (*Olea europaea* L.), and grapes (*Vitis vinifera* L.) as the main crops (DGA, 2012).

Rain-fed crops are commonly managed under either cereal–fallow rotations or monocropping together with intensive tillage (56% of rainfed surface is intensively tilled). However, land managed under conservation tillage (i.e., NT) instead of intensive tillage has increased in recent years, representing 20% of land surface currently (DGA, 2012). Within irrigated crops, corn (47%), barley (22%), and wheat (17%) are commonly grown as part of a rotation (GEA, 2000). Surface irrigation and sprinkler irrigation systems are most widely used at similar rates (MAPAMA, 2013). Regarding tree crops, the most generalized practice is minimum soil tillage, representing 55% of woody crop surfaces (DGA, 2012).

2.2 The RothC Model

The RothC model (Coleman and Jenkinson, 1996) requires a small number of easily available input data and has been widely used to simulate the impact of agricultural land management on SOC changes (Coleman et al., 1997; Falloon and Smith, 2002; Johnston et al., 2009). As a summary, the

RothC model divides the SOC into five fractions: four of them are active, and one is inert (i.e., inert organic matter). The active pools are decomposable plant material (DPM), resistant plant material (RPM), microbial biomass, and humified organic matter. The decomposition of each pool (except inert organic matter) is governed by first-order kinetics (yr^{-1}), characterized by its own turnover rate constant (10 for DPM, 0.3 for RPM, 0.66 for microbial biomass, and 0.02 for humified organic matter) and modified by factors related to air temperature, soil moisture, and vegetation cover, which are main input parameters to run the model. RothC does not include a plant growth module, and thus plant C inputs to soil have to be entered as exogenous inputs to the model. Incoming plant C is split between DPM and RPM, depending on the DPM/RPM ratio of the particular incoming plant material or organic residue. Both of them decompose to produce microbial biomass, humified organic matter, and evolved CO_2 . For most agricultural crops and improved grasslands, a DPM/RPM ratio of 1.44 is used (i.e., 59% of the plant material is DPM and 41% is RPM; Jones et al., 2005).

The model uses a monthly time step to calculate total SOC and its different pools on a years-to-centuries timescale. The climate input parameters include monthly average air temperature, monthly precipitation, and monthly open-pan evaporation. Other input parameters are soil clay content, monthly C input from plant residues or exogenous organic matter (e.g., manure), and monthly information on soil cover, whether the soil is bare or covered by plants.

2.3 Input Datasets and Spatial Layer Linkages

The Aragon autonomous community covers 21 agricultural regions. Information on agricultural land uses was obtained from the Corine Land Cover. We distinguished five main classes of land cover: rainfed arable land, irrigated arable land, orchards, olive groves, and vineyards. The last three classes were compressed into a general class of woody crops. Soil data were obtained from a recent assessment at the national level (López Arias and Grau Corbí, 2005). In this assessment, among others, variables such as soil texture and SOC to the 30-cm soil depth were analyzed and spatially represented for the entire Spanish area (Rodríguez Martín et al., 2009). For our study, a total of 309 georeferenced points were selected (Supplemental Fig. S1). We set up an equal-interval classification (five intervals) and a mean value of each interval for SOC stocks and clay content properties layers (Supplemental Fig. S2) to overcome the large variability in both SOC stocks ($25\text{--}151 \text{ Mg ha}^{-1}$) and clay content (14-30%) across the area studied.

Climate change data, corresponding to $50\text{-km} \times 60\text{-km}$ grids, were produced by the Meteorological State Agency using a regionalization technique explained in Brunet et al. (2008). We simulated SOC changes for the period of 2010 to 2100 under four climate change scenarios and one baseline scenario. The latter consisted of historical average monthly temperature and precipitation

data of more than one decade. The four climate change scenarios were obtained from two atmosphere-ocean global circulation models, ECHAM4 and CGCM2, forced by two Intergovernmental Panel on Climate Change (IPCC) Special Report on Emissions Scenarios, A2 (medium-high emissions scenario) and B2 (medium-low emissions scenario) (Nakicenovic et al., 2000; more details can be found in the section “Climate Change Scenarios” below). Furthermore, potential evapotranspiration for each decade from 2010 to 2100 was estimated monthly using air temperature according to the Hargreaves method (Allen et al., 1998).

The overlay of climate grids on the spatial layers of agricultural land uses and clay content through ArcMap 10. 2. 2 (MaDGIC, 2014) resulted in 1337 individual polygons. Mean SOC stocks for initialization purposes were assigned to the spatial units through a query operated in Microsoft Access.

2.4 Model Running and Parametrization

Carbon inputs derived from plants and animal manure application were estimated. To explore the maximum potential for SOC sequestration in the study area, simulations were performed assuming that all plant residues were returned to the soil. Within rainfed and irrigated crops, annual average crop yield values for the 2003 to 2013 period were obtained from the Agricultural Statistical Yearbook of Aragon (DGA, 2012). Total aboveground biomass was estimated from crop yields using average harvest index (HI) values obtained from different studies performed in the study area (Daudén and Quilez, 2004; Daudén et al., 2004; Moret et al., 2007; Berenguer et al., 2009; Yagüe and Quilez, 2010a, 2010b; Álvaro-Fuentes et al., 2013; Erice et al., 2014; Plaza-Bonilla et al., 2014). After an extensive literature review of the study conditions, HI values were set to 0.42 and 0.38 for rainfed and irrigated crops, respectively. We assigned a HI of 0.50 to corn, assuming its different morphology compared with the other irrigated crops. Belowground plant residues were estimated from shoot/root ratio. Similarly to HI, a literature review was performed to obtain mean shoot/root values representative of the study area (Lohaus et al., 1998; Vamerli et al., 2003; Plaza-Bonilla et al., 2014). The final mean shoot/root values were fixed to 4.12, 7.66, and 2.21 for rainfed crops, irrigated crops, and corn, respectively. For both irrigated and rainfed crops, we assumed that 50% of C inputs occurred in the month of harvest and the remaining 50% in the three previous months.

For woody crops, pruning residues were also estimated using data obtained from studies performed in Spain or in similar Mediterranean conditions (Di Blasi et al., 1997; González et al., 2005; Nieto et al., 2010; Velazquez-Martí et al., 2011; Aguilera et al., 2015). The final pruning values considered were 1.52, 2.90, and 4.80 Mg ha⁻¹ for olive groves, vineyards, and orchards, respectively. We assumed that 70% of the C inputs occurred in the pruning months and the remaining 30% during

the four previous months. Carbon inputs from animal manure application were based on the studies of Sanz-Cobeña et al. (2014) and Pardo et al. (2017) and were calculated as follows: dry matter excretion rates ($\text{kg location}^{-1} \text{ yr}^{-1}$) for livestock were first obtained from the National Inventory Report (MAPAMA, 2011) and then subsequently multiplied by the livestock population of each animal category for 2008 and for each agricultural region (MAPAMA, 2009). Outdoor grazing animals' excreta were assumed to reach mainly grasslands, which were not included in the present study. Consequently, to estimate manure flows applied to croplands, we deducted animal excretion during grazing from the total excreta by applying the grazing factor proposed by the Spanish National Inventory (UNFCCC, 2014). Finally, manure flows applied to cropland dry matter was converted to C by assuming 80% content of volatile solids and 55% of C content in volatile solids. (Adams et al., 1951). We assumed that C inputs of animal manure were applied only to arable land (irrigated and rainfed).

To run the model, monthly C inputs derived from plant residues and from animal manure application were assigned to each of the spatial units according to land use. The splitting ratios for DPM and RPM (DPM/RPM) were assigned differently for each agricultural system. We assumed 49% DPM, 49% RPM, and 2% humified organic matter for manure and 59% DPM and 41% RPM (DPM/RPM = 1.44) for both irrigated and rainfed crops based on Coleman and Jenkinson (1996). Regarding woody crops, and to refine the data, we considered a better resistance to degradation than cropping systems (50% DPM and 50% RPM, DPM/RPM = 1), since woody crops contains higher lignin content.

Water inputs from irrigation were also taken into account and added to monthly precipitation in the weather file of the model and managed as baseline conditions. Average irrigation doses of the different crops applied in the baseline climatic scenario were obtained from Lecina et al. (2010), and rates were calculated considering the different cropping areas. For the climate change scenarios, we chose to keep the same irrigation patterns and rates as in the baseline climatic scenario. Although we acknowledge the robustness that an adjustment of the irrigation strategies could bring to climate change scenarios (Zhao et al., 2015), we decided to have a conservative assumption (i.e., unchanged irrigation) due to large uncertainties (Wada et al., 2013) associated with choosing a specific irrigation adjustment (Klove et al., 2014). Indeed, for this region (the Ebro region), there have been many studies that have shown large variation in the range (3–20%) of irrigation need predictions (Iglesias and Minguéz, 1997; Jorge and Ferreres, 2001; Döll, 2002; Fischer et al., 2007; Rey et al., 2011; von Gunten et al., 2015). The large uncertainty in these estimates is not only due to climate models and scenarios used (García-Vera, 2013), but also to factors in relation with future human impacts on land use changes and demands (Pulido-Velázquez et al., 2015). For example, the future choice of crop

types by farmers as a response to climatic change or social and economic factors (von Gunten et al., 2015) is likely to play an important role in constraining future irrigation water demand (Wada et al., 2013).

RothC initialization requires C pools to be sized. For regional-scale simulation, C pool quantification is infeasible. As an alternative method, we used the pedotransfer functions established by Weihermüller et al. (2013), which have been shown to be helpful in initializing the reactive pools of the RothC model. These functions are based on easily available variables such as SOC stocks and clay content, which, for this study, were derived from Rodríguez Martín et al. (2016).

Since the regional simulation is computationally intensive and time consuming due to the combination of a large number of runs for each polygon and a large number of polygons, we developed a VBA (Visual Basic for Applications)-based program in Excel to simulate changes in SOC stocks simultaneously for the different polygons for the period of 2010 to 2100.

2.5 Climate Change Scenarios

Climate change projections suggest that Mediterranean Spain is likely to become one of the areas in Europe that will be more severely affected by climate change. Christensen et al. (2007), for example, showed that the highest climate change impact would affect the Mediterranean part of Spain, with a temperature raise of $> 6^{\circ}\text{C}$. As a summary of the climate data used in this study, all climate change scenarios predict a decrease in precipitation and an increase in mean air temperature in the following order (Fig. 1, Fig. S3): CGCM2-B2 $<$ CGCM2-A2 $<$ ECHAM4-B2 $<$ ECHAM4-A2. Although there is an overall decrease in monthly precipitation of 10.6 and 9.5 mm under ECHAM4-A2 and ECHAM4-B2 scenarios and 3 and 4 mm under CGCM2-B2 and CGCM2-A2 scenarios, respectively, the average monthly temperature rises from 2°C under CGCM2-B2 to 7°C under ECHAM4-A2. Not only do climate change scenarios show lower annual precipitation, but they also indicate different annual precipitation distribution than the baseline scenario (Fig. 1). The climate models producing the largest changes in climate (ECHAM4-B2 and ECHAM4-A2) predict a significant decrease in precipitation during the typical precipitation season (spring and autumn) (Fig. 1). The decadal distribution pattern of annual precipitation is significantly modified among the climate change scenarios compared with the current climatic conditions (Fig. S3). However, the annual and decadal temperature distribution are not modified by climate change (Fig. 1, Fig. S3). Finally, when we compare the climate projections used in our study for the different Aragon climatic regions, we can observe that the Ebro depression in the central area of Aragon showed the highest temperatures under climate change conditions and the smallest average precipitation decrease

compared with the other two mountainous areas (the Pyrenees mountains in the north and the Iberian mountains in the south, Table S1).

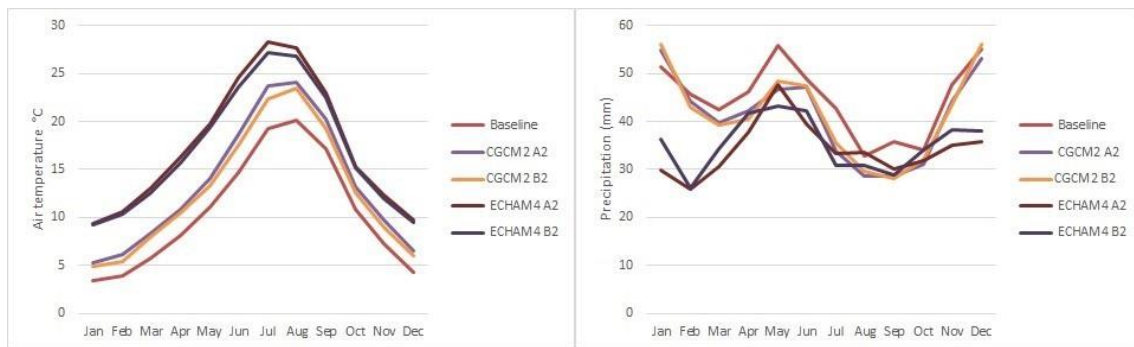


Fig. 1. Mean monthly air temperature and precipitation distribution for the different climate scenarios during the 2010-2100 period in the Aragon region

2.6 Soil Management Scenarios

To enhance SOC stocks under climate change conditions during the simulation period of 2010 to 2100, we simulated alternative soil management scenarios, such as NT and vegetation cover for rainfed and woody crop systems, respectively. For NT practices, we considered two livestock future projections for livestock numbers and, thus, for animal manure. Then, we analyzed the effect of these practices (Supplemental Table S2) under the most extreme climate change scenario (ECHAM4-A2).

No-tillage has been shown to be an interesting strategy for rainfed systems of Aragon in terms of SOC increase (Álvaro-Fuentes et al., 2008). However, the NT effect of reducing organic matter decomposition rates cannot be directly simulated with the RothC model. Therefore, we indirectly considered NT practices by accounting the impacts of NT on plant residue data (Nemo et al., 2017). On the basis of experimental studies from the zone (Lampurlanés and Cantero-Martínez, 2006; Plaza-Bonilla et al., 2017), we made the assumption that plant C input at sowing was greater under NT (60%) than under deep or minimum tillage.

Since future projections for livestock production are subject to a large degree of uncertainty (Thornton, 2010; van Grinsven et al., 2015), we explored the potential range of livestock change by both an increase and a decrease in animal numbers ($\pm 20\%$ animal production). Both scenarios are supposed to match potential demand-driven trends, the first by rising income and urbanization of the population, and the second if the population has a concern over eating animal products. A linear increase or decrease in animal numbers for the 2010 to 2030 period and a stable situation for the 2030 to 2100 period were assumed for this study.

The use of vegetation cover in olive groves could be an efficient strategy to increase SOC sequestration and reduce erosion impacts (Gómez-Muñoz et al., 2014). This strategy is especially

relevant in the context of Mediterranean olive groves, as it has been confirmed by some studies already (e.g., Álvarez et al., 2007). Accordingly, for woody crops, we simulated soil management consisting of a natural vegetation cover and assumed a residue of dry biomass of $1570 \text{ kg ha}^{-1} \text{ yr}^{-1}$ based on Gómez-Muñoz et al. (2014) and monthly C inputs of 0.06 Mg ha^{-1} (assuming 45% C content in dry matter).

3 Results and Discussion

3.1 Regional Soil Organic Carbon Changes under Climate Scenarios

Results showed an increase in total SOC stocks in Aragon for the 2010 to 2100 period under baseline and climate change conditions (Fig. 2). To explore the potential for SOC sequestration, simulations were performed under the assumption that all plant residues were returned to the soil. Therefore, the observed increase can be partly explained by the biomass returns and C input levels considered during the simulation period, mainly due to pruning in woody crop systems (Sofa et al., 2005), crop residues in both rainfed and irrigated systems (Powlson et al., 2011), and the climate change conditions in both mountainous zones. Comparing SOC content evolution among climate scenarios, the greatest enhancement in SOC content was observed in the baseline scenario (assuming no climate change conditions), with SOC values increasing from 49.5 (2010) to $75.2 \text{ Mg C ha}^{-1}$ (2100). In comparison, in all the climate change scenarios, the potential for SOC sequestration decreased, with the lowest SOC increase found in the two ECHAM4 climate scenarios (A2 and B2, 68.5 and 68 Mg C ha^{-1} , respectively), (Fig. 2). For CGCM2-A2 and CGCM2-B2 scenarios, intermediate SOC values were observed at the end of the simulation period (2100), with 70.2 and $70.8 \text{ Mg C ha}^{-1}$, respectively (Fig. 2). The lowest SOC sequestration rates found in ECHAM4-A2 and ECHAM4-B2 scenarios (Fig. 2) were associated climatically with the greatest decline in precipitation rates and rise in temperature (Supplemental Table S3).

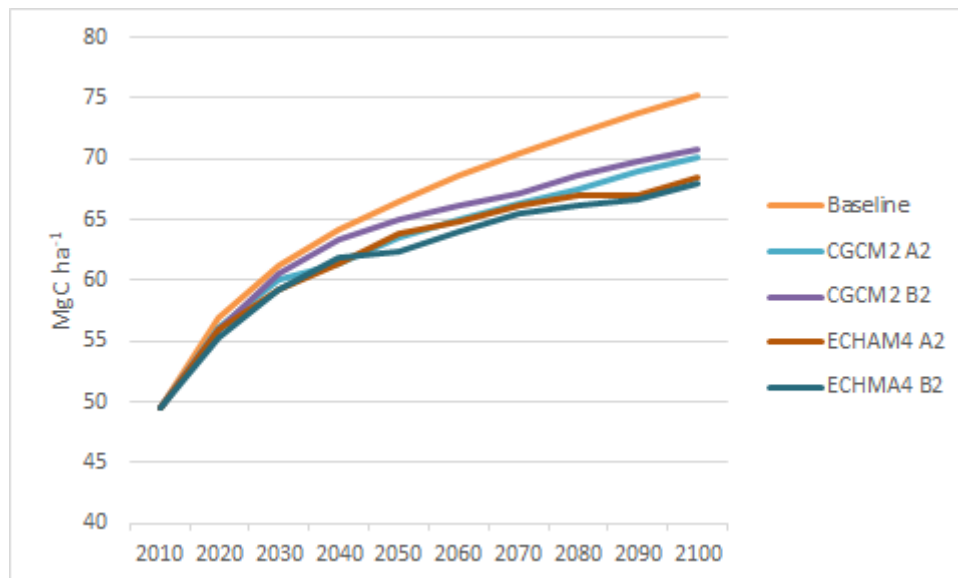


Fig. 2. Soil organic carbon (SOC) content evolution under the Baseline scenario and the four climate scenarios tested (CGCM2 A2, CGCM2 B2, ECHAM4 A2, ECHAM4 B2) during the 2010-2100 period at the 0-30 cm soil layer in the agricultural surface of the Aragon region

It is worth noting that ECHAM4-A2 showed slightly larger SOC contents than ECHAM4-B2, mainly in the last five decades of the simulation period (Fig. 2). In that period, average temperatures were consistently higher under the ECHAM4-A2 scenario than under ECHAM4-B2, whereas precipitation rates were slightly lower (Fig. S3), thus suggesting the combined effect of both climatic variables on soil moisture deficit and consequently on SOC evolution. In line with our results, Smith et al. (2005) underlined the combined effects of climate change on SOC dynamics as a balance between temperature increase and soil moisture variation for the entire European agricultural surface.

The climate change conditions of the scenarios considered resulted in slight differences in SOC sequestration rates. The estimated decrease in potential SOC sequestration rates under climate change conditions could be mainly associated with the higher temperature predicted in the climate change scenarios (Table S3), which could have triggered greater decomposition rates (Davidson and Janssens, 2006). Hence, the effect of average temperature increases of 6 to 7°C (Fig. 1a) in the whole Aragon region would be linked to the lowest SOC estimated under ECHAM4 scenarios. However, lower precipitation and reduced soil moisture can slow down SOC decomposition (Skopp et al., 1990), thus counteracting the higher temperature influence. These two combined effects would explain the slight differences in SOC evolution observed between ECHAM4 and CGCM2. In the CGCM2 scenarios, the average temperature rise was moderate (2–3°C), but the precipitation decrease was also moderate, which involves a smaller restriction on SOC decomposition rate due to soil moisture deficit. As a result, potential SOC sequestration rates simulated in the CGCM2 scenarios were just slightly higher than in the ECHAM4.

The spatial distribution of SOC levels in 2100 in the ECHAM4-A2 scenario showed lower SOC contents than those found under baseline conditions (Fig. 3). For example, in the humid zones (Pyrenees and Iberian zones, located in the north and south parts of Aragon, respectively), the model predicted a decrease of total surface with SOC contents $>97 \text{ Mg C ha}^{-1}$ in 2100 (Fig. 3). Similarly, in the central region of the study area, where the drier conditions prevailed ($<400 \text{ mm}$ precipitation), the area with SOC levels $<57 \text{ Mg C ha}^{-1}$ also increased (Fig. 3). These results provide an indication of the effects of warming on SOC dynamics.

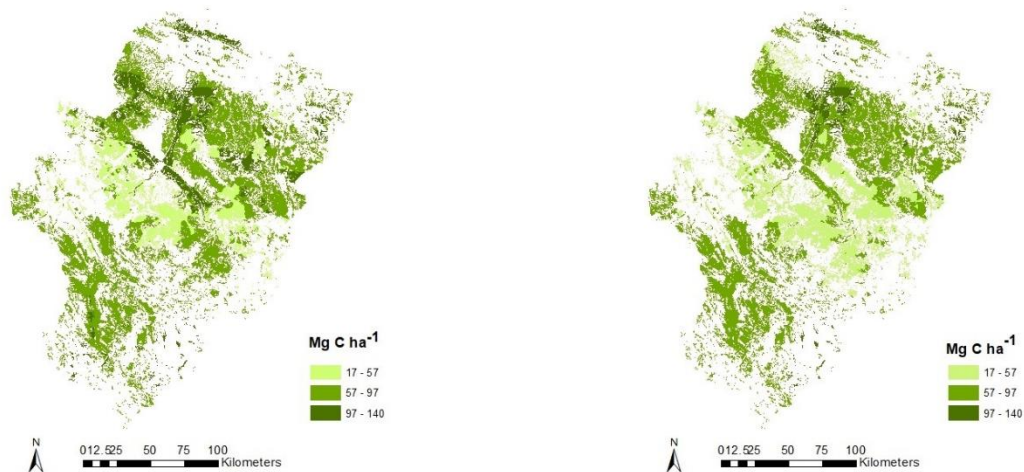


Fig. 3. Spatial distribution of soil organic (SOC) content in 2100 under the Baseline scenario (on the left) and the ECHAM4-A2 climate change scenario (on the right) at the 0-30 cm soil layer in the agricultural surface of the Aragon region

Our results suggest that the decrease in soil moisture seems to constrain soil microbial activity, this effect being very clear in rainfed cropping systems. In fact, the RothC water balance results showed that, for about half of the year, microbial activity was reduced up to 90% compared with the rest of the year. Compared with another study applied in the same area (Álvaro-Fuentes et al., 2012a) and using a different model (Century), however, this moisture effect was the major factor controlling SOC dynamics. The difference between results from different models is likely explained by differences in two parameter values. The rate modifier for temperature parameter value is always

higher in RothC than in the Century model, and the opposite is found for the rate modifier for moisture value (Falloon and Smith, 2002). The underlying uncertainty in the response of soil C to soil moisture is generally attributed to the associated uncertainty in the relationship between soil moisture and soil microbial processes (Falloon et al., 2011). This relationship is complex and depends on several processes, particularly oxygen diffusion and biochemical processes. In RothC, the soil moisture reduction depends on soil clay content, precipitation, and evaporation rate (Bauer et al., 2008), which underestimates the influence of other possible factors. A better understanding of the relationship between soil moisture and SOC decomposition is needed to reduce this uncertainty and improve our confidence in SOC changes under climate change predictions.

According to variability within climate change scenarios and considering the different agricultural systems modeled, irrigated crops showed the highest SOC increase at the end of the simulation period for all climate scenarios (Table 1). For all climate scenarios, rainfed crops showed smaller increases in C sequestration than irrigated crops (Table 1). This could be associated with the limited amount of harvest residue in rainfed conditions, especially after drought periods in semiarid Mediterranean areas (Navarrete et al., 2009), and with the higher water supply in irrigated crops, mainly by irrigation, as it increases SOC stocks through greater plant residue (Gillabel et al., 2007).

Table 1. Changes in soil organic C content during the period of 2010 to 2100 for different agricultural systems and under five different climate scenarios (baseline, CGCM2-A2, CGCM2-B2, ECHAM4-A2, and ECHAM4-B2) in the 0- to 30-cm soil layer

Agricultural classes†	Baseline	CGCM2-A2	CGCM2-B2	ECHMA4-A2	ECHMA4-B2
	Mg C ha ⁻¹				
RC	21.4	16.7	17.6	17.1	16.7
IC	46.2	39.5	39.1	29.4	28.2
OR	20.5	16.3	17	16	16.1
OG	-4	-6.3	-6.1	-7.5	-7.4
V	5.3	3	3.3	4.7	5

† RC, rainfed crops; IC, irrigated crops; OR, orchard; OG, olive groves; V, vineyard.

The smallest SOC increase was observed in the vineyard and olive grove cropping systems for all the climate scenarios. Indeed, olive groves showed SOC losses under all climate change scenarios, reaching -7.5 Mg ha⁻¹ (Table 1) under the ECHAM4-A2 scenario. In Mediterranean areas, olive plantations are traditionally cultivated in non-irrigated soils with low fertility, with low planting densities and intensive tillage, making them more susceptible to SOC losses (Nieto et al., 2010).

The highest SOC stock can be explained by greater C input in irrigated crops than in rainfed and woody crops, and a lower temperature in the baseline scenario versus climate change conditions. It therefore seems likely that the model is sufficiently sensitive to the C input and temperature.

Climate change could alter C inputs, as changes in temperature, precipitation, and atmospheric CO₂ levels could affect net primary production (Falloon et al., 2007). The RothC model does not include changes in these C net primary production-related inputs (Meersmans et al., 2013), constraining the reliability of SOC stock changes estimations. In Mediterranean conditions, it is unlikely that crop yield or C inputs would increase under climate change conditions with higher temperature and lower water availability (Wan et al., 2011). Higher atmospheric CO₂ has the potential to increase crop yield as a result of enhanced crop photosynthesis (Högy et al., 2009; Álvaro-Fuentes et al., 2012a) and an increase in water use efficiency via lower stomatal conductance (Morgan et al., 2004). However, plant quality and quantity respond not only to concentrations of CO₂ but also to changes in temperature and precipitation patterns (driven by changes in the occurrence of extreme climatic events or changes in average conditions), and stressors such as ozone concentration or salinity. The extent to which these variables can affect crop yield will depend on complex interactions between these variables, nutrient availability, type of species (e.g., C₃ vs. C₄ species, herbaceous vs. woody species, etc.), and the indirect effect of climate change on forage pests and diseases. Moreover, although out of the scope of this study, some possible adaptation practices including improvement of crop varieties, implementation of technology change, and adjusting the harvest date could offset the declining trend of crop yields and hence of C inputs.

3.2 Soil Organic Carbon Changes under Management Scenarios

Considering the warmest climate change scenario (ECHAM4-A2), the two NT scenarios stored almost more than twofold the SOC stored in the control during the study period (Fig. 4). Both livestock scenarios showed similar levels of SOC sequestered, with 43 and 42.8 Mg C ha⁻¹ for the increased and decreased livestock projections, respectively (Fig. 4). Hence, the decrease or increase in manure (20% until 2030) did not significantly affect the SOC content in agricultural soils of Aragon during the period of 2010 to 2100. This might be explained by the fact that the amount of animal manure assumed in our study region was much lower than the total C inputs from the plant residues.

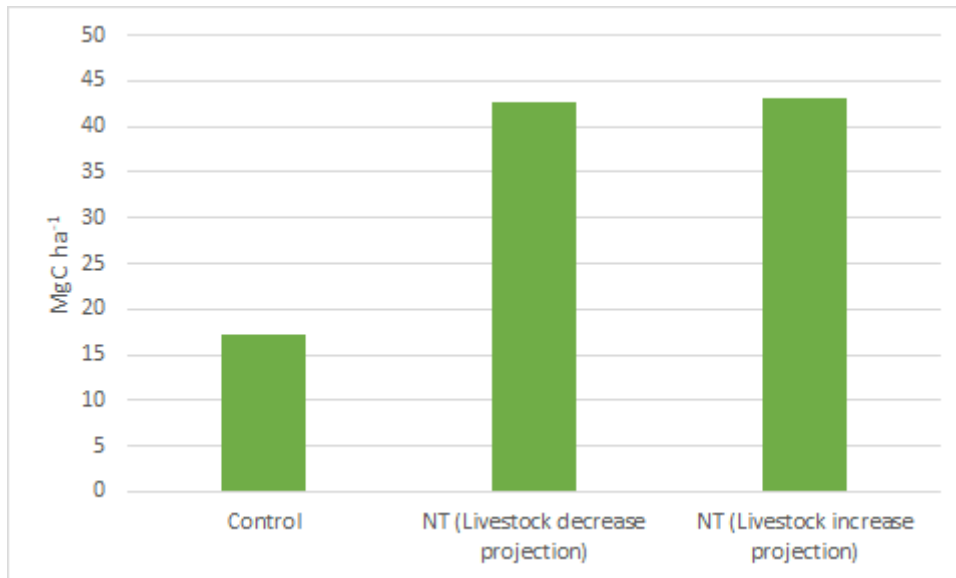


Fig. 4. Changes in soil organic carbon content for the 0-30 cm soil layer in the rain-fed crop (RC) system between the control scenario and the two no-tillage (NT) scenarios (livestock decrease and increase) under the ECHAM4 A2 climate change scenario during the 2010-2100 period

Navarrete et al. (2009) found an annual average sequestration rate ranging between 0.4 and 0.5 Mg C ha⁻¹ under Mediterranean conditions and NT soil management, which is similar to the values that we have observed in this study (0.47 Mg C ha⁻¹) over the simulated 90-yr period. The average annual sequestration rate achieved in our case study is included in the range values of the mentioned study, since in our case, we are assuming total incorporation of residues while considering the warming conditions of the ECHAM4-A2 climate change scenario. Similarly, referring to woody crop systems, vegetation cover under the ECHAM4-A2 climate change scenario quadrupled the SOC sequestration rate compared with the unchanged soil management (control) (Fig. 5).

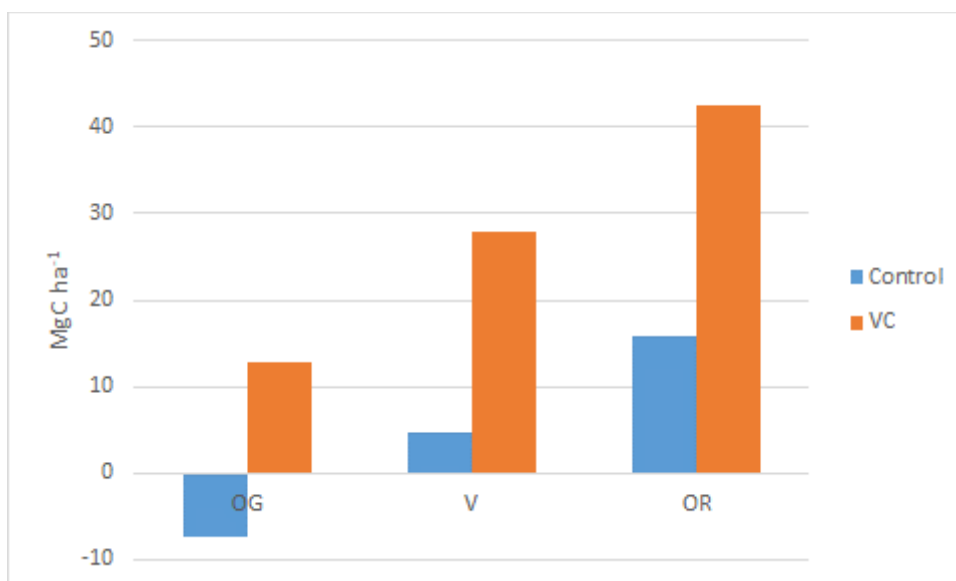


Fig.5. Changes in soil organic carbon (SOC) content for the 0-30 cm soil layer in the woody crop (WC) systems (orchards, OR; olive groves, OG; and vineyards, V) between the control scenario and the vegetation cover (VC) scenario under the ECHAM4 A2 climate change scenario during the 2010-2100 period

According to these findings, both soil management practices of NT in rainfed and vegetation cover crops in woody crops are effective in enhancing SOC stocks in Mediterranean Spain under climate change conditions (Fig S4).

3.3 Model Evaluation

We evaluated the model results against measured data only for the baseline SOC stock predictions, as the RothC model is not able to simulate crop yields. Considering that such SOC data are scarce at the scale of the studied region (Aragon scale), we used the data from Álvaro-Fuentes et al. (2009), which measured SOC sequestration rates in field experiments in Aragon. Under the baseline scenario, in rainfed systems, the RothC model simulated SOC sequestration rates of 21.4 Mg C ha⁻¹ during a period of 90 yr, which is equivalent to 0.23 Mg C ha⁻¹ yr⁻¹ (Table 1). We observed a slight difference between mean simulated and measured values (0.23 and 0.18 Mg C ha⁻¹ yr⁻¹, respectively). This is due to the fact that we are considering the maximum potential of the SOC sequestration assuming a total incorporation of C inputs, thus proving that RothC model results agree reasonably with the expected and measured ranges. However, future research efforts should be made using more and larger, reliable datasets (e.g., under more management practices and cropping systems) to increase our confidence in the RothC results under different management practices and cropping systems.

Climate change is expected to affect crops responses in many and complex ways (e.g., average yields, plant quality). For example, whereas water shortage has undoubtedly a negative effect on yield, this can be partly offset, as has been found in common crops like wheat (Challinor et al. et al., 2014; Ferrise et al., 2016), by photosynthetic stimulation and enhanced water use efficiency through stomatal closure as a result of enhanced CO₂ atmospheric concentration.

For the Iberian Peninsula, most studies that do not consider adaptation measures predict a reduction in crop rainfed dry matter yields for all climate change projections (e.g., 20%: Ciscar et al., 2014), being especially severe for the end of the 21st century (e.g., wheat: Hernandez-Barrera et al., 2017) due to increased drought duration and intensity.

Provided that attempting to robustly predict changes in crop yields (i) is out of the scope of this study, (ii) involves complex methodologies and uncertain assumptions (e.g., a limited set of available field data), and (iii) depends on other farmer decisions (e.g., technology improvements: Iglesias and

Quiroga., 2007; changes in varieties, planting times, irrigation, and residue management: Challinor et al., 2014), we assumed that climate change did not affect crop yields and, subsequently, the amount of C plant residue inputs to the soil compared with the baseline scenario. In recognition of this limitation, for illustration purposes, we made a simple scenario test to see the SOC model response to changes in crop yields in representative rainfed cropping (wheat) systems in our study region. For this exercise, we used the range of potential yield change ($\pm 30\%$) found in the study of Iglesias et al. (2000) and explored the SOC changes as a result of steps of 10% change under CGCM climate change scenarios. Results from this test indicate that each 10% change in dry matter yields resulted in 5% change in SOC storage, compared with the control scenario (with no C input change) at the end of the simulation period (Fig. S5). These results certainly confirm the importance that future studies attempt to focus on C input effects variations associated with uncertainty in dry matter yields in climate change scenarios.

