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Title: Carbon saturation and assessment of soil organic carbon fractions in Mediterranean rainfed olive orchards under plant cover management

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Abstract: Olive groves are undergoing a marked change in the way that inter-row land is managed. The current regulation and recommendation encourages the implementation of plant cover, mainly to improve soil fertility and reduce erosion. However, there is no quantitative information on the dynamics and pools of soil organic carbon (SOC) fractions of different protection levels of the plant-residue-derived organic carbon (OC). This study was conducted to provide a range of annual OC inputs in commercial olive oil groves under natural plant cover, to assess the influence of the annual application of aboveground plant cover residues on unprotected and physically, chemically and biochemically protected SOC. In addition, we tested the carbon saturation hypothesis under plant cover. Ten olive oil orchards under plant cover management (PC), together with five comparable bare soil olive oil orchards (NPC) were selected and annual aboveground natural plant residues and SOC pools were sampled and quantified. Annual aboveground plant cover biomass and OC production in PC olive orchards averaged 1.48 t dry-weight (DW) ha-1 and 0.56 t C DW ha-1, respectively with a great variability among sites (coefficient of variation of about 100 %). SOC concentration in PC orchards was, on average, 2.8 (0 - 5 cm soil) and 2.0 (5 - 15 cm) times higher than in bare soils of NPC, and the pool of protected SOC in the top 15 cm was 2.1 times higher in the PC (17.9 mg C g-1 □ 5.7) (±standard deviation) compared to NPC (8.5 mg C g-1 □ 2.9) olive orchards. Linear or saturation type relationships between each SOC fraction and total SOC content for the range of SOC of the commercial olive oil orchards were statistically indistinguishable, and thus linear models to predict SOC accumulation due to plant cover in olive orchards are suitable, at least for the studied range of SOC. Overall, at regional scale where olive oil groves represent a very high proportion of the agricultural land, the use of plant cover appears to be a promising practice that promotes protection of the SOC, thus improving SOC sequestration.

Dear Editor, Dr.Surinder Saggar,

We appreciate the time and constructive comments that the editor and reviewers dedicated to our manuscript.

Attached to this cover letter, we have provided our responses to the comments of the reviewers and given a detailed description of the changes carried out during revision. Briefly, we have accepted most of the reviewers' suggestions, and we have made clearer in the text a number of issues raised by the reviewers.

In addition, we have modified the Introduction to include specific hypotheses. To address the reviewers' comments, we have fully revised the Discussion in order to make it clearer and shorter. Furthermore, in order to increase the robustness of our results and explanations, some new references have been included in the manuscript.

The final version of the manuscript has been reviewed by a native English speaker (co-author: Pete Smith) for linguistic / grammatical correctness.

The changes we have made have improved the quality of the manuscript, and we hope that it now suitable for publication in Agriculture, Ecosystems and Environment. Please do not hesitate to contact us in case of further queries.

Looking forward to your response,

Yours truly,

J.L Vicente-Vicente & Roberto García-Ruiz, corresponding authors, on behalf of the authors.

Responses to reviewer's comments

REVIEWER 1

Mayor comments

"The title does not sound and need more specific regarding the research theme".

We do not fully agree with reviewer comment at this respect. We believe title describes the research topic well. We investigated soil organic carbon fractions under plant cover in Mediterranean rainfed olive oil orchards, which is precisely the description in the title.

However, being aware of the reviewer's concern regarding the title, we have changed the title as follows: "**Carbon saturation and assessment of soil organic carbon fractions in Mediterranean rainfed olive orchards under plant cover management".**

For the introduction, I suggest that after literature review, a specific hypothesis should be put forwarded to test in the present study.

The hypotheses of the study have been included in the last paragraph before explaining the aim of the study in the Introduction section (L. 117-120).

In the Material and Methods section, the authors defined too much "carbon" term, such as carbon (C), organic carbon (OC), soil organic carbon (SOC), total carbon (TC), Total soil organic carbon (TOC), which may easily confuse the readers.

We appreciate this comment and we agree with the reviewer's concern. Total soil organic carbon (TOC) has been substituted by "total SOC". And TC has been eliminated. We hope that after this change, the manuscript is clearer.

Also, some definitions are not clear. For example, L173-174, the authors should tell the readers the differences between organic matter and SOC. I understanding that the organic matter was estimated by the SOC multiplying the factor of 1.724 in most case.

Soil organic matter was quantified directly and was not an estimation based on SOC. This was explained in section 2.3 of the submitted manuscript"…organic matter content was estimated according to Nelson and Sommers (1982) by weight loss after ignition". On the other hand, SOC was also determined directly (L. 179-180) "….after digesting the soil samples with dichromate and sulphuric acid following the method proposed by Anderson and Ingram (1993)".

For the L212, I wander what the differences between the TOC and SOC

We agree with this and other reviewer comments on some confusion among C, OC, TC, TOC and SOC. We have reworded those sentences in which some of these terms appear to make the abbreviations consistent. We now use only SOC.

For the discussion section, much work still needs to improve the quality of manuscript. For example, L284, this subsection seems a little redundant and the authors should further squeeze the contents by focusing on the theme

According to the suggestion of the reviewer, we have reworded this and other subsections of the discussion section to reduce and/or remove redundancy and to provide a better focus.

L 317-318, this relationship may be very weak but I suggest that the authors should provide some specific data (r =?, p = ?, n=?).

Data has been included (r = 0.41, p= 0.24, n = 10)

Also, in many places, some statements should be support by citing some literature work (e.g., L327-L328; L353-356; L419-423; L449-451).

After careful reading, we agree with the reviewer comment and have added references as detailed below.

L 327-328. We appreciate this comment. However, we have already included some references such as Castro et al. (2008), and Guzmán and Foraster (2011) (L. 323-324). Therefore, we do not believe that additional references are required here.

L. 353-356. The reference is given a few lines after this sentence. "The presence of many different wild plant species in the plant cover communities also introduces a greater diversity of carbon compounds into the soil, some of which may be more resistant to decomposition (Tiemann et al., 2015)". Furthermore, in this context, we remark that "the formation of microaggregates within macroaggregates is increased after the incorporation of plant cover residues (Six et al., 2000). The release of biogenic products and other binding agents, such as polysaccharides and root exudates (Puget and Drinkwater, 2001), during the incorporation and relatively-rapid decomposition of the residues of the plant cover may have promoted the solid-phase reaction between organic matter and clay and silt particles, leading to the formation of stable microaggregates (Golchin et al., 1994).
Therefore, we believe that the idea of the formation of microaggregates after plant residues incorporation is well explained through the references included in the manuscript.

L. 419-423. We really appreciate the suggestion of the reviewer, and in this line an important review carried out by Barré et al. (2014) has been added.

Barré, P., Fernandez-Ugalde, O., Virto, I., Velde, B., Chenu, C., 2014. Impact of phyllosilicate mineralogy on organic carbon stabilization in soils: incomplete knowledge and exciting prospects. Geoderma, 235–236:382–395.

L. 449-451 We agree with the reviewer, and we have included the reference of Six et al. (2002).

L427, in this subsection, the authors cited too much work by Stewart et al. (2007), but in fact the data of present study seemed to not be line with the C saturation type proposed by Stewart et al. (2007). Thus, more explanations are needed. I wander whether it is necessary to cite too much work by Stewart et al. (2007).

Unfortunately, we did not find other studies assessing C saturation using the SOC fractionation of Six et al. (2002). In this sense, the study of Stewart et al. (2007) was the best to be compared our results.

We agree with the reviewer that results of our study do not show saturation behaviour for the different SOC fractions, whereas the study of Stewart et al. (2007) did it for some of the SOC fractions. However, we believe that in the discussion it is clearly explained "The fact that in our study the physically and chemically protected SOC did not showed saturation could be likely due to the relatively low range of total SOC of our study compared to that of Stewart et al. (2007) (i.e., 5.1 to 96.1 mg C g-1). Indeed, in the long-term agroecosystem experiments of Stewart et al. (2007), the number of fractions fitting the C saturation model within each site was directly related to maximum SOC content. Thus, SOC saturation in these fractions might does occur but that it is not always seen in agricultural field experiments since the range of OC input levels use to be too small for the saturation tendency to be showed".

Nevertheless, and according to the reviewer comment, we have deleted some of the references to the Stewart et al. (2007) study.

TABLES AND FIGURES

For the fig.1, I suggest that the Frequency distributions of aboveground biomass and organic carbon should be fitted with the Gaussian function to check whether these data are normally distributed or not.

Figure 1 shows the actual frequency distribution of annual production of aboveground biomass of olive oil groves under a natural plant cover. We did not analyse for normality (Gaussian function) as we only use in the manuscript the mean value and an indicator of the dispersion of the data around the mean of the annual production of aboveground biomass.

For the figures 2 and 3, the average values with the same letters indicates no significantly differences between SOC fractions rather than management types within same soil layers. Please, check.

The reviewer is correct. We apologize for these mistakes. In fact, the statistical analysis has been done between depths instead of between managements. Therefore, in the figure captions the words “between managements” have been substituted by “between depths” in the figure 2 and 3.

For the figures 5 and 6, the comparison between the management types for the given SOC fraction is done by the T test if the data are normally distributed. The authors should clearly state the results of normality test and then the method used for the comparison.

For all the comparisons and statistics, were checked previously for homocedasticity and normality. This was shown in the submitted version in M&M 2.5 section.

“The effects of the presence of plant cover on total and SOC fractions for the two different depths were assessed using two-way ANOVA (management and depth as factors). Previously, tests of homoscedasticity and normality were carried out. These analyses were done using the IBM SPSS Statistics 20 software”.

Fig. 7, I suggest that some lines for the best-fit linear or saturation model for each fraction should be added in the plot

We agree with the reviewer and results for both models, linear and saturation, have been included for each soil organic carbon fractions in the Figure 7.

