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Soil organic matter dynamics in Portuguese natural and sown rainfed grasslands

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ABSTRACT

Soil organic matter (SOM) is a particularly important parameter in soil management, especially in mineral soils in Mediterranean and semi-arid countries where its concentration is low. In these conditions, increasing SOM concentration has several agronomic and environmental benefits, ranging from increase in water holding capacity to soil protection and carbon sequestration. We develop a model to express the short-term trend of SOM increase in grasslands as the balance between input and mineralization. This model is calibrated using five years of soil analyses from eight locations. In each location there were either two or three plots with the different grassland systems considered: sown biodiverse permanent pastures rich in legumes (SBPPRLs), fertilized natural grasslands (FNGs), and (un-improved) natural grasslands (NGs). SBPPRL are a new system consisting in the use of plant biodiversity to increase pasture productivity and resilience. So far, they exist mostly in Portugal.

We use statistical calibration to adjust an asymptotic curve to the data and obtain the model parameters. Under the assumption of equal mineralization rates across grassland systems, we find that the expected steady-state long term SOM concentration in undisturbed SBPPRL is higher than in NG and FNG. Fertilization does not significantly increase SOM input, and so the trend in SOM is equal for NG and FNG. In 10 years, there is an average increase of 0.21 percentage points per year in SBPPRL. In turn, SOM increases in FNG and NG are 0.08 percentage points per year.

1. Introduction

Since the beginning of the domestication of livestock by humans, fields of herbaceous plants, named in this sense as “pastures” or “grasslands”, have been used to feed animals (Suttie et al., 2005). In Portugal, there are now around 1.5 million hectares of grasslands (INE, 2010), divided between three grassland systems: natural grasslands (NGs), fertilized natural grasslands (FNGs), and sown biodiverse permanent pastures rich in legumes (SBPPRLs). These three types of pasture correspond to three different degrees of intensification.

NG is by far the most used grassland system in Portugal. It consists of either fallow stages from long cereal rotations, or spontaneous vegetation in previous croplands which have since been converted to areas for livestock feed. NG typically has no specific management, except for occasional operations to control shrub growth. The most widely used operation is tillage.

The only difference between NG and FNG is that the latter is fertilized. The species and varieties of spontaneous grasses and legumes are the same, but fertilization increases productivity. As a consequence, shrub control has to be done more frequently. Advocates of FNG claim, though, that fertilization is a compromise between productivity and natural values. Furthermore, methods of shrub control other than tillage will benefit the soil nutrient recycling system. Shrubs have deeper roots than grasses and legumes, and therefore access nutrients in deeper layers of soil to grow. Control operations will then shred their aboveground biomass. This biomass remains on the ground and is incorporated in the first layer of soil, which is then used by pasture plants to grow.

However, some other farmers and agricultural scientists believe that fertilization alone does not provide the best results in terms of plant productivity and animal feed quality. Advocates of sown pastures believe that the introduction of specific species or varieties,

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either absent or in lesser percentage in spontaneous grasslands (as, for example, some varieties of legumes) will establish a functioning ecosystem with complementary ecological niches and improve production.

This line of thought led to the development of the SBPPRL system in Portugal in the 70's decade. SBPPRL consist of diverse mixes of up to twenty different species or varieties of seeds, and are rich in legumes. Commonly SBPPRL are more productive than natural grasslands, and are also richer in number of species (Carneiro et al., 2005). There are fewer gaps in plant cover throughout the plots, since species variability ensures that the species most suited for each spatial condition will thrive. Even though there is a well documented experience with the use of sown pastures (FAO/CIHEAM, 2008), this specific system only exists in Portugal and, to a lesser degree, Spain and Italy. There are many studies on the role of biodiversity in productivity, but SBPPRL remain the only widespread large-scale application of what may be called "biodiversity engineering".

The seed mix is designed specifically for each location after soil analysis. Species in the mix is adapted to soil physical and chemical characteristics, as well as to local climate conditions, and therefore there is no single representative mix. However, some very common sown species in SBPPRL are *Trifolium subterraneum*, *Trifolium michelianum*, *Ornithopus* spp., *Biserrula pelecinus*, annual *Medicago* spp., and grass species of the genera *Lolium*, *Dactylis* and *Phalaris*. The mixes of sown species are often enriched with seeds from spontaneous plants such as *Plantago* spp., *Vulpia* spp. and *Bromus* spp. (David Crespo, personal communication). Legumes are thus very common in these mixtures and cover more than 50% of first-year SBPPRL (Carneiro et al., 2005). As pasture settlement progresses, legumes increase and eventually dominate. Percentage of legumes in the plant cover of a mature SBPPRL (more than 5 years) is around 25–30%. Legumes are inoculated with bacteria of the genus *Rhizobium* which induce nitrogen-fixing nodules in the roots of legumes. The fixated atmospheric nitrogen is then used by grasses what makes the overall system self-sufficient in terms of nitrogen.

The higher plant productivity of SBPPRL implies increased atmospheric carbon capture through photosynthesis. Part of the biomass produced is stored in soils due to the high density of yearly-renewed roots. Storage is in the form of non-labile soil organic carbon (SOC), which is part of the soil organic matter (SOM) pools. SOM pools are also increased by leaves' senescence, and by animals returning undigested fibre to the soil.

Therefore, SOM is the key parameter in soil management (Bot and Benites, 2005). Its importance stands out in mineral soils of Mediterranean and semi-arid countries, where SOM concentrations are generally low. Even in those conditions, pastures provide particularly high and stable SOC pools. Guo and Gifford (2002) indicate that SOC stocks decline after land use changes from pasture to cropland by 59%, and increase after land use changes from crop to pasture by 19%. Martens et al. (2004) show that for systems undisturbed for 130 years, pastures' soils had 25% more carbon than cropped soils. Improvements such as fertilization and sowing in pastures increase SOM accumulation by increasing pasture productivity.