3.4 Qualitative Analysis of Uncertainty

Uncertainty related to this work may be ascribed to the model applied, the initial size characterization of SOC fractions, and the nonavailability of some data at the temporal or spatial levels. Indeed, for most of the parameters (e.g., inputs of irrigation water, HI), modeling was performed according to the most common practices of the study area. Despite RothC performance shown in many studies (Coleman et al., 1997; Falloon and Smith, 2002; Falloon et al., 2006), we have to take into account some limitations of the model and of the procedure applied. The model uses a monthly time step to calculate total SOC, which may overlook some processes of SOC changes occurring at daily timescale. The proposed regional analysis is based on a spatial division of the agricultural Aragon territory into different geographical areas that share a set of specific parameters (e.g., climate, soil properties, and land use). The model runs each one of the spatial units independently so that possible interaction (e.g., water and soil erosion, horizontal flows) between neighboring polygons is ignored (Paustian et al., 1997). Regarding erosion, RothC does not consider the C lost by this phenomenon, which may become a limitation in certain areas where soil loss is more accentuated (Martínez-Mena et al., 2008). Changes in soil management and C input values throughout the study period were not considered, which brings another source of uncertainty. However, assuming constant soil management conditions during simulation period allows isolated analysis of the impact of climate change on SOC sequestration. Finally, regional climate change projections are subject to several sources of uncertainty associated with global general circulation models and regionalization techniques.

4 Conclusions

This study has shown significant variations in the SOC sequestration capacity among the different agricultural systems in a representative Spanish area under climate change conditions. Regarding the different agricultural systems tested, whereas irrigated crops resulted in largest SOC sequestration potential even under the most extreme scenario (ECHAM4), rainfed crops, vineyards, and olive groves showed the lowest potential. These differences are probably due to low productivity of certain rainfed agricultural systems that led to a reduction in harvest residue matter, suggesting that C inputs must be the greatest SOC driver.

According to comparisons among climate change scenarios, temperature increase and rainfall decrease will generally lead to a decline in SOC content. Indeed, ECHAM4 scenarios predicted the greatest impacts on SOC sequestration for the next 90 yr due to a high temperature increase.

No-tillage, in the case of rainfed crops, and vegetation cover, for olive groves and other woody crops, were the alternative management strategies to alleviate climate change effects and SOC loss. These changes in management enhanced the amount of SOC sequestered and were found to be effective strategies in reducing CO₂ emissions and increasing soil potential to sequester C under future climate change conditions.

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CHAPTER 2: ESTIMATING SOIL ORGANIC CARBON CHANGES IN MANAGED TEMPERATE MOIST GRASSLANDS WITH RothC

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Abstract

Temperate grassland soils store significant amounts of carbon (C). Estimating how much livestock grazing and manuring can influence grassland soil organic carbon (SOC) is key to improve greenhouse gas grassland budgets. The Rothamsted Carbon (RothC) model, although originally developed and parameterized to model the turnover of organic C in arable topsoil, has been widely used, with varied success, to estimate SOC changes in grassland under different climates, soils, and management conditions. In this paper, we hypothesise that RothC-based SOC predictions in managed grasslands under temperate moist climatic conditions can be improved by incorporating small modifications to the model based on existing field data from diverse experimental locations in Europe. For this, we described and evaluated changes at the level of: (1) the soil water function of RothC, (2) entry pools accounting for the degradability of the exogenous organic matter (EOM) applied (e.g., ruminant excreta), (3) the month-on-month change in the quality of C inputs coming from plant residues (i.e. above-, below-ground plant residue and rhizodeposits), and (4) the livestock trampling effect (i.e., poaching damage) as a common problem in areas with higher annual precipitation. In order to evaluate the potential utility of these changes, we performed a simple sensitivity analysis and tested the model predictions against averaged data from four grassland experiments in Europe. Our evaluation showed that the default model's performance was 78% and whereas some of the modifications seemed to improve RothC SOC predictions (model performance of 95% and 86% for soil water function and plant residues, respectively), others did not lead to any / or almost any improvement (model performance of 80 and 46% for the change in the C input quality and livestock trampling, respectively). We concluded that, whereas adding more complexity to the RothC model by adding the livestock trampling would actually not improve the model, adding the modified soil water function and plant residue components, and at a lesser extent residues quality, could improve predictability of the RothC in managed grasslands under temperate moist climatic conditions.

1 Introduction

Temperate grasslands, which cover 1.25×10^9 ha globally, are important sinks of SOC, containing approximately 12% of the global SOC pool (Lal 2004). Changes in grassland management (e.g., stocking rate, fertilisation) are frequent in temperate conditions affecting SOC dynamics (Soussana et al. 2004; Conant et al. 2017; Eze et al. 2018a). Grasslands ecosystems under temperate moist conditions are subject to processes that may differ from arable systems in regards with SOC sequestration. In particular, below-ground plant residues in grasslands provide important C inputs for soil C sequestration (Sainju et al. 2017): Grassland species allocate more C below-ground

than cereals (Pausch and Kuzyakov 2018) and below-ground C has longer residence time than above-ground C (Cougnon et al. 2017). Moreover, rhizodeposition is an important source of C inputs (Kuzyakov and Schneckenberger 2004), which is rarely quantified and still remains the most uncertain component of soil C fluxes in terrestrial ecosystems (Pausch and Kuzyakov 2018).

Furthermore, grazing and wheeling by vehicles can cause damage soil and vegetation structure by trampling and poaching, both affecting plant production, and the potential amount of C inputs causing soil C loss (Ma et al. 2016; Eze et al. 2018a). Under temperate moist conditions, precipitations are high and winters are relatively mild with a relatively long growing season, susceptible to poaching (Tuohy et al. 2014). Poaching is a common soil damage problem of livestock treading which has not been extensively simulated in grazing ecosystems (Miao 2016). Also, temperate moist climatic conditions imply that soils are wet-saturated during certain wet periods in which decomposition of organic matter is limited (Moyano et al. 2013).

Therefore, improving the methods to estimate SOC stock changes in managed grasslands is key to obtain reliable estimates of SOC (Herrero et al. 2011) and determine the real contribution of livestock to the net global greenhouse gas emissions.

Recent research in temperate grasslands has shown that grasslands can act either as C sink (Ma et al. 2015; Eze et al. 2018b) or source (Abdalla et al. 2018) depending on how animals, vegetation, soil, climate, and management practices interact with each other (Graux et al. 2013; Rees et al. 2013; Soussana et al. 2013). To study the long-term responses of SOC changes in grasslands, we can use both data from long-term field trials (Skinner and Dell 2015; Gourlez de la Motte et al. 2018) and simulation models. Models allow to obtain complementary information to trials, for example, hypothesis forming or/and to predict long-term responses of grasslands to climate change and management (FAO 2018). For strategic studies, e.g. assessing potential of grasslands to sequester SOC in a region, simple soil models, e.g. RothC (Coleman and Jenkinson 1996), ICBM (Andren and Katterer 1997), C-Tool (Taghizadeh-Toosi et al. 2014) and Yasso07 (Tuomi et al. 2009) are most useful as they require a limited and easily available input data. The RothC model, originally developed for arable soils, is one of the models that has been most widely validated and effectively used for different cropland and grassland systems at different spatial scales (Poeplau and Don 2013; Senapati et al. 2013; Smith et al. 2014).

In general, RothC showed a good performance under grassland ecosystems (Falloon and Smith 2002; González-Molina et al. 2011). Studies using RothC for grassland ecosystems have required specific initialization (Liu et al. 2011; Nemo et al. 2017) using information from long term grassland experiments (Cagnarini et al. 2019). On the other hand, there are also several limitations to RothC particularly under managed moist grasslands. For instance, RothC presented a limitation considering

management (Brilli et al. 2017). Despite the number of possible interactions in grassland systems, e.g., between plant, soil and animals, RothC simplified the effects of different management affecting some of these processes on grasslands and indirectly simulates grazing activity by altering the amount of total plant C inputs. As for animals C inputs, RothC offers default quality values for C inputs from grazing animals or manure applications. Moreover, the model does not consider the trampling effect on soil physical conditions related to grazing (Smith et al. 2014). Besides, under temperate moist climatic conditions, RothC model is unable to adequately predict C dynamics in waterlogged soils (Falloon et al. 2006), which imply oxygen limitation and thus a decline in decomposition rate (Moyano et al. 2013). Furthermore, as a general limitation, regarding plant residues, RothC does not differentiate between above- and below-ground C inputs (Nemo et al. 2017).

Considering the model limitations, we aimed to introduce modifications to RothC and assess the ability of the proposed modifications to predict the measured SOC stocks from intensive grassland sites under moist climatic conditions. To adapt RothC to temperate moist managed grassland, we hypothesized that the aforementioned factors (i) could be easily implemented in RothC, (ii) significantly affect SOC changes and (iii) could improve RothC predictions of SOC changes. To evaluate the suggested modifications, we assessed the model performance against published experiments through a stepwise approach, as well as its sensitivity to the main modifications.

2 Materials and methods

2.1 RothC model overview

The RothC -26.3 (Coleman and Jenkinson 1996) model divides the SOC into five fractions, four of them are active and one is inert (i.e., inert organic matter, IOM). The active pools are: decomposable plant material (DPM), resistant plant material (RPM), microbial biomass (BIO) and humified organic matter (HUM). The decomposition of each pool (except IOM) is governed by first-order kinetics, characterized by its own turnover rate constant and modified by environmental factors related to air temperature, soil moisture and vegetation cover, which are the main input parameters to run the model. Incoming plant C is split between DPM and RPM, depending on the DPM: RPM ratio of the particular incoming plant material or organic residue. Both of them decompose to produce BIO, HUM and evolved CO₂. The proportion that goes to CO₂ and to BIO + HUM is determined by the clay content of the soil which is another input to the model.

The model uses a monthly time step to calculate total SOC and its different pools changes on years to centuries time scale.

2.2 RothC tested modifications

Four modifications were proposed and tested in this study to the RothC excel version (“Rothc_single_layer_4_active_pools_Feb_2013”): (i) extensions of soil water content function up to saturation; (ii) enlargement of C input pools to account for the diversity of applied exogenous organic matter (EOM) from ruminant excreta; (iii) affiniton of plant residue components and quality variability; and (iv) the trampling/poaching effect of grazing animals.

2.2.1 Soil water saturation in RothC

RothC contains a minimum rate modifying factor of 0.2, when soil moisture is at minimum moisture capacity (i.e., at the extreme of water limitation). However, no correction is applied under water saturation and when soil is oxygen limited. In order to represent the reduction in the decomposition rate above field capacity (Moyano et al. 2013; Yan et al. 2016; Han et al. 2019), the rate modifying factor for moisture was assumed to follow a linear decline trend until a minimum rate of 0.2 (20%), at saturation conditions, as suggested by (Smith et al. 2010) in the ECOSSE model.

ECOSSE soil moisture function was derived from SUNDIAL and RothC models under anaerobic and aerobic conditions and based on Rothamsted field experiment (Flattery et al. 2018). The model was designed for use across a range of land uses, and water contents are included up to saturation (Smith et al. 2010). It was evaluated under European conditions and showed a good performance (Dondini et al. 2017).

Soil water contents at saturation and field capacity conditions are estimated by considering soil properties related to soil texture as described by (Raes 2017).

The conversion from soil water content to soil moisture deficit (SMDi, mm) used in RothC referred to (Farina et al. 2013) (Supplementary information A).

2.2.2 Exogenous organic matter diversity (EOM)

Exogenous organic matter partition for the RothC model was estimated by Peltre et al. (2012), based on an indicator of potential residual organic C in soils (IROC), which is derived from Van Soest fractions and the proportion of EOM mineralized during 3 days of incubation. Similarly, Mondini et al. (2017) improved the prediction of SOC stocks in amended soils by fitting the RothC partitioning pools of different EOM to the respiratory curves. Such adjustment of the partition of EOM into RPM, DPM and HUM entry pools of RothC provided a successful fit and had been reproduced in other studies (Pardo et al. 2016). However, the above-mentioned studies have summed up all the different animal excreta into one category and did not distinguish excretions from different animal types (e.g.,

ruminants, pigs...). In order to capture the specific characteristics of ruminant excreta, we developed a methodology based on Pardo et al. (2017) as illustrated in Fig. S1. In this study, Pardo et al. (2017) proposed a partition of the C inputs from excreta into RothC pools based on the relationship between lignin content (Van Soest fractions) and anaerobic biodegradability, estimated as follows (Eq. (1)):

$$B = 0.905 \times \exp(-0.055 \times \text{lig}(\%)) \quad (1)$$

Where B is biodegradability and Lig is lignin content as % of Volatile Solids (VS).

The Van Soest fractions are then partitioned into the pools of RothC based on its degradability, represented by the parameter B (i.e, lignin, holocellulose and solubles). A fraction of lignin is allocated into the HUM pool, representing the most resistant material. The rest of the lignin and the most resistant fraction of holocellulose and solubles are assigned to the RPM, while the most labile fraction of holocellulose and solubles are allocated to DPM.

This is expressed as VS %, following the equations.

$$HUM = \text{Lig} \times (1 - B) \quad (2)$$

$$RPM = \text{lig} \times B + (\text{Holocellulose} + \text{Solubles}) \times (1 - B) \quad (3)$$

$$DPM = (\text{Holocellulose} + \text{Solubles}) \times B \quad (4)$$

The Van Soest fractions were derived from literature review for animal excreta of ruminants. As a result of this review, we identified large variability in animal excreta's fractions (lignin: Fig. S2 and soluble: Fig. S3). These differences were associated to a diverse array of factors and especially those in relation with the animal diet composition (e.g., high concentrate diet generally would imply lower lignin content in the ruminant's excreta). For the ruminant excreta quality to the RothC entry pools, we used as input to the above questions an average value for the different fractions considered (data from Fig. S2 and Fig. S3) (Table 1). Additionally, in a separate exercise, we evaluated how the impact of uncertainties of these fraction values could lead to uncertainties on the SOC results. For this exercise, both extreme values (i.e., maximum and minimum) were assessed using a sensitivity analysis (See Sensitivity analysis section).

Table 1. Ruminant excreta quality and its fitting to the RothC entry pools (based on scientific literature review)

	RothC Pools		
	HUM	RPM	DPM
Ruminant excreta	0.1	0.6	0.3

2.2.3 Plant residue: components and quality

The RothC model does not distinguish between above- and below-ground plant residues. We hypothesise that accounting for month-to-month changes in plant residue quality may improve RothC predictions under wet conditions, while not adding too much complexity to the modelling approach. Regarding plant C inputs distribution, RothC is known to be relatively insensitive to the distribution of C inputs through the year (Smith et al. 2005).

Model users generally use above-ground residues as surrogate for total plant C inputs and do account less for root inputs in RothC (Nemo et al. 2017). Here we separated the plant residue C inputs into three components (i.e., above-ground residues, below-ground residues and rhizodeposits). The structure of C input derived from plant residues in RothC modified model is as illustrated in Fig. S4. Parting from above-ground biomass, we used root to shoot (R:S) ratio to estimate below-ground biomass (when its value is not available). We assumed N fertilisation as the main driver for R:S ratio in grasslands as many studies have proved the strong dependence of the latter on N inputs (Poeplau 2016; Sainju et al. 2017). We referred therefore to Poeplau (2016) equation for RothC C input parameterisation under temperate grasslands in order to consider the fertilisation effect on the R:S ratio:

$$R:S = 4.7375 e^{-0.0043 \cdot N \text{ input}} \quad (5)$$

Where R:S is the Root: Shoot ratio and N input is nitrogen fertilisation expressed in kg N ha⁻¹ year⁻¹.

Unlike in annual croplands, in perennial grassland ecosystems, below-ground C biomass does not correspond to the below-ground residue. Instead, below-ground residues correspond to 50% of the total below-ground C biomass (Poeplau 2016) since the average annual root turnover of grasslands has been estimated to be 50 % in the temperate zone (Gill and Jackson 2000).

Regarding rhizodeposition estimation, we referred to an extensive literature review in which net rhizodeposition-to-root-ratio from grasslands was estimated to be 0.5 (Pausch and Kuzyakov 2018).

We assumed a C concentration of 45 % of the plant biomass (Kätterer et al. 2012).

Plant residue quality (biochemical composition), as one of the main drivers of decomposition, is represented in the RothC model by the DPM:RPM ratio (i.e., ratio of rapidly and slowly decomposing pools), which can be obtained by optimization to obtain the best fit according to different land use types. For instance, for most agricultural crops and improved grasslands, RothC uses a DPM: RPM ratio of 1.44 (i.e. 59% of the plant material as DPM and 41% as RPM). For unimproved grasslands and scrubs (including Savannas) a ratio of 0.67 is used (Coleman and Jenkinson 1996). Plant residue

quality is variable in time and depends on several factors (e.g., maturity stage, climate variables and nitrogen fertilisation) (Buxton 1996; Ball et al. 2001).

In order to fit the DPM: RPM ratio to the specific conditions of temperate grasslands, including its variability over the year, we used the stepwise chemical digestion (SCD) method (Van Soest 1970), already used by (Gunnarsson et al. 2008; Borgen et al. 2011). For simplicity, we assumed that the DPM pool could be approximated to the C in the Neutral Detergent Soluble (NDS), and the RPM pool as the C in the Neutral Detergent Fiber (NDF) (i.e., holocellulose and lignin fractions).

Regarding below-ground plant material quality, the quantity of lignin itself is the main potential driver of differential degradation between both above- and below-ground plant components (Rasse et al. 2005). Therefore, we added up the difference of lignin percentage of ~ 8% (between above- and below-ground parts) to get the below-ground RPM pool, referring to (De Neergaard et al. 2002).

The DPM pool is then derived by subtraction according to the equation:

$$DPM(\%) = 100 - RPM(\%) \quad (6)$$

Finally, we assumed that the C inputs derived from rhizodeposition are transferred to DPM of the RothC because of the expected rapid decomposition of this labile substance by rhizosphere microorganisms (Klosterhalfen et al. 2017).

2.2.4 Animal trampling effect: Poaching

We hypothesise that accounting for animal trampling may improve RothC predictions, while not adding too much complexity to the modelling approach. The trampling effect generally depend on stocking density, soil moisture content, soil texture, and the presence/ absence of a protective vegetation cover (Bilotta et al. 2007). Apart from the stocking rate, the remaining factors were reflected in the RothC default model. In this context, we developed a simple poaching modification based on available data obtained from temperate moist grassland studies (Phelan et al. 2013; Tuñon et al. 2014; Tuohy et al. 2014), considering that our modelling should be validated apart. The main objective of introducing the poaching effect was to predict the level of soil damage and its impact on plant C inputs as a function of soil moisture, soil compaction and degradation under grazing conditions (e.g. under different stocking rates) (Fig. S5). Soil moisture is estimated in RothC using the Soil Moisture Deficit (SMD) value that considers rainwater inputs and soil texture properties (i.e., clay content). According to Herbin et al. (2011) and Piwowarczyk et al. (2011), we used SMD as a proxy for soil moisture to predict when soil water conditions are likely to lead to hoof damage. For simplification reasons, we assumed water saturation conditions from SMD = -10 mm onwards (according to the soil moisture modification), as a condition of poaching occurrence as in Scholefield

and Hall 1985 and Tuohy et al. (2014). Livestock density has an effect on the level and extent of treading damage (Nie et al. 2001; Tuohy et al. 2014) illustrated by hoof print depth and soil surface deformation (Tuñon et al. 2014). Depending on the soil surface deformation of a treading event, the pasture production is reduced (Phelan et al. 2013) and thus its plant C input (Eze et al. 2018a) (Fig. S4, Fig. S5). The main equations related to the conceptual diagram of poaching modification are described in Supplementary information A.

As the poaching effect in temperate grazing systems seems to cause only short-term reduction in pasture plant production but there is a relatively fast recovery to these damages (Black 1975; Tuñon et al. 2014), we considered that plant C input reduction due to poaching effects would only occur in months when soil was prone to poaching.

2.3 Study sites and input datasets

2.3.1 Study sites description

In order to validate the proposed modifications, we used data from four studies of European managed grasslands having temperate conditions and being characterized by precipitations > 1000 mm, and grass and clover mixture. The grassland sites (Laqueuille intensive grazing grassland, Oensingen intensive cutting grassland, Easter Bush intensive grazing grassland and Solohead dairy research farm) (Table 2) were mainly selected from the FLUXNET program (<http://www.fluxnet.ornl.gov/>; (Baldocchi 2008)). Information on geographic and climatic characteristics, soil properties and management of the different sites are listed in the Table 2. More details are provided in Supplementary information B.

2.3.2 Input data for the model and main assumptions

Plant carbon inputs in the different sites were estimated depending on the available data using the method described in the section “Plant residues: Components and quality”. For the Laqueuille site, average above-ground C residue was obtained from available measured data and it represented 20% of above-ground C standing biomass (Table 2). We used the R:S ratio to estimate below-ground biomass from average measured above-ground standing biomass. Below-ground C residues were assumed to be 50% of the below-ground C biomass (Poeplau 2016) (Table 2). For the Oensingen site, average above- and below-ground C biomass were obtained from Ammann et al. (2009). We used the same assumption as Poeplau (2016) for cutting grasslands, assuming that 30% of the above-ground biomass is not harvested, and that only 50 % of that fraction is turned over annually and

becomes available for soil organic matter formation (Schneider et al. 2006) (Table 2). To estimate below-ground C residue, we used the same assumption as commented for Laqueuille site (Table 2). The same assumptions were considered for the grazing Easter Bush site. From the average measured above-ground biomass we assumed only 20% as residues as in the Laqueuille grazing site and the same hypothesis for the below-ground C residue as in the other previous sites (Table 2). For Solohead dairy research farm, we used as input the available measured data of above- and below-ground C biomass and used the same assumption for above- and below-ground C residues as all the previous sites (Table 2). Finally, for the rhizodeposition as commented previously, we used an annual net rhizodeposition-to-root ratio of 0.5.

The proportions of plant C input added to the soil in each month were set as the pattern of inputs typical of European grasslands suggested by Smith et al. (2005). Referring to plant residue quality we ascribed RPM and DPM pools related to NDF and NDS, respectively for each plant residue component (as described in the sub-section “Plant residues: components and quality”).

The C animal excreta in Laqueuille grazing grassland were derived from Vertès et al. (2019) referring to the C intake grass-based rations, as the management is a continuous grazing from May to end of October without additional feed supply (Klumpp et al. 2011). Therefore, we estimated the C animal excreta as 32 % of the measured C intake using average values for the simulation period 2004-2012 (Vertès et al. 2019). Annual C derived from cattle slurry in Oensingen site were estimated from Ammann et al. (2007) as an average of the provided years. Carbon input from grazing animal excreta was estimated the same as in Laqueuille site, while annual C input derived from organic fertilisation for Easter Bush was deduced from Jones et al. (2016) during the period 2004-2010 as 0.32 Mg C ha⁻¹yr⁻¹. The same method was used to estimate annual total N fertilisation and annual stocking rate of this site. For Solohead dairy research farm, C input derived from animal excreta were calculated the same as in Laqueuille site and all other input data were estimated as average annual from the same study (Necpálová et al. 2013).

The different input data to the model regarding management, soil properties to estimate soil water content at saturation and field capacity conditions (Raes 2017), as well as grass type to characterise the plant residue quality for the different study sites are illustrated in Table 2.

Table 2. Location, climate, soil properties, management type and input data to the model of the grassland study sites (available through the European Fluxes Database Cluster: <http://www.europe-fluxdata.eu> (except Solohead farm))

Site name and references	Laqueuille (Klumpp et al. 2011)	Oensingen (Ammann et al. 2009)	Easter Bush (Skiba et al. 2013; Jones et al. 2016)	Solohead farm (Necpálová et al. 2013)
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Country	France	Switzerland	United Kingdom	Ireland
Altitude (m)	1040	450	190	150
Latitude	45° 38' N	47° 17' N	55° 52' N	52° 51' N
Longitude	02° 44' E	07° 44' E	03° 02' W	08° 21' W
Mean air temperature (°C)	7	9	9	10.6
Mean annual precipitation (mm)	1012	1263	1031	1017
Simulation period	2004-2012	2004-2011	2004-2011	2004-2011
Grassland type	Intensive semi-natural permanent grassland	Intensive permanent grassland	Intensive permanent grassland	Intensive permanent grassland
Management (Mowing/Grazing frequency)	-Grazing by heifers (May to October)	-Grass mowing (4 times a year) -No grazing	- Grazing all year round by cattle and sheep	-Grazing by dairy cows February to November -Mowing
Annual production (t ha ⁻¹ yr ⁻¹)	7.1	7.5	5.6	13.5-14.7
Stocking rate (LSU ha ⁻¹ yr ⁻¹)	~1	-	0.83	~2
Total N fertilisation	Mineral fertilisation in three splits: 210 kg N ha ⁻¹ yr ⁻¹	Solid ammonium nitrate or cattle slurry at the beginning of each growing cycle (after the previous cut): 214 kg N ha ⁻¹ yr ⁻¹	Ammonium nitrate fertiliser was applied to the field 3-4 times per year, usually between March and July (~ 229 kg N ha ⁻¹ yr ⁻¹)	N fertiliser ~183 kg N ha ⁻¹ yr ⁻¹
SOC in the topsoil (Mg C ha ⁻¹ yr ⁻¹)	114 ± 1.48 (20 cm depth) in 2004 125.8 (20 cm depth) in 2008 121±2.35 (20 cm depth) in 2012	64.7 (20 cm depth) in 2004 68.3±1.6 (20 cm depth) in 2011	93.26 (30 cm depth) in 2004 87.87 (30 cm depth) in 2011	137±6.5 (30 cm depth) in 2004 142.8±7.14 (30 cm deth) in 2008 148.8±7.16 (30 cm deth) in 2009 149.2±9.7 (30 cm depth) in 2011

Site name and references	Laqueuille (Klumpp et al. 2011)	Oensingen (Ammann et al. 2009)	Easter Bush (Skiba et al. 2013; Jones et al. 2016)	Solohead farm (Necpálová et al. 2013)
Soil properties	The soil is an Andosol (20% clay, 53% silt and 27% sand) with 11% carbon and 18% organic matter.	The soil is classified as Eutri-Stagnic Cambisol (FAO, ISRIC and ISSS, 1998) developed on clayey alluvial deposits. Clay contents between 42% and 44% induce a total pore volume of 55% and a fine pore volume of 32% (permanent wilting point)	The soil type is an imperfectly drained Macmerry soil series, Rowanhill soil association (Eutric Cambisol) with a pH of 5.1 (in H ₂ O) and a clay fraction of 20-26%.	The predominant soils are poorly drained gleys (90%) and grey-brown podzolics (10%) with a clay loam texture and low permeability (28% clay, 35% silt)
Grass type	Grass clover mixtureThe dominant grass are <i>Dactylis glomerata</i> , <i>Trisetum flavescens</i> , <i>Poa pratensis</i> and <i>Agrostis capillaris</i>	Grass clover mixture	>99% rye grass (<i>Lolium Perenne</i>) and < 0.5% white clover (<i>Trifolium repens</i>)	rye grass and white clover (20 to 25%)
Estimated/Measured R:S ratio	1.92 (Estimated)	1.46 (Measured)	1.77 (Estimated)	0.88 (Measured)
Measured Above-ground C (t C ha ⁻¹)	0.89	1.3	0.5	4.8
Estimated/Measured Below-ground C (t C ha ⁻¹)	1.71 (Estimated)	1.9 (Measured)	0.88 (Estimated)	4.2 (Measured)
Plant residue components (t C ha ⁻¹)	C _a = 0.19; C _b =0.86; C _r =0.86	C _a = 0.2; C _b =0.95; C _r =0.95	C _a = 0.1; C _b =0.44; C _r =0.44	C _a = 0.9; C _b =2.1; C _r =2.1
Biomass quality	NDF _a ranges from 0.55 to 0.67 NDF _b ranges from 0.63 to 0.75 NDF _r =0	NDF _a ranges from 0.56 to 0.68 NDF _b ranges from 0.64 to 0.76 NDF _r =0	NDF _a ranges from 0.55 to 0.69 NDF _b ranges from 0.63 to 0.77 NDF _r =0	NDF _a ranges from 0.51 to 0.64 NDF _b ranges from 0.59 to 0.72 NDF _r =0
C input derived from ruminant excreta (t C ha ⁻¹ yr ⁻¹)	0.54	0.47	0.75	2.3

NDF_a, Neutral Detergent Fiber corresponding to resistant above-ground plant material; NDF_b, Neutral Detergent

Fiber corresponding to resistant below-ground plant material; NDF_r, Neutral Detergent Fiber corresponding to

rhizodeposits

C_a, above-ground plant C input; C_b, below-ground plant C input; C_r, plant C input corresponding to rhizodeposition.

2.3.3 Model initialisation and running

For RothC initialisation, since radiocarbon measurements are costly and rarely performed routinely, we used the pedotransfer functions established by Weihermüller et al. (2013) to estimate all active C pools from initial provided measured SOC stocks. The initial IOM pool, according to these pedotransfer functions was set to match the equation proposed by Falloon et al. (1998):

$$IOM = 0.049 TOC^{1.139} \quad (7)$$

We modelled SOC dynamics from the different study sites using a stepwise approach. First, we used the default RothC version (RothC_0) and, subsequently we progressively added the different modifications tested (Table 3): (i) ruminants excreta (RothC_1 modification); (ii) plant residue components and its characteristics (RothC_2 modification); (iii) saturation conditions for soil water function (RothC_3 modification) and (iv) soil poaching (RothC_4 modification).

Soil organic carbon stocks were simulated at 20 cm depth for Laqueuille and Oensingen and at 30 cm depth at Easter Bush and Solohead dairy farm.

Table 3. RothC versions tested in the study with the modification included in each version

RothC version	Modifications
RothC_0	RothC default version
RothC_1	RothC_0 + ruminant excreta characteristics
RothC_2	RothC_1 + plant residue characteristics and its variability
RothC_3	RothC_2 + saturation conditions for soil water function
RothC_4	RothC with all modifications: RothC_3 + inclusion of poaching effect

2.4 Model evaluation

We used different performance indices and threshold criteria based on Smith and Smith (2007) (Table S1). The ability of each modification to improve SOC dynamics simulation was evaluated using the root mean square error (RMSE), mean difference of simulations and observations (BIAS) and the model efficiency (EF) (Table S1).

2.4.1 Sensitivity analysis

Several studies have indicated that the RothC model is most sensitive to C inputs (Gottschalk et al. 2012; Stamati et al. 2013; Riggers et al. 2019). In our study, analyses were performed to test the sensitivity effect on SOC changes of the different modifications (other than C inputs) implemented in the model, using RothC_4. Model sensitivity was expressed as an index, which considered different input values related to the modifications (i.e., plant residues quality, ruminant excreta quality and soil moisture up to saturation) from minimum to maximum (Table 4) and then the output values were analysed according to the following index (Smith and Smith 2007).

$$\text{Sensitivity index} = \frac{\max(Pi) - \min(Pi)}{\max(Pi)} \quad (8)$$

Where max (Pi) is the maximum output value and min (Pi) is the minimum output value.

We used NDF as a proxy for RPM in relation with plant residues quality (Table 4), assuming that NDF varies from 30 to 70% as minimum and maximum values based on 15 papers (Table S2). We used the lignin fractions (% VS) as a proxy for EOM in relation with ruminant excreta quality assuming minimum and maximum values from literature values shown in Table 4. Similarly, for soil moisture variation, we tested minimum (0.2) and maximum values (1) of the rate modifying factor for moisture (Table 4).

Table 4. Model modified components used for the sensitivity analysis and their interval values

Modified component	Proxy	Interval for possible values
Plant residues quality (e.g., perennial grass)	NDF	30 – 70 %
Quality of ruminant excreta (e.g., cattle slurry)	Lignin	9 – 28%
Soil moisture up to saturation	Rate modifying factor for moisture	0.2 – 1

3 Results and discussion

3.1 Measured versus simulated SOC stocks

All four sites showed, in general, a similar pattern of annual SOC stocks with the RothC default version (i.e., RothC_0) as well as with the four modified versions (Fig 1). In all four sites, the lowest simulated SOC stocks were observed in the default model version (RothC_0). RothC_0, for Laqueuille, Oensingen and Solohead sites, simulated that SOC was reduced during the time of the experiment (Figs 1a, 1b and 1d), which was the opposite trend that measurements showed. For example, in the Laqueuille intensive grassland, SOC stocks predicted by the RothC_0 version decreased from 114 to 102 Mg C ha⁻¹ whereas measured values increased from 114 to 121 Mg C ha⁻¹ (Fig 1a).

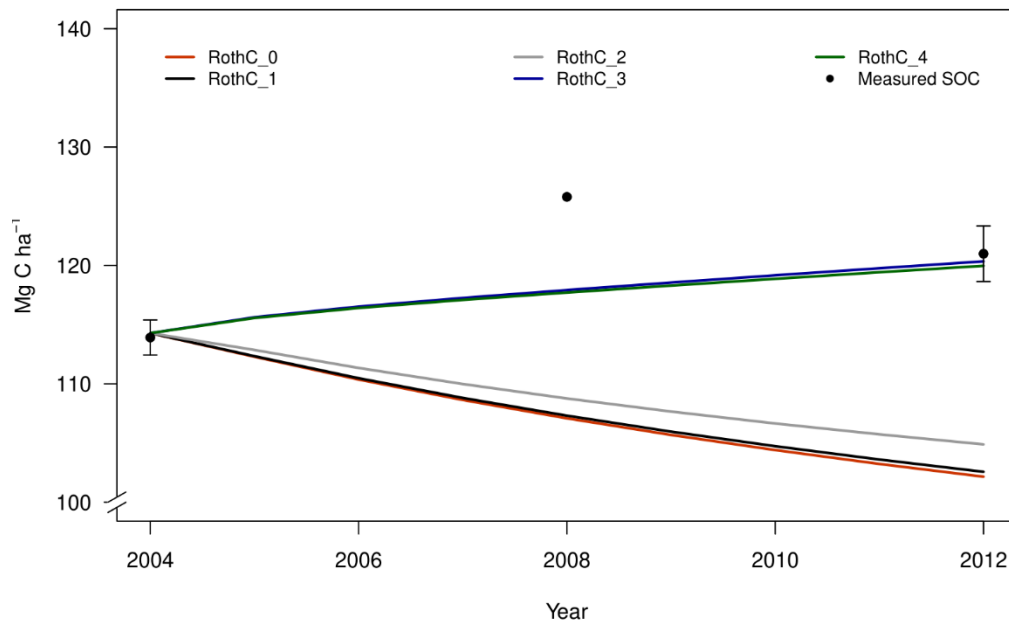
By implementing changes to account for ruminant excreta quality (RothC_1) on the study sites, the model resulted in a slight increase in SOC in time. Moreover, this SOC increase was lower than that simulated by RothC_2 (Fig 1). Changes in the modification of plant residues (RothC_2) resulted in greater SOC increased values in time when compared with the previous modification (RothC_1) (Fig 1). The lower effect of the simulation of animal excreta characteristics in RothC_1 could be explained by the higher quantity of plant residues while adding the rhizodeposition component together with above- and below-ground components in RothC_2.

By introducing the soil moisture modification in RothC (RothC_3), the model simulated an increase in SOC stocks which, trend-wise, differs from the RothC_0 model, but coincides with measured data (Fig 1). For example, SOC stocks at the end of the simulation period in 2011 reached 88.38 Mg C ha⁻¹ (RothC_3) compared to 83.7 Mg C ha⁻¹ (RothC_0) in the Easter Bush intensive grazing grassland (Fig 1c). Soil moisture modification at saturation reduces decomposition rates values for very wet soils conditions. In fact, the 4 sites used in our study have soil water saturation during many months of the year (with an average of 8 months).