REVIEWER 2

No measurements were made of soil bulk density (BD) and hence the C sequestration results cannot be presented on an equivalent mass basis. Differences in the depth of any cultivation under the two systems compared may have lead to differences in soil BD thereby slightly distorting comparisons of C sequestration based on a common sampling depth (0 – 15 cm). For example, the lack of

correlation between C inputs and SOM cited in lines 236-238 may, to some exteng, be due to some of the additional C inputs on PC farms being incorporated below de sampling depth.

In the revised version of the manuscript the soil bulk density has been included and shown in table 1. On the other hand, we agree with the reviewer's concern that differences in bulk density might explain differences in SOC stocks estimated from SOC concentrations. Nevertheless, after considering bulk density, differences in SOC between covered and uncovered olive oil orchards remain. We also agree with the reviewer comment on the possible contribution of some organic carbon in PC plots moving below the sampling depth. We have added a sentence to make this clearer.

This dilution may mean that SOC throughout the soil profile was even greater in the PC soils, compared with the NPC, than suggested by sampling to only 15 cm (e.g. lines 265-266). I do not think this is a major issue as the object of the paper is to report C additions and hence the potential for C sequestration. Therefore, the authors can address this issue by referencing likely differences in soil BD under the two systems and hence the difference between potential and actual C sequestration per unit of soil mass.

We agree with the reviewer's concern. We have addressed this issue by taking into account bulk density.

BULLET POINTS

Do the models referred to estimate SOC accumulation to depth taking account of equivalent mass?

We are not sure of the reviewer point. We tried to assess to what extent C saturation hypothesis is verified under plant cover management in commercial olive oil groves. If concentration of a given protected organic carbon pool (mg C g-1 of specific fraction) shows saturation behaviour at high SOC, means that soil have a maximum capacity to protect SOC in this fraction. If it is not the case and concentration of a give protected organic carbon fraction (mg C g-1 of specific fraction) shows linearly with SOC, then the saturation hypothesis should be rejected at least for the range of SOC assayed.

Linear or saturation model refers to linear or saturation curves between soil organic carbon concentration (mg C g-1 fraction) in a specific soil organic carbon fraction and soil organic carbon content (mg C g-1 soil)

MODERATE POINTS

Please, indicate the species that comprised the plant cover and whether there were any differences in the species among the orchards.

Unfortunately, we did not analyse for species composition of the communities of natural plant cover in olive oil orchards. This was not our objective. In addition, species composition of the communities of the inter-row area of olive groves shows very high spatial and intra and inter-annual variability, mainly due to intra and inter annual pattern of precipitation and other landscape features (Laila, 2015). After a visual appreciation of the plots while sampling, most abundant species belong to Graminaceae (mainly*Lolium sp*. *Hordeum sp*., and *Avenafatua* or *Avena sativa*), Brassicaceae (*Brassica sp*., *Lobularia sp*., and *Aurinium sp*.), Asteracea, such as *Chrysanthemum sp*., and some legumes such as *Medicago sp*. and *Vicia sp*.

Nevertheless, carbon and nitrogen contents and C-to-N ratio of the natural plant cover residues were relatively homogeneous. In the case of the organic C content of the biomass residues the mean value was 37.4% (±2.1) (Coefficient of variation = 5.6 %), whereas for the nitrogen content it was 2.3% (±0.46) (Coefficient of variation = 20.1).

Therefore, despite the suspected differences in species composition of the natural plant cover of the PC plots, they were relatively homogeneous in terms of plant residue quality.

Laila, 2015. PhD entitled "Agroecological transition of the olive oil groves: a study case" (https://dialnet.unirioja.es/servlet/tesis?codigo=20760).

Can the large differences in crop residues be attributed to any differences in the plant cover species?

We believe that main differences in the annual production of biomass of the natural plant cover are attributable to differences in landscape features (e.g. north versus south facing), soil fertility and variability in the microclimatic conditions among sites.

Was there any association between tree density and SOC content?

The tree density was very similar in the 10 sites (between 95 to 120 trees ha^{-1}). Therefore, we believe that differences in tree density do not play a major role in the differences in SOC, at least for the range of tree density sampled.

Line 75, please indicate the approximate size of this proportion

We have reworded the sentence. Therefore, "Significant proportion of total agricultural production" has been substituted with "about 60% of the total olive orchards surface".

Lines 140, 249, you mean "comprised", not "compromised".

We appreciate this comment and the words have been corrected with the suggestion of the reviewer.

MINOR POINTS

All these mistakes have been corrected:

Line 135, replace "were" with "was"

Line 138, replace "lower" with "less"

Line 208, replace "site" with "sites"

Lines 273 and 380, replace "managements" with "management"

Line 278, replace "models" with "model"

Line 291, "throughout" I suggest you mean "through"

Line 380, it is either "plant residues serve" or "plant residue serves"

Line 443, delete "a" before "saturation"

Line 447, replace “finding” with “findings”

Line 456, replace “showed” with “show”

REVIEWER 3

The introduction is a bit wordy and it could be shortened.

According to the reviewer comment we have shortened the introduction section by removing some non-essential sentences.

The discussion section is rather long and some reduction may improve the readability of the ms and highlight the author’s results. To this regard, especially in the section 4.1 and 4.2, the authors should do a greater effort to discuss and interpret their results beside those of the wide literature reported.

According to the suggestion of the reviewer, we have reworded these subsections of the discussion section to reduced or removed redundancy to better focus on the topic.

SPECIFIC COMMENTS

Line 95….Recently found that….

We have corrected the mistake. Effectively, “than” must be substituted with “that”

L.136-137. Please, indicate the region or province and the location of the sites.

The provinces of the location of the different sites have been included, and also the region (Andalusia). In L. 192 we have remarked that the sites were located in the provinces of Granada and Jaén.

We did not include the location of each of the 10 sites (for instance in a map), since we consider that this information is not relevant for the analysis of the results. Table 1 already shows the main characteristics of the soils of the 10 different sites, and we believe there are already many tables and figures. Nevertheless, if reviewer and editor

think that manuscript would benefit of the location of olive farms in a map (new figure), we have no problem showing it.

L.138. On what series did you calculate MAP? Please, add this information to the text.

This value corresponds to the average precipitation (last 15 years) of different meteorological stations of Granada and Jaén provinces close to the olive oil farms location. This information has been added in the revised version of the manuscript.

L. 140 Change "compromised" with "occupied"

Done.

L.141 Were the soils different among the ten sites? Please, add info on soil types and parent material from which soils developed in the different sites.

The main characteristics of the soils are shown in the Table 1. We appreciate the comment of the reviewer, but we consider that we include enough information about soil features in Table 1. However, in order to better clarify the soil features of the ten different sites we have specified in the M&M section that soils in these ten sites are placed under similar parent material features (marls).

L. 143. In terms of climate, …

The mistake has been corrected.

L.180. dispersion in water?

The dispersion and the fractionation were carried out under wet conditions. It has been clarified in the text.

L.186. What was the Nacl concentration and the density of the solution used for the density flotation?

The density of the NaCl concentration was 1.3 g/cm^3.

Since in the tables and figures the different pools were reported as unprotected, chemically, physically and biochemically protected, the authors should report at

the end of the fractionation procedure the fractions belonging to the different pools (although this has been reported in table 2). This helps the reader to follow the presented results.

We added the information of the different SOC fractions in Table 2 only to describe the fractions we obtained with the fractionation method. However, according to the reviewer's suggestion, we have added at the end of the fractionation procedure the fractions belonging to the different pools.

L.202-217. Please, rewrite in a clearer way this part

In this part, some words have been changed in order to clarify the theoretical framework of the methodology used, and also to include the suggestions made by other reviewers. We hope it is now clearer.

L. 249. Change "compromised" with "represented".

Done

L. 266. Please, add the units to the values.

Units in this part have been included.

L. 298 Each year

We wrote "on one year" because the biomass production was recovered only in one year. Changes to "in one year" instead.

L. 309 Please, add reference

We acknowledge this suggestion and we included the reference of Baldock (2007).

L 314 This value ……..in this case "this value" seems to be referred to 80%. Anyway, the experiment of Vasquez et al lasted about half time that Gómez-Muñoz.

We acknowledge this suggestion. We have removed the reference of Vazquez et al. (2003) because we consider that it does not give any additional important information. Therefore, the suggestion of the reviewer is not necessary in the new version of the manuscript.

L 334 and tables. Please, report in all the tables the meanings of the abbreviations used. In the same way, this should be along the text; for example, the reader probably does not remember what site is CT.

We agree with the reviewer comment regarding the need for explanation of the meanings of some of the abbreviations in tables and in the main body of the manuscript. These have been described in the text and tables 1 and 3.

L 285-341. This part is, in my opinion a bit long. Further, maybe major room should be given to the effect of the rhizodeposition processes on soil C, since it is the main flow of C into the soil caused by herbaceous cover, even in the sub superficial horizons

This part has been reworded according to the comments and suggestions of the other reviewers.

L. 396-399. This part is not clear and a bit speculative. Please, explain why is increased the formation of micro aggregates within macro aggregates is increased. In my opinion, you can't assume that the formation of micro within macro aggregates increased in PC, since the amount of the stability of the aggregate classes were not measured, and micro and macro aggregates were separated during the fractionation procedure.

We agree with the lack of clarity of the idea that we wanted to expose with these sentences. SOC concentration in the < 53 mm fraction (silt+clay) which was isolated from the 53 – 250 mm size fraction (microaggregates) was almost 4 times higher in PC compared to NPC plots. This suggests that the amount of SOC chemically protected within microaggregates is promoted with the incorporation of plant cover residues. In the following sentence, we tried to discuss this result according to the most accepted theory: "The release of biogenic products and other binding agents, such as

polysaccharides and root exudates (Puget and Drinkwater, 2001), during the incorporation and relatively-rapid decomposition of the residues of the plant cover may have promoted the solid-phase reaction between organic matter and clay and silt particles, leading to an increase in the chemically protected SOC within microaggregates and to the formation of stable microaggregates (Golchin et al., 1994)". Next, we tried to explain how, with the support of a study, the fact that an increase in SOC in the silt+clay fraction isolated from the microaggregates might lead to an increase in the stability of microaggregates: "This result is in line with those of Garcia-Franco et al. (2015) who found that the proportion of microaggregates, and their stability, within small macroaggregates increased after green manuring together with reduced tillage."