Increasing SOM improves soil nutrient availability and water holding capacity, thus increasing plant productivity and reducing surface runoff of water, which in turn decreases sediment loss and soil erosion (EEA, 2004). Decreasing water runoff and soil erosion have positive effects even outside the plot. Sediments, nutrients, organic matter and pesticides carried in water contribute to silting, eutrophication and contamination of surface waters. These effects are known, but their true costs are still hard to estimate. Nitrogen fixation by legumes eliminates the need for nitrogen fertilizers, whose production is highly energy demanding, and therefore responsible for high greenhouse gas emissions. Finally, both increased stocking rate allowed by the higher animal carrying capacity and reduced fertilizer use increase the economic viability of the farms. This is particularly important because socio-political and economic conditions are barriers to the successful implementation and management of pasture systems which provide environmental services (Neely et al., 2009).

Increasing SOM, nutrient availability and water in soils provides both mitigation and adaptation to climate change. SOM accumulation through an increase of SOC is the mechanism through which carbon is sequestered in grassland soils. This is particularly important for Portugal, being one of the few countries to elect the "Grassland Management" voluntary activity, in the framework of the Land Use, Land Use Change and Forestry (LULUCF) activities, now named Agriculture, Forestry and Other Land Uses (AFOLU), under Article 3.4 of the Kyoto Protocol. This choice was made mainly because of the implementation of the SBPPRL system in Portugal. However, there is currently no study published on the potential of the SBPPRL system to increase SOM. According to IPCC guidelines, Tier 1 approaches imply that one sequestration factor is attributed, regardless of related field data. The present work should be a first step: we study how much, on average, SBPPRL increase SOM, in relation to the baseline, which are natural grasslands.

In this paper we develop a model to determine the average trend of SOM concentration in NG, FNG and SBPPRL. Our main objective is to determine the average SOM accumulation potential in each grassland system. We hypothesise that the variation in SOM over time is the balance between SOM input and output in a plot. The implication of this model is that SOM asymptotically reaches a long-term equilibrium.

The dynamic parameters in this model are the SOM input and the mineralization rate. In order to estimate the values of these parameters, we calibrate the model statistically using field data. Data was collected from 2001 to 2005 in several locations in Portugal, during two demonstration projects. Project AGRO 87, "Sown biodiverse permanent pastures rich in legumes – a sustainable option for degraded land use" (Carneiro et al., 2005) collected samples in six farms. At the same time, Project PAMAF 4073, which was continued as Project AGRO 71, "Recovery and improvement of Alentejo's degraded soils using grasslands" collected samples from two additional farms. We filled-in some missing data, since some samples were not collected.

We use two statistical methods for calibration: one where all parameters are specific for each grassland system, and one where there is only a specific SOM input. We then compare the dynamics of the three systems in 10 years. Finally, we validate the results obtained and draw some conclusions. In order to do so, we compare our results with other data and studies.

2. Method

2.1. Characterization of the plots

Data was obtained from rainfed pastures in eight farms in Portugal from 2001 to 2005 (Table 1, location in Fig. 1). Plot areas ranged from 5 to 15 ha. Each plot's soil and landscape type was approximately homogeneous, in terms of soil and previous use. These pastures were not isolated test sites. They were located in private land currently used by farmers for animal production. Prior to the beginning of the projects, plots were used in a system of long cereal/fallow rotations – one year of crop production for each five to seven years of fallow (which was used as a "natural pasture" featuring spontaneous herbaceous plants). In Farm #1 the NG plot was fertilized in 2002, and so the NG system was lost. In Farms #7 and #8 (Project Agro 71), FNG were not studied. Almost no samples were collected in 2002.

Table 1
Soil and site characterization in the sites of Projects Agro 87 (farms 1–6) and Agro 71 (farms 7 and 8).

Farm No.	Farm	Location	Soil original material	Texture
1	H. Cabeça Gorda	Vaiamonte	Gneiss	Loam
2	H. Monte do Mestre	São Vicente	Limestone	Loamy clay
3	H. Claros Montes	Pavia	Granite	Loamy sand
4	H. Refróias	Cercal	Schist	Loamy sand
5	H. Cinzeiro e Torre	Coruche	Sandstone	Sand
6	Quinta da França	Covilhã	Granite	Loamy sand
7	H. Monte da Achada	Castro Verde	Schist and Greywacke	Sandy loam
8	H. Corte Carrilho	Mértola	Schist	Loamy sand

The main meteorological characteristics of test sites #1–#5 and #7–#8 are similar, with average daily temperature of 15.5–16.8 °C (APA, 2009; approximate mean figures for the period of 1930–1970) and yearly precipitation in the years from 2001 to 2005 of around 200–750 mm yr^{-1} (SNIRH, 2009). Farm #6 is the only one in Central Portugal, and shows lower average daily temperature (11.3 °C) and higher precipitation (as high as 1195 mm from October 2003 to September 2004).

Fertilization was used in SBPPRL and FNG depending on soil needs in each sampling sites, and was determined according to the results of initial soil analysis (Carneiro et al., 2005). Both grassland systems were subjected to the same fertilization rates during all years of the project. The difference during the installation of SBPPRL is that, previous to sowing, plots were tilled in the upper layer of soil, and a phosphate and potassium fertilizer (superphosphate 18% or 0:21:21) was used (ranging from 200 to 450 kg ha^{-1} in all farms). Limestone was added in SBPPRL if soil pH was lower than 5.3 to lower acidity to optimum levels for legumes (1 t ha^{-1} in farms #5 and #7, and 2 t ha^{-1} in farms #3, #4 and #6). Other micronutrients were added, like zinc sulphate copper sulphate or borax. Molybdate (a salt of molybdic acid) was added together with the seeds. SBPPRL were installed using 30 kg ha^{-1} of seeds.