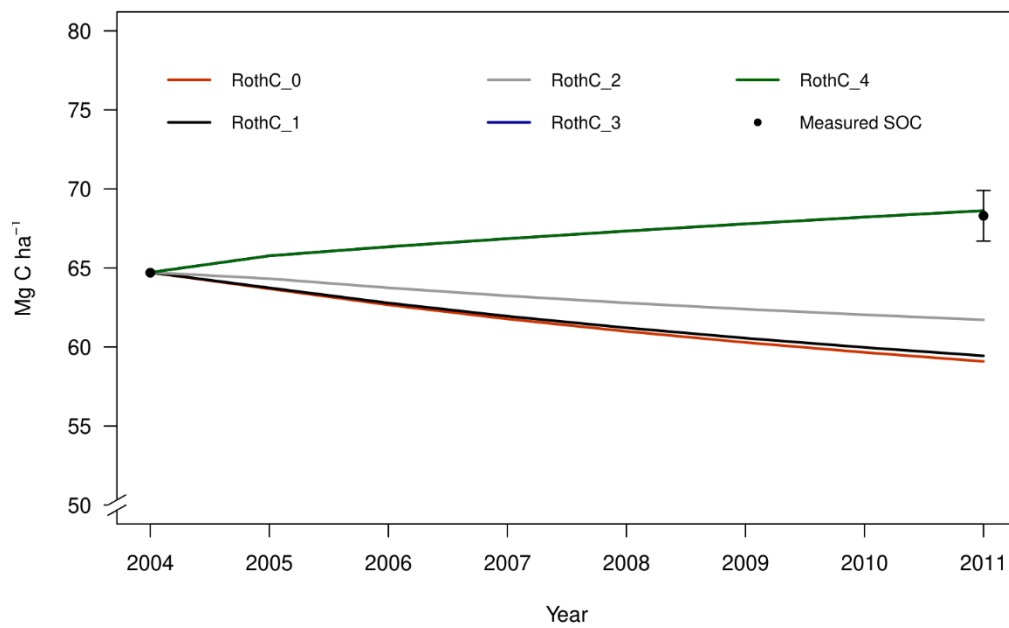
Including the poaching effect (RothC_4), resulted in slightly reduced SOC stocks compared with RothC_3, specially for the Solohead site (Figs 1a, 1c and 1d). This reduction in SOC stocks in RothC_4 compared with the RothC_3 version could be explained by the reduction in plant C inputs due to poaching that typically occurs at saturation conditions (Menneer et al. 2005; Eze et al. 2018a). In general, the highest predicted SOC stocks values and the closest to the measured values at the end of the simulation period resulted after RothC_3 and RothC_4 simulations (Fig 1). For Laqueuille grassland intensive site, RothC_3 and RothC_4 were able to match the general trend of SOC increase (between 2004 and 2012) and the SOC stocks at the end of the simulation period, but not the change

of SOC stocks corresponding to the year 2008. However, SOC simulation for Solohead research farm, using RothC_3 and RothC_4 modified versions were within the range of measured data of SOC stocks (Fig 1).

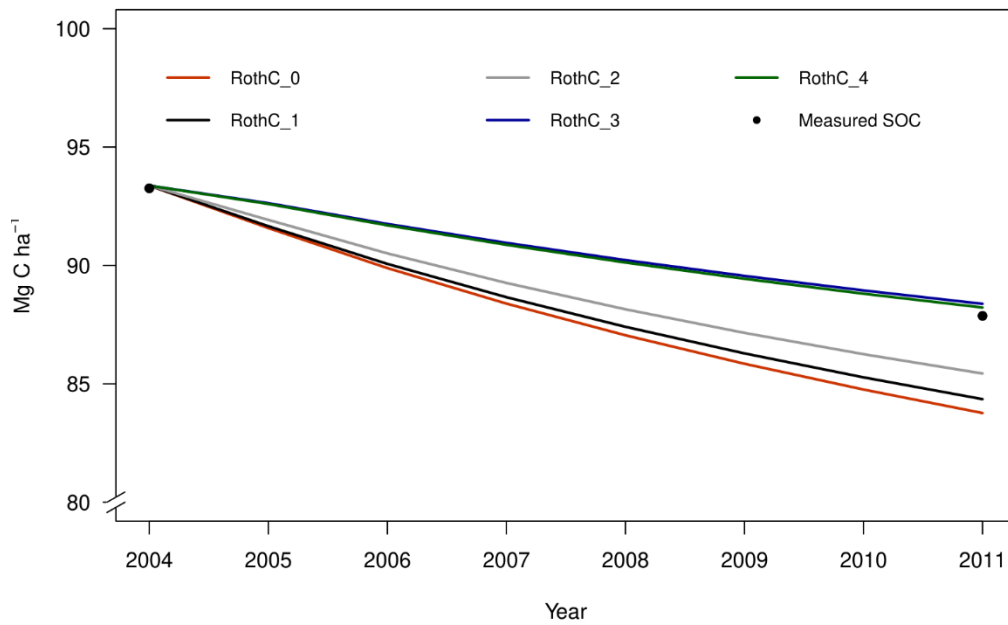
(a)



(b)



(c)



(d)

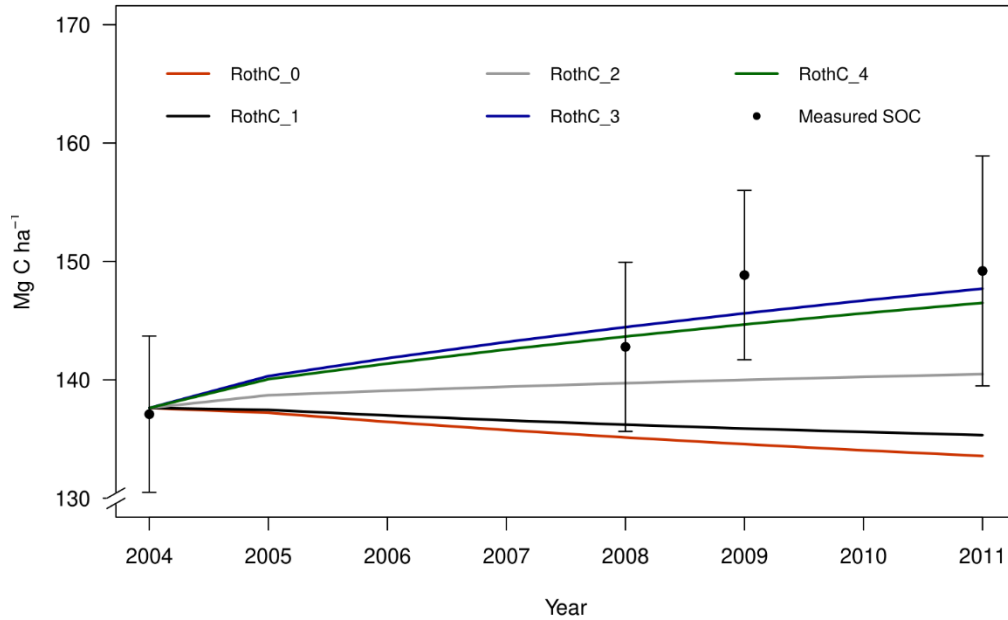


Fig. 1. Measured and simulated annual SOC stocks (Mg C ha^{-1}) using the default RothC model (RothC_0) and the modified RothC versions (RothC_1; RothC_2; RothC_3; and RothC_4) for the different validation sites: (a) Laqueuille intensive grazing grassland; (b) Oensingen intensive cutting grassland; (c) Easter Bush intensive grazing grassland; and (d) Solohead dairy research farm

3.2 Model performance

In general, the RothC default version (RothC_0) showed a good performance with an EF value of 78% (Table 5). However, the different cumulated modifications presented enhanced the predicting performance of RothC for these specific sites. In particular, simulated SOC stocks using the RothC_3 and RothC_4 versions almost matched measured values (Fig 2) achieving model efficiencies of 99% and 98% (Table 5). Therefore, these two modifications accurately predicted SOC changes.

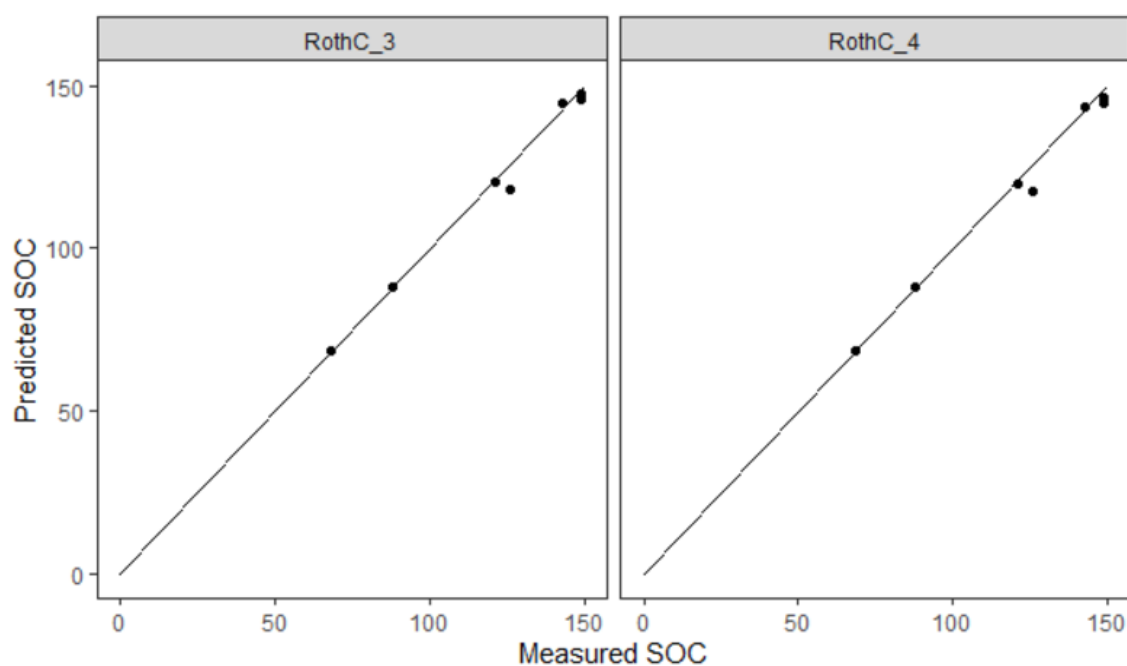


Fig. 2. Measured versus predicted values of SOC stocks at the end of the simulation period using RothC_3 and RothC_4 model versions for the different study sites

The negative bias (reaching -18.8 in Laqueuille site) and the higher RMSE values obtained in RothC_0 compared with the different RothC modified versions indicated that the default RothC version underestimated SOC stocks, especially in the Laqueuille and Solohead sites, which presented the highest SOC content (Table 5). This confirmed the fact that the RothC model is unable to adequately predict soil C dynamics in organic or waterlogged soils (Falloon et al. 2006). In this context, adding the modification of the soil moisture function in RothC_3 reduced the bias and the RMSE (Table 5) and improved the general trend of SOC stocks compared with the default version RothC_0 in all simulated sites (Fig 1). RothC_0 assumes high decomposition rates with high soil moisture, but it does not consider the cessation of the decomposition process which occurs in high wet soils close to saturation conditions (Das et al. 2019), frequent in temperate moist grasslands. The inclusion of the ruminant excreta quality in the model only slightly improved the SOC predictions in

RothC_1 compared to RothC_0 (Table 5). In this context, Heitkamp et al. 2012 and Mondini et al. (2017) emphasised the importance of modifying the quality of residues to improve the model performance, concluding that the adjustment of DPM:RPM ratio led to better model performance than the use of default DPM: RPM values provided by the model.

Comparing RothC_1 and RothC_2 versions, it could be deduced that integrating quantity and quality distinction of plant residue in RothC_2, as a primary source of SOC (Castellano et al. 2015), improved SOC predictions. Adding the modification of plant residues in terms of quantity and quality contributed to improve SOC simulation compared to the modification of specifying animal excreta quality. The improvement showed by plant residues modification, particularly in Solohead Research farm, could be explained by the higher sensitivity of the model to C inputs quantity compared to C inputs quality and the importance of including rhizodeposition together with above- and below-ground components in plant C input quantification. Indeed, as a fundamental source of C inputs, rhizodeposition was recommended to be added to the different plant residue components in SOC models (Rumpel and Kögel-Knabner 2011; Klosterhalfen et al. 2017), particularly RothC (Balesdent et al. 2011).

The poaching effect is assumed to reduce plant productivity and the potential amount of C inputs to the soil (Eze et al. 2018a) and thus causing SOC loss (Ma et al. 2016). Consequently, the poaching modification included in the RothC_4 version predicted reductions in SOC stocks compared to the RothC_3 version (Fig 1a, 1c and 1d). The reduction in SOC stocks is explained by the lower C inputs during the months when grazing occurs under saturation conditions. Only in the case of the Easter Bush site, the poaching modification contributed to improve SOC predictions in the RothC_4 version (Table 5, Fig 1c). A possible explanation to this improvement in the SOC predictions is that the soil in Easter Bush site is poorly drained and grazing by ruminants occurs all year round and thereby highly susceptible to poaching. In the same context, Vuichard et al. (2007) enhanced the original PASIM grassland constructing a simple and empirical model of the detrimental impact on vegetation of trampling by grazing animals by removing at each time step a fixed proportion of the above- ground biomass. However, it is important to point out the complexity of the poaching effect, as it induces more impacts other than the detrimental vegetation impact which are beyond the scope of our study. In this context, Russell and Bisinger (2015) pointed out the inconsistency and limitation of the studies dealing with the grazing effect on SOC. Therefore, more robust experiments are needed in order to define the severity of the poaching effect according to soil moisture saturation, livestock density and soil type.

Therefore, particularly, RothC_3 showed the best agreement (Table 5, Fig 2), as the effect of the poaching modification added in RothC_4 is minimal and uncertain. In this sense, the poaching

modification could be of major importance under heavy stocking rates or overgrazing management associated to SOC loss (Dlamini et al. 2014).

Table 5. Root mean square error (RMSE) and mean difference of simulations and observations (BIAS) of SOC stocks (Mg C ha⁻¹) for each model version and grassland intensive site and model efficiency (EF) and RMSE across sites

Site	Performance test	RothC_0	RothC_1	RothC_2	RothC_3	RothC_4
Laqueuille	BIAS	-18.77	-18.45	-16.56	-4.26	-4.55
	RMSE	15.21	14.95	13.43	4.53	4.67
Oensingen	BIAS	-9.22	-8.86	-6.58	0.32	0.32
	RMSE	13.49	12.97	9.64	0.47	0.47
Easter Bush	BIAS	-4.10	-3.52	-2.43	0.51	0.35
	RMSE	4.67	4.00	2.77	0.58	0.40
Solohead	BIAS	-12.52	-11.13	-6.88	-1.02	-2.00
	RMSE	8.85	7.89	5.03	1.55	1.98
All sites	RMSE	11.36	10.75	8.66	2.77	3.01
	EF	0.78	0.80	0.87	0.99	0.98

Testing the model performance based on each of the individual modifications for the different sites allowed improving our understanding of its impact to the model (Table 6). Soil moisture up to saturation conditions in the soil water function of RothC showed the best performance compared with the other modifications (Table 6). The modification of RothC water function at saturation conditions fit to the temperate moist climatic conditions, since the different study sites showed saturation conditions most of the year. However, the poaching effect alone contributed to reduce SOC stocks and thus the model performance, since the poaching effect is related to water saturation conditions (Tuohy et al. 2014). The enhancement in the model performance showed by the quality of ruminant excreta depends on its quantity. Indeed, the BIAS reduction with ruminant excreta quality modification compared with the default version (Table 5 and 6) was more important in the grassland sites with major ruminant excreta application (e.g., Solohead research farm). However, the plant residue modification showed a higher improvement compared with the ruminant excreta quality as it implies an increase in C inputs with the inclusion of the rhizodeposition component.

Table 6. Root mean square error (RMSE) and mean difference of simulations and observations (BIAS) of SOC stocks (Mg C ha⁻¹) for each specific modification (i.e., soil moisture up to saturation, ruminant excreta quality, plant residue, poaching effect) to the model and grassland intensive site and model efficiency (EF) and RMSE across sites

Site	Performance	Soil moisture up to saturation	Ruminant excreta quality	Plant residue	Poaching
Laqueuille	BIAS	-7.27	-18.45	-16.88	-18.91
	RMSE	6.31	14.95	13.69	15.32
Oensingen	BIAS	-2.95	-8.86	-6.94	-
	RMSE	4.32	12.97	10.16	-
Easter Bush	BIAS	-1.29	-3.52	-3.02	-4.21
	RMSE	1.47	4.00	3.44	4.79
Solohead	BIAS	-7.17	-11.13	-8.27	-13.21
	RMSE	5.23	7.89	5.97	9.31
All sites	RMSE	5.51	10.75	9.19	12.19
	EF	0.95	0.80	0.86	0.46

However, testing the model based on the combined effect of soil moisture up to saturation and poaching effect showed a great performance compared with the effect of excreta and plant residues for the different sites with a RMSE of 5.96 compared with 8.66 (Table 7). The modifications of soil moisture up to saturation and poaching effect reduced the BIAS compared with animal excreta and plant residue modifications for the different study sites, except for the Solohead research farm. This could be explained by the fact that the latter received higher C inputs derived from animal excreta and plant residues and lower water saturation conditions compared with the other sites (Table 2). The modifications of soil moisture up to saturation and plant residues presented the best performance among all sites (Table 7). Particularly, the plant residues modification implied an accounting for rhizodeposition component and thus a significant increase in C inputs compared with the minimum proportion of plant residues reduction induced by the poaching effect of grazing animals at saturation conditions. Therefore, the model modification with the greatest positive impact was soil moisture up to saturation (Table 6 and 7). However, the impact of plant residues and ruminant excreta modifications depends on the C input quantity (Table 6 and 7). The poaching effect could not be considered without taking into account the soil moisture saturation modification, as it showed a lower performance than the default model version (Table 5 and 6).

Table 7. Root mean square error (RMSE) and mean difference of simulations and observations (BIAS) for the combined modifications (soil moisture up to saturation and poaching; ruminant excreta and plant residues; soil moisture saturation and plant residues) in Mg C ha⁻¹ to the model and grassland intensive site and model efficiency (EF) and RMSE across sites

Site	Performance test	Soil moisture saturation and Poaching effect	Ruminant excreta and plant residues	Soil moisture saturation and plant residues
Laqueuille	BIAS	-7.79	-16.56	-4.58
	RMSE	6.67	13.43	4.67
Oensingen	BIAS	-2.95	-6.58	-0.05
	RMSE	4.32	9.64	0.07
Easter	BIAS	-1.44	-2.43	-0.10
Bush	RMSE	1.64	2.77	0.11
Solohead	BIAS	-7.96	-6.88	-2.45
	RMSE	5.76	5.03	2.24
All sites	RMSE	5.96	8.66	3.12
	EF	0.94	0.87	0.98

3.3 Sensitivity analysis

A sensitivity analysis of RothC_4 was performed to assess the robustness of the modifications (plant residues quality, ruminant excreta quality and soil moisture up to saturation) made in the different model versions presented. In general, RothC_4 seems to be more sensitive to C input quantity than to quality and to soil moisture function, particularly at saturation conditions.

The sensitivity analysis performed for resistant plant residues pool with the RothC_4 version showed a sensitivity index varying between 0.8% for the Easter Bush site and 2.6% for Oensingen and Solohead research farm (Table 8). Although the model was not very sensitive to the quality of plant residues, adding this modification enhanced the results depending on the quantity of plant residues (Table 8). In this context, according to other studies (Shirato and Yokozawa 2006; Borgen et al. 2011; Heitkamp et al. 2012), specifying plant C input quality depending on residues partitioning instead of using the default RothC ratio for DPM and RPM should enable more reliable modelling of SOM dynamics. In order to ensure the sensitivity of the model to the plant C inputs in terms of

quantity, we assessed its sensitivity to the R:S ratio based on our extensive literature review for temperate grassland species (Table S3). The sensitivity shown by the model to plant residues was higher than the sensitivity to the plant residues quality (Table S4).

In relation to the sensitivity of the RothC_4 version to the animal excreta quality, the values of sensitivity index obtained for the different experiments were in general low (between 1.1% and 3%) (Table 8). So, the use of average value for the different animal excreta fractions does not really impact the results, as we implemented in EOM modification. As for plant residues, the greatest value for the Solohead research farm could respond to the higher C inputs derived from animal excreta that received Solohead research farm as compared to the other sites. In order to focus on RothC_4 sensitivity to animal excreta quality with relation to its quantity, we assumed an annual C input derived from animal excreta of about 2.5 t C ha⁻¹ distributed between March and June for the remaining sites that receive smaller amount of organic fertiliser. As animal excreta quality in the RothC model is connected to its quantity, the sensitivity index of animal excreta quality increased as its quantity increased (Table S5). In this context, according to Mondini et al. (2018), RothC displayed a moderate sensitivity to variations in animal excreta quality, more specifically the ratio between decomposable and resistant pools.

Sensitivity index regarding soil moisture modification was higher compared with the other modifications reaching, for example 12.8% in the Laqueuille site (Table 8). The variation in the sensitivity among the different study sites depend on their soil properties. Therefore, the modified model is sensitive to the rate modifying factor for soil moisture up to saturation under temperate moist climate conditions. In this context, Bauer et al. (2008) concluded that reliable prediction of carbon turnover requires that the soil moisture together with the soil temperature reduction functions of the model need to be valid for the environmental conditions.

Table 8. Sensitivity index of varying resistant plant residues fraction, lignin content corresponding to animal excreta quality and the rate modifying factor for moisture from its minimum to maximum values in RothC_4 for the different study sites

Site	Plant residues quality (Resistant fraction)			Animal excreta quality (Lignin content)			Rate modifying factor for soil moisture		
	Output (min)	Output (max)	Sensitivity index	Output (min)	Output (max)	Sensitivity index	Output (min)	Output (max)	Sensitivity index
Laqueuille	118.6	120.4	1.5%	119.1	120.4	1.1%	104.6	120	12.8%
Oensingen	67.2	69	2.6%	68	69	1.4%	61.6	69.7	11.6%
Easter Bush	87.6	88.4	0.8%	87.3	88.7	1.6%	85.3	89.6	4.8%
Solohead	143.8	147.6	2.6%	143.6	148.1	3%	139.4	150.4	7.3%

3.4 Sources of uncertainty and research needs

Although RothC_3 and RothC_4 simulations performed well in simulating SOC changes for the selected sites, there were limitations related to the uncertainty of, both, model inputs and modifications, and to the limitation of the data used for validation.

Regarding model inputs, uncertainty was mainly related to the lack of detailed measured data of C inputs derived from plant and/or animal origin. In this study, we used the average of available measured values (details can be found in the section “Input data for the model and main assumptions”). However, measured C inputs is not always available, so its value could be supplied via linkage with another model, considering the grazing effect (case of plant residues). It is important to point out that previous studies running RothC in grassland ecosystems overestimated C inputs (Nemo et al. 2017) and there is a lack of detailed information on how plant residues were estimated and/or assumptions regarding their conversion to C inputs (Nemo et al. 2017). In particular, the estimation of below-ground C inputs is another major source of uncertainty for SOC modelling (Keel et al. 2017). Indeed, belowground C inputs depend on multiple factors, including site-specific agronomic practices and the response of plant genotypes to them, and direct measurements of belowground C inputs is a challenging issue (Cagnarini et al. 2019). For instance, if we estimate R:S ratio according to equation (5) with the measured values in Oensingen site, we found close values of 1.9 and 1.5, respectively. However, for the Solohead research farm, the values were more different with a measured R:S ratio of 0.88 compared to an estimated value of 2.1. Moreover, the use of pedotransfer equations for initialising SOC pools, as an alternative for soil physical fractionation, may represent another source of uncertainty (Van Looy et al. 2017). Indeed, although the reliability of pedotransfer equations, they could reveal some errors which are in the range of measurement error for SOC (Weihermüller et al. 2013).

Regarding model modifications, a linear decline in the rate modifying factor for soil moisture was assumed under saturation conditions, like in the ECOSSE model, as there was not sufficient evidence to suggest a more refined relationship as indicated by Smith et al. (2010). However, the effect of soil moisture on SOC dynamics is complex and non-linear (Batlle-Aguilar et al. 2010), interacting with temperature effect (Lee et al. 2018). Improvements could be achieved by using a more refined function based on robust field experiments in order to better represent the different grassland sites. Furthermore, our estimations of animal excreta quality, based on literature review, are not conclusive and further refinements based on experiments could be made as, for example, to account for animal intake quality to predict its excreta quality. Regarding the poaching effect modification, based on the literature review we made, the number of long-term experiments under temperate moist region is limited. Moreover, due to the complexity of the soil damage (i.e., poaching) in which many factors

could be involved (i.e., soil, animal, plant) (Tuñon 2013), it is difficult to generalise our findings. The lack of usable, mechanistic simulation models of soil deformation under hooves and wheels is partly due to the lack of appropriate conceptual understanding and theory of the complex soil mechanical processes involved as well as the shortage of good and relevant experimental data (Scholefield and Hall 1985).

Our equations and values suggested for the different modifications are representative for the conditions of moist temperate intensive grasslands and other site-specific equations, that are tailored to the objective study site, could be used.

In our study, simulations of the different modifications were compared to measured data of different study sites. However, field measurements also have deviations, which are source of uncertainty as they are used as the scale to evaluate model performance (Chen et al. 2017).

As future improvements, measurement of the different input data to the model (e.g., plant residues) would maximise the accuracy of estimations. However, this technique involves time, cost and labour (Catchpole and Wheelert 1985). As an alternative non-destructive method, combining the process-based model RothC with machine learning techniques can successfully help infer additional information from incomplete data sets (Morais et al. 2019). For instance, the machine learning algorithms based on remote sensing data, such as the Artificial Neural Network as a powerful empirical modelling, could improve the estimation of above-ground biomass with higher accuracy (Yang et al. 2018; Gao et al. 2020).

For future work, our modifications could be reproduced and /or refined to improve assessments of SOC changes in managed grasslands under temperate climatic conditions not only at a plot level but also at regional level since grassland systems continue to be understudied at broader scales (Gottschalk et al. 2012; Morais et al. 2019).

4 Conclusions

This study adapted the RothC model to managed grasslands under temperate moist conditions. The proposed modifications to the model considered the incorporation of distinction for plant residues components (i.e., above- and below-ground residues and rhizodeposition) in terms of quantity and quality and distinction for ruminant excreta quality, the extension of soil moisture up to saturation conditions and, finally, the introduction of the livestock damaging effect (i.e., poaching) on plant residues under water saturation conditions. The moisture response modification and the partition of C inputs derived from plant residues components improved the model predictability, but plant residues and ruminant excreta quality modifications improved the model predictability at a lesser

extent. Finally, poaching simulation did not improve the model, since it results in complex and multi-factorial effects in these temperate grasslands. These modifications do not impair the performance of the model under temperate conditions. Indeed, they represent a broadening in the capability of the RothC model to simulate managed grassland under temperate moist conditions. It must be kept in mind that although there was good agreement between results from modified model and measured data from different studies, validating against more sites would greatly improve model confidence.

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CHAPTER 3: REGIONAL MODELLING OF SOIL ORGANIC CARBON CHANGES AND GREENHOUSE GAS EMISSIONS IN GRASSLANDS ASSOCIATED TO CATTLE DAIRY PRODUCTION IN NORTHERN SPAIN

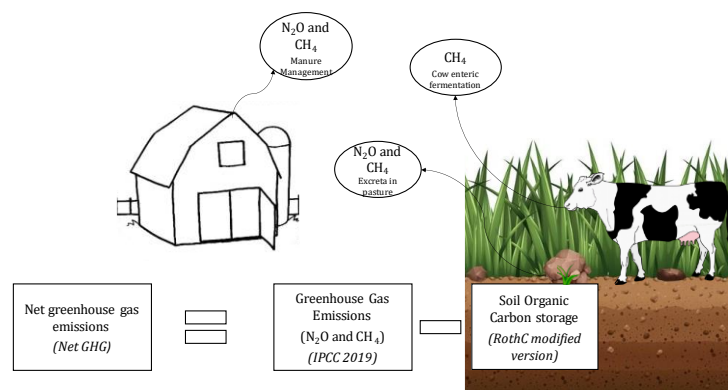
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Abstract

Grasslands are one of the most widespread vegetation types worldwide, providing several ecosystem services and predicting the regional net greenhouse gas emissions (Net GHG) on grasslands has become of great interest.

In this study, we aimed to assess regional soil organic carbon (SOC) change, over 30-yr period (1981-2010), and annual greenhouse gas (GHG) balance in 405,000 ha of moist temperate Spanish grasslands associated to dairy cow production. To achieve this aim, we used an integrated modelling framework consisting in geographic information systems (GIS), the RothC model to simulate SOC changes in managed grasslands under moist temperate conditions and Tier 2 recent IPCC methodologies to estimate emissions.

Results showed an average regional SOC change rate of $0.16 \text{ Mg C ha}^{-1} \text{ year}^{-1}$, related with the initial SOC and livestock density. Average net GHG was positive, contributing to global warming by $5.6 \text{ Mg CO}_2\text{-e ha}^{-1} \text{ year}^{-1}$. Livestock density was the main factor affecting net GHG emissions in the grasslands associated to dairy production of Northern Spain. We determined a threshold of livestock density below which there is no SOC accumulation of 0.95 LU ha^{-1} and a threshold of approximately 0.4 LU ha^{-1} above which net GHG emissions per livestock unit are reduced.

In conclusion, our study confirms the importance of dairy cow grazing systems to preserve and/or enhance SOC stocks in grasslands of Northern Spain. It is therefore crucial to optimise the livestock density over a large variety of feed intake and alternative manure management mitigation options to reduce the net GHG emissions.

1 Introduction

Grasslands are one of the most widespread vegetation types worldwide, occupying 70% of the world's agricultural area (Whitehead et al. 2018) and represent an important ecosystem which provides key services to human society (Eze et al. 2018; Ma et al. 2015), including food production and soil C sequestration (Klumpp and Fornara 2018).

However, decadal fluctuations have occurred in the GHG balance of grasslands, with values ranging from a net GHG source of $0.6 \pm 1.3 \text{ Gt CO}_2\text{-e year}^{-1}$ in the 1970s to a source of $1.8 \pm 0.7 \text{ Gt CO}_2\text{-e year}^{-1}$ during the 2000s (Chang et al. 2021). Within the livestock sectors, which ties with grasslands via grass feeding, the cattle dairy sector, emitting about 3.2% of the global anthropogenic GHG emissions (Gerber et al. 2013), is a major contributor to the total GHG emissions. Important sources of direct GHG emissions from dairy farms include CH_4 enteric fermentation and, CH_4 and

N₂O from manure storage and handling, and crop and pastureland. At the same time, grassland soils can act as carbon sinks, which, in grassland-based livestock systems, can partly offset the climate impact of cattle production. For example, in Canadian beef cattle systems, two thirds of direct CH₄ and N₂O emissions were offset by SOC accrual (Liang et al. 2020). Quantifying the net balance between soil organic carbon (SOC) stocks in grasslands and GHG emissions is therefore important to assess the climate impact of grassland-based livestock systems (Conant et al. 2017).

Spain is one of the seven major producers of cow milk in the EU (EUROSTAT 2019). Of the 156 million tonnes that the EU is estimated to produce per year, Spain provides 5.1%, having provided 7.2 million tonnes in 2019. The dairy farming activity is mainly located in regions of the Northern Atlantic zone of Spain, where grasslands are very productive as a consequence of adequate climatic conditions of frequent rainfall and cool temperature (Smit et al. 2008). Whereas the number of cows in milking in Spain has been substantially reduced by over 20% in the last years, the level of production of milk has increased due to improvements in productive and reproductive management (MAPA, 2019). Moreover, these improvements have resulted in a reduction in the national inventory of GHG in direct CH₄ emissions for the dairy cattle of 25% and 36% for enteric fermentation and manure management, respectively (Cortés et al. 2021; UNFCCC 2021).

Alongside the national GHG inventory, there have been studies using a life cycle assessment (LCA) framework and varied methodologies to estimate the C footprint of milk from dairy cattle in Northern Spain. Del Prado et al. (2013) assessed the C footprint of milk and farm C balance from 17 surveyed commercial confined dairy farms in the Basque Country (Northern Spain) using a combination of models (e.g., grassland model NGAUGE: Brown et al. 2005). Laca et al. (2019) analysed C footprint from two different dairy systems (semi-confinement and pasture-based) in Asturias (Northern Spain) using the Greenhouse Gas Protocol. Ibidhi and Calsamiglia (2020) estimated C footprint, excluding emissions from transport and purchased feeds, in twelve Spanish dairy farms selected from three regions in Spain using the Integrated Farm System Model (IFSM). To our knowledge, however, there are no regional estimations of SOC and GHG emissions in Northern Spain grasslands under moist temperate climatic conditions. In order to assess the regional net direct GHG emissions of managed grassland-based dairy cattle systems of Northern Spain (at the grassland soil and barn level), an integrated modelling framework consisting in geographic information systems (GIS), the RothC model to simulate SOC changes and Tier 2 IPCC methodologies to estimate the CH₄ and N₂O emissions from enteric fermentation, manure storage and handling, and grassland soils were used.

2 Materials and methods

2.1 Study area and dairy systems characterization

The simulated area includes all provinces of the Spanish Autonomous Communities of Galicia, Asturias, Cantabria, the Basque Country and Navarra (Fig. S1). The climate is mainly European Atlantic with annual mean rainfall greater than 1000 mm and air temperature of about 12-14°C per year.

In the studied area, the surface of land covered by permanent grasslands and forage crops are 1,388,007 ha (Fig. S2) and 265, 217 ha, respectively (ESYRCE 2019). These two surfaces correspond to the 63% and 12% of the utilised agriculture area, respectively (ESYRCE 2019).

Dairy cattle production in Northern Spain accounts for 60% of milk production in the Spanish Country (MAPA 2016). The usual cattle breed is Holstein-Friesian. In order to best characterise the diversity of farm management in this area, we identified different typologies of cattle dairy farming based on two recent reports (Flores-Calvete et al. 2016 and MAPA 2019) and gathered input data to define each typology. The different typologies of annual diets composition and management for the lactating dairy cows among the different regions of our study area are described in “Cows´diet” sub-section. Average annual dairy production of the study region per farm is about 233 tonnes of milk (Table S1) but regionally, it varies across the different Northern Spanish regions as shown in Table S1 (Flores-Calvete et al. 2016). The common management strategy consists in pasture management for the whole year of heifers and dry cows (except in winter) while confining of the lactating cows for most of the year, feeding them both annual forage crops (often maize silage) and concentrates (MAPA 2019) (See “Cows´Diet” sub-section).