We made the mistake of mixing microaggregates with macroaggregates when we wanted to discuss the fact that in PC plots, SOC chemically protected within microaggregates was increased respect to NPC olive oil farms

We have reworded these sentences to make it clearer.

L. 417-425 This part seems to be highly speculative rather than based on the results of the authors, that do not present any mineralogical data. Indeed, the fact that clay particles are strongly negative charged is not always true, as the clay charge depends from the clay minerals comprising the soil colloidal fraction

We clearly found that both organic carbon content linked to silt-clay (mg C g-1 of soil in the < 53 µm) and organic carbon concentration of the silt-clay fraction (mg C g-1 of silt-clay) were significantly higher in soil of the PC plots. We did not search for the intimate mechanism. With this paragraph (lines 417-425) we tried to provide the most accepted hypothesis which might explain the enrichment of organic carbon chemically protected under a natural plant cover. We agree with the reviewer comment regarding that clay charge depends on the composition of the tetrahedral and octahedral sheets of the clay, and there are some (rare) cases in which net charge of the clay particles are neutral. However, even neutral charge clay particles might have a net negative charge due to predominant pH. In our studied soils, soil pH was generally higher than 8.0 and

thus a net negative charge of clay particles is quite likely. Nevertheless, we have reworded the sentence to deal with the reviewer's concern.

REVIEWER 4.

I just have a concern with the objective three – to elucidate if the relationship between SOC and organic carbon fractions follow s a linear or a saturation curve over the range of SOC measured. I have doubts if the methodology used allowed to be conclusive regarding C saturation. The C input was limited – Annual aboveground biomass production in the plots varied from an average of 0.65 to 2.53 t ha-1 during a maximum time of 12 years. With that, I believe that C saturation was not 0065pected as the results showed with linear adjustments. In order to reach C saturation a higher C input would be required or a longer time of addition. Anyway, the discussion and conclusion are right and the manuscript can be accepted in the present form.

We partially agree with the reviewer's comment in this respect. For a given annual input of organic carbon some time is needed to reach the new equilibrium. The time taken is highly dependent on climatic conditions, landscape features and management among others. In our study, we have assumed that SOC under a specific entry of organic carbon is close to steady state. The same approach was undertaken in similar studies, for instance, that of Stewart et al. (2008).

Highlights

- Annual aboveground organic carbon production in olive orchards with plant cover averaged 0.56 t C ha^{-1}.
- The pool of protected soil organic carbon was 2.1 times higher orchards with plant cover compared to those with no plant cover.
- Linear models to predict soil organic carbon accumulation due to plant cover in olive orchards are suitable.

Carbon saturation and assessment of soil organic carbon fractions in Mediterranean rainfed olive orchards under plant cover management

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Abstract

Olive groves are undergoing a marked change in the way that inter-row land is managed. The current regulation and recommendation encourages the implementation of plant cover, mainly to improve soil fertility and reduce erosion. However, there is no quantitative information on the dynamics and pools of soil organic carbon (SOC) fractions of different protection levels of the plant-residue-derived organic carbon (OC). This study was conducted to provide a range of annual OC inputs in commercial olive oil groves under natural plant cover, to assess the influence of the annual application of aboveground plant cover residues on unprotected and physically, chemically and biochemically protected SOC. In addition, we tested the carbon saturation hypothesis under plant cover. Ten olive oil orchards under plant cover management (PC), together with five comparable bare soil olive oil orchards (NPC) were selected and annual aboveground natural plant residues and SOC pools were sampled and quantified. Annual aboveground plant cover biomass and OC production in PC olive orchards averaged 1.48 t dry-weight (DW) ha^{-1} and 0.56 t C DW ha^{-1}, respectively with a great variability among sites (coefficient of variation of about 100 %). SOC concentration in PC orchards was, on average, 2.8 (0 – 5 cm soil) and 2.0 (5 – 15 cm) times higher than in bare soils of NPC, and the pool of protected SOC in the top 15 cm was 2.1 times higher in the PC (17.9 mg C g^{-1} ± 5.7) (±standard deviation) compared to NPC (8.5 mg C g^{-1} ± 2.9) olive orchards. Linear or saturation type relationships between each SOC fraction and total SOC content for the range of SOC of the commercial olive oil orchards were statistically indistinguishable, and thus linear models to predict SOC accumulation due to plant cover in olive orchards are suitable, at least for the studied range of SOC. Overall, at regional scale where olive oil groves represent a very high proportion of the agricultural land, the use of plant cover appears to be a promising practice that promotes protection of the SOC, thus improving SOC sequestration.

Keywords

Soil organic carbon, soil organic carbon fractions, plant cover, olive orchards

1. Introduction

Soils are the largest carbon (C) reservoir of the terrestrial C budget (Lal, 2004), representing about 2500 Pg C (1500 of soil organic carbon and 950 of inorganic forms) (Lal, 2008). Therefore, even a relatively small increase or decrease in soil C content due to changes in land use or management practices may result in a significant net exchange of C between the soil reservoir and the atmosphere (Houghton, 2003). Conversion of natural ecosystems to agroecosystems causes a significant depletion of the soil organic carbon (SOC) pool (Lal, 2004), mainly because C output exceeds the input and this is exacerbated when soil degradation is severe. Therefore, agricultural soils have the potential to sequester C from the atmosphere with proper management. Thus, policy makers face the challenge of developing and implementing effective SOC accretion strategies for agriculture, which requires identification of the best management practices for each agroecosystem. A number of agricultural management strategies are known to sequester soil C by increasing C inputs to the soil and enhancing various soil processes that protect C from microbial turnover. However, uncertainties about the extent and permanence of C sequestration in these systems remain (Six et al., 2002).

Most experimental studies to date have focused on the impacts of specific agricultural management practices on SOC dynamics have been performed under extensive cereal and irrigated crops in temperate (Virto et al., 2011; Dimassi et al., 2014) or Mediterranean areas (Álvaro-Fuentes et al., 2009; López-Garrido et al., 2011). However, very few studies have been carried out under rain-fed tree crops in semiarid areas, such as olive oil orchards, where these crops represent about 60% of the total olive orchard surface (e.g. Nieto et al., 2013).

The incorporation of cover crops (i.e. green manure) into the soil of a given cropping system

72 is considered a promising sustainable management practice to reduce soil erosion risk (Alliaume
73 et al., 2014; Francia-Martínez et al., 2006; Gómez et al., 2009), while compensating soil C losses
74 derived from land-use change and tillage in agricultural fields (Gómez-Muñoz et al., 2014;
75 Milgroom et al., 2007; Ramos et al., 2010). This is an important issue in Southern Spain, where
76 regional authorities introduced a policy of Good Agricultural and Environmental Practice in
77 olive farming, which consists of linking the subsidy for cultivating the olive crop to the
78 requirement of provide/permit additional cover plants under certain circumstances (e.g. mean
79 slope over 7 %).

80 Plant cover in olive oil orchards is mainly comprised of natural vegetation which is allowed to
81 emerge spontaneously in autumn and winter along the middle of the orchard lanes, covering up
82 to approximately one-third of the surface. The plant cover should be eliminated in late March or
83 early April, before it starts competing for water and it is then usually disrupted, mainly by
84 mechanical mowing and/or herbicides. Plant residues may be left on the soil surface or
85 mechanically mixed into the top centimetres of soil by tillage. Both approaches are currently
86 used and are realistic land management options. Most previous studies related to the
87 effectiveness of cover plants in olive orchards have been designed to evaluate the effects of this
88 practice in mitigating soil erosion (e.g. Gómez et al., 2004), but to a lesser extent to evaluate the
89 dynamics of C cycling associated with it (Castro el al., 2008). Vicente-Vicente et al. (2016)
90 recently found that plant cover in woody crops (olive and almond orchards and vineyards)
91 significantly contributed to SOC accumulation with annual rates averaging 1.1 t C ha^{-1}.

92 The amount of plant residue and the degree of SOC decomposition are key factors in the
93 formation and stabilization of aggregates, which in turn improve soil structure and drive SOC
94 sequestration (Haynes and Beare, 1996). However, no studies in olive groves have been done to
95 determine the effects of plant covers on different SOC pools and to elucidate how SOC interacts
96 physically and chemically with aggregates, as well as with mineral particles.

The physical protection of organic carbon (OC) by aggregates (Denef et al., 2001, 2007) and the physico-chemical stabilization are considered to be important mechanisms of SOC stabilization (Krull et al., 2003; Marschner et al., 2008; Garcia-Franco et al., 2014). The study of different protected SOC fractions is a key element in the reliable assessment of soil C dynamics and can be used as an early indicator of soil changes caused by management practices (Six et al., 2002). The identification of these fractions will improve our understanding of how aggregates stabilize and store SOC (von Lützow et al., 2007), helping us to select the best sustainable land management practices with regard to the enhancement of SOC sequestration in Mediterranean areas.

On the other hand, according to the theory of soil C saturation proposed by Stewart et al. (2007), the potential for SOC stabilization has a limit, and as the SOC approaches its saturation level, the increase in SOC stock becomes smaller despite increasing C input rates. Stewart et al. (2007) found that SOC saturation does occur, but that it is not always seen in agricultural field experiments since the range of C input levels is often too small for saturation to be shown. Several other studies also support the theory that soils can become C saturated (Chung et al., 2008; Six et al., 2002).