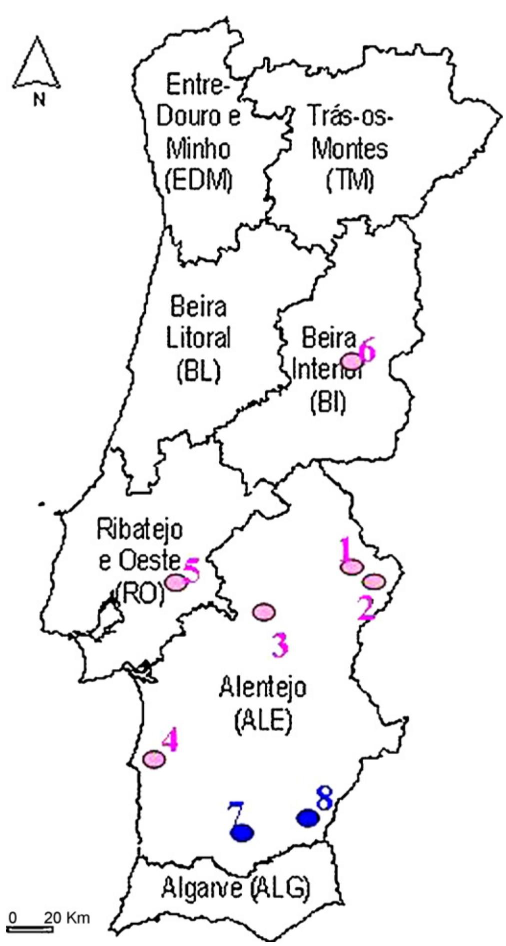

Fig. 1. Map of Portugal, with the indication of the sampling sites of Projects Agro 87 (farms 1–6) and Agro 71 (farms 7 and 8).

Throughout the project, farmers registered the number of animals put on each plot each day. That information was then averaged as cattle units (CU) in a year (also considering the days when there were no animals in the pasture) (Carneiro et al., 2005). One CU is the equivalent of one adult cow (a steer corresponds to 0.6 CU and an ewe to 0.15 CU). Results have shown that SBPPRL support higher stocking rates. The average stocking rate between 2001 and 2004 was 1.0 CU yr^{-1} in SBPPRL and 0.43 CU yr^{-1} for FNG and NG. In 2004–2005 figures are much lower because of a severe drought in Portugal, but they were still higher for SBPPRL (0.36 CU yr^{-1} against 0.14 CU yr^{-1} for FNG/NG).

2.2. SOM data

SOM determination begins with collection of soil samples. One composite sample was collected in each plot. Each composite sample was obtained from the mix of a variable number of sub-samples collected throughout each homogeneous plot, in order to be representative of the average SOM in the plot. The samples were collected and analysed by Laboratório Químico-Agrícola Rebelo da Silva (LQARS), which is the Portuguese Government's official soil laboratory.

In general, SOM is composed of living organisms (bacteria, fungi, plant roots and animals), dead animal and plant tissues in several stages of decomposition but still recognizable, and a complex mixture of decomposed, modified or reprocessed material called humus (which is usually 60–80% of all SOM) (Bot and Benites, 2005). In the preparation of the sample for analysis, any living organisms, as well as gross animal and plant material, are removed from the sample. Therefore, results in this paper for SOM refer only to humus and some minor organic material.

Sample laboratorial analysis begins with the entire sample being spread on a tray and dried overnight, at 35–37 °C. The sample was crumbled mechanically and passed through a 2 mm stainless steel sieve. The sieved material is the 'fine soil' subject to analysis. Samples were then analysed for several parameters, such as pH, nitrogen, phosphate and potassium levels, lime requirement and SOM concentration. It is the latter parameter that we use in this work.

From 2001 to 2004, results for SOM concentration were obtained by the wet oxidation method. This method consists in the digestion of organic carbon by sodium dichromate, followed by colorimetric determination on a molecular absorption spectrophotometer at 640 nm (Carter, 1993). From 2005, a dry combustion method was used (including for samples collected in 2005). It consists of the determination of total carbon by dry combustion, according to ISO Standard 10694, using a CNS elemental analyzer (Rodeghiero et al., 2009). Organic carbon is determined indirectly after correction of the total carbon concentration for the carbonates present in the soil

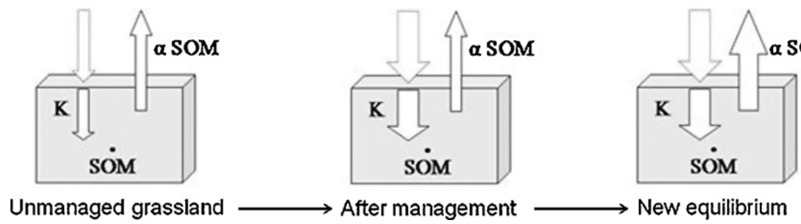

Fig. 2. Different estimation procedures used for the SOM dynamic model. K, α – parameters in Eq. (2); SOM – soil organic matter.

sample. Both methods measure SOC concentration, which is then multiplied by 1.724 (assuming that 58% of organic matter is carbon) to obtain the corresponding SOM concentration. Since two different methods were used, an unpublished study was conducted by LQARS to obtain an equivalence factor between results. This study guarantees that the results are consistent. Final results for SOM are presented in mass percentage (%) units, equal to grams of SOM per 100 g of soil.