2.2 Change in SOC stocks

We used a modified version of the RothC model (Jebari et al. 2021) adapted to simulate SOC changes in managed grasslands under moist temperate climatic conditions. The RothC (Coleman and Jenkinson 1996) model divides the SOC into five fractions, four of them are active and one is inert (i.e., inert organic matter, IOM). The active pools are: decomposable plant material (DPM), resistant plant material (RPM), microbial biomass (BIO) and humified organic matter (HUM). The decomposition of each pool (except IOM) is governed by first-order kinetics, characterized by its own turnover rate constant and modified by environmental factors related to air temperature, soil moisture and vegetation cover, which are the main input parameters to run the model. Incoming plant C is split between DPM and RPM, depending on the DPM:RPM ratio of the particular incoming plant material

or organic residue. Both of them decompose to produce BIO, HUM and evolved CO₂. The proportion that goes to CO₂ and to BIO + HUM is determined by the clay content of the soil which is another input to the model. The model uses a monthly time step to calculate total SOC and its different pools changes on years to centuries time scale.

The modifications of the modified model version we used consisted in (i) considering plant residues components and its quality variability across the year, (ii) established entry pools that account for the ruminant excreta as a specified exogenous organic matter and (iii) water contents up to saturation in the soil water function (Jebari et al. 2021).

For RothC initialisation we used the pedotransfer functions established by Weihermüller et al. (2013) to estimate all active C pools. The initial IOM pool was set to match the equation proposed by Falloon et al. (1998):

$$IOM = 0.049 SOC^{1.139} \quad (1)$$

The assessment of SOC stock changes of dairy cows' grasslands from 1981 to 2010 was based on spatial units (i.e., municipalities), using GIS (i.e., ArcMap 10.2). We developed a VBA (Visual Basic for Applications)-based program in Excel to simulate changes in SOC stocks for the 1981-2010 period at municipalities level simultaneously. We used this approach since the regional simulation is computationally intensive and time consuming due to the combination of a large number of runs.

2.2.1 Input data

Climate data

Monthly average temperature and precipitation data for the different Northern municipalities of Spain were extracted from the Spanish State Meteorological Agency (AEMET 2012) for the range 1981–2010. Monthly potential evapotranspiration was estimated using Thornthwaite equations (Thornthwaite 1948).

Soil properties

Soil properties data were obtained from a previous assessment at the national Spanish level (Rodríguez Martín et al. 2016). In this assessment, soil texture and SOC from the 0-30 cm soil depth were analysed and spatially represented for the entire Spanish area (Rodríguez Martín et al. 2016). It is worth to notice the large variability in SOC stocks (32 – 241 Mg ha⁻¹) and clay content (6 – 30%) across the study area (Figs. S3 and S4).

Soil water content at saturation and field capacity conditions were deduced from FAO estimations considering soil properties related to soil texture (Raes 2017). Soil textural classes, used to estimate

soil moisture function under soil water saturation, were derived from the European Soil Data Centre (Ballabio et al. 2016).

Plant residues

Grassland associated to dairy cow production in Northern Spain are commonly based on grass-white clover swards; mainly ryegrass with about 5-10% of white clover (*Trifolium repens*. L.). We assumed 5% white clover in our simulated swards.

Using available records of estimated grass dry matter production (kg ha^{-1}) of municipalities in our study area (Doltra et al. 2019; Baizán et al. 2021), we generated a simple linear regression model using climate data (i.e., temperature and precipitation) as explanatory variables since climate is considered the most important driver for perennial pasture production. Results of dry matter production estimation from the linear regression varied between 6308 and 11363 $\text{kg ha}^{-1}\text{year}^{-1}$.

In order to estimate below-ground biomass from above-ground biomass values, we used a value of four for root to shoot (R:S) ratio, typical for temperate grasslands (Mokany et al. 2006). Regarding plant residues, it was assumed that 65% of above-ground biomass is harvested or consumed by dairy cows as average value of Soussana and Lemaire (2014) and Poeplau (2016) and only 50% of the remaining fraction (i.e., of 35%) is turned over annually and becomes available for soil organic matter formation as above-ground residue (Schneider et al. 2006). Similarly, 50% of below-ground biomass was assumed as below-ground residue as the annual root turnover is about 50% in the temperate zone (Gill and Jackson 2000). Regarding rhizodeposition estimation, we referred to a ratio, typical for grassland species, between net rhizodeposition and below-ground biomass of 0.5, as in Pausch and Kuzyakov (2018) review. Finally, we assumed a C concentration of 45% of the plant biomass (Kätterer et al. 2012).

Cows' diet

Given the non-availability of the different feeding systems at a more refined spatial scale, we referred to the regional main diet typologies according to Flores-Calvete et al. (2016) for the lactating dairy cows sub-category (Table S2) and to MAPA (2019) for the remaining dairy cows' sub-categories (i.e., heifers and dry dairy cows) (Tables S3 and S4). The common management strategy consists in heifers and dry cows grazing for most of the year (75%) while lactating cows are housed for most of the year (77-90%). The feeding regime of lactating dairy cows consists of mainly annual forage crops (e.g., maize silage) and concentrates (31-40%) (MAPA 2019) (Table S2). The different nutritive values were identified taking into account the ingredients offered for the different typologies. Crude protein varies between 17 and 19% for lactating cows while average digestibility is of 71%.

The feeding regime of dry dairy cows and heifers typical of our study area considering our simulation period consisted of lower concentrates percentage (25%) as illustrated in Tables S3 and S4. The crude protein reached only 13.5% for both dry dairy cows and heifers, while the digestibility 65% and 66% for dry dairy cows and heifers, respectively.

The Dry matter intake varied between 15 and 17 kg DM animal⁻¹ day⁻¹ for lactating dairy cows of the different typologies and was estimated of 7 and 8.6 kg DM animal⁻¹ day⁻¹ for dry cows and heifers, respectively.

Estimation of C inputs to the soil via C balance

Soil C inputs from manure included the excretion of grazing animals and the application of managed manure. The C flows from manure can be estimated, using a mass-balance approach, from the fraction of the diet DM consumed (C ingested animals) that is not digested, and thus excreted as faecal material and combined with urinary excretion.

$$\text{Carbon inputs from animal manure} = \text{C ingested}_{\text{animals}} - \text{C in milk}_{\text{animals}} - \text{C in body weight change}_{\text{animals}} - \text{C in CO}_2 \text{ resp}_{\text{animals}} - \text{C in CH}_4 \text{ enteric}_{\text{animals}} - \text{C in CH}_4 \text{ manure management} - \text{C in CO}_2 \text{ manure management}$$

C ingested_{animals}: equals the fraction of the diet consumed, which refers to the C that is contained in the dry matter intake estimated as IPCC (2019) methodology.

C in milk_{animals}, C in body weight change_{animals} and C in CO₂ resp_{animals}: equals the fraction of the digested fraction retained, which is used for milk production, for growth and animal respiration, respectively.

C in CH₄ enteric_{animals}: equals to the fraction emitted from animal enteric fermentation as indicated in Supplementary Information B (IPCC 2019).

C in CH₄ manure management: CH₄ emissions from management and grazing dairy cows were calculated annually as detailed in Supplementary Information B according to IPCC (2019).

C in CO₂ manure management: CO₂ emissions from manure management, derived from the ratio used by Pardo et al. (2017) from CH₄ manure emissions.

Manure C input per livestock unit was then multiplied by livestock units for each category per municipality and divided by average dairy cows holding area according to the Agricultural Census (INE 2009) to get tonnes of C excreted ha⁻¹ year⁻¹.

2.2.2 Spatial layer linkages

We referred to the municipalities with grasslands associated to dairy production, according to the National Statistical Institute (INE 2009), as spatial units. Monthly average climate data were assigned to the different spatial units according to their proximity to the meteorological stations, using GIS (ArcMap 10.2). Regarding soil properties, we obtained the statistic mean of SOC stocks and clay content for each municipality spatial unit (Rodríguez Martín et al. 2016) through ArcMap, in order to generate precise values of all contained pixels of each municipality. Soil textural classes were also extracted and ascribed to the different municipalities through ArcMap.

2.2.3 Uncertainty analysis: Monte Carlo simulation

A Monte Carlo simulation was used to assess the sensitivity of the results of SOC stocks to uncertainties in the estimation of certain parameters. This was done by constructing probability density functions (PDF) for the most relevant model parameters and input variables considered to be uncertain. As we aimed to explore the potential of management practices to increase SOC stocks, specific attention was paid to evaluate the influence of C input as the main driver of SOC sequestration (See Results section). Monte Carlo simulation was performed iteratively (1000 times) to sample random values for C inputs using normal distribution, with the aim to explore potential deviation for SOC stocks by combining plant residues and animal excreta. The approach used to define the uncertainty was as follows: We referred to plant dry matter production values as a proxy for plant residues; We selected a range of maximum and minimum values based on a sample of measured and reported data of dry matter related to the study area; A normal distribution around the mean value was assumed and the range of maximum and minimum values was assumed equal to the 95% confidence interval (Table S5). A similar approach was applied to estimate the PDF of the C inputs from animal excreta assuming a maximum and minimum value range and a standard deviation value (Table S5).

At a large geographical scale, Monte Carlo requires many model runs and enormous computational time. Thus, nine municipalities over our study area were selected for the uncertainty analysis, which were able to well represent the spatial distribution. Municipalities considered were close to meteorological stations to minimize uncertainties derived from climate data.

2.3 Greenhouse gas emissions

We used recent IPCC refined Tier 2 methodology to estimate direct GHG emissions (i.e., CH₄ and N₂O emissions) (IPCC 2019) and the latest European Monitoring and Evaluation Programme

(EMEP) methodology to estimate ammonia (NH₃) volatilization and nitrate (NO₃) leaching from manure storage and grassland soils at municipality level (EMEP 2019). Ammonia and NO₃ leaching are not GHG but they were considered as precursors of N₂O (indirect N₂O). The methodology relies on enhanced characterisation of animal population, assumed diet characteristics and manure management for the estimation of emissions. We multiplied the different emission factors by their correspondent number of each sub-category of dairy cows (i.e., lactating dairy cows, dry cows and heifers) for the different municipalities of our study area. The typologies which characterized the predominant practices of each region of our study area (details on grazing practices, dietary information and feed quality) according to animal type, physiological status, age, growth rate, activity level and production were drawn from (MAPA 2019 and Flores-Calvete et al. 2016). The explanation of the methodology used to estimate the CH₄ and N₂O emission factors is included in Supplementary Information B.

In order to aggregate the effect on the climate of the different forms of GHG we used the global warming potential metric for a 100-year time horizon (GWP100) based on latest values from IPCC (2014). The net emissions equivalent to CO₂ (CO₂-e) was calculated as a balance between the overall annual GHG CO₂-e fluxes calculated at the field and barn scale (CH₄ and N₂O) and the estimated long-term soil C gains (i.e., average annual SOC accumulation over 30 years) expressed as CO₂-e (Eq. 2):

$$GHG/yr (CO_2-e) = CO_2-eN_2O + CO_2-eCH_4 - CO_2-e_{CO_2}(SOC \text{ change}) \quad (2)$$

Where CO₂-eN₂O is the nitrous oxide emission and CO₂-eCH₄ is the methane emission calculated according to IPCC (2019) in Mg CO₂-e ha⁻¹ per year; eCO₂ is the multiplier between molar weights of CO₂, carbon (44/12); SOC change corresponds to the change in SOC stocks (Mg C ha⁻¹ year⁻¹).

3 Results and discussion

3.1 Regional changes in SOC stocks

3.1.1 SOC change rate

Modelled annual SOC change rate for grasslands in dairy cattle systems of Northern Spain municipalities showed an average of 0.16 Mg C ha⁻¹ year⁻¹ at 30 cm depth between 1981 and 2010 (Fig. 1), which is in the range of SOC change rates values found in other studies for moist temperate European grasslands (Ma et al. 2015). For example, in Belgium grasslands an average SOC change rate of 0.45 Mg C ha⁻¹ year⁻¹ was found for the period 1955-2005 (Goidts and van Wesemael 2007).

The different values found between our study and the Goidts and van Wesemael (2007) study may be explained by the higher manure application in Belgium grasslands (mainly during the first decades of the study period before regulating the manure application) compared with applications in Northern Spain region.

The range of SOC change rate values found (from -0.95 to 3.24 Mg C ha⁻¹ year⁻¹) is within the range of previous existing studies compiled from temperate grasslands both at regional and plot level (Table S7). Most municipalities (about 82%) showed SOC stock change rates between -0.5 and 0.66 Mg C ha⁻¹ year⁻¹ (Fig. 1) and less than 6% of municipalities presented SOC change rates higher than 1 Mg C ha⁻¹ year⁻¹ (Fig. 1). The little SOC change in most of the spatial units could be explained by the fact that grasslands were generally undisturbed and that SOC accumulation is dependent on C inputs (Horwath and Kuzyakov 2018).

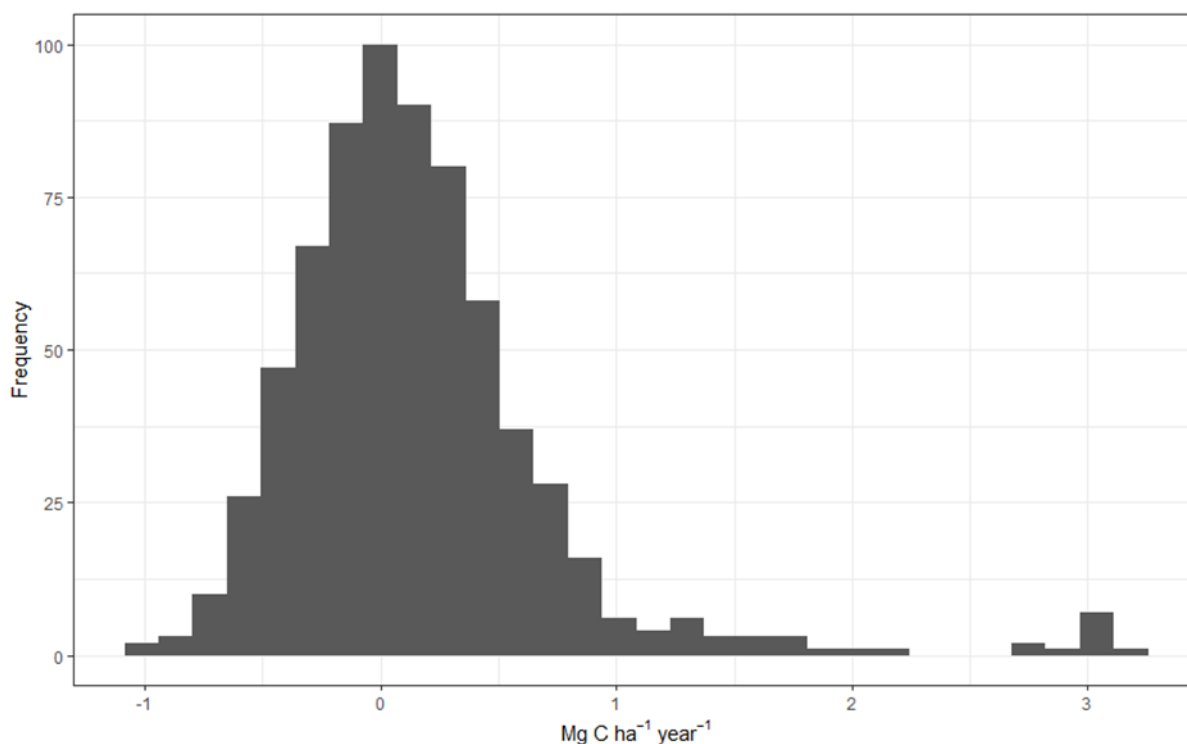


Fig. 1. Histogram of annual soil organic carbon change rate (in Mg C ha⁻¹ year⁻¹) for dairy production grasslands of Northern Spain municipalities

The highest rates of SOC change rates were observed in the grasslands located in the Southeastern part of the study area (Fig. 2). The climate in this region has a marked influence from the Mediterranean with a mean annual precipitation of 731 mm, which results in lower initial SOC stocks (49 Mg C ha⁻¹). Furthermore, production systems are characterised by intensive dairy farming with large C inputs derived from dairy cow excreta (up to 500 kg N ha⁻¹ year⁻¹). These factors resulted in high SOC change rates (Fig. 2). In contrast, the lowest SOC change rates were observed in areas with high mean annual precipitation (>1500 mm), initial high SOC stocks (171 - 223 Mg C ha⁻¹) and low

animal density with low animal excreta ($<30 \text{ kg N ha}^{-1} \text{ year}^{-1}$). At the same time, the model predicted SOC loss in certain grassland areas with initial SOC content higher than 91 Mg C ha^{-1} and low livestock densities (Fig. 2).

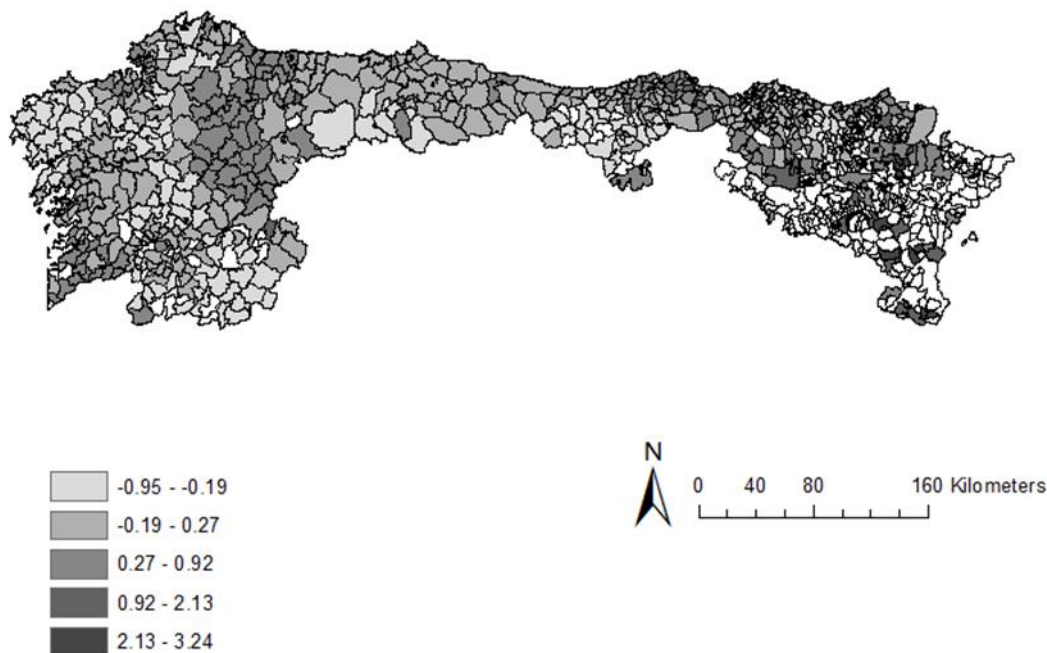


Fig. 2. Soil Organic Carbon stock change rates ($\text{Mg C ha}^{-1} \text{ year}^{-1}$) of dairy cows' grasslands in Northern Spain municipalities

3.1.2 Relationship between SOC change rate and different factors

The relationship between SOC change rate and different plant, climate and soil variables, as well as the interrelation among all variables, were analysed by stepwise linear regressions (Table 1) and correlation analyses (Table S8). The five variables analysed (C inputs, initial SOC, soil texture, mean annual temperature and mean annual precipitation) were significantly related with SOC change rate. Two variables showed the highest relationship and explanation of variance (about 81%) with the SOC change rate: the C inputs (positively related) and the initial SOC content (negatively correlated) (Tables 1 and S8), which is, in fact, in line with the findings from existing studies of long-term evolution of SOC stocks at regional scale in similar European grasslands (e.g., Bellamy et al. 2005; Saby et al. 2008; Goidts and van Wesemael 2007). The relationship between SOC change rate and the initial SOC content might illustrate the fact that soil organic matter dynamics tend to reach equilibrium (Goidts and van Wesemael 2007), since SOC accumulation capacity is limited. The clay content presented a significant positive weak correlation with the SOC change rate as in Goidts and

van Wesemael (2007) study (Table S8). This could be explained by the RothC structure which takes into account the clay component, which affects soil organic matter decay rates. The temperature and precipitation showed weak and negative correlation with SOC change rate in our study area. However, in our study we found that mean annual precipitation was positively correlated with initial SOC content similar to other studies performed in Northern Spain (Calvo De Anta et al. 2015). Furthermore, sites with high mean annual precipitation and high initial SOC tended to show lower SOC rates than sites with lower mean annual precipitation and low initial SOC levels (Meyer et al. 2016).

Table 1. Stepwise linear regression between annual SOC change rate (Mg C ha⁻¹ year⁻¹) and the different model input variables

	(1) SOC _r Coefficient	(2) SOC _r Coefficient	(3) SOC _r Coefficient	(4) SOC _r Coefficient	(5) SOC _r Coefficient
C input	0.3990 ^{***}	0.3506 ^{***}	0.3567 ^{***}	0.3722 ^{***}	0.3924 ^{***}
SOC_i		-0.0094 ^{***}	-0.0099 ^{***}	-0.0104 ^{***}	-0.0088 ^{***}
Clay			-0.0084 ^{***}	-0.0114 ^{***}	-0.0108 ^{***}
MAT				-0.1735 ^{***}	-0.1696 ^{***}
MAP					-0.0031 ^{***}
Constant	-1.7624 ^{***}	-0.3657 ^{***}	-0.1693 [*]	2.2728 ^{***}	2.2673 ^{***}
No. of Observations	690	690	690	690	690
R-Squared	0.580	0.808	0.818	0.913	0.935
F Statistic	949.437	1442.748	1031.095	1794.303	1954.323

*** p<0.01, ** p<0.05, * p<0.1

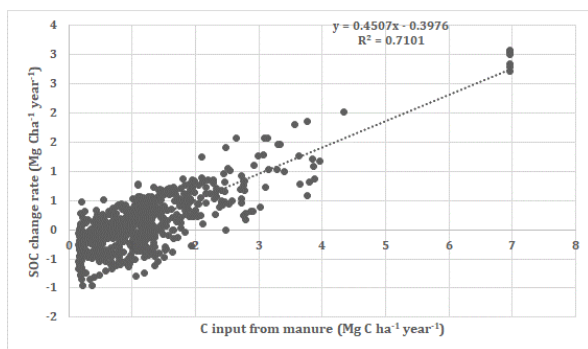
SOC_r is the annual SOC change rate (Mg C ha⁻¹ year⁻¹); SOC_i is the initial SOC content (Mg C ha⁻¹ year⁻¹); Clay is the soil clay percentage (%); C input is the C input derived from vegetation and animal manure (tC ha⁻¹ year⁻¹); MAT is mean annual temperature (°C) and MAP is annual precipitation (cm).

Overall, C input was the main controlling factor of SOC changes in Northern Spain dairy cattle grasslands, particularly C inputs derived from dairy manure, as they presented higher variability (0.16 and 6.96 Mg C ha⁻¹ year⁻¹) and proportionality to SOC change rate, compared with plant residues (a range value between 2.4 and 4.3 Mg C ha⁻¹ year⁻¹) (Fig. 3a). Our results were in agreement with Fornara et al. (2016) who identified the importance of manure application to increase SOC stocks in grassland systems at longer time scales. In our study, it was observed SOC accumulation, when dairy manure exceeds 0.88 Mg C ha⁻¹ year⁻¹ (Fig. 3a). Particularly, average SOC change rate would

decrease to $-0.3 \text{ Mg C ha}^{-1}$ in the absence of C inputs from dairy cows according to our simulation (Fig. S5).

Manure application rates were related with livestock density since we assumed that is not economically viable to export manure 30 km away from the municipality (Fealy and Schröder 2008) (Fig. 3b). Consequently, variations in livestock density directly affected manure rates and SOC stock changes as it has been shown in studies with grazing animals under different intensity (e.g., Mcsherry and Ritchie 2013). We found a threshold of livestock density of 0.95 LU ha^{-1} (Fig. 3b) (corresponding to the dairy manure amount of $0.88 \text{ Mg C ha}^{-1} \text{ year}^{-1}$), from which level there was always positive rates of SOC changes (Fig. 3a). Other studies have shown that this relationship is not linear and tends to get to a plateau or even an inverted u- shape trend (Ward et al. 2016). However, our study did not show this limiting threshold by manure input excess, which either is a limitation of the RothC model, or we did not reach to a saturated SOC level.

(a)



(b)

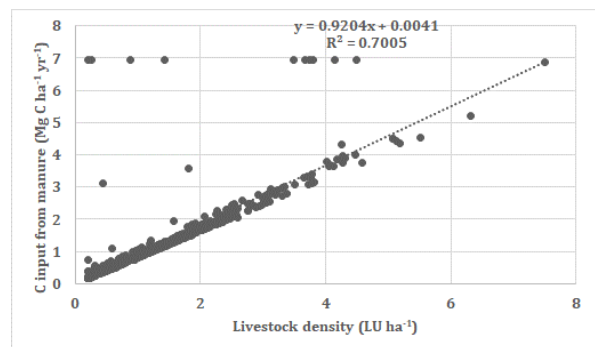


Fig. 3. (a) Soil Organic Carbon change rate in relation to C inputs from dairy manure for grasslands associated to dairy production in Northern Spain (b) C input derived from dairy manure in relation to livestock density for grasslands associated to dairy production in Northern Spain

3.2 Net GHG emissions expressed as CO₂-e

We found that average net GHG emissions associated with the cattle dairy system (i.e., grassland and barn level) excluding the pre-farm phases (e.g., feeds) and farm energy use of the study area was always positive. The estimated net GHG emission rates ranged from -9.8 to $22.4 \text{ Mg CO}_2\text{-e ha}^{-1} \text{ year}^{-1}$ (average value of $5.6 \text{ Mg CO}_2\text{-e ha}^{-1} \text{ year}^{-1}$) (Fig. 4a) and from -1.4 to $1.7 \text{ kg CO}_2\text{-e L Milk}^{-1} \text{ year}^{-1}$ (average value of $0.8 \text{ kg CO}_2\text{-e L Milk}^{-1} \text{ year}^{-1}$) (Fig. 4b).

Our average estimation of net GHG emissions per ha is within the range of some of the values reported for dairy grasslands under comparable temperate climate. However, published net GHG

values in these conditions are very diverse. For example, whereas Fornara et al. (2016) and Koncz et al. (2017) estimated a net GHG emissions per ha for dairy farming between 4.8 to 6.8 Mg CO₂-e ha⁻¹ year⁻¹ (Ireland) and 4.75±1.44 Mg CO₂-e ha⁻¹ year⁻¹ (Central–Eastern Europe), which is in line with our results, Del Prado et al. (2013) and Pirlo and Lolli (2019) estimated higher values of 7.8 and more than 8 Mg CO₂-e ha⁻¹ year⁻¹, in Northern Spain and Italy, respectively. On the other side of the net GHG values, our results were higher than Graux et al. (2012) who found a net GHG of 2.7–2.8 Mg CO₂-e ha⁻¹ year⁻¹ for French grassland-based dairy cattle systems. Chang et al. (2015) estimated the GHG balance for European grasslands using the process-based biogeochemical model ORCHIDEE-GM and found a net GHG sink. The study included both extensively and intensively managed grasslands by mowing and grazing regimes and did not account for the C export through milk products and liveweight gain which may partly explain the difference. The large differences between studies can be explained by the variety of production system across the different studies, as well as the different methodologies used for the estimation of the net GHG.

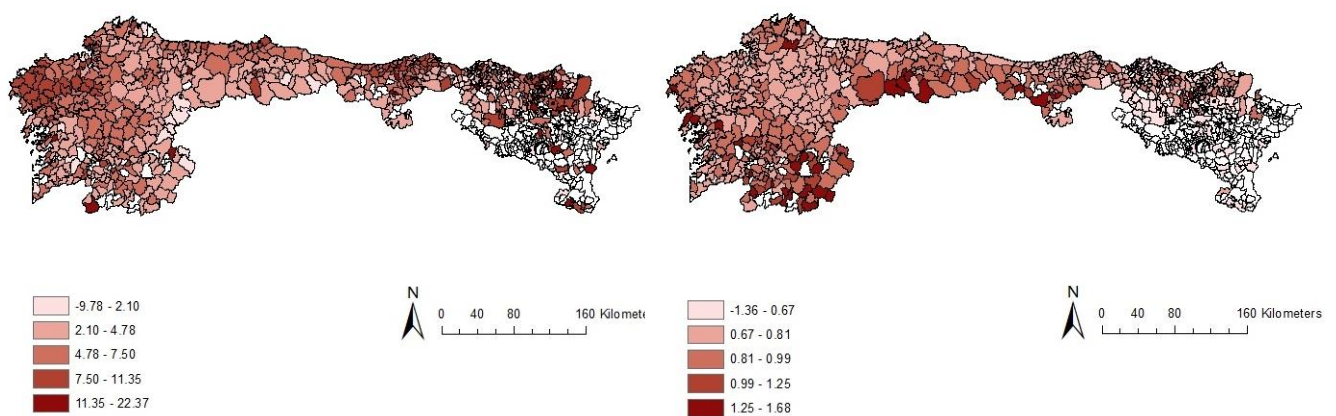


Fig. 4. Net GHG emissions per area in Mg CO₂-e ha⁻¹ year⁻¹ (a) and per litre of milk in kg CO₂-e year⁻¹ (b) per municipalities in Northern Spain

We estimated that SOC storage contributes to offset an average of 9% overall GHG emissions, which is in the lowest range established by Fornara et al. (2016). However, this result should be taken with caution, as part of emissions derived from the feed (i.e., concentrates and silage) was not considered in our assessment. This feed produced elsewhere may have come from cropping systems

that, not only may have emitted large non-CO₂ GHG emissions, but they also may have led to some SOC release, which may compensate this sink activity (Powlson et al. 2011).

Total GHG emissions varied between 1.1 and 34.3 Mg CO₂-e year⁻¹ per ha and between 0.6 and 1.3 kg CO₂-e year⁻¹ per litre of milk (Fig. 5). As expected, the largest share of emissions of the grassland-based dairy systems of Northern Spain were derived from the enteric fermentation (with an average of almost 60%). The second largest source of GHG emissions was CH₄ manure management (average of 18.6%) and N₂O soil emissions (average of 17.5%) came third.

Regarding N₂O soil emissions, the mean of our estimates among the different municipalities showed an average value of 2.04 kg N₂O-N ha⁻¹ year⁻¹ (corresponding to an average N input of 207 kg ha⁻¹).

The resulting methane Conversion Factor (MCF) values (CH₄ emitted per kg of volatile solid) for the different spatial units of our study were lower than that for locations under warm conditions specified in IPCC (2019) (from 15 to 22%).

Average estimated NH₃ volatilization and NO₃ leaching, which are precursors of N₂O, were 19.3kg N ha⁻¹ year⁻¹ (corresponding to 8 Mg CO₂-e ha⁻¹ year⁻¹) and 34 kg N kg ha⁻¹ year⁻¹ (corresponding to 14.2 Mg CO₂-e ha⁻¹ year⁻¹); respectively. NH₃ emissions presented 61% of total ammoniacal Nitrogen (TAN) from manure applied to the grasslands in the range of Sommer et al. (2019) review. The annual N leaching was also in the range of reported values in Lüscher et al. (2014) review for livestock- based grasslands with white clover in Europe (losses of 28–140 kg N ha⁻¹).

(a)

(b)

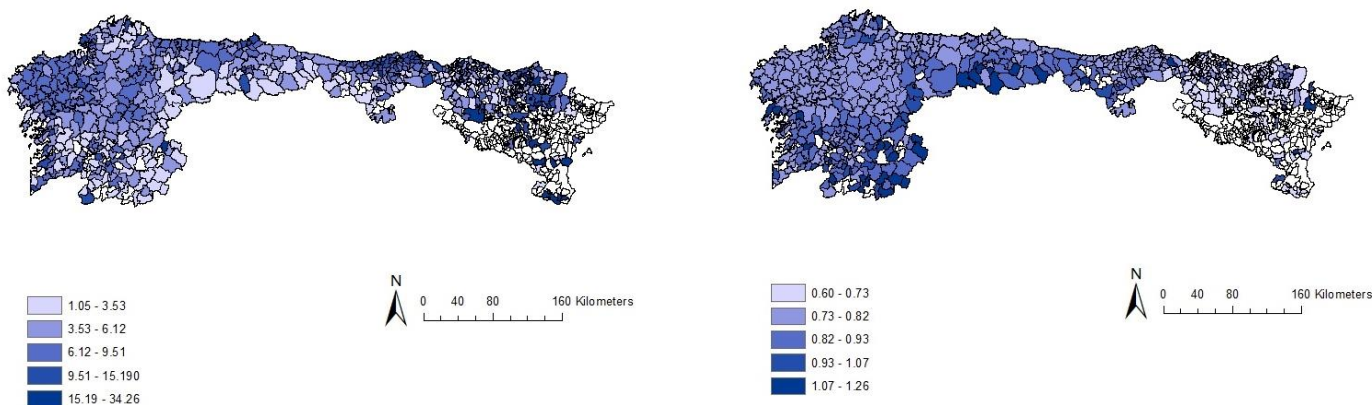


Fig. 5. GHG emissions per area in Mg CO₂-e ha⁻¹ year⁻¹ (a) and per litre of milk in kg CO₂-e year⁻¹ (b) per municipalities in Northern Spain

Relationship between GHG and different farm parameters

Relationships between GHG emissions and different farm parameters, related with management, productivity and diet typology, were assessed at the province level and as a function of litre of milk or ha (Fig. 6). GHG emissions when expressed per unit of area (ha) were different from those per L of milk (Fig. 6) as Salvador et al. (2017) study. This could be explained by the fact that expressing emissions per ha only does not appropriately reflect the effect different dairy systems can have on milk production (O'Brien et al. 2011).

As expected, milk production per animal proved to be the main factor affecting GHG emissions per litre of milk ($R^2 = -0.83$; $p < 0.003$) (Fig. S6) (Lorenz et al. 2019; Drews et al. 2020). Assumed feed quality also significantly affected GHG emissions, resulting in lower emissions per litre of milk in those areas with larger proportion of concentrate feed (e.g., concentrate feed: $R^2 = -0.58$; $p = 0.08$) (Fig.S6) (Morais et al. 2018). Therefore, total GHG emissions per litre of milk unit favoured intensive systems in terms of milk production, livestock density and feeding quality (Fig. S6). Our approach to evaluate the climate impact of different farm intensities, nevertheless, has several limitations since lower enteric CH₄ emissions from more intensive livestock would generally be offset, at least in some degree, with greater C footprints of imported feed or larger use of on-farm energy (electricity and gasoil). Moreover, some of the SOC stored in more intensive livestock densities would not be mitigating climate change as it may be led by C inputs from imported feed, indirectly (Powlson et al. 2011). These elements, feed C footprint, energy use in the farm and the built-up of SOC due to external C inputs via feed that has contributed to SOC depletion elsewhere were not included in the study.

On the other hand, total GHG emissions per ha favoured extensive production systems, according to the correlation results with feed quality (i.e., level of concentrate in the diet) and livestock density (Fig. S6). Indeed, the higher the livestock density the greater the estimated GHG per ha in our study ($R^2 = 0.88$; $p = 0.009$) (Fig. S6). This contrast with some studies, e.g. under tropical conditions, which favoured climatic factors effect (Ruggieri et al. 2020) over the livestock density. In general, GHG emissions and SOC change rate for the simulated spatial units were conditioned by livestock density (Fig. 6a). The distribution of net GHG per ha was almost proportional to the distribution of SOC change rate (Figs. 2 and 4a). Only few municipalities presented an opposite trend of net GHG (i.e., negative values) and high SOC change rate. These municipalities presented low livestock densities and thus lower CH₄ enteric emissions, being influenced primarily by livestock density (Liebig et al. 2010; Schönbach et al. 2012), while receiving higher C inputs of dairy manure from nearest municipalities. Therefore, increasing livestock density and thus C input as management choice to improve C accumulation may increase GHG emissions (Fig. 6a) (Soussana et al. 2010). In

this sense, practices intended to offset GHG emissions using C sequestration must therefore consider impacts on other GHGs like N₂O and CH₄ (Graux et al. 2012). For instance, the effect of different livestock densities can be explored over a wide variety of cattle feed intakes and for alternative manure management options (Graux et al. 2012).