The role of plant cover in fruit tree cropping systems on SOC sequestration at regional scale might require the use of models. The elucidation of linear or saturation relationships across a typical range of C inputs, due to the presence of plant cover and protected SOC fractions, is important to accurately predict the potential for C sequestration under this management.

We hypothesized the following: (1) Spontaneous plant cover increases the total SOC content compared to the non-cover management; (2) this increase is mainly due to an increase in the most labile SOC fractions; and (3) there is a maximum capacity limit for SOC accumulation for some fractions, especially those related to the silt and clay content.

The main purpose of this study was to assess the effectiveness of plant cover for enhancing soil C sequestration in semiarid rain-fed olive oil orchards, to promote changes in existing conventional agronomic practices from a climate change mitigation perspective. Specifically, the objectives were: (1) to determine the variability of annual aboveground OC input due to the presence of a plant cover; (2) to assess the effects of plant cover residue addition to the soil on SOC accretion and SOC fractions of different protective levels (unprotected and physically, chemically and biochemically protected); and (3) to elucidate if the relationship between total SOC and SOC fractions follows a linear or a saturation curve over the range of SOC measured.

2. Material and methods

2.1. Site description and experimental design

Ten olive orchards, in which a plant cover (PC thereafter) was left to grow in the inter-row area each year during the last twelve years, were selected in different sites (CA1, CA2, CT, MO, LO, DE1, DE2, PE, JA and AL) of Jaén and Granada provinces (Andalusia, southern Spain) in soils over marls with the same parent material. Mean annual precipitation in the area was 446 mm (average value from different meteorological stations in Granada and Jaén locations) about 10 % less than the 25-y average. Aboveground plant cover biomass in all orchards was mechanically mowed each year during March and plant residues were left on the soil surface. Typically, plant cover comprised between 30 and 60 % of the whole olive oil farming area. Soils in these orchards differed in a range of characteristics, some of which are shown in Table 1. Five out of the ten PC olive orchards were paired with a nearby (within a distance of tens of meters) comparable olive orchard (in terms of climate, orientation, slope, soil properties and farming characteristics such as tree density and age), except for the lack of plant cover during at least the last 12 years. In these olive orchards with bare soil (NPC, thereafter), plants were controlled by

mechanical mowing and/or applying pre-emergence herbicides in the autumn. Thus, differences between these five pairs of olive oil farming were attributed primarily to the presence or absence of the plant cover during autumn to the end of March and to the management related to the control of plants. All olive orchards presented a tree density of between 90 – 120 trees per hectare, aged 35 to 45 years, and trees were distributed in a regular arrangement typical of fruit trees, with a canopy cover typically of between 40 – 70 % of the orchard area.

2.2. Soil and aboveground plant cover biomass sampling

In each of the ten PC olive orchards, aboveground annual plant cover biomass produced in 2010 was randomly determined several days before mowing (between the end of March to early April) by randomly throwing five woody frames (50 cm x 50 cm) in the inter-row area and subsequently measuring the dry weight of the aboveground plant biomass collected.

Soil below the frames used to collect aboveground cover plant biomass was also sampled. At each of the sampling point, a 50 cm x 50 cm pit was opened and soil samples were taken at depths of 0 – 5 and 5 – 15 cm. In the NPC olive orchards, 0 – 5 cm and 5 – 15 cm deep soils of the inter-row area were collected in the same day and in the same way that comparable PC olive orchards. Soil samples were transported to the laboratory in air-tight containers in the same day of collection.

2.3. Soil and aboveground plant cover biomass analysis

Aboveground plant cover biomass was dried (60 ^{o}C, 5 days), weighed, ground with a ball mill and analysed for total SOC and nitrogen in a CHN auto-analyser (Carlo Erba NA200, Milan, Italy).

Soil samples were air-dried and sieved through a 2 mm sieve. Particle size distribution was determined by the pipette method (Gee and Bauder 1986). Soil available potassium and soil

labile phosphorus content were analysed according to Grant (1982) and Olsen and Sommers (1982), respectively. Bulk density (BD) was determined according to Blake and Hartge (1986). Soil cation exchange capacity (CEC) was analysed according to Rhoades (1982). Soil organic matter content was quantified according to Nelson and Sommers (1982) by weight loss after ignition. SOC was determined after digesting the soil samples with dichromate and sulphuric acid following the method proposed by Anderson and Ingram (1993).

2.4. Soil carbon fractionation

Separation of the various soil C pools was accomplished by a combination of physical and chemical fractionation techniques in a simple, three-step process modified from Stewart et al. (2009). Briefly, after a first step consisting in the partial dispersion and physical fractionation of the soil in wet conditions to obtain three size fractions (> 250 μm, 53 – 250 μm and < 53 μm), a69 second step, which involved further fractionation of the 53-250 μm fraction previously isolated, was followed. The > 250 μm, 53 – 250 μm and < 53 μm fractions isolated after the first step corresponded to the coarse non-protected particulate organic matter (cPOM), microaggregate (μagg) and easily dispersed silt and clay (dSilt+dClay) fractions, respectively. In this second step a density flotation with sodium chloride was used to isolate fine non-protected POM (LF). After removing the fine non-protected POM, the heavy fraction was dispersed overnight by shaking with 15 glass beads and passes through a 53 μm sieve, separating the microaggregated-protected POM (> 53 μm in size, iPOM) from the microaggregated-derived silt- plus clay-size fractions (μSilt+μClay). The third step involved the acid hydrolysis of each of the isolated silt+clay-sized fractions. The silt+clay-size fraction from both the density flotation (μSilt+clay) and the initial dispersion and physical fractionation (dSilt+dClay) were subjected to acid hydrolysis as described by Plante et al. (2006). Acid hydrolysis consisted of incubating the

193 samples at 95 ^{o}C for 16 h in 25 ml of 6 M HCl. After hydrolysis, the suspension was filtered and
194 washed with deionized water over a glass-fibre filter. Residues were oven-dried at 60 ^{o}C and
195 weighed. These fractions represent the non-hydrolysable C fractions (NH-dSilt+dClay and NH-
196 μSilt+μClay). The hydrolysable C fractions (H-dSilt+dClay, H-μSilt+μClay) were determined
197 by difference between the total organic C content of the fractions and the C contents of the non-
198 hydrolysable fractions.

199 This three-step process isolates a total of 12 fractions and it is based on the assumed link
200 between the isolated fractions and the protection mechanisms involved in the stabilization of
201 organic C (Six et al., 2002). The unprotected pool includes the cPOM and LF fractions, isolated
202 in the first and second fractionation steps, respectively. The physically protected SOC consists of
203 the SOC measured in the microaggregates. It includes not only the iPOM but also the
204 hydrolysable (H-μsilt+clay) and non-hydrolysable (NH-μsilt+clay) SOC of the intermediate
205 fraction (53 – 250 μm). The chemically and biochemically protected pools correspond to that
206 hydrolysable (H-dsilt+clay) and non-hydrolysable (NH-dsilt+clay) SOC in the fine fraction (< 53
207 μm), respectively. For further information, Table 2 shows a description and the significance of
208 each of the analysed fraction.

209 Total SOC and OC of each of the soil fractions were determined after digesting the soil
210 samples, previously grounded with a ball mill, with dichromate and sulphuric acid following the
211 method proposed by Anderson and Ingram (1993).

212 The SOC concentration was used as a balance between C input and decomposition, to
213 normalize across sites, as sites differ in aboveground plant cover biomass C input, decomposition
214 rate and field management. This approach has been demonstrated to be useful for normalising
215 SOC fractions (Stewart et al., 2007), and it has been showed that at steady state a whole soil that
216 shows a linear increase in C with respect to C inputs will also exhibit linearity between total
217 SOC concentration and SOC concentrations of the C fractions. Thus, we used total SOC as a

proxy for C input to determine if the different fractions of SOC were influenced by C saturation. A soil fraction exhibiting a linear relationship between total and fraction SOC is interpreted as not being influenced by C saturation, while fraction exhibiting an asymptotic relationship shows evidence for C saturation.

2.5. Statistical analysis

The effects of the presence of plant cover on total and SOC fractions for the two different depths were assessed using two-way ANOVA (management and depth as factors). Previously, tests of homocedasticity and normality were carried out. These analyses were done using the IBM SPSS Statistics 20 software.

Pearson correlation coefficients were used to test the relationship between total SOC and aboveground plant cover productivity and C input due to plant residues.

3. Results

3.1. Carbon inputs due to aboveground plant cover productivity

NPC orchards did not produce any plant biomass, as the surface was kept free of vegetation. Annual aboveground plant cover biomass production in the PC plots varied greatly from an average of 0.65 t dry-weight (DW) ha^{-1} found at Cortijo Tobazo (CT) site to 2.53 t DW ha^{-1} at the Jaén (JA) site, with an overall mean of 1.48 t DW ha^{-1} (Figure 1a and Table 1). OC content of the aboveground plant cover biomass on dry weight basis of the whole set of the studied orchards averaged 37.4 % (coefficient of variation of 3.6 %, data not shown). The annual input of OC due to residues of the aboveground plant cover biomass averaged 0.56 t DW ha^{-1}, with a minimum and a maximum of 0.24 and 1.0 t DW ha^{-1}, respectively (Figure 1b). Neither the

aboveground plant cover biomass production nor OC input were significantly correlated with the top 5 cm SOM ($r = -0.53$ to -0.55; $p > 0.05$) or SOC ($r = -0.60$ to -0.62; $p > 0.05$) contents.