There are few results available for 2002, in a set obtained from 2001 to 2005. Since our regression models (whose equations we show in the next section) use consecutive pairs of points ($SOM_{i,t-1}$, $SOM_{i,t}$), two pairs are almost always missing: (2001, 2002) and (2002, 2003). Furthermore, FNG are missing more pairs of points than SBPPRL and NG. Therefore, whenever there is a missing value between two others, we calculate the geometric average (the "growth rate" of SOM) of the two observations. For example, assuming that SOM_{t-1} is missing, while SOM_{t-2} and SOM_t are not, we calculate the missing value as:

$$SOM_{t-1} = SOM_{t-2} \cdot \left(\frac{SOM_t}{SOM_{t-2}}\right)^{1/2}. \quad (1)$$

2.3. SOM dynamic model

As the thorough review done by Falloon and Smith (2009) shows, other models in the literature intend to explain inter- and intra-annual variability in SOM. To explain such variability, they are required to use environmental variables, such as climate and soil type. Out of the 33 models reviewed and assessed by these authors, only one (O'Brien, 1984) had a yearly time step and no meteorological and management variables. But the O'Brien model considered interactions with plants, and had a completely different objective than ours. However, in our case, it is the SOM trend we wish to estimate and not the interannual variation of SOM levels. Our objective is to calibrate a time series to capture the trend of SOM dynamics in the three grassland systems.

Therefore, we use a simple mass balance model for SOM dynamics, calibrated using field data. The model states that the mass percent balance of SOM is the difference between input and mineralization:

$$\frac{dSOM_t}{dt} = K_i - \alpha SOM_t, \quad (2)$$

where SOM is the SOM concentration (percentage points, equal to $g_{SOM}/100\,g_{soil}$) at time t, K is the SOM input, and α is the organic matter mineralization rate. We assume that the mineralization rate does not depend on the grassland system, since we are assuming that in a steady-state it is a fixed fraction of the SOM pool.

The schematic representation of the model is shown in Fig. 2. Steady-state grasslands balance SOM input (from plants) and mineralization. Changes in management (such as fertilization or sowing) create a transient state during which SOM accumulates. The increase in production increases SOM input, but mineralization has not yet adjusted to the new situation. As years pass, mineralization also increases, and a new steady-state is reached.

We solve Eq. (2) by integrating it between $t - \Delta t$ and t:

$$SOM_t = \frac{K_i}{\alpha}\left(1 - e^{-\alpha\Delta t}\right) + e^{-\alpha\Delta t} SOM_{t-\Delta t}. \quad (3)$$

Therefore, the general solution for Eq. (2) has a saturating exponential form. This means that SOM accumulation is limited by an upper bound. In pastures, if there are no land use conversions or other management activities, disregarding climate effects, SOM reaches a long-term equilibrium.

Note that we assume that SOM input (parameter K) is a function of the grassland system. Therefore, at least part of the SOM input has to reflect grassland productivity. But, as the inspection of the SOM analysis results in Table 2 will show, farms with high initial SOM still increased their SOM concentration by a relatively high percentage, regardless of the pasture type. To capture both these effects, we separate K_i in a fixed term K_i' (which is a function of grassland system, not of representative local conditions), and a variable part (which is a linear function of the initial SOM concentration, being the proportionality parameter a):

$$K_i = K_i' + aSOM_0. \quad (4)$$

Eq. (4) shows that we are using initial SOM as a proxy for representative conditions of the location. This approach is justified by the fact that natural soil and climate conditions, as well as the history of the field, determine the initial SOM concentration. This slightly changes the model. Substituting Eq. (4) in Eq. (3), we obtain the

Table 2
SOM concentration in each grassland system for each experimental site (0–10 cm).

Farm No.	Grassland system	SOM (%)				
		2001	2002	2003	2004	2005
1	SBPPRL	1.55	**2.17**	3.05	3.60	3.80
1	FNG	1.30	**1.84**	2.60	3.40	3.00
2	SBPPRL	1.75	**2.15**	2.65	2.70	5.40
2	FNG	1.95	**2.42**	3.00	4.50	3.50
2	NG	1.95	**2.29**	2.70	4.00	4.00
3	SBPPRL	**0.45**	0.73	1.20	1.63	1.60
3	FNG	**0.68**	**0.86**	1.10	1.40	2.00
3	NG	**0.92**	**1.01**	1.10	1.20	1.15
4	SBPPRL	3.40	3.08	5.10	4.60	5.60
4	FNG	3.80	**4.23**	4.70	5.40	5.60
4	NG	3.80	**4.23**	4.70	5.60	–
5	SBPPRL	0.65	**0.81**	1.00	1.28	1.50
5	FNG	0.55	**0.78**	1.10	1.15	1.25
5	NG	0.55	**0.61**	**0.68**	0.75	0.55
6	SBPPRL	1.82	**2.09**	2.40	2.18	2.70
6	FNG	1.75	**2.25**	2.90	2.70	2.70
6	NG	1.75	**2.33**	3.10	2.40	–
7	SBPPRL	0.55	0.83	1.14	1.60	–
7	NG	1.10	1.20	1.20	1.33	–
8	SBPPRL	0.80	1.40	1.54	2.08	–
8	NG	0.84	1.06	1.10	1.45	–
Average	SBPPRL	1.50	1.51	2.26	2.46	3.43
	FNG	1.87	–	2.57	3.09	3.01
	NG	1.67	1.13	2.32	2.39	1.90

NG – natural grasslands; FNG – fertilized natural grasslands; SBPPRL – sown biodiverse permanent pastures rich in legumes; SOM – soil organic matter; missing data was filled in using the geometric average of the increase rates, and is shown bold; when result is "–", data could not be filled-in.