With our analysed data we can establish a potential livestock density (approximately > 0.4 LU ha⁻¹), from which the net GHG emissions (expressed as CO₂-e per unit of LU) would be smallest (Fig. 6b). This result contrasts with some studies, e.g., grasslands under semiarid continental climate, where moderate livestock densities (i.e., 0.39 animal ha⁻¹), lead to the smallest GHG emissions levels (Liebig et al. 2010).

(a)

(b)

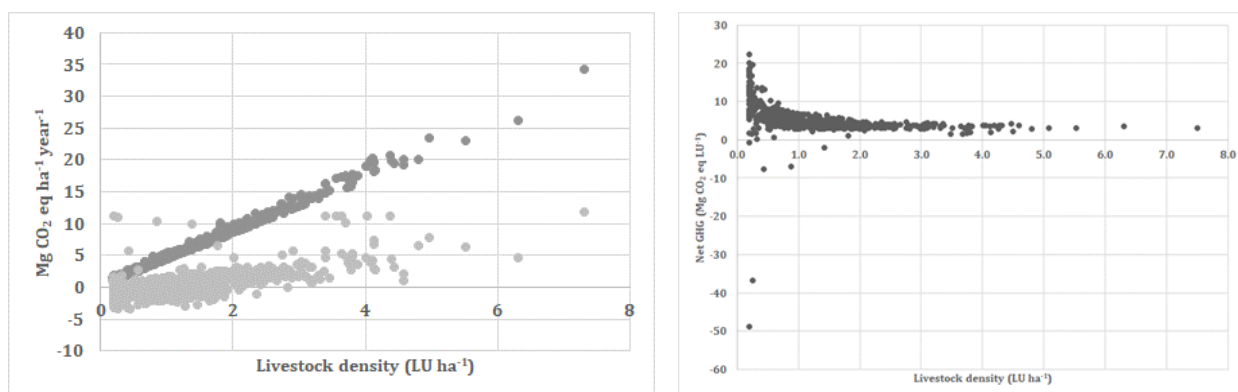


Fig. 6. Relationship of livestock density with SOC change rate (light grey) and total GHG emissions (dark grey) (a), Net GHG emission per livestock unit in relation to livestock density (b)

We show the importance that livestock density as a management tool can have on the environmental sustainability of grasslands through its impact on net GHG emissions as in McGinn et al. (2014). It is also crucial to point out the importance of concentrate reduction referring to the diet quality. The concentrate level of the diet related to intensification of dairy production could lead to significant carbon leakage not captured in our estimation (e.g., land use change...) which correspond to higher emissions (Styles et al. 2018). Moreover, although beyond the scope of our study, concentrate reduction could imply a milk production decline but at a low rate per saved kilogram of concentrate (Leiber et al. 2017).

3.3 Sources of uncertainty

In our study, C inputs derived from animal excreta and from plant residues were identified as main driver for SOC change. In order to quantify the uncertainty in C inputs, a Monte Carlo approach

was conducted to estimate SOC change (30 years) for selected municipalities. The mean of the range of possible SOC stocks depending on the uncertainty (and variations) of the C inputs was close to our simulated values (Table S6). Therefore, our findings of SOC accumulation could be interpreted as a good indicator of possible SOC storage in our study area.

Regarding GHG emissions, emissions from calves were not estimated as their contribution to GHG emissions is the lowest (Mc Geough et al. 2012). Moreover, our study did not account for energy requirements or extra-costs in terms of GHG of the future products. Furthermore, N₂O emission factor calculations were based on IPCC Tier 2 simplified equations, although the component processes of nitrification and denitrification are highly complex and depend on several soil and environmental factors (Farquharson and Baldock 2008).

In general, despite limitations, our findings could be interpreted as a valuable indicator of net GHG emissions of grasslands associated to dairy production in our study area.

4 Conclusions

This work is the first modelling study of net GHG at regional scale for grasslands associated to dairy production in moist temperate Spain. We found a positive contribution of these grassland systems to global warming. Soil organic carbon was able to offset 9% of GHG emissions. However, this mitigation potential could be smaller when total GHG emissions, including the pre-farm phases such as feeds and farm energy use, are estimated. Our study confirms the importance of dairy cows to preserve and enhance SOC stocks. Livestock density was the main factor affecting net GHG emissions associated to grassland sites of dairy production in Northern Spain. Therefore, we recommend to “optimise” the livestock density in order to offset GHG emissions and enhance SOC accumulation. Regarding feed quality, our findings favour a reduction in the level of concentrates and thus an extensification of dairy production systems as it contributes to higher GHG emissions per ha and probably per milk production units when emissions from imported feed are accounted for. Finally, our study illustrated the importance of considering all GHG emissions as well as the interaction between C and N cycles. This is crucial in order to study livestock density effect, as a management practice for both SOC accumulation and GHG reduction, over a wide variety of cattle feed intakes and alternative manure management options.

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**CHAPTER 4: POTENTIAL IMPACTS OF CLIMATE CHANGE ON SOIL
ORGANIC CARBON AND NET GHG EMISSIONS OF GRASSLANDS
ASSOCIATED TO DAIRY PRODUCTION IN NORTHERN SPAIN**

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Abstract

Climate change is projected to significantly affect soil organic carbon (SOC) dynamics and probably greenhouse gas (GHG) emissions derived from livestock production systems. The ability of soil grasslands to store carbon (C) as the climate changes is uncertain. Moreover, there is a need to project GHG emissions under climate change scenarios to get the net greenhouse gas emissions (net GHG) while taking SOC sequestration in consideration.

In this modelling study, we adopted a modified version of RothC model to simulate future (2010–2100) SOC and IPCC refined guidelines to estimate GHG emissions in more than 405 000 ha of moist temperate Spanish grasslands associated to dairy production. We assessed the climate change effect on the net GHG emissions under different climate scenarios (baseline, RCP 4.5 and RCP 8.5) and the potential of alternative manure management practices to reduce the climate change effect. The results showed a decrease in average SOC and an increase in GHG emissions under both climate change scenarios compared with the baseline reference. The greatest losses of SOC stocks and increases of GHG emissions were found mainly under the RCP 8.5 climate change scenario. Effects of changes in temperature and precipitation showed that, for SOC, the temperature effect predominates over the precipitation effect. The average net GHG emissions associated with the dairy farm system of Northern Spain was then positive contributing to the global warming potential with average value of 5.8 and 6.2 Mg CO₂-e ha⁻¹ year⁻¹, under the RCP 4.5 and RCP 8.5, respectively.

Under climate change conditions, implementing specific manure management practices, namely the anaerobic digestion, were predicted to mitigate the climate change impacts and reduce the net GHG for the grassland systems associated to dairy production in Northern Spain.

1 Introduction

Grassland-based dairy cattle farming is one of the main land uses in the Northern moist temperate Spain (Galicia, Asturias, Cantabria, Basque Country and Navarra), comprising about 1.2 million ha of permanent grasslands. Frequent rainfall and cool temperature conditions are key factors that promote plant growth and production and make grasslands of this region be very productive (Smit et al. 2008). Accordingly, they account for 60% of milk production in Spain, about 2.7% of the milk produced in the European Union (EU-28) (EUROSTAT 2017) and have an economic value of 1300 million euros (MAPA 2016).

Grasslands in Northern moist temperate Spain present a significant potential for soil organic carbon (SOC) storage compared to other land uses (Ganuza and Almendros 2003). However, the C

footprint of the grassland-based dairy production in Northern Spain under current climatic conditions, which was assessed by several studies using mainly Life Cycle Analysis, has shown that milk production has net GHG emissions associated (Del Prado et al. 2013; Laca et al. 2020). In general, dairy production systems are important sources of direct GHG from enteric fermentation (CH_4), manure storage and handling (CH_4 and N_2O), and crop and pasture land (mainly N_2O) (Gerber et al. 2013). A large proportion of total GHG emissions was associated with CH_4 output (27%–49%) from dairy farms in Northern Spain, where a large percentage comes from manure management after enteric fermentation (Del Prado et al. 2013). The intensification of dairy production systems favoured potential CH_4 and N_2O emissions originated from slurry management (Petersen 2018). Dairy cows (including lactating dairy cows, dry dairy cows, heifers and calves) are projected to produce by 2025 around 30% of total agriculture greenhouse gas (GHG) emissions in the EU-28 according to projections carried out with the CAPRI model (European Commission 2015).

Global climate models project the global mean temperature to increase by 1 to 5.7°C for the year 2100, compared to 1850-1990 period (IPCC 2021). Based on climate predictions, pluvial flooding will increase in Northern Europe and hydrological and agricultural/ecological droughts in the Mediterranean (IPCC 2021). Climate parameters have been proven to play a crucial role in the soil mechanisms controlling SOC decomposition (Paul 1984; Conant et al. 2008). In this context, the response of SOC contents to climate change has been widely investigated at different scales and both positive (e.g., Álvaro-Fuentes et al. 2012) and negative responses (e.g., Smith et al. 2005) have been reported.

In temperate grasslands, climate change might have a significant impact on net greenhouse gas exchange and potential climate feedbacks (Eze et al. 2018). For instance, CH_4 emissions from manure management increase exponentially with increasing temperatures (IPCC 2019). Consequently, more efforts should be implemented on manure management to reduce potential climate change effects on GHG emissions from dairy production systems. However, due to system interactions, mitigation practices that reduce emissions in one stage of the manure management process could increase emissions elsewhere (Montes et al. 2013). For this purpose, simulation models may be suitable tools. At the end, ecosystem models are frequently the only available tool to study climate change related issues and other long-term effects (FAO 2010). Indeed, modelling long-term SOC changes is essential to assess future CO_2 emission patterns and the evaluation of management options for GHG mitigation (Adhikari et al. 2019). There are several studies in which SOC changes in grassland systems under climate change conditions have been simulated at different spatial scales and using different simulation tools, for example RothC (e.g., Wiesmeier et al. 2016; Xu et al. 2011) and Century (e.g., Lugato et al. 2014; Álvaro-Fuentes et al. 2012). However, in moist temperate systems, the

contribution of grassland-based livestock systems to global warming under climate change has been less studied. Graux et al. (2012) studied the potential net GHG emissions for different grassland-based dairy livestock systems in France and showed different trends depending on the management.

To our knowledge, this is the first attempt to predict the impact of climate change on net GHG emissions at a subnational level in dairy grassland systems of Northern Spain. Our main objective was to provide emission/sink estimates of the three major GHG (methane, CH₄; nitrous oxide, N₂O; and carbon dioxide, CO₂) at a regional scale and under different projected future climate scenarios. Once this objective was completed, different alternative manure management options to mitigate climate change effects were evaluated. To achieve the objectives, we used a modified version of RothC (Jebari et al. 2021), adapted to managed grasslands under moist temperate climatic conditions, to estimate SOC dynamics. Furthermore, the recent IPCC refined Tier 2 methodology was used to estimate direct GHG emissions (i.e., CH₄ and N₂O emissions) (IPCC 2019) and the latest European Monitoring and Evaluation Programme (EMEP) methodology to estimate ammonia (NH₃) volatilization and nitrate (NO₃) leaching from manure storage and grassland soils (EMEP 2019). Finally, we applied medium-low and high representative concentration pathways (RCP) climate scenarios (medium-low RCP 4.5; high RCP 8.5) and a baseline scenario specifically built for the conditions of Northern Spain.

2 Materials and methods

2.1 Study area

The studied region comprises the grasslands associated to dairy production in North Spain, with a total surface of 405,000 ha (Fig. S1). The climate is mainly moist temperate, with annual mean rainfall ranging from 800 to 3000 mm and air temperature of about 12-14°C per year.

Grassland ecosystems of dairy production in Northern Spain are commonly based on grass (mainly ryegrass) with around 5% of white clover (*Trifolium repens* L.) swards. The common management strategy consists in grazing for most of the year (75%) of heifers and dry cows while confining of the lactating cows for most of the year (77-90%), feeding them both annual forage crops (often grass or maize silage) and concentrates (31 – 40%) (MAPA 2019). Urine and faeces of lactating dairy cows are generally excreted in the stable. This manure is stored as liquid (slurry) in tanks or lagoons. However, excreta from dry dairy cows and heifers are generally mixed with straw and other bedding and handled as solid manure (farmyard manure, FYM). All details about the study area and dairy systems characterisation could be found in Chapter 3.

2.2 SOC change

We used a modified version of the RothC model (Coleman and Jenkinson 1996) adapted to managed grasslands under moist temperate conditions to estimate SOC change (from 2010 to 2100) at a municipality spatial unit for grasslands associated to dairy production of our study area. The main modifications of the model version used are explained in detail in Jebari et al. (2021).

The RothC -26.3 (Coleman and Jenkinson 1996) model divides the SOC into five fractions, four of them are active and one is inert (i.e., inert organic matter, IOM). The active pools are: decomposable plant material (DPM), resistant plant material (RPM), microbial biomass (BIO) and humified organic matter (HUM). The decomposition of each pool (except IOM) is governed by first-order kinetics, characterized by its own turnover rate constant and modified by environmental factors related to air temperature, soil moisture and vegetation cover, which are the main input parameters to run the model. Incoming plant C is split between DPM and RPM, depending on the DPM:RPM ratio of the particular incoming plant material or organic residue. Both of them decompose to produce BIO, HUM and evolved CO₂. The proportion that goes to CO₂ and to BIO + HUM is determined by the clay content of the soil, which is another input to the model. The model uses a monthly time step to calculate total SOC and its different pools changes on years to centuries time scale. As commented before, we used a modified RothC version, adapted to livestock-based grasslands under moist temperate climatic conditions. The main modifications consisted in (i) considering plant residues components and its quality variability across the year, (ii) established entry pools that account for the ruminant excreta as a specified exogenous organic matter and (iii) water contents up to saturation in the soil water function. More detailed description of the different modifications could be found in Jebari et al. (2021).

For RothC initialization, C pools were estimated according to Weihermüller et al. (2013), based on clay content obtained from Rodríguez Martín et al. (2016) and SOC stocks for the year 2010 obtained from simulation results of a previous paper of the same authors. The initial IOM pool was set to match the equation proposed by Falloon et al. (1998) (Eq. 1):

$$IOM = 0.049 SOC^{1.139} \quad (1)$$

We developed a VBA (Visual Basic for Applications)-based program in Excel to simulate changes in SOC stocks simultaneously for the different spatial units (i.e, municipalities) for the period of 2010 to 2100 due to the combination of a large number of runs for each spatial unit of the regional simulation.

2.3 Input datasets

2.3.1 Soil properties

Soil texture at 30 cm depth was provided spatially (as a raster layer) from Rodríguez Martín et al. (2016). Our study area presented a large variability in clay content (6 – 30%). The statistic mean of clay content for each municipality was obtained through ArcMap 10. 2. Initial SOC stocks were extracted for each municipality from a previous work of the same authors in which SOC stocks were simulated in the same study area and for the period 1981-2010, using the modified version of RothC adapted to managed grasslands under moist temperate conditions (Jebari et al. 2021).

Soil water content at saturation and field capacity conditions were deduced from FAO estimations considering soil properties related to soil texture (Raes et al. 2017). Soil textural classes, used to estimate soil moisture function under soil water saturation conditions, were derived from the European Soil Data Centre (Ballabio et al. 2016). They were extracted and ascribed to the different municipalities through ArcMap.

2.3.2 C input derived from plant residues and animal manure

Climate change is known to affect the plant growth and production through the interaction of different factors (i.e., temperature rise, precipitation change and atmospheric CO₂ enrichment) (Gamage et al. 2018) which, in turn, it is influenced by management practices (Petersen et al. 2021). In agricultural soils, several studies have assumed C input increases under climate change (e.g., Smith et al. 2005; Graux et al. 2012). However, this last assumption might be rather optimistic given rising evidence for negative effects of climate change on plant growth (Wiesmeier et al. 2016). Therefore, the possibility of stagnation or even the reduction of C inputs should be considered in SOC projections (Wiesmeier et al. 2016). Although climate change would affect the Atlantic zone of Northern Spain (with higher temperature and CO₂ concentration and lower water availability) according to regional data of our study area, the impact on grassland productivity was rather uncertain and was not quantified. Given the negligible effect of climate change on plant production in the Atlantic region of Europe (Dellar et al. 2018) and for simplicity, we considered that the effect of climate change on plant production and soil management was non-existent.

We used the same assumptions to estimate C input derived from plant residues and the same mass-balance approach (by subtracting gross C production from total C ingestion by livestock) to predict C input derived from animal manure as in our previous work (See Chapter 3). Urine and faeces of lactating dairy cows are generally excreted in the stable as liquid manure (slurry). However, excreta

from dry dairy cows and heifers are stored as solid manure. For simplicity purposes, we did not consider inputs from feeding waste and bedding materials and we did not consider any change in the manure amount.

2.4 Greenhouse gas emissions

We estimated both direct emissions (i.e., CH₄ and N₂O emissions) and indirect emissions (i.e., precursors of N₂O: ammonia (NH₃) volatilization and nitrate (NO₃) leaching from manure storage and grassland soils) for grassland-based dairy cattle in Northern Spain at a municipality level. Direct GHG emissions were estimated using recent IPCC refined Tier 2 methodology (IPCC 2019) and indirect emissions were estimated according to the latest European Monitoring and Evaluation Programme (EMEP) methodology (EMEP 2019). In order to estimate the total emissions per spatial unit (i.e., municipality), we multiplied the different emission factors by their correspondent number of each sub-category of dairy cows (i.e., lactating dairy cows, dry cows and heifers) for each spatial unit of our study area. The data utilized were obtained from several existing datasets and reports (MAPA 2019 and Flores-Calvete et al. 2016), according to the typologies which characterized the predominant practices of each region of our study area (details on grazing practices, dietary information and feed quality).

In order to aggregate the effect on the climate of the different forms of GHG we used the global warming potential metric for a 100-year time horizon (GWP100) based on the IPCC fifth assessment report (IPCC 2014). The net emissions equivalent to CO₂ (CO₂-e) was calculated as a balance between the overall GHG CO₂-e fluxes estimated at the field and barn scale (CH₄ and N₂O) and the estimated long-term soil C gains (i.e., SOC accumulation) expressed as CO₂-e (Eq. 2):

$$Net\ GHG/yr\ (CO_2-e) = CO_2-eN_2O + CO_2-eCH_4 - CO_2-e_{CO_2}(SOC\ change)\ (2)$$

Where CO₂-eN₂O is the nitrous oxide emission and CO₂-eCH₄ is the methane emission calculated according to IPCC (2019) in Mg CO₂-e ha⁻¹ per year; eCO₂ is the multiplier between molar weights of CO₂, carbon (44/12); SOC change corresponds to the change in SOC stocks (Mg C ha⁻¹ year⁻¹).

2.4.1 Methane emissions

CH₄ derived from enteric fermentation

We implemented Tier 2 refined methodology to estimate enteric fermentation as in equation (3). The methane conversion factor (Y_m) was estimated according to the feeding typology reflected for each dairy cows' sub-category depending on neutral detergent fiber (NDF) and digestibility of the annual feed ration. Gross energy intake was calculated as outlined in the IPCC Tier 2 methods (IPCC

2019) according to the feeding typology for each dairy cows' sub-category. Then, the emission factor was multiplied by the associated dairy cows' sub-category number for each municipality of our study area.

$$EF = \frac{GE \cdot Ym}{55.65} \quad (3)$$

Where EF: emission factor (kg CH₄ head⁻¹ year⁻¹); GE: gross energy intake (MJ head⁻¹ year⁻¹); Ym: methane conversion factor (% of GE in feed converted to methane); 55.65: energy content of methane.

CH₄ emissions derived from manure management

As manure is managed in multiple systems considering municipalities of our study area, manure EF were allocated to the dominant storage systems (i.e., manure of lactating dairy cows is stored as slurry with natural crust, while manure of dry dairy cows and heifers is handled as solid storage). Indeed, emissions from manure management depend not only on management system characteristics but also on manure characteristics (i.e., Volatile Solids (VS)), which were estimated based on feed intake and digestibility, used to estimate enteric fermentation EF (Eq. 3). In order to determine the Methane conversion factor (MCF) for slurry, we referred to the IPCC suggested model. The MCF model requires monthly air temperature profiles as well as the average number and timing of the emptying of manure storages. The VS and maximum methane producing capacity for residues, based on IPCC guidance and percentage of excreted VS handled as a liquid are also additional input parameters. The model calculations run for three years, in order to ensure VS available has stabilized on an annual basis. In our study, the MCF model was run for the different municipalities of our study area. The average value for each municipality was then multiplied by VS to get CH₄ emissions from manure management (Eq. 4).

$$EF = VS \cdot \left[0.24 \cdot 0.67 \cdot \left(\frac{MCF}{100} \right) \cdot ARMS \right] \quad (4)$$

Where EF: annual CH₄ emission factor for dairy cows (Kg CH₄ dairy cow⁻¹ year⁻¹); VS: volatile solid excreted for dairy cows (Kg dry matter dairy cow⁻¹ year⁻¹); 0.24: maximum methane producing capacity for residues produced by lactating dairy cow, it is of 0.18 for heifers and dry cows (m³ CH₄ kg⁻¹ of VS excreted); 0.67: conversion factor of m³ CH₄ to kg CH₄; MCF: methane conversion factor for each residue management system (%); ARMS: fraction of dairy cow's residues handled using animal excreta management system.

2.4.2 Nitrous oxide emissions

We used Tier 2 of IPCC (2019) methodology to estimate N₂O emissions produced, directly and indirectly, during the storage and treatment of manure as well as direct and indirect soil N₂O emissions (derived from animal excreta, applied fertilisers and deposited dung and urine from grazing dairy cows to the pastures, crop residues and pasture renewal). Reported N₂O emissions are generated using N excretion results and emission factors for N₂O emissions, as well as volatilization and leaching factors; with total related N₂O emissions equalling the sum of direct and indirect emissions.

Management information of fertilization and amounts of N fertilizers were obtained from expert knowledge of most common local dairy farmer practices.

2.5 Climate change scenarios

We simulated SOC dynamics and GHG emissions of dairy grasslands in Northern Spain for the period of 2010 to 2100 under two climate change scenarios and one baseline reference scenario. The climate change scenarios correspond to RCP 4.5 and RCP 8.5. The RCP 4.5 scenario represents a medium-low emission scenario with stabilization of CO₂ emissions from 2050 onwards. The RCP 8.5 scenario represents a high emission scenario with stabilizing CO₂ emissions post-2100 (Meinshausen et al. 2011). These two scenarios have been used widely to evaluate the potential impact of climate change on the environment (e.g., Di Vittorio et al. 2014; Li et al. 2015). The baseline scenario consisted of historical average monthly temperature and precipitation data of several decades. The climate data correspond to 12.5 km grids and were produced by the Spanish Meteorological State Agency using a regional downscaling under the project CORDEX (AEMET 2017) and with climate data obtained from the global climate model HadGEM2 and the regional circulation model CCLM 4.8.17 (Kotlarski et al. 2014 ; Casanueva et al. 2016).

We estimated average monthly temperature and precipitation for each decade and each municipality under the different climate scenarios. Whereas potential evapotranspiration for each decade from 2010 to 2100 was estimated according to Thornthwaite equations (Thornthwaite 1948), using average decadal climate data for all climate scenarios.

Compared with the baseline reference, the average annual temperature under RCP 4.5 and RCP 8.5 increased by 1.5 and 1.75 °C, respectively, until 2050 (Table 1). During the period 2050-2100, it increased even further by 2.8 and 4.2 °C, respectively (Table 1). However, average annual precipitation showed a decrease under both climate change scenarios (RCP 4.5 and RCP 8.5) at the end of the simulation period 2100 by 126 and 254 mm under RCP 4.5 and RCP 8.5, respectively.

Table 1. Projected climate changes (mean annual precipitation and air temperature) under the climate change

scenarios (RCP 4.5 and RCP 8.5) over 2050 and 2100 compared with the corresponding values in the baseline data of the study area

Climate scenario	Time period	Mean	annual	Mean	annual
		precipitation (mm)		temperature (°C)	
Baseline	2010 - 2050	1161.81		11.94	
	2050 - 2100	1161.81		11.94	
RCP 4.5	2010 - 2050	1184.62		13.42	
	2050 - 2100	1035.86		14.72	
RCP 8.5	2010 - 2050	1142		13.69	
	2050 - 2100	907.57		16.17	

The changes in the decadal distribution of climate data of temperature and precipitation are shown in Fig. 1. Temperature showed the same pattern for both climate change scenarios with a significant increase for the last five decades under both climate change scenarios specially RCP 8.5 (with an increase of 35.4% compared with 23.6% under RCP 4.5) compared with the baseline reference (Fig. 1). However, the changes in average monthly decadal precipitation were often substantial and showed considerable variation between all climate scenarios. Moreover, both climate change scenarios showed lower precipitation at the end of the simulation period by 18 and 34% under RCP 4.5 and RCP 8.5, respectively (Fig. 1 and Table 1).

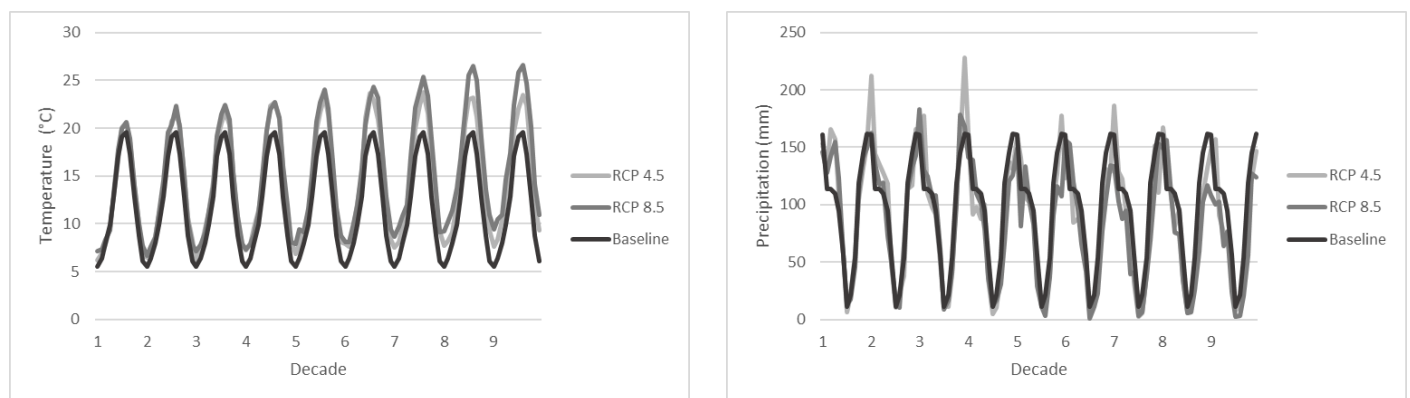


Fig. 1. Future projections of average decadal temperature (on the left) and average decadal precipitation variation (on the right) for moist temperate Spanish region under the climate change scenarios (RCP 4.5 and RCP 8.5) and the reference baseline scenario

2.6 Manure related management scenarios

A total of four main manure management scenarios were generated to assess their climate change mitigation potential. A first reference scenario was produced in which it was assumed a natural crust for the slurry storage system. This slurry is removed and applied to grasslands all year long except in summer. This reference scenario was compared with the next alternative GHG mitigation scenarios: (i) the presence of a cover (i.e., a rigid structure that covers the slurry, impermeable to water, and gasses) for the slurry storage, (ii) the removal of the slurry during different seasons of the year and (iii) anaerobic digestion (AD).

2.6.1 Cover for slurry storage

Covers are a potential mitigation measure that can be implemented on liquid manure storage facility and they are produced with materials of natural origin (e.g. clay aggregates), synthetic origin (e.g. plastic, and rubber), and composites of both (VanderZaag et al. 2008). Compared to uncovered conditions, nearly all cover types have been capable of substantially reducing NH_3 emissions (Berg et al. 2006).

We implemented then the scenario of a rigid cover for the slurry storage system to reduce total N_2O emissions derived from manure storage. In this context, we used IPCC (2019) and EMEP (2019) equations for the cover manure management.

2.6.2 Slurry removal

A direct way to avoid GHG emissions is to reduce the time manure is stored. Indeed, frequent manure application reduces GHG emissions from storage due to the shorter retention times and the reduced surface area (Aguirre-Villegas and Larson 2017). We considered scenarios of avoiding manure application whether in Autumn or in Winter together with the reference scenario (i.e., avoiding manure removal in Summer). However, avoiding manure application in Spring is not relevant, as grasslands need to be fertilized during Spring season. Moreover, emptying a manure tank in the Spring before warmer temperatures begin presents another opportunity to reduce GHG emissions in slurry storage systems (Novak and Fiorelli 2009).

Methane Conversion Factor model, using IPCC refined method, was useful for evaluating those scenarios with different retention times over the year, while taking into account the monthly temperature under the baseline and the climate change scenarios.

2.6.3 Anaerobic digestion

The anaerobic digestion is a naturally occurring process in which microbial organisms break down organic materials (i.e., manure) in the absence of oxygen to produce biogas, which is primarily a mix of methane (CH₄) and carbon dioxide (CO₂). Biogas can be combusted to produce electricity or thermal energy for heating applications, upgraded to be injected into a natural gas pipeline, or compressed to be used as transportation fuel. The remaining fraction after digestion (known as digestate) can be used as fertilizer as it maintains the nutrient contents of the initial feedstock. Manure processing as AD help mitigating GHG emissions from manure and energy (Aguirre-Villegas and Larson 2017). Emission reductions from energy come from the displaced emissions that biogas-based electricity has when replacing grid electricity (Ebner et al. 2015). Reductions from manure are mostly from the capture of CH₄ during digestion which is then converted to CO₂ during combustion, as well as the reduction of carbon available to produce CH₄ in storage (Aguirre-Villegas and Larson 2017).

During the AD process, there is a fraction of C that is released to the atmosphere, instead of being applied to the soil (Pardo et al. 2017). In this context, a distinction between emissions from manure in barns and outside storage facilities is important for assessing effects of AD, where mainly posttreatment emissions are affected (Peterson et al. 2018). Therefore, we included specific C and N cycling effects of AD process using Pardo et al. (2017) methodology, as they are usually neglected (Meier et al. 2015).

3 Results and discussion

3.1 SOC change

The trends in SOC over the simulation period are shown as the annual change rate of SOC (Mg C ha⁻¹, 0-30 cm) per municipality spatial unit. In average, the model predicted that climate impacts (under climate change conditions) on dairy grasslands soils tend to decrease SOC stocks in Northern Spain. During the two-time horizons (2050, 2100), the baseline reference scenario, showed median SOC change rates of 0.42 and 0.25 Mg C ha⁻¹ year⁻¹ (Fig. 2). Both climate change scenarios RCP 4.5 and RCP 8.5 showed a close median SOC change rate of 0.22 and 0.19 Mg C ha⁻¹ year⁻¹ until 2050, respectively (Fig. 2). However, the median SOC change rate was lower by 2100, reaching 0.007 Mg C ha⁻¹ year⁻¹ under RCP 4.5 and even a loss of -0.05 Mg C ha⁻¹ year⁻¹ under RCP 8.5 (Fig. 2). Our findings were in agreement with Smith et al. (2005), who found that SOC decomposition becomes faster in regions where temperature increases greatly, and soil moisture remains high enough to allow decomposition (Fig. 1). Similarly, Lugato et al. (2014) predicted an overall increase of SOC stocks

according to different climate-emission scenarios up to 2100 for European agricultural soils. But, the declining of SOC stocks under future climate change found in certain conditions is in line with some other SOC projections for agricultural soils (Senapati et al. 2013; Wiesmeier et al. 2016). For example, Xu et al. (2011) modelled SOC changes with the RothC in eight Irish grassland soils from 2021 to 2060 assuming constant C inputs and two different initialization methods. They estimated a decrease of SOC stocks by 2 to 6% for different climate change scenarios.



Fig. 2. Median and range (defined by minimum, maximum and upper/lower quartiles) of annual SOC change rate under the baseline scenario and the climate change scenarios (RCP 4.5 and RCP 8.5) during the time horizon 2010-2050 (on the left) and the time horizon 2010-2100 (on the right)

Modelled SOC storage with RCP 4.5 were substantially higher than the SOC stocks of the RCP 8.5 (with up to 4-fold decrease in SOC stocks) (Fig. 2). Under the baseline reference, average SOC stocks increased by 26.7% at the end of the simulation period. However, average annual SOC change rate increased by 11.8% under RCP 4.5 (annual SOC change rate of 0.15 Mg C ha⁻¹ year⁻¹), and only 7.7% (annual SOC change rate of 0.10 Mg C ha⁻¹ year⁻¹), under RCP 8.5 at the end of the simulation period (Fig. 2). Our findings are higher than Zhang et al. (2017), who simulated SOC stock changes under climate change conditions for grasslands using the DNDC model and the same climate change scenarios as our study. They found a significant reduction on SOC stocks compared to the baseline of 4.14 % and 4.25% under RCP4.5 and RCP8.5, respectively. The difference with our study is partly explained by the different assumptions and pedoclimatic conditions with our study. Smith et al. (2005) predicted a slight decrease by 1% or even a slight increase of 1.6% in SOC levels for European grasslands between 1990 and 2080, under climate change conditions, which are much smaller changes than our results indicate. These smaller or even opposite changes in grassland SOC levels compared

with the previous study might be also attributed to differences in soil and climate conditions covered by the study of Smith et al. (2005).

Dairy grasslands in Northern Spain will potentially act as C source or sink depending on C input application and climate effect interaction together with soil properties (Fig. 3). In particular, under all three climate scenarios, declines in the SOC change rate were more observed in regions with high initial SOC contents (Fig. S2 and Fig 3) and lower C input applications (Fig. S3 and Fig. 3).