3.2. *Soil organic carbon fractions of olive orchards with plant cover*

SOC content in the top 5 cm of the inter-row soils of PC orchards ranged from 11.5 to 44.8 mg C g^{-1} and, as expected, these values were higher, about a 50 % on average, than those found in the 5 – 15 cm depth soil (Table 3 and Figure 2). Mean unprotected and physically protected SOC of the top 5 cm of soils (10.0 and 5.2 mg C g^{-1}, respectively) were significantly higher than average values obtained for the 5 – 15 cm (5.3 and 3.6 mg C g^{-1}, respectively). These differences were not observed for the chemically and biochemically protected SOC. However, SOC density (i.e. mg C g^{-1} fraction) of unprotected, and physically and chemically protected fractions were significantly higher in the top 5 cm (Figure 3). The biochemically protected fraction did not show differences in both concentrations per gram of soil or per gram of fraction between depths.

Unprotected SOC comprised a relatively high proportion of the total SOC with values ranging from 16.6 to 57.3 % (average of 33.0 %) and from 6.8 to 56.3 % (average of 24.4 %) for 0 – 5 and 5 – 15 cm soil depths, respectively (Figure 4 and Table 3). The differences between depths were significant for the percentage of the unprotected fraction (Figure 4). However, the contribution of the physically, chemically and biochemically protected SOC to the total SOC did not differ significantly with depth (Figure 4). The contribution of biochemically protected SOC was significantly and negatively correlated ($r = -0.55$; $p < 0.05$) with total SOC.

3.3. Effects of the organic carbon input due to aboveground plant cover biomass on total and SOC fractions

Total SOC contents in soils of the PC orchards were significantly higher than in the paired NPC orchards, and this was true for both, 0 – 5 cm (2.8 times on average) and 5 – 15 cm (2.0

264 times on average) soil depths (Figure 5 and Table 3). A similar trend was found for the
265 unprotected, and physically, chemically and biochemically protected pools, which were on
266 average 4.5, 2.7, 3.2 and 1.9 times higher, respectively, in the top 5 cm and 2.7, 2.0, 3.0 and 1.8
267 times higher, respectively, in the 5 – 15 cm layer of the soils of the PC than in the NPC olive
268 orchards (Figure 5). Protected SOC in the top 15 cm was 2.1 times higher in soils of the olive oil
269 orchards with plant cover (17.9 mg C g^{-1} soil ± 5.7) than in the comparable olive oil orchards
270 with bare soils (8.5 mg C g^{-1} soil ± 2.9).
271 The higher Total SOC in olive orchards under a plant cover treatment was mainly due to the
272 higher OC concentration per gram of fractions, mainly for unprotected, and physically and
273 chemically protected fractions (Figure 6).
274
275 *3.3. Relationship between soil organic carbon fractions and total soil organic carbon: test for*
276 *soil organic carbon saturation hypothesis*

277 Relationship between total SOC (mg C g^{-1} soil) and SOC concentration (mg C g^{-1} fraction) for
278 the different fractions was tested pooling all sites, depths and management data. The unprotected
279 SOC was best fitted to a linear function when the 0 – 5 and 5 – 15 cm soil depth samples were
280 pooled (r^2 = 0.86, $p < 0.05$, N = 90) (Figure 7a). However, for the physically and chemically
281 protected SOC pools, both linear and saturation functions showed similar regression coefficients
282 (Figures 7b and 7c), and therefore they were indistinguishable. The biochemically protected
283 SOC pool did not show significant regression either to a linear nor a saturation curve type
284 (Figure 7d), and remained relatively similar independently on the SOC content. There were not
285 significant differences between the predicted values of the linear and saturation curves for the
286 range of total SOC observed in this study.

287

4. Discussion

4.1. Annual organic carbon input under plant cover and soil organic carbon stocks in olive orchards

Our data of annual production of aboveground plant cover biomass are in the range of 1.0 and 4.0 t DW ha^{-1} obtained by Repullo et al. (2012) in a plant-covered olive oil orchard of Córdoba (Spain) during a period of three agricultural years, but were lower than the biomass entering to the soils due to crops residues or seeded cover crops of grain crops (Allmaras et al., 1998). Relatively low annual aboveground plant cover biomass production in rain-fed olive oil orchards is not surprising since the inter-row area of the olive oil farming is neither fertilised nor irrigated.

On average for the ten PC olive oil orchards, 0.56 t ha^{-1} of OC was left on the soil surface on one year. This average is within the range of 0.2 to 0.7 t C ha^{-1} yr^{-1} estimated by several researchers (Freibauer et al, 2004; Hutchinsonet al., 2007) as the potential for C sequestration under scenarios of application of crops residues. However, the extent to which the input of OC derived from plant cover increases the soil C stock in the inter-row of olive orchards will ultimately depend on decomposition rate of that OC. Decomposition rate depends on many factors including plant biomass quality (e.g. C-to-N ratio and lignin and polyphenol contents); edaphic and environmental conditions and aboveground plant residues management (e.g. biomass clearing method and residue displacement) (Kumar and Goh, 2000). A relatively high decomposition rate of the cover plant residues could be expected as plant C-to-N ratio was relatively low (average 17.1; min 14.3 and max 24.0) (Baldock, 2007). On the other hand, it should be noted that plant residue decomposition is expected to slow down when they are left on soil surface (Cooper et al., 2006), as is usually the case for olive orchards. Gómez-Muñoz et al. (2014) found that about 20 % of the added plant cover residue in an olive orchard remained in the litter bags after 343 days, indicating that the other 80 % was decomposed or entered into the soil as < 1mm (mesh size) particle fragments.

We did not find a relationship between annual aboveground plant cover OC production and the 0 – 5 cm pool of SOC ($r = 0.41$, $p = 0.24$, $N = 10$). Many long-term agroecosystem field experiments with different levels of annual OC inputs, show that SOC stocks was linearly related or followed a saturation behaviour, with the annual amount of OC inputs (e.g. Kong et al., 2005; Paustian et al. 1997; Stewart et al., 2007). However, our results were not unexpected since plant-OC production was measured in a single year and the pool of SOC is the result of the accumulated balance between OC inputs and decomposition during many years. Moreover, the relationship between levels of OC inputs and SOC stock observed by the above researchers was only clear for a large range of OC inputs (i.e. from 1 t C to more than 5 t C) and in our study the highest annual aboveground plant OC production was about 1 t C. In addition, inter-annual plant cover biomass production of olive orchards has shown between 4-fold (Guzmán and Foraster, 2011) and one order (Castro et al., 2008) of variation, mainly driven by the high inter-annual variability in precipitation typical of Mediterranean regions. In addition, SOC decomposition rate might differ among sites of different pedo-climatic properties, resulting in different SOC stocks for a similar level of annual aboveground plant cover residue OC input. For instance, SOC in the Cortijo Tobazo site was the highest, but the OC input of the aboveground plant residues at this site was the lowest. Finally, the lack of relationship might also be due to the fact that C input *via* roots of the plant cover might represent a significant input of OC, and it cannot be ruled out that part of the input of OC has been mobilized below the sampling depth; neither mechanism was unaccounted for in this study.

In any case, the effects of the presence of plant cover on SOC stocks was clear when comparing SOC in the five paired PC – NPC olive oil orchards. In four out of the five paired comparisons between 9.0 and 16.1 more t C ha^{-1} was stored in the top 15 cm of the soils of the PC, whereas at Cortijo Tobazo, the difference was of 29.3 t C ha^{-1}. These values were similar to

337 the 8.4 – 15.0 t C ha^{-1} more SOC storage in the top 15 cm of an olive oil orchard under a cover
338 plant treatment compared to a plant cover-free plot (Castro et al., 2008).
339 The higher SOC stock in soils under the plant cover treatment might be due, not only to the
340 annual OC input of the plant residues, but also to a decrease in SOC losses by soil erosion
341 (Gómez et al., 2009). In addition, the diversity of wild spontaneous plant cover might have an
342 important impact on SOC accrual by improving the ability of soil microbial communities to
343 rapidly process plant residues and protect them into aggregates, and by introducing greater
344 diversity of OC compounds into the soil, some of which may be more resistant to decomposition
345 (Tiemann et al., 2015).
346 Assuming an annual average of plant-aboveground OC input of about 0.56 t C ha^{-1}, and that
347 20 % remains in the soil after one year, for the 1.5 million hectares of olive oil groves of
348 Andalusia, about 168 000 t C could accumulate annually into the soils. This estimate has many
349 uncertainties, but highlights the significance of the implementation of this technically and
350 economically viable practice on potential for C sequestration, at least at regional scale. In
351 addition, for the five PC – NPC comparisons soil CEC, exchangeable K, labile P and K were
352 between 1.5 to 2.0 and 1.1 to 1.8 times higher in the 0 – 15 cm soils of PC olive oil orchards.
353 Thus, the benefits of a plant cover in olive groves could lead not only to C sequestration, but
354 could also to improve soil properties, resulting in better fertility.

355

356 *4.2 Unprotected and protected SOC fractions under natural plant cover of olive oil orchards*

357

358 In our study unprotected, and physically and chemically protected fractions were significantly
359 higher in soils with plant cover (figure 6). The highest increase was achieved for the cPOM (the
360 coarse non-protected SOC) fraction due to an increase in the SOC concentration (e.g. mg C g^{-1}
361 fraction) of this fraction (between 2.5 to 7.3 times higher than that of the uncovered plots). This

was not unexpected, as recently derived, partially decomposed spontaneous plant residues together with seeds and microbial debris, such as fungal hyphae and spores that are not closely associated with soil minerals constitute the unprotected SOC pools (Six et al., 1999; Six et al., 2002). As this pool is sensitive to management practices and, consequently, highly influenced by future soil management, it should not be considered as a pool of SOC sequestrated in the long term. Indeed, many early studies have found that the LF and POM are relatively easily decomposable and are greatly depleted upon cultivation (e.g. Cambardella and Elliott, 1992; Solomon et al., 2000), indicating their relatively unprotected status.