general expression of the model:

$$SOM_{i,t} = \frac{K_i'}{\alpha}\left(1-e^{-\alpha\Delta t}\right)+\frac{a}{\alpha}\left(1-e^{-\alpha\Delta t}\right)SOM_{i,0} + e^{-\alpha\Delta t}SOM_{i,t-1}. \quad (5)$$

We also test the hypothesis that SOM dynamics for SBPPRL is different in the first year. There are several reasons that justify this approach. First, SBPPRL plots are tilled in the first year, and thus there is increased SOM mineralization for $t=1$. In the first year plants blossom only from the seeds which were sown, and therefore SBPPRL produce less biomass than in the following years (Carneiro et al., 2005). It is only from the second year on that a seed bank is created (much larger than the amount of seeds initially sown) from which the pasture permanently blossoms every year. Furthermore, first-year plant roots do not fully decompose and integrate SOM pools until later years. There is a lag between the increase in production and the increase in input.

The model is calibrated estimating a regression equation in which SOM_t is the dependent variable and SOM_{t-1} and SOM_0 are the independent variables. Only the constant (independent) term depends on the grassland system and, in the case of SBPPRL, if it is a first year observation. We introduce this first-year dummy in the parameter K' for simplicity purposes, and also because part of the effect is in SOM input.

Therefore, the constant term may be calculated as the sum of four dummy variables. The dummy $d_i=1$ if observation regards grassland system i and $d_i=0$ otherwise. The model we estimate is

$$\begin{aligned} SOM_t \;=\; & C_{1,K^{t=1}_{SBPPRL}}d_{1,K^{t=1}_{SBPPRL}} + C_{1,K^{year>1}_{SBPPRL}}d_{1,K^{year>1}_{SBPPRL}} \\ & + C_{1,K_{FNG}}d_{1,K_{FNG}} + C_{1,K_{NG}}d_{1,K_{NG}} \\ & + C_2 SOM_0 + C_3 SOM_{t-1} \end{aligned} \quad (6)$$

This model is estimated using 78 filled-in observations and has six regression constants $(C_{1,i}, i=\left\{K^{t=1}_{SBPPRL}, K^{t>1}_{SBPPRL}, K_{FNG}, K_{NG}\right\}; C_2; C_3)$. These six constants can be used, combining Eqs. (5) and (6), to obtain the six model parameters, which are $K_i'=f(i)$ (considering $i=\left\{SBPPRL(t=1),\ SBPPRL(t>1),\ FNG,\ NG\right\}$), α and a. The equivalence is

$$\begin{cases} \alpha = \dfrac{-\ln C_3}{\Delta t} \\ a = \dfrac{\alpha C_2}{1-e^{-\alpha\Delta t}} \\ K_i' = \dfrac{\alpha C_{1,i}}{1-e^{-\alpha\Delta t}}, \quad i=\left\{SBPPRL(t=1),\ SBPPRL(t>1),\ FNG,\ NG\right\} \end{cases} \quad (7)$$

We estimate the regression constants using software SPSS Statistics 17.0 with an Ordinary Least Squares (OLS) method and a stepwise regression (Verbeek, 2001).

2.4. Application and validation of the SOM model

The procedures referred in this section are a simplified approach intended only for validation of some parameters obtained.

To determine the grassland system in which the increases in SOM was highest, we calculate SOM increases in all systems starting from the same arbitrary initial SOM. Since we need a scenario for initial SOM, we assumed a starting hypothetical concentration of 0.87%, which is the average SOM concentration in soils in the region of Alentejo, which has highest area of pastures (LQARS, unpublished). We also apply the model to the initial SOM concentrations measured in each farm. This procedure tests the validity of the model by comparing measured and calculated results.

All results in this paper are shown in percentage points (pp), equal to $g_{SOM}/100\,g_{soil}$. However, for comparison purposes, we need to determine the carbon equivalent to SOM increases. Stated in another way, we find the equivalent to 1% SOM in terms of $t\,C\,ha^{-1}$.

The average soil bulk density (BD) in Portuguese soils is $1.48\,g\,cm^{-3}$, according to the Harmonized World Soil Database (HWSD; Fischer et al., 2008).[1] A 1 pp increase in SOM means that there is an increase of $0.0148\,g_{SOM}\,cm^{-3}$ of soil. Since the soil samples were collected at up to 10 cm, the SOM mass per unit may then be subsequently determined per unit area. Using this procedure, we find that 1 pp increase in SOM is equivalent to the storage of $14.8\,t\,SOM\,ha^{-1}$.

We can then convert K' from % (in mass) per unit of time into $t(SOM)\,ha^{-1}$, which is then converted to equivalent plant production. In order to do so, we assume that only humus is captured in SOM analysis, which is at most 80% of belowground biomass in pastures (Bot and Benites, 2005). IPCC (1997) indicates 2.8 as the default root to shoot ratio (R:S) for semi-arid grasslands. This value is consistent with the R:S of 0.5 to 4.8 in grazed pastures, which is the range of the comprehensive data for several regions gathered by Coupland (1976). Dividing K' by 80% and then by R:S, we obtain an estimate of aboveground production.

Using this rationale, we also find the equivalent of SOM increases in terms of carbon sequestration. In order to do so, we had to assume a conversion factor between SOM and soil organic carbon (SOC). Since approximately 58% of SOM is SOC (IPCC, 1997, 2003), and both are measured as $g/100\,g_{soil}$, then the mass of SOC is also 58% the mass of SOM. Therefore, 1% of SOM increase corresponds to the sequestration of $8.58\,t\,C\,ha^{-1}$. This is used when comparing our results to other studies.