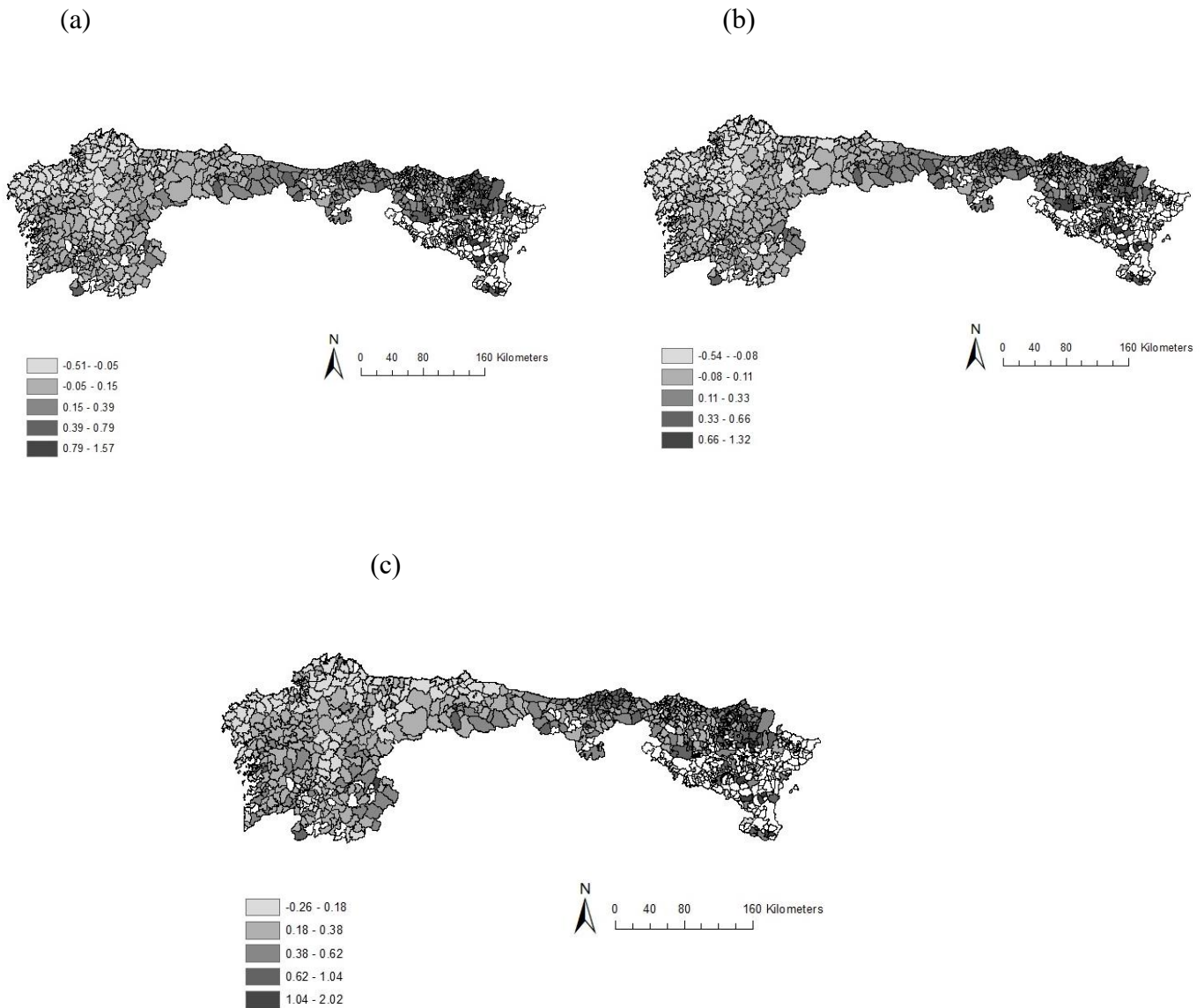


Fig. 3. Soil Organic Carbon stock change rates (Mg C ha⁻¹ year⁻¹) of dairy cows' grasslands in municipalities of Northern Spain under RCP 4.5 (a) and RCP 8.5 (b) climate change scenarios and the reference baseline scenario (c)

In our study area, grasslands located in the Southern part have a marked influence from the Mediterranean Sea and they are also characterised by a more intensive management and, in turn, higher C input additions. In our simulation study, the SOC stock change of this region increased under climate change at a higher rate (i.e., 1 Mg C ha⁻¹ year⁻¹) than other similar studies under the same

climatic conditions (Francaviglia et al. 2012). Therefore, application of manure is likely to increase SOC stocks (Whitehead et al. 2018; Kühnel et al. 2019).

In our study, the effect of temperature predominates over precipitation similarly to Stergiadi et al. (2016) and Zhang et al. (2017) (Fig. 4). Under moist temperate climate, soil temperature regulates microbial activity by a larger extent as soil moisture (Karlsson et al. 2016; Hursh et al. 2017). Under the RCP 4.5 scenario, with a 20% temperature increase, the annual SOC change rate decreased by 58%, while with a 35% temperature increase under RCP 8.5 the annual SOC change rate decreased by 73% (compared with the baseline reference). Therefore, when soil moisture is not limiting, as is the case of our study area increasing temperatures will accelerate SOC decomposition (Smith et al. 2005; Muñoz-Rojas et al. 2015).

Opposite to our findings, Kerr and Ochsner (2020) hypothesized that future soil moisture conditions, rather than precipitation or air temperature, may be the key determinant of climate change–SOC feedback effects at temperate grassland sites. The rate modification by temperature is identical for each SOM pool in RothC despite indications for varying temperature sensitivity of labile and stable SOM pools (Wiesmeier et al. 2016). According to results from laboratory incubations and long-term experiments the more stable SOM pools are indeed more temperature sensitive (Leifeld and Fuhrer 2005). Therefore, the decline in SOC stocks in our findings could be much higher, if we consider stable SOM pools react more sensitively to warming than more labile SOM pools.

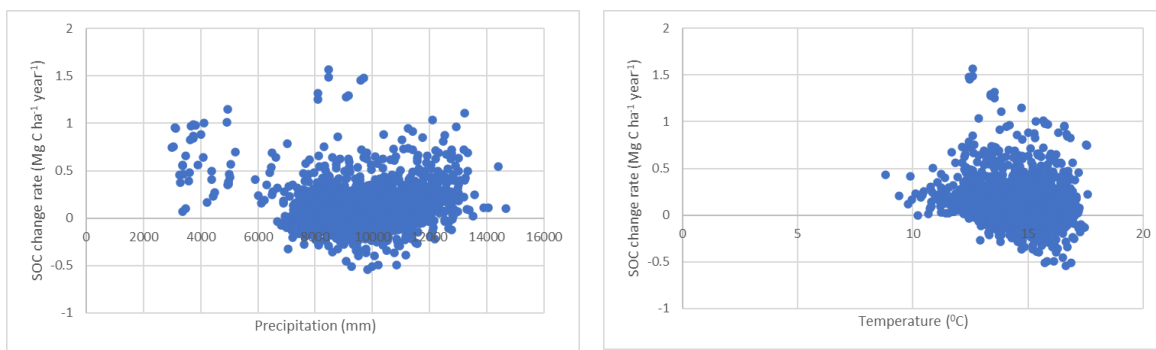


Fig. 4. Soil organic change rate as a function of average annual precipitation (on the left) and average annual temperature (on the right) under climate change conditions (RCP 4.5 and RCP 8.5)

3.2 Net GHG emissions

We found that average net GHG emissions associated with the dairy farm system (i.e., direct emissions from field and barn level) of Northern Spain were positive for the different municipalities, under the different climate scenarios, thus contributing to the global warming. The spatial distribution of Net GHG emissions maintained the same pattern among the different climatic scenarios (Fig. 5). Unlike the SOC storage (Fig. 3), the municipalities characterised by higher C input and thus higher

livestock density (Fig. S3), presented higher net GHG emissions associated to higher enteric fermentation and manure management emissions (Fig.3, Fig.5). Together with SOC decomposition, higher temperature induced an increase in Methane Conversion Factor (MCF) up to an average of 21.6 and 23.5% under the climate change scenarios RCP 4.5 and RCP 8.5, respectively, compared with only 16.2% under the reference baseline scenario. This increase in MCF resulted in higher CH₄ emissions from manure management (Table 2). Therefore, climate change conditions of higher temperature enhanced SOC loss and CH₄ emissions derived from manure management explained the higher net GHG emissions under both climate change scenarios (i.e., RCP 4.5 and RCP 8.5) compared to the baseline scenario (Fig.5).

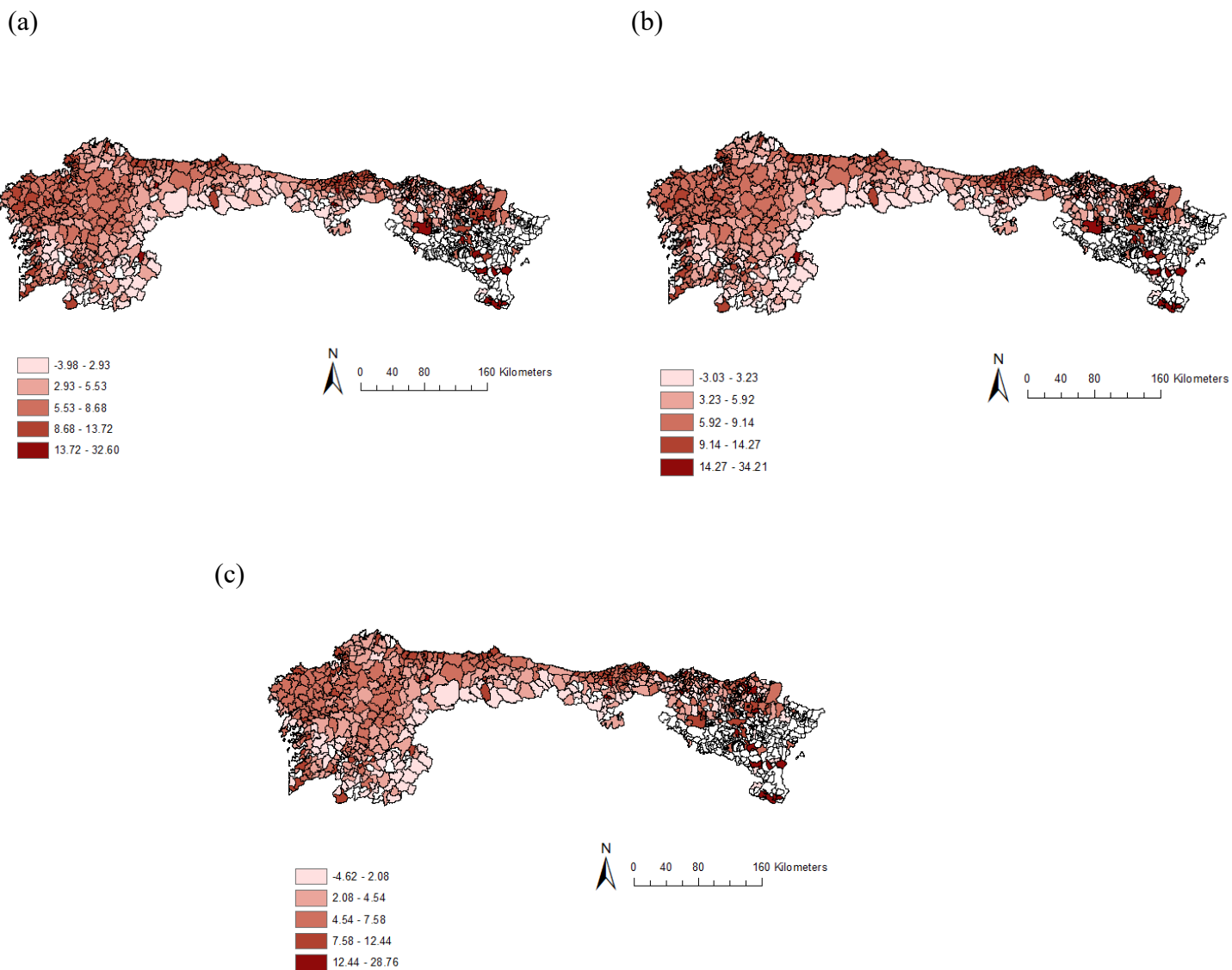


Fig. 5. Net GHG emissions per area in Mg CO₂-e ha⁻¹ year⁻¹ under RCP 4.5 (a), RCP 8.5 (b) and the baseline reference (c)

The estimated net GHG for the dairy farming of the different municipalities under RCP 4.5 ranged from -4 to 33 Mg CO₂-e ha⁻¹ year⁻¹ (with average value of 5.8 Mg CO₂-e ha⁻¹ year⁻¹) (Fig. 5). Net

GHG emissions under RCP 8.5 varied between -3 and 34 Mg CO₂-e ha⁻¹ year⁻¹ (average value of 6.2 Mg CO₂-e ha⁻¹ year⁻¹) (Fig. 5). On the other hand, the net GHG under the reference baseline scenario showed an average value of 4.7 Mg CO₂-e ha⁻¹ year⁻¹. Graux et al. (2012), evaluating French grassland based dairy systems under SRES A2 forcing conditions, showed an increase in net GHG in extensively managed grassland systems and a reduction in net GHG in intensively managed grassland systems (where SOM decomposition acceleration is compensated by enhanced net primary production).

Therefore, according to our modelling approach, climate change effects of temperature and precipitation induced an increase in net GHG emissions, basically because of the increase in SOC storage loss and CH₄ manure emissions (Table 2).

Table 2. Average CH₄ and N₂O emissions, total greenhouse gas emissions, SOC storage and Net GHG expressed in Mg CO₂-e ha⁻¹ year⁻¹ for the grasslands associated to dairy production different in Northern Spain

Climate scenario	CH ₄	from	CH ₄	from	CH ₄	from	N ₂ O	from	N ₂ O	from	GHG	SOC	Net
	enteric fermentation		manure management		grassland soil		manure management		grassland soil		emissions	accumulation	GHG
Baseline	3.88		1.09		0.01		0.24		0.85		6.07	1.37	4.70
RCP 4.5	3.88		1.41		0.01		0.24		0.85		6.40	0.57	5.82
RCP 8.5	3.88		1.57		0.01		0.24		0.85		6.55	0.37	6.18

3.3 Manure management scenarios to reduce GHG emissions

3.3.1 Rigid Cover

We found an average 19% reduction in N₂O emissions with the cover among the different climate scenarios (Table 3). As a result, reducing NO₃ leaching and NH₃ volatilisation should also contribute to reductions in total N₂O emissions (Chadwick et al. 2011). We based our estimations on emission factors for production systems, together with average annual temperature (IPCC 2019; EMEP 2019). Our approach may present certain degree of uncertainty since, during storage, microbial activities in manure might be affected by local climatic conditions (Peterson et al. 2013).

3.3.2 Slurry removal

Increasing the time of manure storage increases the period during which CH₄ is emitted as well as the emission rate. Hence, we explored the potential for CH₄ reduction while comparing the reference scenario with alternative scenarios of removing slurry referring to different seasons of the year (See “Manure related management scenarios” subsection), and therefore to different climatic conditions (Table 3).

Results showed that the low emitting scenario consisting of removing slurry all seasons except in the Winter resulted in a reduction in CH₄ emissions from manure management up to 28% (Table 3). Although out of the scope of our study, avoiding manure application in Winter would help to avoid potential eutrophication since frequent application during precipitation events or snowmelt could lead to runoff, leaching and loss of valuable nutrients (Aguirre-Villegas and Larson 2017). Removing slurry during all the year except in Autumn showed the lowest reduction among the alternative practices with more than 11%, as the temperature conditions in the storage system are higher than the Winter season. However, these emissions could increase during application as more ammoniacal N and VS are available to promote N₂O, CH₄, and NH₃ release (Chadwick et al. 2011; Jokela et al. 2017).

Our approach of MCF model (IPCC 2019) accounted for timing and length of storage, manure composition, and monthly temperature variations, and retention time in the barn. In this context, Peterson et al. (2018) stressed out the time as a key variable as management decisions influence storage conditions on a daily basis, and storage time before or after manure treatment (Peterson et al. 2018).

According to our results, GHG emissions from manure management can be further reduced by combining the manure management practices of rigid cover while removing the manure all year long except in winter (Table 3).

Table 3. Net GHG (Mg CO₂-e ha⁻¹ year⁻¹) for a combination of climate scenarios (Baseline, RCP 4.5 and RCP 8.5) and manure management practices (M0, M1, M2 and M3) and their effect on N₂O or CH₄ emissions from manure management (%)

Scenario	Range of Net GHG	CH ₄ reduction from manure management (%)	N ₂ O reduction from manure management (%)
Baseline - M0	-4.62 – 28.76	-	-
Baseline - M1	-4.63 – 28.50	-	19
Baseline - M2	-4.7 – 26.28	28.45	-
Baseline - M3	-4.65 – 27.55	11.75	-
RCP 4.5 - M0	-3.98 – 32.60	-	-

RCP 4.5 - M1	-4.00 – 32.34	-	19
RCP 4.5 - M2	-4.08 – 29.33	27.8	-
RCP 4.5 - M3	-4.02 – 30.86	11.22	-
RCP 8.5 - M0	-3.03 – 34.20	-	-
RCP 8.5 - M1	-3.04 – 33.95	-	19
RCP 8.5 - M2	-3.14 – 30.69	27.2	-
RCP 8.5 - M3	-3.08 – 32.26	11.35	-

M0, Reference Manure management scenario (Slurry removed all year long except in Summer, where the slurry storage system is with natural crust); M1, Slurry storage system with rigid cover; M2, Slurry removed all year long except in Winter; M3, Slurry removed all year long except in Autumn.

3.3.3 Anaerobic digestion

Average SOC increase showed a reduction under AD management scenario for both future climate change conditions (Fig. 6). This reduction was a result of lower C input derived from excreta, as part of it was converted to CH₄ and CO₂ in the storage system (Petersen et al. 2013). Moreover, emissions after land application increased about 17% under AD scenarios, due to the higher manure total ammoniacal nitrogen (TAN) (Aguirre-Villegas et al. 2019) (Fig. 6). However, AD helped avoiding 95% of CH₄ emissions derived from manure management that would have otherwise occurred (Fig. 6). Aguirre-Villegas et al. (2015) highlighted emissions of manure management on dairy systems can be reduced by more than 40%, which is in line with our findings. Therefore, the net GHG of the AD manure management scenario under both RCP 8.5 and RCP 4.5 was reduced by 22.8% and 21.5%, respectively (Fig.6). Particularly, the net GHG corresponding to the AD management scenario was then equivalent to the net GHG under the baseline reference scenario. The mitigation potential achieved under AD manure management within our study is higher than the findings of Peterson et al. (2018), as we used a different modelling approach. The anaerobic digestion has many other benefits that are not analysed in this study, such as the production of renewable energy, and the avoided use of fossil-based energy on-farm, which promotes the sustainability and profitability of dairy farms (Aguirre-Villegas et al. 2019). Moreover, injection of digestate during land application is an effective management practice to reduce NH₃ emissions, which at the same time increases N availability and reduces GHG emissions (Aguirre-Villegas et al. 2019). Although the benefits of AD are numerous, it is a capital intensive technology that might only be justified at large farms (Aguirre-Villegas et al. 2019).

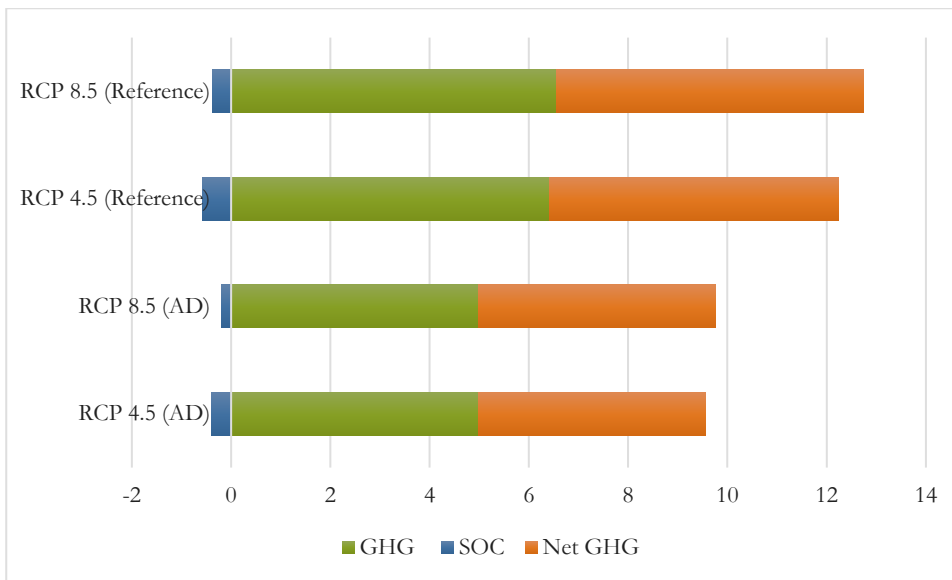


Fig. 6. Net balance of the reference scenario and alternative manure management scenarios (Anaerobic digestion) under both RCP 4.5 and RCP 8.5 climate change scenarios during the period 2010-2100

We did not consider mitigation scenarios to avoid soil N₂O emissions derived from manure application in our study, as the IPCC methodology refers to emission factors rather than detailed climate-based equations for N₂O soil emissions. Referring to the scientific literature, there is several measures to reduce soil N₂O emissions: For instance, N₂O emissions are considerably reduced if the amount of N applied with the manure corresponds to the amount necessary for optimal pasture growth. In this context, nitrification inhibitors have a potential to reduce N₂O emissions, as well as N leaching from manure or fertilizers, and the reduction may be as high as 40 to 50% according to some meta-analyses (e.g., Qiao et al. (2015)).

Although out of the scope of our study, the diet choice was found to be the main factor controlling the C footprint of cattle dairy production in Northern Spain (Del Prado et al. 2013). In this context, the 3-Nitrooxypropanol (NOP) showed to be is a promising CH₄ inhibitor according to meta-analysis conducted by Dijkstra et al. (2018). NOP also contributed to reduce 11.7% of total emissions for the dairy production systems according to the life cycle analysis conducted by Feng and Kebreab (2020).

3.4 Limitations

The originality of our study stands on the assessment of the net GHG emissions of grassland-based dairy systems at regional level under climate change and alternative manure management mitigation scenarios. However, our study involved some limitations that should be highlighted. Regarding SOC dynamics estimation, uncertainty related to this work may be ascribed to the model applied, and the non-availability of some data at the temporal or spatial levels (See Chapter 3). Indeed,

the proposed regional analysis is based on a spatial division of the grassland systems for dairy production in Northern Spain territory into different spatial units (i.e., municipalities) assuming homogeneity of a set of specific parameters (e.g., soil properties). Changes in soil management and C input values throughout the study period were not considered and may need to be refined. In this context, many studies (e.g., Dondini et al. (2018) and Hewins et al. (2018)) stressed the climate impacts on plant productivity and C input amounts. In particular, the plant growth is vulnerable to shifts in temperature and precipitation (Emadodin et al. 2021). Furthermore, the RCP scenarios were used to provide the possible changes in climate in this study, but as a long-term climate projection, the uncertainty in the projected climate will increase as the time span increases (Moss et al. 2010). Particularly, projected rainfall presented the factor with the most variability between climate scenarios and the primary source of uncertainty in SOC response (Meyer et al. 2018). Finally, we assumed that the grassland type was the same in the different scenarios. However, the grassland community structure could be altered under both grazing and climate change (Koerner and Collins 2014). The assumption that grassland community structure remains stable in the simulation could induce uncertainty while more research is required to clarify the specific responses of plant communities to climate change (Ghahramani et al. 2019).

Regarding GHG estimation under climate change projections, apart from uncertainties induced from our main assumptions (See Chapter 3), there are uncertainties that could be related to IPCC Tier 2 methodology (Clark 2017). For instance, N₂O emission factor calculations based on IPCC Tier 2, did not account for refined environmental regulators for instance on daily or monthly basis, which may modify emissions of applied nitrogen. Moreover, the potential impact of climate change on N₂O mitigation strategies remains speculative and requires further research (Griffis et al. 2017). In this context, a multi-model ensemble could improve the predictions (e.g., NGAUGE (Brown et al. 2005; Del Prado et al. 2006) and DNDC (Li et al. 1992; Giltrap et al. 2010)).

Finally, our assessment of the net GHG is limited to grassland-based systems and did not account for mixed forage systems including maize silage, for example. The analysis described in this paper cannot be considered a full life cycle assessment as our estimation for the net GHG of dairy grassland systems excluded pre-farm phases (e.g., feeds) and farm energy use.

4 Conclusions

We found that average SOC of grassland-based dairy livestock systems in Northern Spain were reduced significantly, compared with the baseline scenario, under future climate change scenarios (particularly under RCP8.5). Furthermore, the variations in SOC were found driven mainly by air

temperature rather than precipitation. Our study showed that future climate change is likely to increase the net GHG of dairy grassland systems in Northern Spain. Together with greater SOC losses, higher temperature induced higher CH₄ emissions derived from manure management.

Based on our findings, combining alternative dairy manure management practices (slurry storage system with rigid cover and the slurry removal all year long except in winter) would help to mitigate the climate change effects and to reduce the net GHG of the grassland-based dairy livestock systems in Northern Spain. The anaerobic digestion was the most effective strategy to mitigate GHG emissions from manure, as it allowed net GHG under both climate change scenarios to equal net GHG under the reference baseline scenario.

Our study illustrates that climate change will impact net GHG emissions of the grassland-based dairy livestock systems in Northern Spain. Furthermore, it emphasises the importance of improving our modelling capabilities, with consideration of off-farm emissions, to provide a clearer picture of the full implication of management practices to mitigate climate change effect.

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GENERAL DISCUSSION

Simulation of changes in soil organic carbon stocks and greenhouse gas emissions in agricultural systems of Spain

Initial soil organic carbon (SOC) content was identified as the predominant environmental variable that influenced SOC. Essentially, for both studied agroecosystems (i.e., Mediterranean croplands and moist temperate managed grasslands), soils with greater initial SOC content displayed lower SOC change rates than soils with low initial SOC content. In this sense, the negative correlation between the SOC change rate and the initial SOC content has been documented in previous studies (Bellamy et al. 2005; Zhao et al. 2013). Furthermore, as discussed in Chapter 3, precipitation was positively correlated with initial SOC content. Therefore, the sites characterised by high precipitation levels and then with high initial SOC contents were associated to lower SOC change rates than sites with low precipitation levels and low initial SOC content (Meyer et al. 2016).

It is important to highlight that carbon inputs were the main driver of SOC changes in both the croplands located in Mediterranean Spain and the moist temperate grasslands located in Northern Spain. A clear example of the main role of C inputs on SOC changes was the higher SOC storage found in irrigated crops compared with rainfed crops in Mediterranean croplands. The high SOC change rate under irrigated crops stressed the key role of irrigation in the productivity of Mediterranean systems (Wriedt et al. 2009) and thus in the production of C inputs. However, although out of the scope of our study, the irrigation could also result in an increase of SOC decomposition favouring soil moisture conditions for C mineralisation (Aguilera et al. 2018). In this context, existing studies reported either a decrease (e.g., Nunes et al. 2007) or an increase (e.g., Montanaro et al. 2010) in SOC associated to irrigation. In this sense, SOC decomposition and its interaction with N₂O emissions is highly linked to the irrigation type adopted (Aguilera et al. 2018).

The lowest levels of SOC in Mediterranean croplands were found in the vineyards and especially in olive groves. The main reason for this finding is that Mediterranean soils dedicated to olive groves are managed under intensive tillage and are prone to erosion (Rodríguez Sousa et al. 2019), and thus to SOC loss. This loss in SOC may represent a great opportunity for the Mediterranean region to achieve significant increases in SOC storage (Francaviglia et al. 2019). In this context, cover crops, as an alternative soil management practice, contributed to quadruple SOC stocks under the extreme climate scenario as outlined in Chapter 1. In the same line, according to López-Vicente et al. (2021) in a Spanish Mediterranean olive grove, the increase in SOC contents in the top 5 cm soil with cover crops reached 88.4 %. Moreover, although out of the scope of this Thesis, the increase in SOC stocks through cover crops in woody crops would prevent soil erosion (Novara et al. 2019).

Under moist temperate grasslands, the importance of C inputs on SOC changes was demonstrated

through livestock intensity (either through grazing or via manure application from housed livestock). The modelling approach setup in this Thesis did not allow to find out the non-linearity of the relationship between SOC stocks and grazing intensity found in other studies (Eldridge and Delgado-Baquerizo 2017). The non-linearity is explained by a limiting threshold of overgrazing from which there is a reduction in SOC stocks. This behaviour is explained by the increase in disturbance and biomass removal lead by overgrazing (Mcsherry and Ritchie 2013). Particularly, poaching, as a soil damage common in moist areas, becomes even more severe at higher grazing densities (Tuohy et al. 2014) resulting in a reduction in grassland productivity (Piwowarczyk et al. 2011) and thus in SOC stocks.

Similarly, livestock density was found to increase GHG emissions per hectare. In this context, livestock density led to increased emissions of N₂O from fertilisation and CH₄ from enteric fermentation similar to other similar studies (Soussana et al. 2013). For instance, at the soil level, soil N₂O emissions may offset SOC sequestration as the changes in SOC turnover feed back into the N cycle (Lugato et al. 2018). A potential livestock density (approximately > 0.4 LU ha⁻¹) was set in Chapter 3 from which the net GHG emissions (expressed as CO₂-e per LU) would be at lowest level. However, Sánchez Zubieta et al. (2021) stressed out the key role of low to moderate grazing intensities as sound grazing management practices to reduce GHG emissions. In addition, as outlined in chapter 3, the level of concentrate feeding must be reduced for the dairy cows as it was found to contribute to higher GHG emissions per hectare and probably per milk production units when C mineralization in the soil during off-farm grain cropping period is taken into account. In this context, higher proportion of concentrates (produced off-farm) in the diet, induced high rates of manure application and N deposition on farmland used for forage production within the system (Bakken et al. 2017).

Climate change effects on soil organic carbon and greenhouse gas emissions in agricultural systems of Spain

In the two studied agroecosystems (Mediterranean croplands and moist temperate grasslands associated to dairy production), SOC stocks decreased under climate change conditions. Our findings were in line with previous studies under temperate climatic conditions (Wiesmeier et al. 2016). Likewise, for both agroecosystems, the relevance of the interaction between temperature and precipitation has been also highlighted in previous studies (Grahammer et al. 1991). For example, Smith et al. (2005) predicted faster SOC decomposition rates in areas where temperature increased and, concurrently, where soil water moisture remained sufficiently high to enable SOC

decomposition.

Findings from Chapters 1 and 4 showed that in the systems studied in this Thesis the effect of temperature predominated over the precipitation, which differs from findings from other studies carried out for moist temperate grasslands of Northern Spain (Guntiñas et al. 2013) and Mediterranean Spanish croplands (Álvaro-Fuentes et al. 2012). Differences among studies reflect the sensitivity of RothC model to temperature (Falloon and Smith 2002) rather than soil moisture, compared with others models (e.g., Century). In this context, a multi-model ensemble for SOC predictions could also reduce model structural uncertainties, as it allows reasonable interpretation of the different factors and parameters of the SOC dynamics modelling (Riggers et al. 2019).

For Mediterranean croplands, the findings from Chapter 1 showed that, under climate change, no-tillage in arable crops and cover crops in woody crops would lead to an increase in the SOC content in agreement with other studies (Álvaro-Fuentes and Paustian 2011; Francaviglia et al. 2019). It is worth to point out that these practices are also cost-effective since they are relatively easy to be implemented by farmers at low cost (Francaviglia et al. 2019).

In this Thesis, it has been shown that climate change conditions and, particularly, the raise in air temperature enhanced SOC loss and CH₄ emissions derived from manure management and consequently induced an increase in net GHG emissions (Chapter 4). In this context, the approach of IPCC methodology allowed to simulate CH₄ emissions derived from manure management through the Methane Conversion Factor model (IPCC 2019). The latest accounted for timing and length of storage, manure composition, and monthly temperature variations for the different climate scenarios. However, the climate change effect was not captured for soil N₂O emissions due to the scarce captured dependence of emission factors on spatial diversity of management, pedoclimatic, soil physical and biochemical conditions (Cayuela et al. 2017).

Following the findings from Chapter 4, the effect of some manure management practices (i.e., rigid cover, slurry removal all year except in winter season, anaerobic digestion) on net GHG was tested under climate change conditions. Since manure contains inorganic nitrogen, water and microbially available sources of carbon, it provides the essential substrates required for the microbial production of N₂O and CH₄, and NH₃ which is an important atmospheric pollutant and precursor of N₂O. But it is also important to point out that these gases may be produced and emitted at each stage of the manure management (e.g., livestock building, manure storage, manure treatment and manure spreading to soil) (Chadwick et al. 2011). Consequently, mitigation practices that reduce emissions in one stage of the manure management process may increase emissions elsewhere (Montes et al. 2013). In the case of anaerobic digestion, the different GHG emissions (i.e., CH₄, N₂O and CO₂) were estimated at different stages (e.g., post digestion and soil application stages) following Pardo et al.

(2017) approach for SIMSWASTE-AD model. In this Thesis, it was shown that the anaerobic digestion was the most effective strategy to mitigate GHG emissions from manure, as it allowed similar net GHG between climate change and reference baseline conditions similar to previous studies (Petersen 2018).

Modelling and research needs

In this PhD thesis it is presented a modification of the soil model RothC. These modifications are presented in Chapter 2 and they were considered based on the scientific literature which, in certain situations, is limited (e.g., poaching effect). In particular, the soil moisture together with the soil temperature reduction functions of the model require greater calibration since they need to be valid for the environmental conditions (Bauer et al. 2008). In this context, Farina et al. (2013) reduced the rate modifying factor for moisture in RothC to improve model performance under Mediterranean dryland conditions. In this Thesis (Chapter 2), a modification was made to the model (i.e., allowing the soil to be under water saturation conditions) and improved its performance for moist temperate conditions. In this context, it was assumed a linear decline in the rate modifying factor for soil moisture, like in the ECOSSE model, as there was not sufficient evidence to suggest a more refined relationship (Smith et al. 2010). More improvements could be achieved, in this respect, by using a more refined function based on robust field experiments, as the soil moisture effect on SOC dynamics is rather complex and presents a lot of uncertainty (Batlle-Aguilar et al. 2010; Falloon et al. 2011).

The RothC model does not simulate management practices (e.g., grazing, tillage). Management data need to be taken into account via their impacts on plant residue data, although it is often difficult to find appropriate information on it (Nemo et al. 2017). For example, in this Thesis, in Chapter 2, the grazing effect was included through livestock density, but in Chapter 1, the tillage effect for the Mediterranean croplands was lacking. Similarly, the representation of erosion on SOC dynamics is another limitation in RothC (Martinez-Mena et al. 2008). In this context, under Mediterranean conditions, soils are prone to land degradation and soil erosion (Muñoz-Rojas et al. 2015).

In the different chapters of this Thesis, it was shown that the C inputs estimation, derived from plant residues, is one of the most decisive parameters in SOC projections. In this context, different studies have overestimated C inputs (Nemo et al. 2017) and there is a lack of detailed information on how plant residues were estimated and/or assumptions regarding their conversion to C inputs (Nemo et al. 2017). In particular, belowground C inputs measurement is challenging and it is strongly influenced by multiple factors (Cagnarini et al. 2019). In modelling studies and specially when using RothC, it is recommended to give a detailed calculation of C inputs and to distinguish between above

and below-ground residues adding the rhizodeposition component (Balesdent et al. 2011).

Under climate change conditions, although an unchanged plant production and C inputs were assumed for simplicity reasons, it is important to fully account for the different factors impacting the C inputs (e.g., future changes in crop and soil management, climate and atmospheric CO₂ changes) (Wiesmeier et al. 2016). Under Mediterranean areas, future projections of C inputs in agricultural soils are even more challenging (Soussana et al. 2013). For instance, under climate change conditions, Spain as other Mediterranean areas, may face scarcity in water availability to meet irrigation requirements and growing food demand (Fader et al. 2016). It is therefore recommended to use of complex plant production models under the different climate change scenarios while considering the aforementioned aspects (i.e., grazing effect, poaching, irrigation technique and the interaction between C and N cycles).