Physically protected SOC was between 1.8 to 10.8 times higher in olive orchards with plant cover. The physical protection exerted by macro- and/or microaggregates on SOC is attributed to the compartmentalization of substrate and microbial biomass (Killham et al., 1993) and, the reduced diffusion of oxygen into macro and, especially, microaggregates resulting in a reduced activity within the aggregates (Sollins et al., 1996). Although the amount of soil aggregates or soil aggregate stability was not measured in our study, it is relatively well documented that plant residues serves, following the decomposition process, as a binding agent to hold soil particles together forming aggregates (Jastrow et al., 1998). Recently, Garcia-Franco et al. (2015) showed after 4 years of green manuring in an almond orchard, that the formation of micro and macro aggregates were promoted. Therefore, the presence of a plant cover and the surface displacement of the plant residues increased the amount of SOC physically protected. In addition, SOC of the silt+clay particles (< 53μm) within micro aggregates (53 – 250 μm) were on average 3.9 times higher in plant covered soil, suggesting that SOC chemically protected within the microaggregates, and eventually stability of the microaggregates, is increased after the incorporation of plant cover residues (Six et al., 2000). This could be explained by the fact that the release of biogenic products and other binding agents, such as polysaccharides and root exudates (Puget and Drinkwater, 2001), during the incorporation and relatively-rapid

387 decomposition of the residues of the plant cover may have promoted the solid-phase reaction
388 between organic matter and clay and silt particles, leading to an increase in the chemically
389 protected SOC within a SOC fraction which is physically protected, and to the formation of
390 stable microaggregates (Golchin et al., 1994). Our results indicate that a significant part of the
391 SOC stabilization is due to physico-chemical protection of OC by mineral particles (Krull et al.,
392 2003; Bronick and Lal, 2005). This result is in line with those of Garcia-Franco et al. (2015) who
393 found that the proportion of microaggregates, and its stability, within small macroaggregates
394 increased after green manuring together with reduced tillage. The higher SOC concentration in
395 both free and silt+clay-occluded SOC in microaggregates of PC olive orchards, relative to NPC,
396 can be beneficial to long-term C sequestration because microaggregates have longer turnover
397 times and higher stability than macroaggregates (Denef et al.,2007; Huang et al., 2010),
398 indicating the potential of this management practice to promote SOC accrual and stabilization.
399 SOC concentration of the silt+clay particles separated by wet sieving in soils covered by wild
400 herbaceous plant community was on average 3.2 times higher than that of soils under NPC. This
401 was not unexpected as it has been long recognised that the addition of organic matter to soils first
402 results in the formation of SOC associations with clay and silt particles (Tisdall and Oades,
403 1982).
404 More recently, Stewart et al. (2009) found that chemically protected SOC comprised an
405 average of 27% of total residue-C stabilized in the soil after addition of wheat residues during
406 2.5 years in a lab controlled experiment. The surfaces of clay particles are usually strongly
407 negatively charged, especially when soil pH is basic as was the case in our studied soils (Barré et
408 al, 2014). As the microbial community processes OC molecules, some of the by-products they
409 produce have strong positive charges. When these molecules make contact with clay particles,
410 they can form strong bonds, effectively protecting the molecules from microbial attack. This
411 form of chemical protection is highly effective and helps to explain why higher SOC and clay

content are correlated worldwide (Jobbagy and Jackson, 2000; Six et al., 2002).

4.3 Protected SOC was not saturated within the range of SOC measured

In this study, it was assumed that SOC concentration is a proxy for soil C input. Stewart et al. (2008) showed mathematically the relationship between the SOC concentrations of individual soil fractions and total SOC concentration, allowing C saturation to be expressed as a function of SOC concentration rather than soil C input. Nevertheless, we acknowledge the limitations to this analysis imposed by using soils from different environments, which will vary in their approximation of steady-state conditions.

Under this assumption, a linear relationship between whole SOC concentration and SOC fraction concentration indicates the lack of C saturation behaviour, whereas fractions exhibiting either an asymptotic relationship are influenced by C saturation.

Across the range of whole SOC concentration that was considered in this study (5.6 to 47.7 mg C g^{-1}), the linear behaviour of the unprotected pool for the combined site data ($r^2 = 0.87$, $p < 0.05$, $N = 90$) did not support the hypothesis of C saturation of this pool. This result is in line with those of Stewart et al. (2008), who found that in all soils of eight long-term agroecosystems experiments and adjacent grassland or forest analysed, the coarse non-protected SOC (cPOM) was best fitted using a linear relationship. The relationship between total SOC and concentration of physically protected pool from microbial attack was similarly fitted to a linear and to saturation curves for the whole set of plots ($r^2 = 0.78$ and 0.77, $p < 0.05$, $N = 90$ for the linear and saturation curves, respectively). In addition, regression coefficients of the linear and saturation curves fitted between the concentration of SOC in the silt+clay soil particles (chemically protected SOC) and whole SOC were indistinguishable ($r^2 = 0.63$ and 0.61, $p < 0.05$, $N = 90$ for the linear and saturation curves, respectively). These results do not agree with other findings. It has been theorised that the relationship between inputs of OC and concentration of

physically and chemically protected SOC should be of saturation type (Stewart et al., 2008). The content of silt+clay particles and the potential for macro and microaggregates formation in a given soil are limited and, thus, the amount of protected SOC throughout these mechanisms should be finite (Six et al, 2002). Stewart et al. (2007) found that when the protective capacity of the soil had been exceeded, further OC additions are not stabilized by these protective mechanisms. Thus, SOC accumulated in the fine and intermediate fractions and relationship between concentration of the physically and chemically protected SOC fractions and total SOC are of saturation type. The fact that in our study, the physically and chemically protected SOC did not show saturation, could be likely due to the relatively low range of total SOC of our study compared to that of Stewart et al. (2007) (i.e., 5.1 to 96.1 mg C g^{-1}). Indeed, in the long-term agroecosystem experiments of these authors, the number of fractions fitting the C saturation model within each site was directly related to maximum SOC content. Thus, SOC saturation in these fractions might does occur but that it is not always seen in agricultural field experiments since the range of OC input levels use to be too small for the saturation tendency to be showed.

SOC concentration of the biochemically protected fraction was not related to total SOC. This result contrasts to Stewart et al. (2007) who showed that biochemically protected SOC showed either a linear or saturation curves. Biochemical protection is acquired through condensation and complexation reactions or through the inherent complex biochemical nature of the organic material (Six et al., 2002), processes which might differ widely among other sites, explaining the lack of relationship in the present study.

Conclusions

Plant cover in olive orchards was a significant annual source (averaging 0.56 t C ha^{-1} yr^{-1}) of OC which might substantially contribute to the transference of atmospheric C into the soil. Indeed, SOC in olive oil orchards after the implementation of plant cover doubled with respect to

462 the usual bare soil management. Most of the SOC gain achieved under plant cover was protected
463 (physically, chemically or biochemically) from microbial activity, and thus contributed to long
464 term SOC sequestration. Therefore, at regional scale, where olive groves represent a very high
465 proportion of the agricultural land, the use of plant cover is a promising practice that promotes C
466 sequestration.

467 For the range of annual OC inputs under a plant cover, and for the total SOC of the commercial
468 olive grove studied, linear or saturation type relationships between total SOC and physically and
469 chemically protected SOC were indistinguishable. While these results do not invalidate the SOC
470 saturation hypothesis, they indicate that models designed to predict SOC sequestration by
471 assuming linearity between annual OC inputs and SOC at steady stay can be applied in olive
472 orchards under a plant cover management.

473

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Tables

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Table 1. Annual aboveground net production of plant cover and main soil characteristics (organic matter, cation exchange capacity, total nitrogen, Olsen phosphorous, available potassium, sand, silt + clay, bulk density and texture) of olive oil orchardswith plant cover (PC) and with no plant cover (NPC) located at Cambil (CA1 and CA2), Cortijo Tobazo (CT), Moraleda (MO), Loja (LO), Deifontes (DE1 and DE2), Pegalajar (PE), Jaén (JA) and Alcaudete (AL).Values show the mean ± standard deviation. Different lowercase letters indicate significant differences among samples into the same location at the $p < 0.05$ level.

Sites	Management	Depth (cm)	Aboveground plant cover productivity (t dry-weight ha^{-1})	OM (%)	CEC (cmol Kg^{-1})	TN (%)	Olsen P (g kg^{-1})	Available K (g kg^{-1})	Sand (%)	Silt+clay (%)	BD (Mg m^{-3})	Soil Texture
CA1	PC	0 -5	1.10±0.37	4.30±0.5a	19.0±0.5a	0.23±0.02a	42.3±9.5a	722±67a	30.9±1.3a	69.0±0.9a	1.19±0.00d	loam clay
CA1	PC	5-15		2.47±0.4b	17.8±1.5a	0.13±0.01b	15.5±3.2b	517±4b	26.1±1.7	73.8±1.2a	1.26±0.02c	loam clay
CA1	NPC	0 -5	-	1.10±0.3c	10.4±0.9b	0.06±0.01c	7.1±0.4b	220±42c	32.5		1.36±0.01b	clay
CA1	NPC	5-15		1.08±0.3c	10.7±0.4b	0.06±0.02c	6.0±1.0b	254±22c	-		1.41±0.06a	clay
CA2	PC	0 -5	0.89±0.44	5.35±1.98a	23.4±4.9a	0.25±0.07a	28.8±11.0a	720±339a	34.4±8.1a	65.5±8.1b	1.17±0.05c	clay
CA2	PC	5-15		2.77±1.35ab	22.9±9.1a	0.14±0.05ab	7.6±2.33b	358±244a	21.2±0.0b	78.8±0.0a	1.27±0.05b	clay
CA2	NPC	0 -5	-	2.17±0.18ab	20.8±1.5a	0.13±0.01b	17.3±2.0ab	455±92a	21.5±1.4b	78.5±1.0a	1.34±0.04a	clay
CA2	NPC	5-15		1.92±0.11b	20.0±2.8a	0.11±0.00b	9.9±2.3b	322±117a	21.8±0.9b	78.1±0.6a	1.35±0.02a	clay
CT	PC	0 -5	0.65±0.15	6.42±0.80a	18.7±0.3a	0.30±0.01a	11.7±0.2a	580±99a	40.7±4.6a	59.2±3.2a	1.13±0.01c	loam clay
CT	PC	5-15		3.00±0.20b	17.3±0.3a	0.13±0.01b	4.6±0.0c	490±21a	33.7±1.7a	66.2±1.2a	1.22±0.02b	clay
CT	NPC	0 -5	-	1.87±0.50bc	11.6±0.5b	0.10±0.01b	8.5±0.9b	275±13b	40.4±0.0a	59.6±0.0a	1.47±0.04a	loam
CT	NPC	5-15		1.33±0.10c	11.4±0.8b	0.07±0.01c	4.5±0.6c	270±21b	40.2±4.3a	59.7±3.0a	1.48±0.03a	loam clay
MO	PC	0 -5	1.50±0.41	3.78±0.70a	14.1±0.2a	0.21±0.04a	30.6±1.1b	371±22a	43.9±0.8a	56.1±0.6b	1.18±0.01b	loam
MO	PC	5-15		1.59±0.10b	12.1±0.2b	0.10±0.01b	15.3±2.4c	305±57ab	39.8±0.7b	60.2±0.5a	1.37±0.02a	loam