3. Results

3.1. Results from soil analyses

Results from soil analyses for SOM concentration are shown in Table 2 (Carneiro et al., 2005). Considering the difference between the first and the last year, the minimum SOM increase for SBPPRL was obtained in Coruche (Farm #5), where the pasture blooming after the first year establishment was poor. The highest increases for SBPPRL were obtained in the more productive gneiss soil in Vaiamonte and the schist soils of Herdade de Refróias. In the latter, SOM at the beginning was already 3%. Farms #7 and #8 increased SOM concentration in SBPPRL by 0.35 pp and 0.43 pp per year. Table 2 also shows filled-in data underlined. Direct comparison with natural (non-fertilized) grasslands at each site shows that increases are usually higher for SBPPRL.

Table 2 also shows averages for SOM concentration in each grassland system and each year (using only original data). In 2005, FNG have SOM concentrations similar to SBPPRL, but they also start from higher initial SOM concentrations. Even though the drought in 2005 had an effect in stocking rates, SOM did not decrease significantly. This means that the lack of water decreased plant production, and therefore also SOM input, but it also decreased SOM mineralization.

All plots were natural pastures before the beginning of the project. Results for SOM in 2001 are previous to the installation of SBPPRL and to the fertilization of FNG making them representative of the initial soil conditions. Therefore, SOM initial value in

[1] See http://www.iiasa.ac.at/Research/LUC/External-World-soil-database/HTML/HWSD_Data.html?sb=4.

Table 3
Average and standard deviation for initial SOM concentration in each farm and grassland system.

Farm	Grassland system	Initial SOM concentration (%)	Average (%)	Standard deviation (pp)	N (#)
1	SBPPRL	1.55	1.43	0.18	2
	FNG	1.30			
2	SBPPRL	1.75	1.88	0.12	3
	FNG	1.95			
	NG	1.95			
4	SBPPRL	3.40	3.67	0.23	3
	FNG	3.80			
	NG	3.80			
5	SBPPRL	0.65	0.58	0.06	3
	FNG	0.55			
	NG	0.55			
6	SBPPRL	1.82	1.77	0.04	3
	FNG	1.75			
	NG	1.75			
7	SBPPRL	0.55	0.83	0.39	2
	NG	1.10			
8	SBPPRL	0.80	0.82	0.03	2
	NG	0.84			
Average of farms		–	1.66	0.14	7
All observations		–	1.66	1.06	18
Average for SBPPRL		–	1.50	0.99	7
Average for FNG		–	1.87	1.21	5
Average for NG		–	1.67	1.17	6

NG – natural grasslands; FNG – fertilized natural grasslands; SBPPRL – sown biodiverse permanent pastures rich in legumes; SOM – soil organic matter.

each location is equal to SOM in the first year, 2001:

$$SOM_0 = SOM_{2001}. \quad (9)$$

The use of SOM_0 as a proxy for representative local conditions (soil parameters, climate conditions, former management), and the use of the first year as the initial SOM concentration, are both justified by visual inspection of results in Table 2 and by statistical results shown in Table 3. SOM_{2001} changes more between farms than between grassland systems, within the same farm.

Table 3 shows that in each farm the standard deviation of the average SOM_0 (for all grassland systems) varies less than in the overall sample (average of all observations for SOM_0). But when we calculate the average $SOM_{i,0}$ for each grassland system i, then we obtain standard deviations similar to that of the whole sample. This fact supports the assessment made from the results in Table 2: the initial SOM concentration is correlated with the farm but not with the type of pasture. It is, therefore, a logical choice for proxy for the specific conditions of a plot in the model.

3.2. Regression results

We used observations in Table 2 as they are shown in order to calibrate the model (percentage values, equal to $g_{SOM}/100\,g_{soil}$). Since we used consecutive pairs of sampling years in the estimation, the number of observations (N) is equal to the number of successive pairs of points in Table 2 for each grassland system.

Regression results for the stepwise regression of Equation (6) are shown in Table 4. Results showed that the statistical fit to base data of the model, measured with the adjusted-R^2 is high, and corresponds to a low root mean standard error (rMSE). However, the coefficients for two variables were not significantly different from zero ($p < 0.05$): the first-year dummy for SBPPRL, $d_{1,K_{SBPPRL}^{t=1}}$, and the dummy for NG, $d_{1,K_{NG}}$. This means that there is no statistical relevance in considering that the input is different in the first year for SBPPRL, and also that the specific input from NG is zero, and so NG only increase SOM depending on the natural qualities of the plot.

We then removed $d_{1,K_{SBPPRL}^{t=1}}$ (which was less significant), and recalculated the model. There was no loss in adjusted-R^2, and $d_{1,K_{NG}}$ was again statistically not-significant ($p < 0.05$). The dummy for FNG, $d_{1,K_{FNG}}$, also became borderline significant (significant for $p < 0.05$ but not for $p < 0.10$). But when we remove $d_{1,K_{NG}}$, then $d_{1,K_{FNG}}$ also becomes not-significant. This means that fertilized plots do not increase their SOM pool significantly more than non-fertilized plots.

Therefore, we obtained a final model with only three regression constants significantly different from zero: one regarding SBPPRL specific SOM input ($C_{1,K_{SBPPRL}}$), one referring to the parameter SOM_0 (C_2), and one referring to the mineralization rate (C_3). This model is similar in terms of statistical fit (adjusted-R^2 and rMSE) to the initial model, but more efficient since it uses 78 observations to estimate 3 parameters (instead of the starting 6).

Note that we also tested the same model using only original observations (removing filled-in values), but the results were the same with minor loss of statistical fit.