In this Thesis, the estimation of GHG emissions was made with the IPCC (2019) Tier 2 methodology (Chapters 3 and 4). Soil attributes (such as pH, soil water content and soil texture), crop types and climate significantly affect GHG emissions. Consequently, these factors should be fully considered (Shakoor et al. 2021). However, the IPCC methodology does not entirely consider them. For example, the estimation for soil N₂O emissions assumed a fix emission factor per climate zone and management type. However, soil N₂O emissions are rather more complex, being controlled by water filled space, soil mineral N content and temperature (Conen et al. 2000). Moreover, the fluxes of each gas are decoupled from each other and the interactions between them cannot be properly accounted. As an example, increasing N fertilization leads to higher N₂O emissions, but there is a concurrent stimulation of plant production, which fosters CO₂ uptake (Vuichard et al. 2007). Nevertheless, given the nonavailability of detailed data at the temporal or spatial levels (e.g., management, microclimate, soil properties) for the net GHG assessment at a regional scale, the use of Tier 2 emission factors rather than complex models is more convenient. In particular, Tier 2 of the 2019 IPCC refinement relies on enhanced characterisation of animal population, diet characteristics and manure management for the estimation of emissions, compared to the 2006 IPCC guidelines.

Likewise, the findings of net GHG emissions from chapters 3 and 4, although they may be important indications for further studies, they illustrated the importance of considering the whole GHG emissions (including off-farm emissions). Furthermore, future research should focus not only on singular mitigation measures but also a combination of options, to promote the development of more practical measures that can be widely adopted for mitigating GHG emissions.

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CONCLUSIONS

The research performed in this Thesis provided an assessment of soil organic carbon (SOC) changes and greenhouse gas (GHG) emissions at a regional scale for different agroecosystems and climatic conditions in Spain (i.e., croplands under Mediterranean climatic conditions and managed grasslands associated to dairy production under Northern moist temperate conditions), using integrated modelling approaches. In this thesis, alternative management practices to reduce/mitigate GHG emissions in the agroecosystems under climate change conditions were also evaluated. In particular, the Thesis provides new insights for SOC modelling using RothC model to fit to managed grasslands under moist temperate climatic conditions and for future research needs. The key conclusions obtained in this Thesis are:

- The soil organic carbon (SOC) stocks of croplands under Mediterranean climatic conditions and managed grasslands associated to dairy production under Northern moist temperate conditions in Spain would be reduced under climate change conditions for the next 90 years, compared to the baseline references. The variations in SOC were found driven by the combined effect of the climatic variables under climate change conditions, particularly, at a higher extent by temperature for the moist temperate grasslands. Carbon inputs were the main driver of SOC changes in both agroecosystems (i.e., the croplands located in Mediterranean Spain and the moist temperate grasslands located in Northern Spain).
- Under Mediterranean croplands, the highest SOC change rates were found in irrigated crops, as a consequence of high C inputs production, and the lowest SOC change rates were found in rainfed woody crops (i.e., olive groves and vineyards). The low SOC change rate under olive groves and vineyards may reflect a great opportunity for SOC storage in Mediterranean croplands. No-tillage, in the case of rainfed field crops, and vegetation cover, for olive groves and the other woody crops, were found to be effective strategies in reducing CO₂ emissions and increasing soil potential to sequester C under future climate change conditions.
- The successful modifications implemented to adapt the RothC model to moist temperate managed grasslands evidenced the importance of fine-tuning the environmental variables (i.e., soil moisture) and the C inputs (particularly plant residues). In this sense, the soil moisture reduction function needs to properly address wet environmental conditions (i.e., water saturation condition). Also, it is key to better adjust the plant residues fractioning into the different C quality components (i.e., above-, below-ground plant residue and rhizodeposits). It was stressed out the importance to validate the modifications using more robust experiments

and to study poaching soil damage, since it results in complex and multi-factorial effects in these moist temperate grasslands.

- Under moist temperate grasslands associated to dairy production, livestock density was found to increase SOC storage at the regional level, as being proportional to C inputs, and net GHG emissions derived from manure and enteric fermentation. The livestock threshold established in this thesis should be optimized in this respect, to reduce net GHG emissions, while considering the interaction between C and N cycles and off- farm emissions.

- As alternative manure management strategies to tackle GHG emissions of the grassland-based dairy livestock systems in Northern Spain, slurry storage system with rigid cover with removal all year long except in winter was suggested. Alternatively, the anaerobic digestion was found as the most effective manure related management practice to mitigate climate change effect, as it allowed net GHG under both climate change scenarios to equal net GHG under the reference baseline scenario.

- For future research, RothC could be integrated with a Nitrogen model to account for the interaction between C and N cycles under the different agroecosystems. The modifications proposed to the RothC to fit to managed moist temperate grasslands could be refined and the implementation of more improvements to the model under Mediterranean conditions is recommended (e.g, tillage effect and erosion phenomenon).

**SUPPLEMENTARY
INFORMATION**

Chapter 1

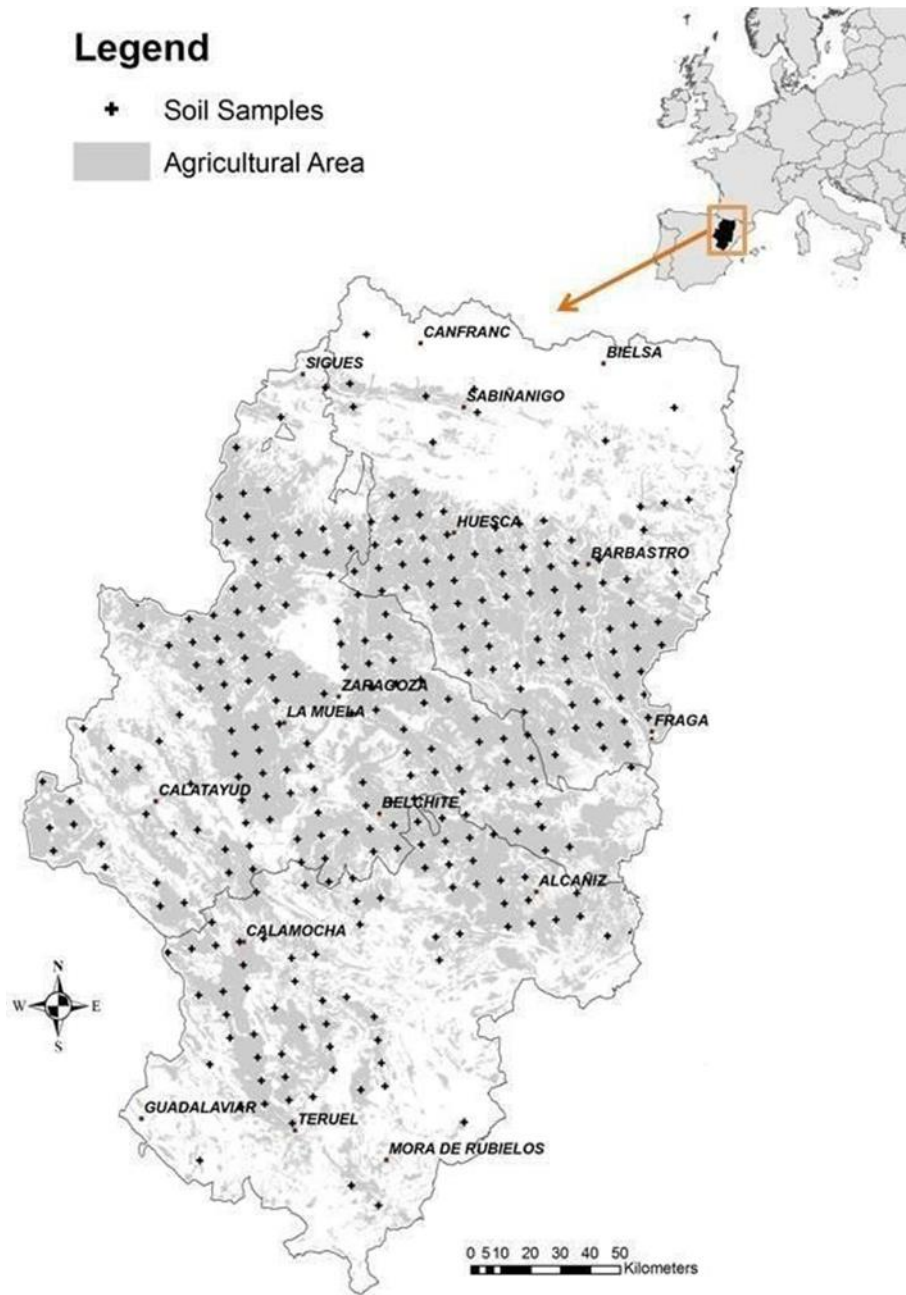


Fig. S1. Map of the Aragon region with the sampling points from the study of López Arias and Grau Corbí (2005)

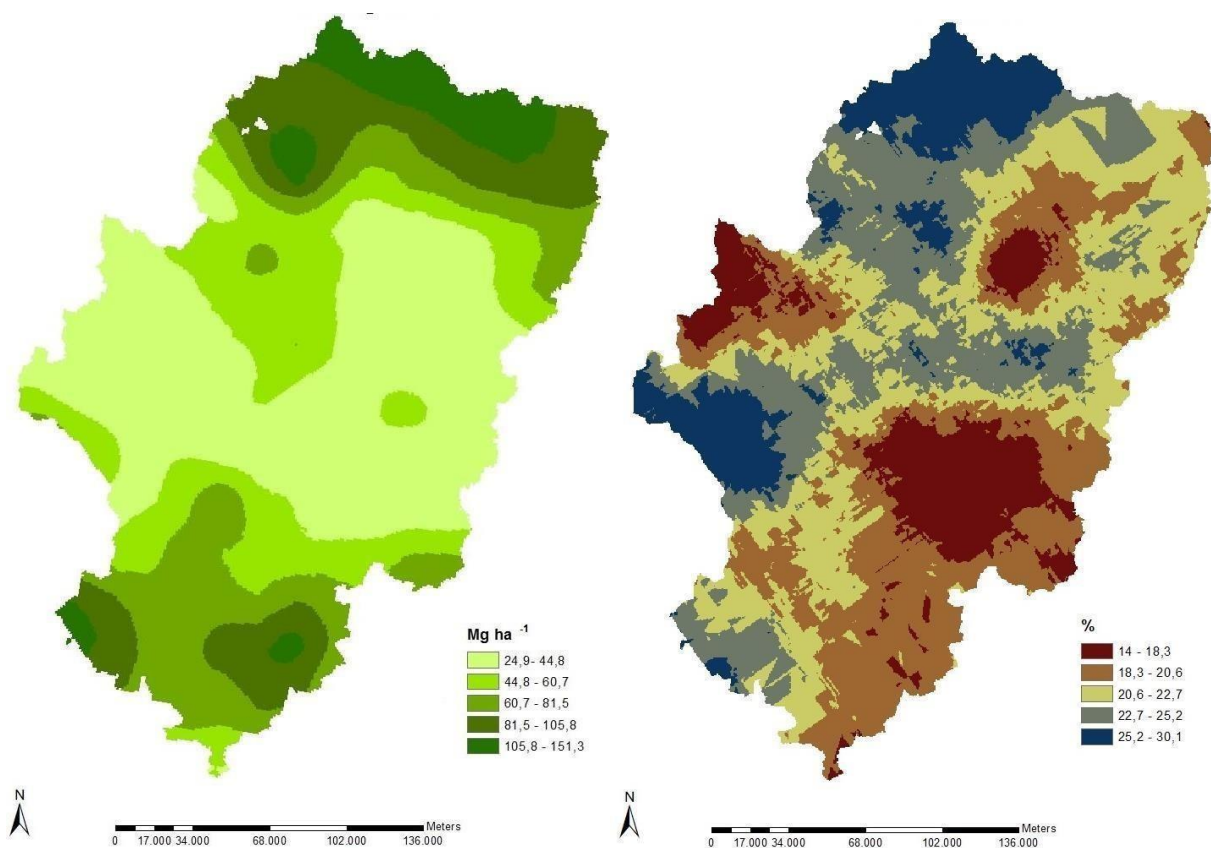


Fig. S2. Distribution of measured soil organic carbon (SOC) content (on the left) and clay percentage (on the right) for the 0-30 cm soil layer in the Aragon region. Data from the study of López Arias and Grau Corbí (2005)

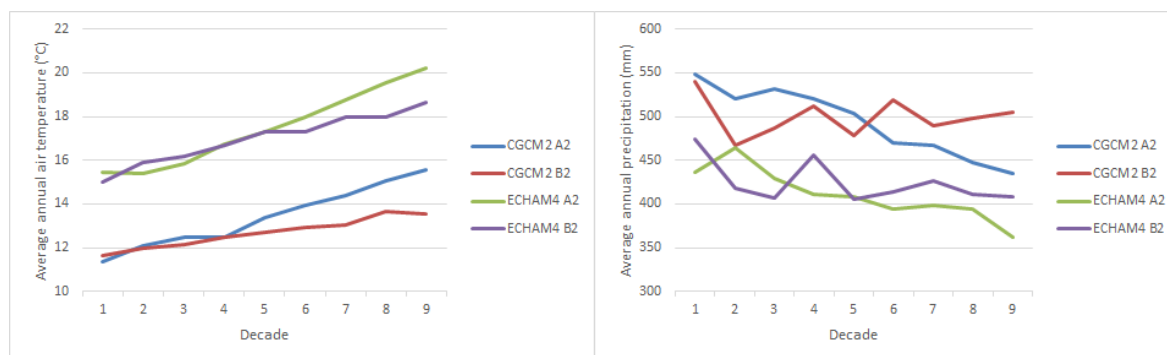


Fig. S3. Decadal variations and long-term trends of annual air temperature (on the left) and annual precipitation (on the right) for the different climate change scenarios during the 2010-2100 period in the Aragon region (decade 1 corresponds to 2010-2020 and decade 9 to 2090-2100)

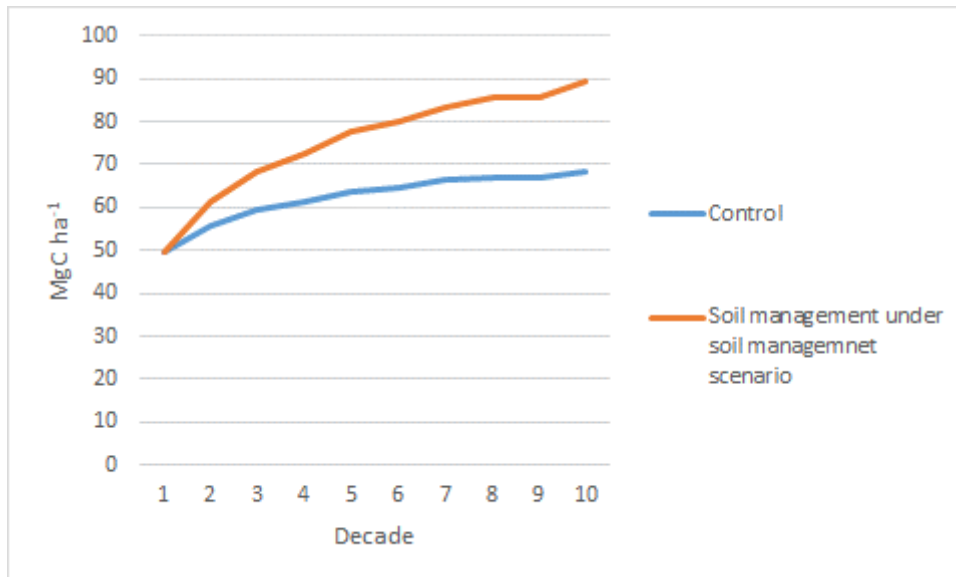


Fig. S4. Decadal changes in soil organic carbon (SOC) content for the 0-30 cm soil layer in the Aragon agricultural soil under ECHAM4 A2 climate change scenario between control and soil management scenarios during the 2010-2100 period (decade 1 corresponds to 2010-2020 and decade 9 to 2090-2100)

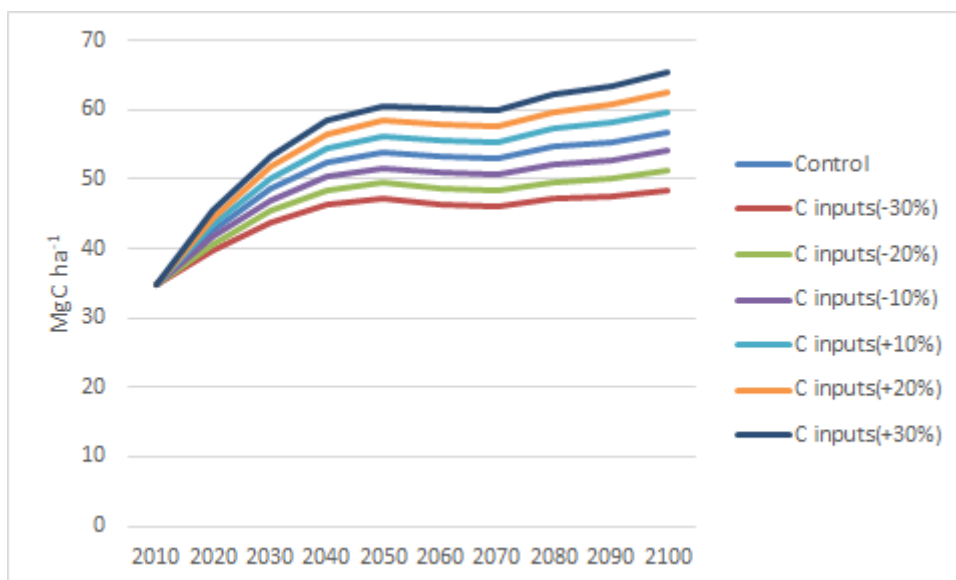


Fig. S5. Results of the sensitivity analysis for C inputs. The y-axis indicates SOC content under the CGCM2-B2 climate change scenario

Table S1. Average annual air temperature and precipitation in selected points of the different zones of Aragon region during the period 2010-2100 under the different climate scenarios

Aragon zone	Climatic parameter	Climate scenario				
		Baseline	CGCM2-A2	CGCM2-B2	ECHAM4-A2	ECHAM4-B2
Ebro depression	Mean annual temperature (°C)	12.6	15.4	14.8	19.5	18.9
	Annual Precipitation (mm)	308.2	288.4	287.9	251.0	254.4
Pyrenees zone	Mean annual temperature (°C)	8.8	11.8	11.1	16.2	15.6
	Annual precipitation (mm)	973.0	925.8	950.5	716.6	749.5
Iberian zone	Mean annual temperature (°C)	9.0	12.1	11.5	16.3	15.7
	Annual precipitation (mm)	573.1	504.9	511.8	409.3	420.5

Table S2. Alternative soil management practices proposed for rainfed crops (RC) and woody crops (WC)

Agricultural system	Soil management practice	Livestock scenario
RC	NT ^a	20% of increase in livestock number for the period 2010- 2030. Stable situation for the period 2030-2100.
		20% of decrease in livestock number for the period 2010- 2030. Stable situation for the period 2030-2100.
WC	VC	No animal manure application

^a NT, no-tillage; VC, vegetation cover

Table S3. Annual SOC sequestration rates at the 0-30 cm soil layer in the agricultural surface of the Aragon region during the period 2010-2100 under climate scenarios

Climate scenarios	Annual SOC sequestration rate (Mg C ha ⁻¹ year ⁻¹)
Baseline	0.28
CGCM2-A2	0.23
CGCM2-B2	0.24
ECHAM4-A2	0.21
ECHAM4-B2	0.20

Chapter 2

Supplementary information A. Model modification

The conversion from soil water content to soil moisture deficit (SMD_i , mm) used in RothC referred to Farina et al. (2013) is given by the following equation (Farina et al. 2013):

$$SMD_i = (WC_i - WC_{fc}) \times 10 \times depth$$

(S1)

Where WC_{fc} is the soil water content at field capacity, WC_i is the soil water content above field capacity.

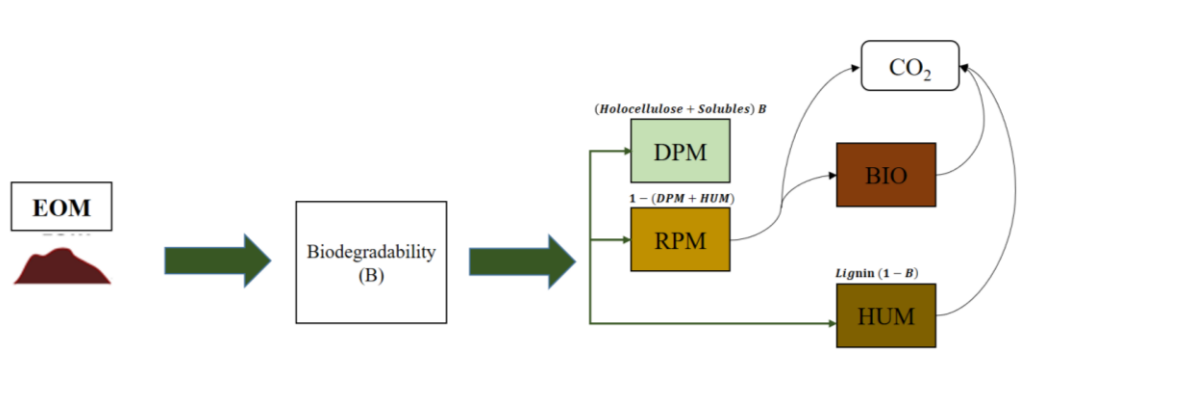
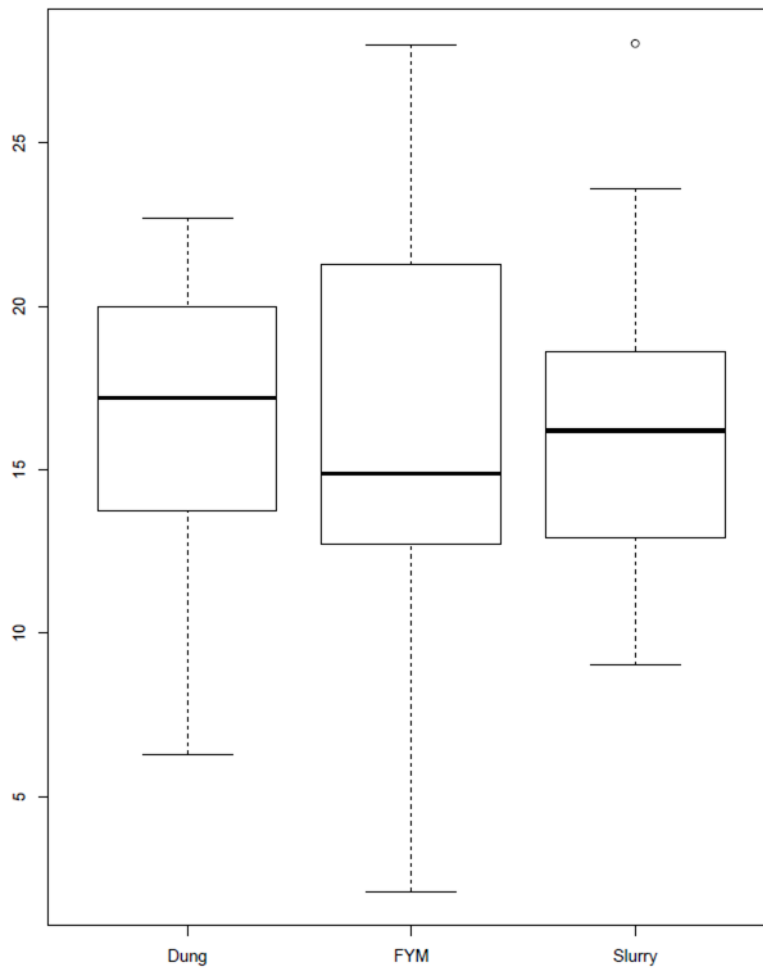
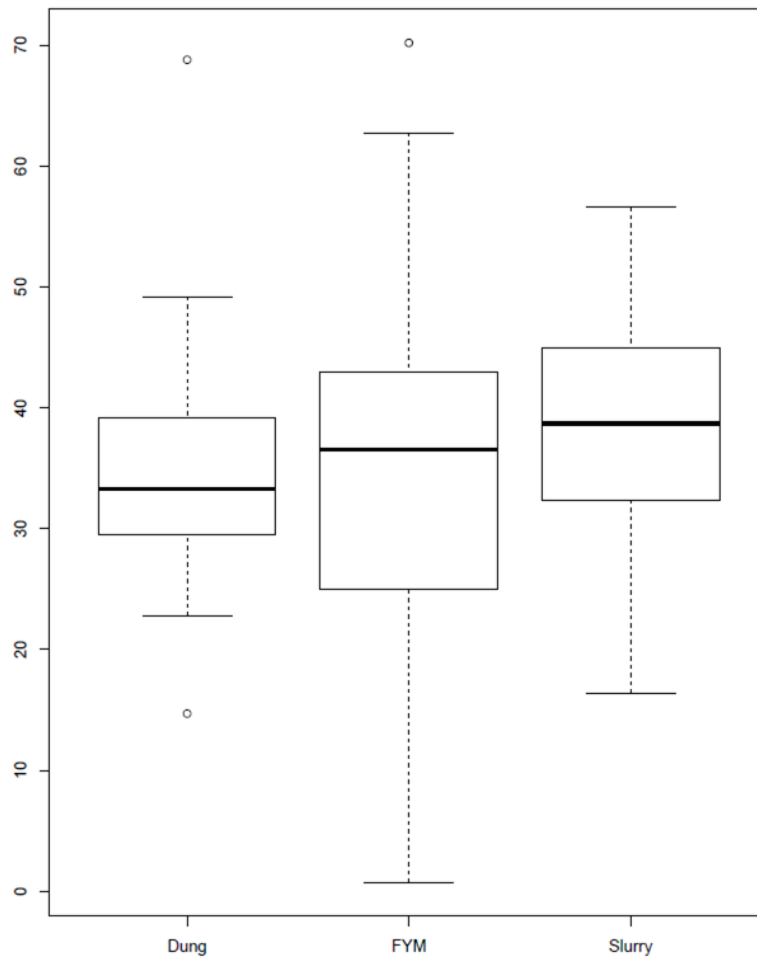


Fig.S1. Structure of C input derived from EOM in RothC modified model. (EOM, exogenous organic matter; DPM, decomposable EOM; RPM, resistant EOM; HUM, humified EOM)



*FYM, Farmacyard manure

Fig.S2. Boxplot displaying the lignin Van Soest fraction variability for the different ruminant residue's types (Dung, Farmacyard manure, slurry), based on literature review findings



*FYM, Farmyard manure

Fig.S3. Boxplot displaying soluble Van Soest fraction variability for the different ruminant residue types (Dung, Farmyard manure, slurry), based on literature review findings

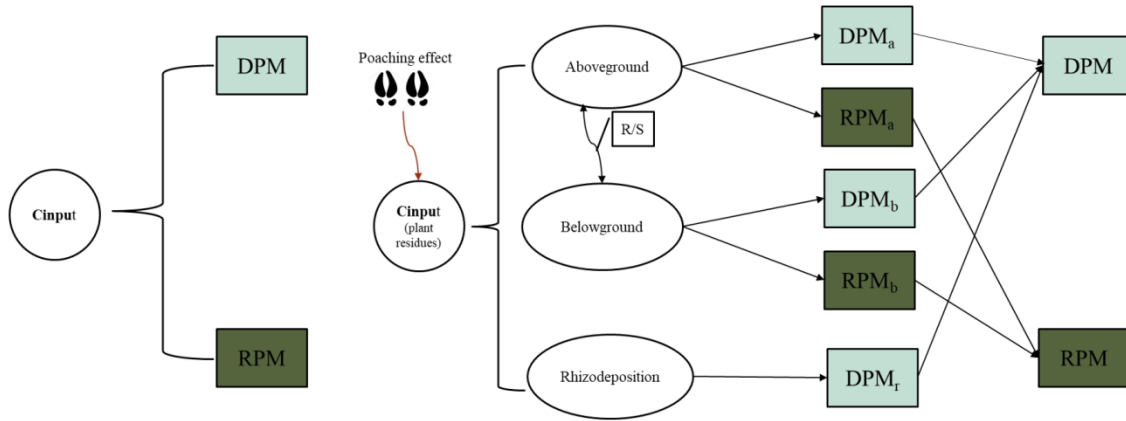


Fig. S4. Structure of C input derived from plant residues in RothC modified model. (DPM: decomposable plant material; RPM: resistant plant material; DPM_a, decomposable above-ground plant material; RPM_a, resistant above-ground plant material; DPM_b, decomposable below-ground plant material; RPM_b, resistant below-ground plant material; DPM_r, decomposable rhizodeposits)

$$\text{Belowground Biomass} = \text{Aboveground Biomass} \times R:S$$

(S2)

$$\text{Belowground Residue} = \text{Belowground Biomass} \times 0.5$$

(S3)

$$\text{Aboveground Residue} = \text{Aboveground Biomass} \times \text{fraction (Harvest or grazing)} \times 0.5 \quad (S4)$$

Equations of cattle poaching effect modification

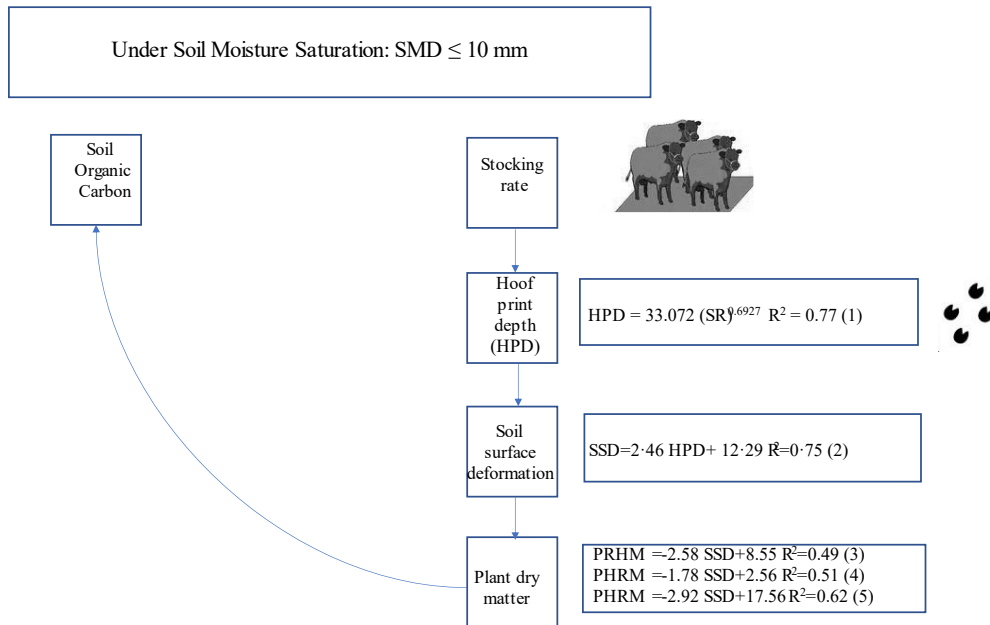


Fig. S5. Conceptual diagram of how the animal trampling effect is simulated to affect SOC dynamics

Hoof print depth (HPD) is function of stocking rate (SR) depending on the soil texture.

HPD is expressed in mm and stocking rate (SR) is expressed in number of cows/ha (Average live weight =550kg).

Equations are deduced from experiments in Tuñon et al. (2014).

For example, for poorly drained soils: $HPD = 33.072 (SR)^{0.6927} R^2 = 0.77$ (S5)

Soil surface deformation (SSD) is significantly correlated with HPD ($SSD = 2.46 HPD + 12.29 R^2 = 0.75$) (A2) (Tuohy et al. 2014). It is expressed in m/m.

The proportional reduction in herbage dry mass (PRHM) following each treading event. This proportion is without unit and was applied for plant C input according to Phelan et al. (2013) equations.

$$PRHM = -2.58 SSD + 8.55 R^2 = 0.49 \text{ (Early-spring turnout with an annual fertilizer input of } 100 \text{ kg N ha}^{-1}\text{)}. \quad (S6)$$

$$PRHM = -1.78 SSD + 2.56 R^2 = 0.51 \text{ (Early-spring turnout with no Fertilizer-Ninput)}. \quad (S7)$$

$$PRHM = -2.92 SSD + 17.56 R^2 = 0.62 \text{ (Late-spring turnout with no Fertilizer-Ninput)}. \quad (S8)$$

Where SSD is expressed in cm/m.

Supplementary information B. Study sites description and input data

1. Study sites description

The Laqueuille intensive site is a semi-natural grazing grassland (2.81 ha). The soil is classified as Andosol (20% clay, 53% silt and 27% sand) (FAO classification). The site was continuously grazed by heifers (1.1 SR/ha/yr) from May to October without additional feed supply, and fertilized with 210 kg N ha⁻¹year⁻¹ (ammonium nitrate) in three splits (more details on Klumpp et al. (2011) and Touhami et al. (2013)). The Oensingen intensive site is cutting grassland. The soil is classified as Stagnic Cambisol (Eutric) (FAO, ISRIC and ISSS, 1998). The field has been sown with grass- clover mixtures since 2001 and is mown 4 times per year and fertilised with 214 kg N ha⁻¹year⁻¹ (as solid ammonium nitrate or liquid cattle manure) at the beginning of each growing cycle (Ammann et al. 2009). The Easter Bush experimental site is under permanent grassland grazing management. The soil is classified as Eutric Cambisol (FAO classification) and is imperfectly drained. Grazing in this site occurs all year round by heifers in calf, ewes and lamb, which always have access to the entire field (more details on Skiba et al. (2013) and Jones et al. (2016)). The Solohead site is a dairy research farm with poorly drained soils. From 2004 to 2011, a typical grassland management involved rotational grazing (Necpálová et al. 2013).

2. Input data for the model and main assumptions

Average monthly temperature and precipitation for Laqueuille, Oensingen and Easter Bush sites were obtained from onsite Meteorological Stations for the periods 2004-2012, 2004-2011 and 2004-2010 respectively. For Solohead dairy farm, climatic data were provided by the Irish Meteorological Service referring to the nearest synoptic station with available climatic data for the simulation period 2004-2011. Monthly potential evapotranspiration was estimated using Thornthwaite equations (Thortwaite 1948) in case of non-availability of data.

Supplementary information C. Modified model performance

Table S1. Model performance measurement indices

Performance measure	Equation	Unit	Value range and purpose
BIAS, mean difference of simulations and observations (Smith and Smith 2007)	$BIAS = \bar{P} - \bar{O}$	Unit of the variable	negative to positive infinity: the closer the values are to 0, the better the model (negative values: underestimation; positive values: overestimation)
RMSE, Root Mean Square Error (Smith and Smith 2007)	$RMSE = \frac{100}{\bar{O}} \times \sqrt{\frac{\sum_{i=1}^n (O_i - P_i)^2}{n}}$	%	0 to positive infinity: the closer the values are to 0, the better the model
EF, Model efficiency (Smith and Smith 2007)	$EF = 1 - \frac{\sum_{i=1}^n (P_i - O_i)^2}{\sum_{i=1}^n (O_i - \bar{O})^2}$	-	Negative infinity to 1 (optimum): the closer the values are to 1, the better the model

P, predicted value; O, measured value; n, number of P/O pairs; i, each of P/O pairs; \bar{O} , mean of measured values; \bar{P} , mean of predicted values.