MO	NPC	0 -5	-	1.14±0.18b	7.0±0.9c	0.07±0.01b	56.2±0.4a	192±47b	39.4±1.7b	60.6±1.2a	1.35±0.02a	loam
MO	NPC	5-15		1.05±0.08b	7.0±0.3c	0.06±0.01b	40.2±8.3b	204±37b	42.8±0.6a	57.2±0.4b	1.36±0.01a	loam
DE1	PC	0 -5	1.07±0.16	7.62±1.20a	17.3±2.6a	0.33±0.04a	26.5±3.1a	364±3a	57.2±3.5a	42.8±2.5b	1.27±0.09b	loamy sand
DE1	PC	5-15		3.25±0.10b	20.1±0.1a	0.15±0.01b	7.4±0.1b	235±57a	44.1±1.7b	55.8±1.2a	1.30±0.02b	loam
DE1	NPC	0 -5	-	6.20±0.21b	19.8±0.3a	0.31±0.01a	30.5±0.8a	531±16a	43.8±1.8b	56.3±1.8a	1.36±0.01a	loam
DE1	NPC	5-15		4.79±2.50b	18.6±2.0a	0.24±0.11ab	21.8±17.3ab	408±223a	46.3±1.2b	53.8±1.2a	1.36±0.01a	loam
LO	PC	0 -5	1.95±0.15	1.72±0.10a	15.4±0.9a	0.10±0.01a	30.3±6.4a	355±71a	28.1±2.7a	71.9±1.9a	1.38±0.03b	loam clay
LO	PC	5-15		1.29±0.07b	15.1±0.5a	0.08±0.00a	26.9±5.1a	284±30a	25.9±0.4a	74.1±0.3a	1.43±0.03a	loam clay
DE2	PC	0 -5	2.04±0.49	2.57±0.20a	16.6±0.5a	0.13±0.01a	46.4±10.5a	337±54a	40.7±2.8a	59.3±2.8a	1.17±0.01a	loam
DE2	PC	5-15		2.31±0.17a	17.2±2.8a	0.14±0.03a	8.7±0.9b	103±4b	38.2±2.8a	61.8±2.8a	1.18±0.02a	loam
PE	PC	0 -5	1.83±0.49	3.34±0.30a	30.8±2.5a	0.19±0.02a	26.9±1.3a	630±127a	23.7±5.3a	76.0±3.7a	1.26±0.01b	clay
PE	PC	5-15		2.56±0.40a	31.3±4.6a	0.14±0.01a	15.15±2.2b	398±11a	19.6±0.6a	80.0±0.4a	1.30±0.01a	clay
JA	PC	0 -5	2.53±1.30	2.62±1.30a	26.0±0.8a	0.14±0.06a	16.8±2.1a	415±28a	17.5±3.5a	82.2±2.5a	1.28±0.01b	clay
JA	PC	5-15		1.19±0.10a	24.0±0.2a	0.08±0.00a	7.6±0.2b	378±39a	12.7±0.3a	87.3±0.2a	1.34±0.01a	clay
AL	PC	0 -5	1.21±0.57	1.80±0.09a	23.7±0.6a	0.10±0.01a	12.1±0.8a	308±4a	28.7±7.0a	71.3±5.0a	1.26±0.03b	clay
AL	PC	5-15		1.31±0.16b	25.7±2.3a	0.08±0.01a	8.0±2.0a	283±88a	17.9±2.5a	82.0±1.7a	1.35±0.03a	clay

Table 2. Main features of the soil organic carbon fractions determined in the PC and NPC olive oil orchards. The fractionation method used was that of Six et al. (1998).

Fraction	Denomination	Particle size	Origin	Type of protection	Description
cPOM	Coarse non-protected POM	> 250 μm	Physical fractionation of the first fractionation step procedure	Unprotected	Mainly compromised of plant residues. but also including seeds and microbial debris. such as fungal hyphae. Some presence of charcoal. A mixture of compounds caused by a regenerating plant residues pool and partial microbial decomposition.Typically high C/N ratio and lignin and with low net N mineralization potential.
LF	Fine non-protected POM	53 – 250 μm	Floating supernatant of the density flotation of the microaggregated fraction	Unprotected	
μaggregate	Microaggregate	53 – 250 μm	Physical fractionation of the first fractionation step procedure	Physically protected	Physical protection exerted by macro or microaggregates attributed mainly to (1) compartmentalization of substrate and microbial biomass and (2) reduced oxygen diffusion into microaggregates.
dSilt+clay	Easily dispersed silt plus clay	< 53 μm	Physical fractionation of the first fractionation step procedure	Chemically protected	C associated with primary organomineral complexes linked to silt plus clay sized particles.
iPOM	Microaggregate-protected POM	53 – 250 μm	Heavy fraction greater than 53 μm of the 53 – 250 μm fraction	Physically protected	POM within microaggregates.
μSilt+Clay	Microaggregated-derived Silt+Clay	< 53 μm	Heavy fraction smaller than 53 μm of the 53 – 250 μm fraction.	Physically protected	C associated with primary organomineral complexes linked to silt plus clay sized particles within microaggregates.

NH–dSilt+Clay	Non-hydrolisable fraction of the easily dispersed silt plus clay	< 53 μm	HCl digestion of the < 53 μm fraction isolated during the physical fractionation of the first fractionation step procedure	Biochemically protected	C chemically recalcitrant in the <53 μm fraction.*
dSilt+Clay	Hydrolisable fraction of the easily dispersed silt plus clay	< 53 μm	Hydrolisable fraction of the < 53 μm fraction isolated during the physical fractionation of the first fractionation step procedure. determined as the difference between the total organic C content of the < 53 μm fraction and the NH-dSilt+clay fraction.	Chemically protected	C associated with primary organomineral complexes linked to silt plus clay sized particles in the <53μm fraction.
NH–μSilt+Clay	Non-hydrolisable fraction of the microaggregate-derived Silt+Clay	< 53 μm	HCl digestion of the heavy fraction smaller than 53 μm of the 53 – 250 μm fraction.	Physically protected	C chemically recalcitrant within microaggregates*
H–μSilt+Clay	Hydrolisable fraction of the microaggregate-derived Silt+Clay	< 53 μm	Hydrolisable fraction of the heavy fraction < 53 μm of the 53 – 250 mm fraction.	Physically protected	C associated with primary organomineral complexes linked to silt plus clay sized particles within microaggregates.
*	Biochemically protected C pool				Occurs due to the complex chemical composition of the organic matter which is an inherent property of the plant residue quality which can be attained during decomposition through the condensation and complexation of decomposition residues. Biochemical resistance to decomposition.

Table 3. Values of total soil organic carbon (SOC) in soils and the amount of organic carbon in the unprotected, and physically, chemically and biochemically protected fractions (mg C g^{-1}) and their contribution (%) to the whole total soil organic carbion in soils with (PC) and without (NPC) plant cover in Cambil (CA1 and CA2), Cortijo Tobazo (CT), Moraleda (MO), Loja (LO), Deifontes (DEI1 and DEI2), Pegalajar (PE), Jaén (JA) and Alcaudete (AL). Values show the mean ± standard deviation. Significant differences between treatments and depths are shown in figures 2. 4 and 5.