Table 5 shows the parameters corresponding to the regression constants in this final version of the model, calculated using Equation (7). Regarding the absolute value of K_i', for SBPPRL it is equal to 0.94 pp yr^{-1}, which is equivalent to an input of 13.9 t ha^{-1} of SOM. Now, we need to transform this value of K into equivalent production. Dividing K by 80% and then by the average R:S of 2.8, we find that aboveground production is 6.2 t C ha^{-1}. Using two extreme R:S, aboveground production would be 3.6–34.7 t C ha^{-1}. The average production falls within the range of dry matter productivity of SBPPRL of 2–9 t ha^{-1} of dry matter (Carneiro et al., 2005).

Table 5 also shows results for K in the specific case of $SOM_0 = 0.87\%$. This value may be used for illustration purposes to determine the average SOM increase in 10 years from each grassland system, as shown in Fig. 3. SBPPRL on average increase their SOM concentration by 0.21 pp yr^{-1}, which is equivalent to 1.78 t C ha^{-1} yr^{-1}. This increase is higher than for FNG and NG (0.08 pp yr^{-1}, equivalent to 0.71 t C ha^{-1} yr^{-1}).

3.3. Assessment of model quality

In order to verify the adjustment to the original data provided by the model, we applied it to each farm. We used parameters in Table 5, and adjusted a model to each plot in each farm, using the plot-specific initial SOM concentration. The initial SOM concentration in each plot corresponds to the first column of data in Table 2.

Table 4
Results of the stepwise regression estimations for each grassland system.

Variables	Adj. R^2	rMSE	$C1, K$								$C2$		$C3$	
			SBPPRL				FNG		NG					
			Coefficient $t = 1$	SE	Coefficient $t > 1$	SE	Coefficient	SE	Coefficient	SE	Coefficient	SE	Coefficient	SE
All	0.971	0.488	0.097	0.187	0.465*	0.132	0.401*	0.135	0.171	0.126	0.577*	0.104	0.561*	0.079
Minus 1st-year	0.971	0.493	–	–	0.365*	0.123	0.254**	0.143	0.095	0.130	0.377*	0.123	0.766*	0.098
Minus NG	0.969	0.485	–	–	0.326*	0.111	0.207	0.128	–	–	0.393*	0.121	0.774*	0.097
Minus FNG	0.969	0.491	–	–	0.258*	0.104	–	–	–	–	0.374*	0.121	0.823*	0.094

NG – natural grasslands; FNG – fertilized natural grasslands; SBPPRL – sown biodiverse permanent pastures rich in legumes; SOM – soil organic matter; C_1, C_2, C_3 – parameters in Eq. (6); rMSE – root mean squared error.
* Variable statistically significant for $p < 0.05$.
** Variable statistically significant for $p < 0.10$.

Table 5
Parameters obtained for each grassland system and respective units.

Grassland system	K' pp yr^{-1}	a yr^{-1}	K (SOM$_0$ = 0.87%) pp yr^{-1}	α yr^{-1}	$1 - e^{-\alpha}$ –
SBPPRL	0.28	0.41	0.64	0.19	0.18
FNG	0.00		0.36		
NG	0.00		0.36		

K', α, a – Parameters in Eq. (6); NG – natural grasslands; FNG – fertilized natural grasslands; SBPPRL – sown biodiverse permanent pastures rich in legumes; pp – percent points.

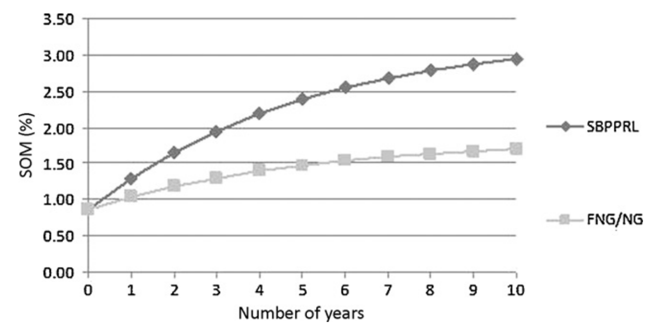

Fig. 3. Simulated SOM concentration in each year, as estimated using data filled-in using geometric averages, starting from 0.87%. NG – natural grasslands; FNG – fertilized natural grasslands; SBPPRL – sown biodiverse permanent pastures rich in legumes; SOM – soil organic matter concentration.

Results are shown in Fig. 4, which plots all observed and modelled results, except points corresponding to initial SOM concentration (which are by construction over the grey line) and except points which were filled-in. The closer the points are to the grey 45° line, the better the fit.

Visual inspection of Fig. 4 seems to show that there is no overall model bias. To verify these hypotheses, we ran a t-test for means for the set of the residues (difference between model predictions and observations). The test shows no evidence to reject the null hypothesis of zero mean ($p < 0.05$). A Phillips–Perron Test to the set of residues also rejects the null hypothesis of a unit root ($p < 0.05$), which means that the series of residuals is stationary. Furthermore, a one series Kolmogorov–Smirnov Test of normality to the series of residuals (ordered using observed SOM) shows that the null hypothesis of the normal distribution cannot be rejected (K–S

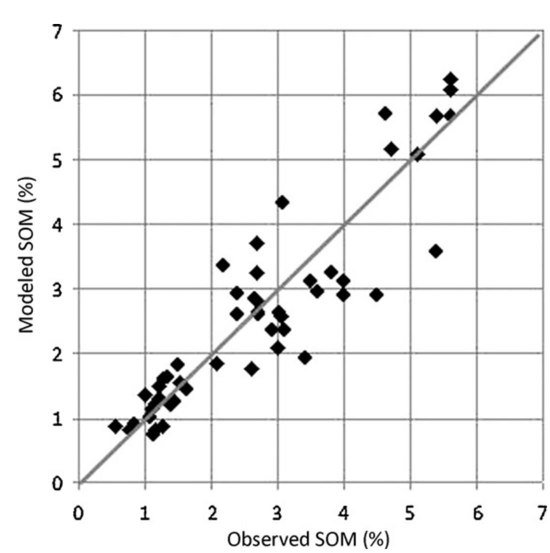

Fig. 4. Observed and simulated SOM concentration for all farms and grassland systems. SOM – soil organic matter.