Supplementary information D. Sensitivity analysis

Table S2. NDF range of perennial ryegrass

NDF min	NDF max	Reference
48.9	52.2	(Boudon and Peyraud 2001)
36.5	53.5	(De Boever et al. 2013)
40.7	49.4	(Elgersma and Søegaard 2016)
45.8	58.5	(Ergon et al. 2016)
43	52.7	(Frandsen 1986)
47.8	57.4	(Küchenmeister et al. 2013)
41.1	54	(Lee et al. 2002)
44	63.2	(Ohlsson et al. 2007)
39	53	(Purcell et al. 2012)
41.4	69.3	(Salama et al. 2017)
38.7	42.5	(Van Vuuren et al. 1993)
39.4	57.8	(Armstrong et al. 1986)
46.1	55.2	(Østrem et al. 2014)
32	47	(Salama et al. 2012)
49.8	57.4	(Sun et al. 2010)

Table S3. R:S ratio for different temperate grassland species (A) and under different Nitrogen supply (B)

(A)

Grass species	Root to shoot ratio	Study
Grass	1.27	(Bessler et al. 2016)
Tall Fescue	0.55	(Bolinder et al. 2002)
Timothy	0.81	(Bolinder et al. 2002)
Ryegrass	0.65	(Bolinder et al. 2002)
<i>Lolium perenne</i>	0.89	(van Eekeren et al. 2010)
<i>Festuca arundinacea</i>	0.72	(van Eekeren et al. 2010)
<i>Dactylis glomerata</i>	0.51	(van Eekeren et al. 2010)
Mixture of <i>Lolium perenne</i> and <i>Festuca arundinacea</i>	0.85	(van Eekeren et al. 2010)
Mixture of <i>Lolium perenne</i> and <i>Dactylis glomerata</i>	0.50	(van Eekeren et al. 2010)
Pasture	0.66	(Kuzyakov 2006)

(B)

Grass or legume	Root to shoot ratio (Low N)	Root to shoot ratio (high N)	Study
Grassland in general		2.4	(Poeplau 2016)
<i>Lolium multiflorum</i>	0.32	0.21	(Henry et al. 2005)
<i>Lolium. perenne</i>	2.59	0.16	(Paterson and Sim 1999)
<i>Festulolium braunii</i>	0.69	0.38	(Mastalerczuk et al. 2017)
Perennial ryegrass	0.34	0.26	(Lehmeier et al. 2010)

Table S4. Sensitivity index of varying R:S ratio from its minimum to maximum values in RothC_4 for the different study sites

Site	Output (max value)	Output (min value)	Sensitivity index
Laqueuille	122.13	114.77	6.3%
Easter Bush	89.28	86.35	3.4%

Table S5. Sensitivity index of varying lignin content corresponding to animal excreta quality from its minimum to maximum values in RothC_4 for the different study sites under C input quantity (derived from animal excreta) scenario of 2.5 t C ha⁻¹ year⁻¹

Site	Output (min value)	Output (max value)	Sensitivity index (%)
Laqueuille (2.5 t C ha ⁻¹)	127.0	133.1	4.7
Oensingen (2.5 t C ha ⁻¹)	74.2	79.5	6.6
Easter Bush (2.5 t C ha ⁻¹)	91.7	96.8	5.3

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Chapter 3

Supplementary Information A: Study area

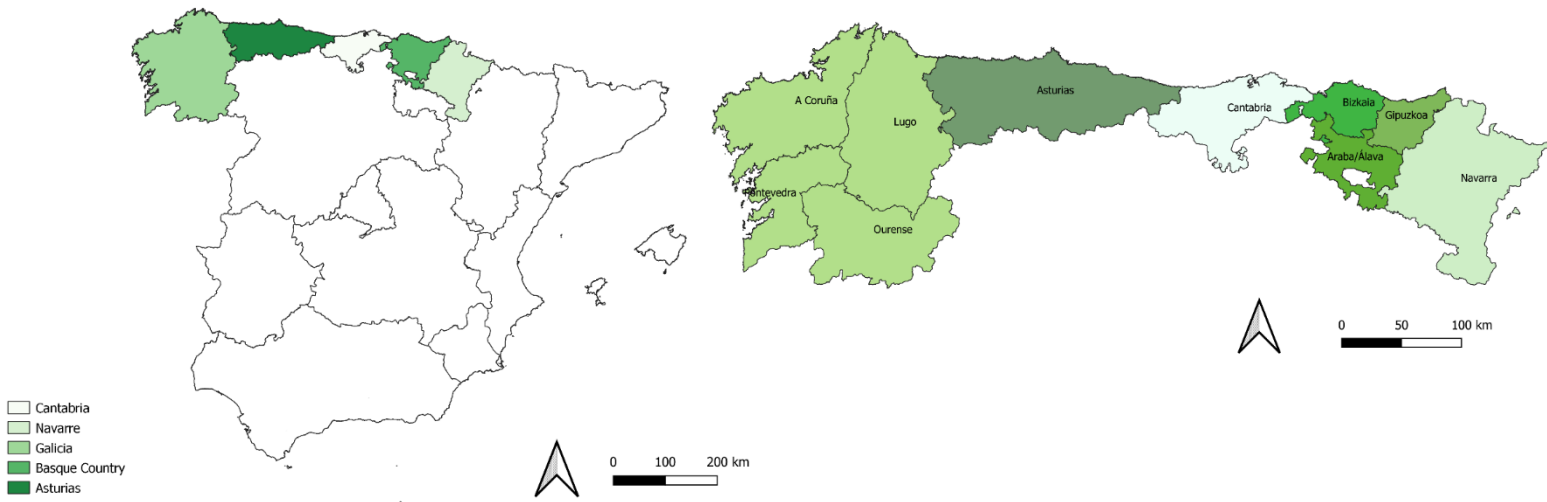


Fig. S1. Autonomous communities of the study area and its localisation

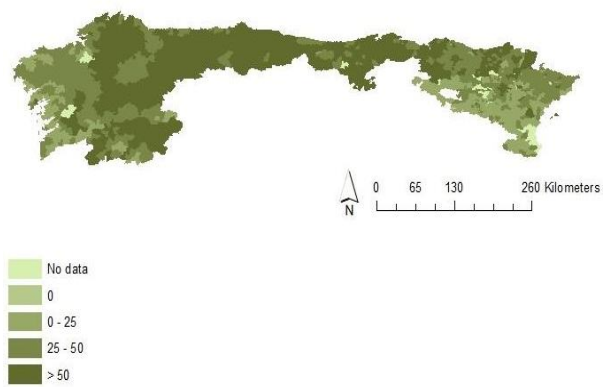


Fig. S2. Percentage of permanent pastures compared to other land uses per municipality in Northern Spain (INE)

Table S1. Dairy production per farm (Average per Autonomous region of Northern Spain)

	kg/farm	kg/lactatin g cow	kg/dairy cow	kg /ha
Asturias	216,863	7,461	6,947	11,015
Cantabria	267,565	7,365	6,949	9,956
Galicia	213,522	7,204	7,083	9,378
Navarra	680,811	8,798	8,584	17,581
Basque Country	425,516	8,492	8,761	12,871

(Flores-Calvete et al. 2016)

Asturias and Cantabria presented the highest percentage of dry matter intake from pasture for lactating dairy cows (22.8 and 20.8 %, respectively) with major percentage of fresh grass in the average annual DM diet (23.7 and 21.1 %, respectively) (Table S2). However, the concentrate portion of average annual DM diet ranges between 31.1% in Galicia and 39.6% in Cantabria (Table S2).

Table S2. Composition of annual feed ration for lactating dairy cows of the different Autonomous Communities of Northern Spain

	Dry matter % for each ingredient of the diet					Dry matter % consumed in	
	grass	grass silage	maize silage	dried forage	concentrates	stable	pasture
Asturias	23.7	21	8.3	15.5	31.4	77.2	22.8
Cantabria	21.1	18.2	6.8	14.3	39.6	79.2	20.8
Galicia	13.1	29.5	15.6	10.7	31.1	86.9	13.1
Navarra	10.4	15.9	22.6	12.2	38.9	89.9	10.1
Basque Country	14.9	20.8	6.2	21.4	36.7	85.2	14.8

(Flores-Calvete et al. 2016)

Table S3. Composition of annual feed ration for dry dairy cows in Northern Spain

Diet	Ingredients	Dry matter % for each ingredient of the diet
Forage (74%)	Cereal straw	37.9
	Ryegrass	62.1
Concentrates (26%)	Corn flour	44.1
	Barley flour	10.3
	Soybean Flour 44	40
	Calcium carbonate	5.6

Table S4. Composition of annual feed ration for heifers in Northern Spain

Diet	Ingredients	Dry matter % for each ingredient of the diet
Forage (76%)	Cereal straw	30.3
	Ryegrass	69.7
Concentrates (24%)	Barley flour	52.7
	Rapeseed meal 33	46
	Calcium carbonate	1.4

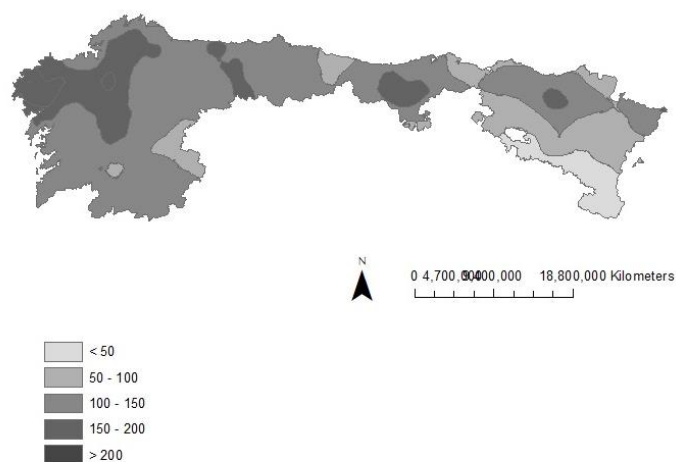


Fig. S3. Soil organic carbon stocks (Mg C ha⁻¹) in the study area

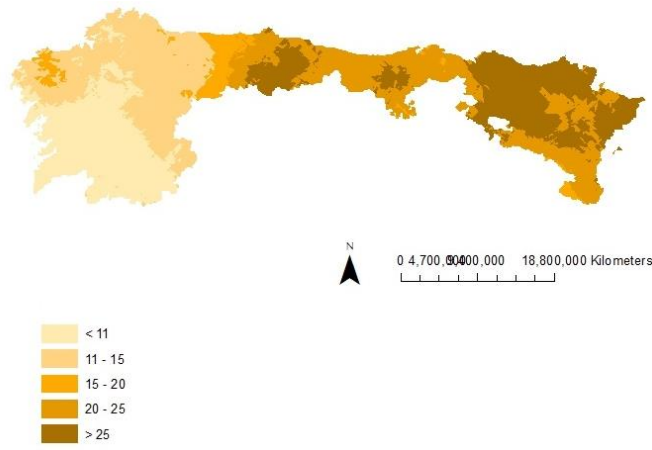


Fig. S4. Clay percentage in the study area

Supplementary Information B: Nitrogen excreta for lactating dairy cows and GHG emissions

1. Nitrous oxide emissions

Nitrogen excreta

According to the recent IPCC Tier 2 refinement, the annual amount of N excreted by each livestock category depends on the total annual N intake and total annual N retention of the animal. We estimated N excretion rates for the different dairy cows' sub-categories (i.e., lactating cows, dry cows and heifers) for each municipality region. For instance, considering lactating dairy cows, annual N intake (i.e., the amount of N consumed annually) depends on the annual amount of feed digested by the dairy cow, and the protein content of that feed. This latter depends on the production level of the lactating dairy cow (e.g., milk production). On the other hand, annual N retention (i.e., the fraction of N intake that is retained by the animal to produce milk) presents a measure of the animal's efficiency of production of animal protein from feed protein.

$$N_{\text{ex}} = (N_{\text{intake}} - N_{\text{retention}}) \cdot 365$$

N_{ex} : annual N excretion rates, kg N dairy cow⁻¹ yr⁻¹;

N_{intake} : annual N intake per head of dairy cow, Kg N dairy cow⁻¹ day⁻¹;

$N_{\text{retention}}$: fraction of daily N intake that is retained by dairy cow.

$$N_{\text{intake}} = \frac{GE}{18.45} \cdot \frac{CP\%}{6.25}$$

GE: gross energy intake of the animal based on digestible energy, milk production, pregnancy, weight and IPCC constants, MJ lactating dairy cow⁻¹ day⁻¹;

18.45: conversion factor for dietary GE per kg of dry matter, MJ kg⁻¹;

CP%: percent crude protein in dry matter;

6.25: conversion from kg of dietary protein to kg of dietary N, kg feed protein (kg N)⁻¹.

$$N \text{ retention} = \frac{\text{Milk} \cdot \frac{\text{Milk PR}\%}{100}}{6.38}$$

Milk: milk production, kg lactating dairy cow⁻¹ day⁻¹;

Milk PR%: percent of protein in milk, calculated as [1.9 + 0.4 • %Fat], where %Fat is an input, assumed to be 4%;

6.38: conversion from milk protein to milk N, kg Protein (kg N)⁻¹.

It is worth to point out that weight gain part is omitted from N retention equation for lactating dairy cows as it is assumed to be null for this animal category.

We used Tier 2 of IPCC (2019) methodology to estimate N₂O emissions produced, directly and indirectly, during the storage and treatment of manure as well as direct and indirect soil N₂O emissions (derived from animal excreta, applied fertilisers and deposited dung and urine from grazing dairy cows to the pastures, crop residues and pasture renewal). Reported N₂O emissions are generated using N excretion results and emission factors for N₂O emissions, as well as volatilization and leaching factors; with total related N₂O emissions equalling the sum of direct and indirect emissions. Periodic pasture renewal was derived from Calvete et al report of 2016 for each region (Flores-Calvete et al. 2016a). Management information of fertilization and amounts of N fertilizers were obtained from expert knowledge of most common local dairy farmer practices.

It is worth to notice that we estimated emissions of NH₃ arise from the excreta of the dairy cow system (deposited in and around buildings housing livestock and collected as liquid slurry, solid manure or litter-based farmyard manure (FYM)) according to EMEP methodology.

2. Methane emissions

CH₄ derived from enteric fermentation

The CH₄ emission factor is based on an estimation of feed intake and a methane conversion factor (Y_m) as IPCC (2019). The methane conversion factor was estimated according to typology of diet ration reflected for each dairy cows' sub-category and region depending on NDF and digestibility of the annual diet ration (IPCC, 2019). Gross energy intake was calculated as outlined in the IPCC Tier 2 methods (IPCC, 2019). In order to estimate total emissions, the emission factor is multiplied by the associated dairy cows' sub-category number and summed for each municipality of our study area.

$$EF = \frac{GE \cdot \frac{Y_m}{100}}{55.65}$$

Where EF: emission factor (kg CH₄ head⁻¹ yr⁻¹); GE: gross energy intake (MJ head⁻¹ yr⁻¹); Y_m: methane conversion factor (% of GE in feed converted to methane); The value 55.65 refers to the energy content of methane.

CH₄ emissions derived from manure management

We estimated CH₄ produced during the production in the house and storage and treatment of dairy cows' excreta (as slurry), and from dairy cows' excreta deposited on pasture whilst grazing. Emissions from manure management depend on animal excreta management system characteristics and manure characteristics (i.e., Volatile Solids (VS)), which were estimated based on feed intake and digestibility, used to estimate enteric fermentation emission factor (IPCC, 2019) (Eq. 3). Manure of lactating dairy cows is stored as slurry, while manure of dry dairy cows and heifers is handled as farmyard manure (FYM). In order to determine the MCF (i.e., Methane conversion factor) slurry, we referred to the IPCC 2019 suggested model (IPCC, 2019). The MCF model requires monthly air temperature profiles as well as the average number and timing of the emptying of manure storages. The VS and maximum methane producing capacity for residues, based on IPCC guidance and percentage of excreted VS handled as a liquid are also additional input parameters. The model calculations run for three years, in order to ensure VS available has stabilized on an annual basis. In our study, the MCF model was run for the different municipalities of our study area. As manure is managed in multiple systems considering municipalities of our study area, manure emission factors were allocated to the dominant storage systems of the different corresponding regions of the study area.

$$EF = VS \cdot \left[0.24 \cdot 0.67 \cdot \left(\frac{MCF}{100} \right) \cdot ARMS \right]$$

Where EF: annual CH₄ emission factor for dairy cows (Kg CH₄ dairy cow⁻¹ yr⁻¹); VS: volatile solid excreted for dairy cows (Kg dry matter dairy cow⁻¹ yr⁻¹); 0.24: maximum methane producing capacity for residues produced by lactating dairy cow, it is of 0.18 for heifers and dry cows (m³ CH₄ kg⁻¹ of VS excreted); 0.67: conversion factor of m³ CH₄ to kg CH₄; MCF: methane conversion factor for each residue management system (%); ARMS: fraction of dairy cow's residues handled using animal excreta management system.

Supplementary Information C: SOC change rate (scenario absence of C inputs from dairy cows)

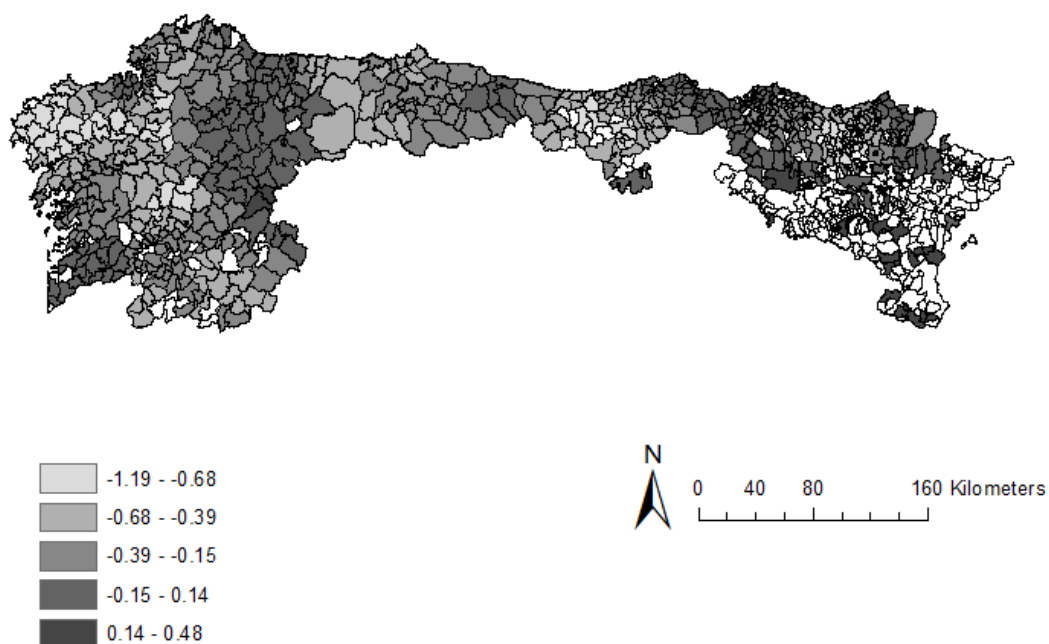


Fig. S5. Soil Organic Carbon stock change rates ($\text{Mg C ha}^{-1} \text{ year}^{-1}$) of dairy cows' pastures in Northern Spain municipalities under the scenario of C inputs from dairy cows = 0

Supplementary Information D: Uncertainty analysis (Monte Carlo simulation)

Table S5. Overview of normal probability distributions of the selected parameters used in the Monte Carlo analysis

	Min	Max	Mean	Standard deviation
Animal excreta ($\text{T C ha}^{-1} \text{ year}^{-1}$)	0	5	2.5	0.5
Plant dry matter ($\text{T C ha}^{-1} \text{ year}^{-1}$)	3.4	11.3	7.4	1

Table S6. Range of variability in SOC stocks (Mg C ha^{-1}) due to C inputs uncertainty for the selected municipalities. The table represents the results of 1000 runs of the modified RothC model with parameters related to C inputs randomly drawn from their expected distribution.

Municipality	Min	Max	Mean	Simulation	Difference
A Coruña	110.1165	126.2282	118.17	123.7637	5.59
Castro de Rei	121.7161	140.5844	131.15	133.7054	2.56
Donostia	109.4063	124.0013	116.70	120.4098	3.71
Ourense	87.04305	101.1538	94.10	98.14938	4.05
Oviedo	103.7383	119.8977	111.82	114.2905	2.47
Santander	109.533	126.5408	118.04	122.8765	4.84
Vitoria-Gasteiz	93.98041	111.2675	102.62	100.3101	-2.31

Supplementary Information E: Soil Organic Carbon change

Table S7. Literature review of SOC change rate at temperate grassland sites using different methods

Soil sequestration rate (Mg C ha ⁻¹ year ⁻¹)	Methodology	Description	Period	Depth (cm)	Location	Reference
0.75	Metanalysis	Ex-arable grasslands of the temperate zone		30	Temperate zone	(Kämpf et al. 2016)
0.3	Measuring and modelling	Cut and fertilized grassland	7		Easter Bush in South-east Scotland	(Jones et al. 2017)
-0.51-8.05	Dynamic chambers	Mineral or organic fertilization		40	South of Edinburgh in Scotland	(Jones et al. 2006)
0.37(± 0.01)	DNDC model	Cutting fertilized grassland		15	Hillsborough, County Down, Northern Ireland, UK	(Khalil et al. 2020)
0.46 (± 0.06)	Measurement	Cutting fertilized grassland		15	Hillsborough, County Down, Northern Ireland, UK	(Khalil et al. 2020)
0.28	Measurement	Fertilization phosphorus (P), and potassium (K)	34	30	Germany and the Netherlands	(Poeplau et al. 2018)
0.13	Measurement	Fertilization nitrogen (N) phosphorus (P), and potassium (K)	34	30	Germany and the Netherlands	(Poeplau et al. 2018)
0.37	Measurement	Increased Fertilization nitrogen (N), phosphorus (P), and potassium (K)	20	30	Germany and the Netherlands	(Poeplau et al. 2018)

-0.07-0.56 (±0.188)	Measurement	Permanent pasture with manure fertilization	35		Park Grass experiment, United Kingdom	(Poulton et al. 2018)
0.02(± 0.01)	Metanalysis	Herbivore exclusion Temperate grasslands	100		Temperate grasslands	(Tanentzap and Coomes 2012)
0.05(± 0.01)	Inventories	Temperate grasslands			Temperate grasslands	(Soussana et al. 2010)
0.22(± 0.56)	C flux balance	Temperate grasslands			Temperate grasslands	(Soussana et al. 2010)
-1.1-0.5	Modelling	Managed grasslands	20	30	France	(Soussana et al. 2004)
0.6-1.5	Measurement	Continuous grazing	2-5	30	Netherlands	(Hoogsteen et al. 2020)
0.6-1.9	Measurement	Lenient strip grazing	2-5	30	Netherlands	(Hoogsteen et al. 2020)
0.3-1	Measurement	Rotational grazing	2-5	30	Netherlands	(Hoogsteen et al. 2020)
2.21	Soil inventories	Semi-natural pasture grazed by heifers	>10 years	60	Laqueuille site France	(Herfurth 2015)
2.29	Net Carbon Storage via Eddy covariance technique	Semi-natural pasture grazed by heifers	>10 years		Laqueuille site France	(Herfurth 2015)
0-8	Review	Temperate grasslands			Temperate grasslands	(Jones and Donnelly 2004)

Table S8. Matrix of correlation

	SOC_r	C input	SOC_i	Clay	MAT	MAP
SOC_r	1.00					
C input	0.76	1.00				
SOC_i	-0.61***	-0.19***	1.00			
Clay	0.14***	0.15***	-0.27***	1.00		
MAT	-0.19***	0.09	-0.07**	-0.09**	1.00	
MAP	-0.26	0.15	0.43***	-0.05**	0.02	1.00

*** p<0.01, ** p<0.05.

SOC_r is the annual SOC change rate; SOC_i is the initial SOC content; Clay is the soil clay percentage; C input is the C input derived from vegetation and animal manure; MAT is mean annual temperature and MAP is annual precipitation.

Supplementary Information F

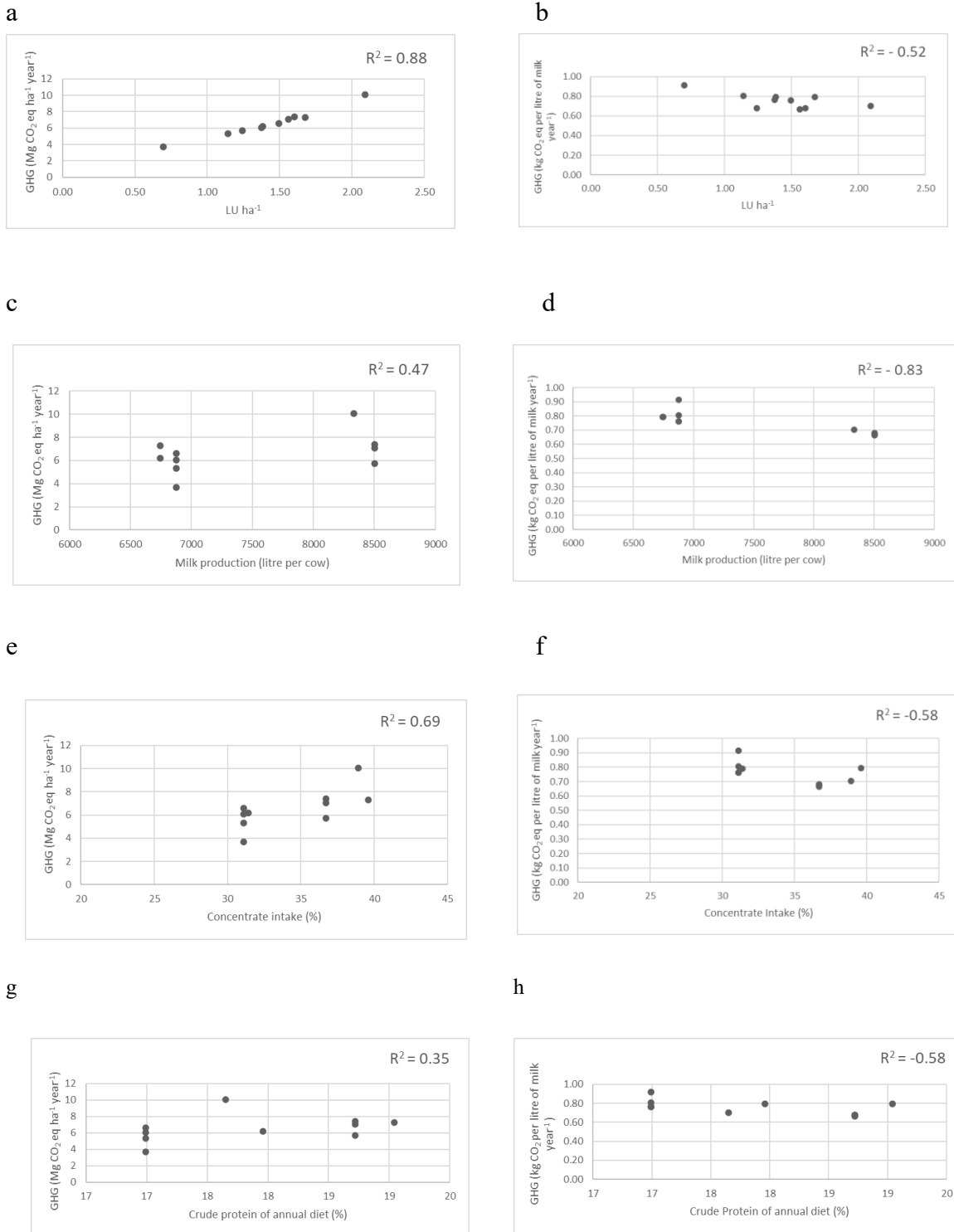


Fig. S6. Greenhouse gas emissions in Mg CO₂-e per ha per year in relation to livestock density (LU. ha⁻¹) (a), milk production (litre per cow) (c), level of concentrate feeding in annual feed ration for lactating dairy cows (e) and crude protein percentage of annual feed ration for lactating dairy cows (g) and in kg CO₂-e per L of milk per year in relation to livestock density (LU. ha⁻¹) (b), milk production (litre per cow) (d), level of concentrate feeding in annual feed ration for lactating dairy cows (f) and crude protein percentage of annual feed ration for lactating dairy cows (h).

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Chapter 4

Supplementary information A: Study area Localisation and characteristics

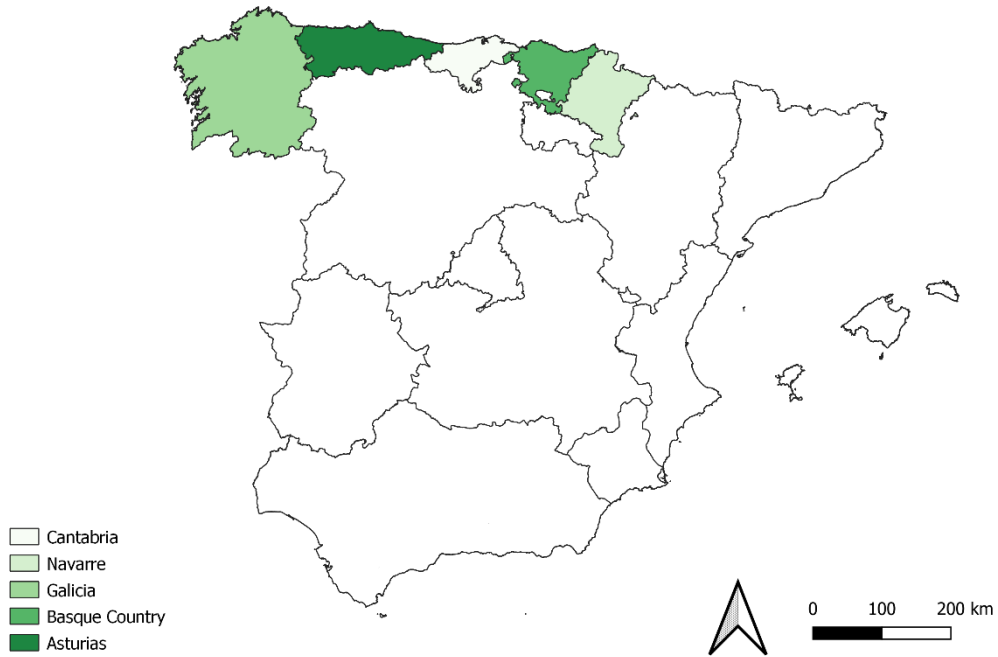


Fig. S1. Autonomous communities of the study area, its localization in Spain

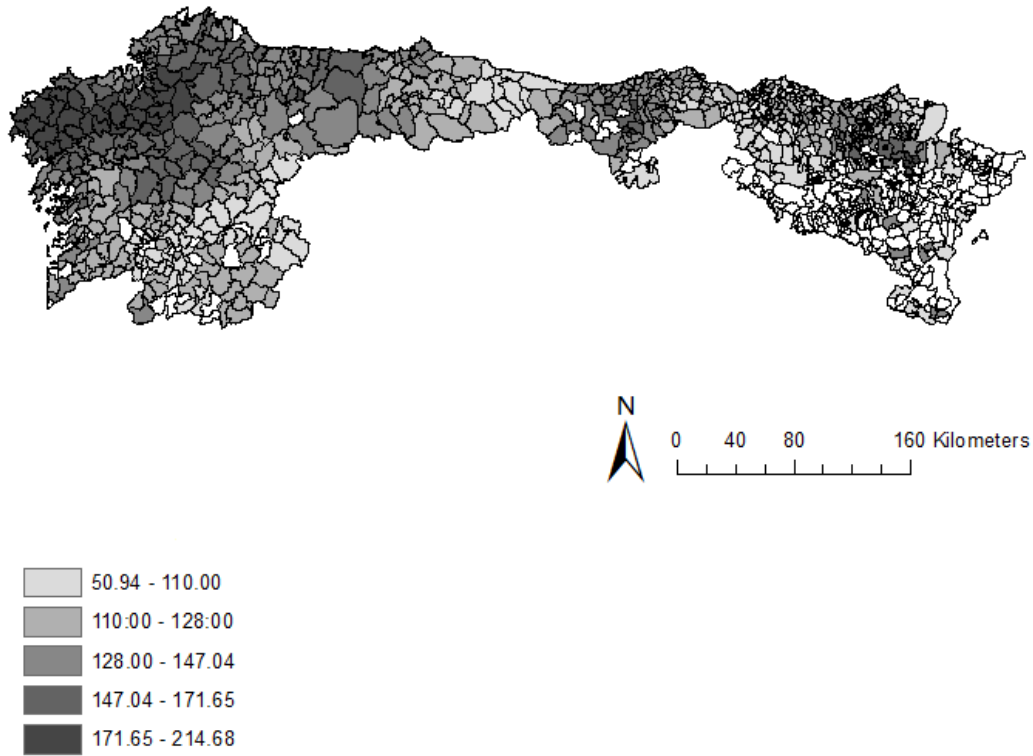


Fig. S2. Initial soil organic carbon content (Mg C ha^{-1}) in 2100 for the different municipalities of our study area (Simulation results of the previous study of the same authors)

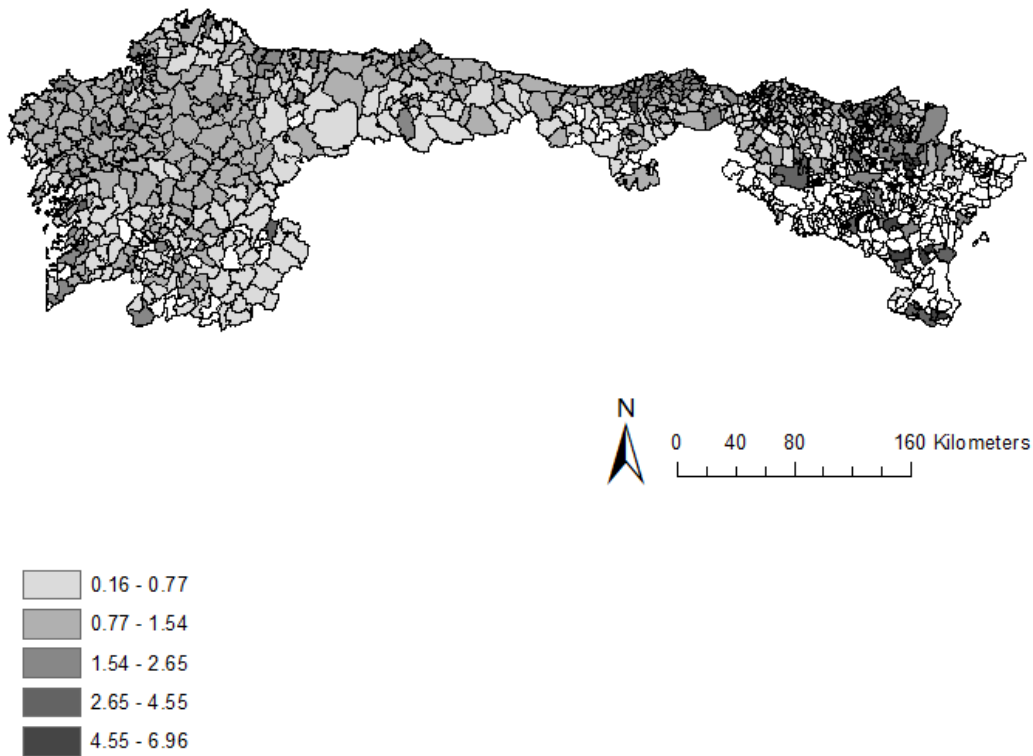


Fig. S3. Carbon input derived from dairy manure (t C ha^{-1}) applied in the grasslands associated to dairy production of Northern Spain per municipality