Z value 0.928, asymptotic sigma 0.356). Therefore, there is no evidence of a spurious regression due to a bias in residuals.

4. Discussion

In this paper, we studied SOM dynamics in sown biodiverse and natural types of rainfed grasslands. SOM concentration is important for many agronomic and environmental reasons. We defined a model to calculate the average trend of SOM increases as a balance between accumulation of organic material in soils and mineralization of the available SOM pool. In time, as SOM increases, it eventually reaches an upper bound. Therefore, in a grassland system with no change in management, the entry and the mineralization eventually cancel each other. Other studies in literature indicate that SOM also increases asymptotically after land use changes (Sollins et al., 1996; West and Six, 2007), as well as in the response to exogenous inputs of carbon or fertilizers (Six et al., 2002, 2004).

The parameters in our model were statistically determined using a method which yielded a significant ($p < 0.05$) fit to the data. The adjustment of the model to the data (measured as adjusted-R^2) is equally high. This is an important result, since we only had soil analyses for the first five years after installation (for one of them, 2002, there is practically no data). Considering that SOM saturation in soils is assessed in the long term (Stewart et al., 2007), the fact that the asymptotic pattern is already picked up in the data is a strong conclusion.

The main objective of the model was to obtain a long-term trend for SOM dynamics in the different grassland systems. We did not wish to obtain a model which predicts year-by-year SOM concentration, but rather to show the average SOM accumulation potential of each system. In this respect, we conclude that SBPPRL increase the soil organic matter pool more than the other types of grasslands. This result is consistent with field observations for stocking rates: SBPPRL produce more biomass, and thus support a stocking rate which is systematically twice or more that of natural pastures.

The increased input in SBPPRL is due to two factors. First, production responds to fertilization, even though this effect alone is not enough to significantly increase the SOM pool, since results for FNG and NG are similar. Second, production responds to the improved seed bank independently of soil characteristics. It is important to notice that SBPPRL in the first-year were tilled and have a small seed bank (the seeds sown). The effect of the productivity loss in terms of SOM in the first-year, however, was not statistically significant.

Table 5 shows results for K in the specific case of $SOM_0 = 0.87\%$. K is a measure of the total input per year. K is higher for SBPPRL than for FNG/NG, which translates to higher SOM increases since the fraction of existing SOM in one year which is mineralized in the next, $1 - e^{-\alpha}$, is constant. The absolute value of K for SBPPRL is approximately double the absolute K for FNG/NG. Since K is SOM input, and it is highly correlated with plant production, this result is consistent with observations for stocking rates, which are also approximately the double for SBPPRL.

The actual difference between SBPPRL and natural pastures may be even greater than we show here. FNG and NG may be overestimated due to the fact that plots were contiguous, since Carneiro et al. (2005) explain that there was some contamination of natural grasslands by sown species.

To our knowledge, there are no other internationally published studies on SBPPRL in Portugal or elsewhere. We find that our results are similar to those found in other preliminary Portuguese studies. Some early results hint that in 10 years SBPPRL increase SOM from 1 to 3% (Crespo, 2004). At Herdade dos Esquerdos, in Vaiamonte (Portalegre, Portugal), following a programme of SBPPRL installation, SOM concentration across the farm increased from between 0.7% and 1.2% in 1979 to between 1.45% and 4.40% in 2003 (Crespo et al., 2004; Crespo, 2006a,b). This SOM increase is higher than that of any natural grassland under any form of management found in the literature.

In a related study, Aires et al. (2008) measured carbon fluxes over a pasture in Southern Portugal, similar to natural pastures in our study. They found that, in 2004–2005 (drought year), pastures emitted 0.49 t C ha^{-1} yr^{-1}, while in 2005–2006 (normal precipitation year) they sequestered 1.91 t C ha^{-1} yr^{-1}. This high intra-annual variability is also captured by our results, since yearly measurements of SOM concentration oscillates around a medium/long-term increasing trend (the trend is given by our model).

Regarding the first year in Aires et al.'s (2008) study, the drought year, it was also a sampling year in our study. Table 2 shows that in FNG and NG there was also a decrease in SOM from 2004 to 2005, which is the period for which Aires et al. (2008) concluded that soils had been emitters. FNG lost, on average, 0.08 pp, and NG lost 0.49 pp. These values are equivalent to a loss of 0.69 t C ha^{-1} yr^{-1}, and 4.20 t C ha^{-1} yr^{-1}, for FNG and NG respectively. When comparing these results to the results of Aires et al. (2008), we see that our approach does not under-estimate emissions.

Still regarding the same period, it is interesting to notice that SBPPRL increased SOM concentration even in the drought year by 0.97 pp on average (Table 2). This indicates that SBPPRL have one further advantage, which is increased resilience in drought years.

Regarding the second sampling year in Aires et al. (2008), it was a normal precipitation year and measured carbon sequestration was 1.91 t C ha^{-1} yr^{-1}. The lowest average comparable carbon sequestration in our study is for FNG and NG, and is equal to 0.71 t C ha^{-1} yr^{-1}, and the highest result is 1.78 t C ha^{-1} yr^{-1} for SBPPRL. Thus results obtained here do not overestimate the potential for sequestration of Portuguese grasslands.

The similarity in results between our work and Aires et al.'s (2008) work also indicates consistency between our method, using soil samples, and theirs, using flux measurements.

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