Site	Management	Depth (cm)	Total SOC (mg C g^{-1})	Unprotected (mg C g^{-1})	Unprotected (%)	Physically Protected (mg Cg^{-1})	Physically Protected C (%)	Chemically Protected (mg Cg^{-1})	Chemically Protected C (%)	Biochemically Protected (mg Cg^{-1})	Biochemically Protected C (%)
CA1	PC	0 -5	33.3±1.1	8.4±0.6	25.3 ±2.4	7.2±0.9	21.7 ±2.0	13.2±0.6	39.6±0.7	4.5±0.3	13.4±1.1
CA1	PC	5-15	21.3±1.4	2.5±0.0	11.9±0.9	4.7±0.6	22.2 ±2.2	10.4±0.9	48.7±1.5	3.7±0.4	17.1±1.2
CA1C	NPC	0 -5	12.9±0.5	4.4±1.9	34.2±13.6	1.8±0.6	13.7±4.3	4.7±1.8	37.2±15.4	1.9±0.05	14.8±0.9
CA1C	NPC	5-15	10.5±3.6	2.3±1.5	20.8±7.6	1.3±0.5	14.1±7.4	5.3±2.9	49.0±9.6	1.5±0.4	16.2±8.4
CA2	PC	0 -5	35.2±8.1	8.6±2.5	24.5±4.6	3.5±2.0	10.5±6.2	16.8±5.2	47.1±4.3	6.4±1.7	17.9±1.0
CA2	PC	5-15	21.2±5.6	3.5±0.9	16.6±1.3	2.2±1.0	10.5±3.5	10.6±2.7	50.2±3.1	4.9±1.5	22.8±2.2
CA2C	NPC	0 -5	14.8±3.5	2.1±0.1	14.8±2.9	3.8±1.1	25.1±1.4	5.7±2.2	37.6±5.3	3.2±0.2	22.5±3.8
CA2C	NPC	5-15	13.3±1.3	1.3±0.4	9.8±2.3	3.0±0.5	23.1±5.5	5.6±1.0	41.6±4.7	3.4±0.4	25.6±1.4
CT	PC	0 -5	44.8±2.5	25.7±2.6	57.3±3.7	7.3±2.1	16.4±5.4	11.2±2.5	24.9±4.3	0.6±0.2	1.4±0.4
CT	PC	5-15	27.0±2.8	12.3±3.0	45.6±9.1	3.5±0.6	13.2±2.6	8.0±2.6	29.3±8.1	3.2±0.2	11.9±1.8
CTC	NPC	0 -5	7.2±1.4	2.3±0.7	32.6±4.9	1.3±0.5	17.3±3.9	2.0±0.6	27.5±3.9	1.5±0.3	22.7±9.5
CTC	NPC	5-15	7.0±1.3	1.8±0.5	25.7±2.8	1.5±0.6	21.6±6.9	0.6±1.0	9.9±16.8	3.0±1.5	42.8±20.0
MO	PC	0 -5	34.6±1.8	14.0±0.4	40.8±2.8	9.0±2.1	26.2±6.3	7.01±1.5	20.3±4.4	4.5±4.0	12.7±10.9
MO	PC	5-15	12.1±1.3	2.9±0.8	23.5±4.8	3.6±0.4	29.7±0.9	0.6±0.1	5.1±0.9	5.0±0.6	41.8±5.2
MOC	NPC	0 -5	13.1±0.8	3.8±0.2	29.4±3.3	4.0±1.1	30.7±9.2	2.5±0.5	19.0±5.0	2.8±0.5	21.5±2.4
MOC	NPC	5-15	12.6±0.4	3.4±0.7	27.0±6.7	2.7±0.4	21.7±3.5	2.6±0.4	20.4±2.8	3.9±0.3	30.9±1.5
DE1	PC	0 -5	21.7±8.8	7.3±3.8	30.5±12.1	5.2±1.7	26.0±9.3	6.2±2.3	29.5±4.8	3.0±1.0	14.0±1.6

DE1	PC	5-15	17.2±1.7	3.8±0.4	22.3±4.7	3.1±1.2	18.3±7.5	6.1±2.3	34.9±9.7	4.2±0.6	24.6±1.8
DE1C	NPC	0 -5	13.3±1.5	3.5±2.0	25.8±12.1	5.7±1.5	43.3±13.7	2.9±0.7	21.9±4.7	1.2±0.2	9.1±0.8
DE1C	NPC	5-15	7.3±0.4	1.6±0.2	22.7±2.3	1.6±0.1	22.1±0.8	2.7±0.2	33.5±0.7	1.6±0.3	21.8±3.5
LO	PC	0 -5	11.5±1.8	2.0±1.1	17.4±7.1	1.8±1.1	15.5±6.9	1.6±0.3	14.4±4.2	6.0±0.7	52.8±10.0
LO	PC	5-15	8.7±1.4	0.8±0.4	8.8±3.9	1.2±0.3	13.9±2.4	4.2±1.3	47.1±8.4	2.6±0.5	30.3±9.8
DE2	PC	0 -5	34.5±2.6	16.1±2.5	46.4±3.6	8.5±1.8	24.8±5.1	6.5±1.3	19.1±4.3	3.3±0.3	9.8±1.2
DE2	PC	5-15	34.3±3.7	15.8±3.7	45.9±5.9	8.6±1.9	25.3±6.3	7.3±2.7	21.2±8.2	2.6±0.5	7.6±1.8
PE	PC	0 -5	21.8±0.7	11.5±0.8	52.6±4.9	2.7±1.9	12.6±8.7	4.9±0.4	22.5±2.2	2.7±0.4	12.3±2.0
PE	PC	5-15	17.3±0.7	9.8±2.4	56.3±11.5	2.0±0.1	11.6±0.8	3.3±1.3	19.5±8.0	2.2±0.6	12.7±4.7
JA	PC	0 -5	19.5±0.5	3.2±0.2	16.6±0.6	1.7±0.7	8.7±3.5	12.6±0.9	64.6±3.4	1.9±0.4	10.1±2.4
JA	PC	5-15	14.1±0.7	1.0±0.2	6.8±1.3	1.4±0.3	10.0±2.3	10.2±0.9	72.0±4.5	1.6±0.1	11.3±0.9
AL	PC	0 -5	21.6±3.1	3.9±1.4	18.8±9.0	4.8±0.8	22.4±1.5	9.8±3.1	44.9±7.8	3.0±0.7	14.0±1.4
AL	PC	5-15	13.8±2.4	0.9±0.3	6.8±2.3	2.1±0.5	15.1±0.8	7.0±1.8	50.6±3.8	3.8±0.3	27.6±3.7

Table 4. Regression coefficients for linear and saturation curves between soil organic carbon concentration in the fractions (mg C g^{-1} fraction) and whole SOC (mg C g^{-1}). NA stands for no significant ($p < 0.05$) regression coefficient. For each of the fraction there was not statistical differences between the values predicted by linear and saturation models.

Fraction	Adjustment
Unprotected	Linear $R^2 = 0.87$ Saturation $R^2 = 0.80$
Physically protected	Linear $R^2 = 0.78$ Saturation $R^2 = 0.77$
Chemically protected	Linear $R^2 = 0.63$ Saturation $R^2 = 0.61$
Biochemically protected	Linear R^2 = NA ($R^2 = 0.0031$) Saturation R^2 = NA ($R^2 = 0.0002$)

Figure 1
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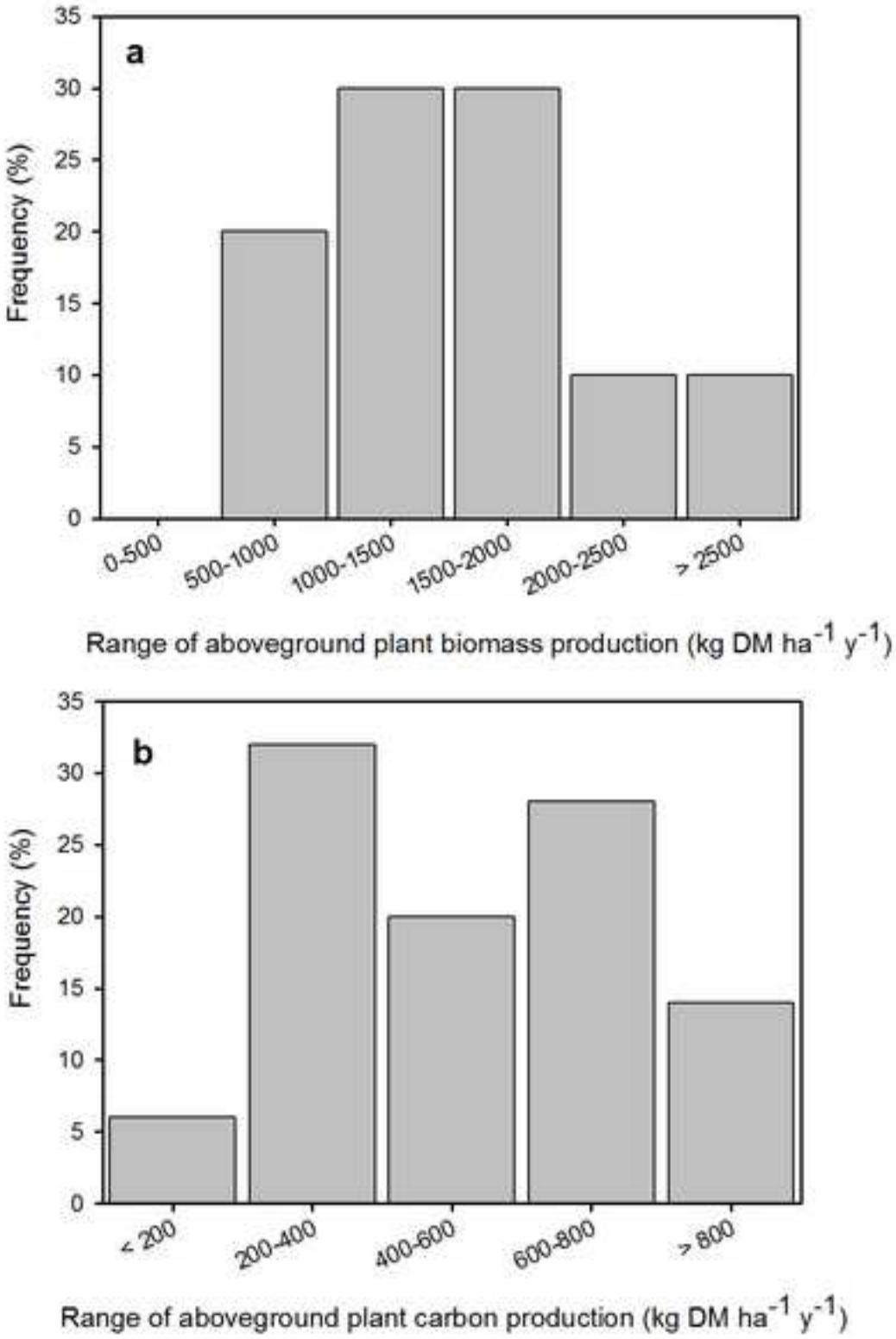

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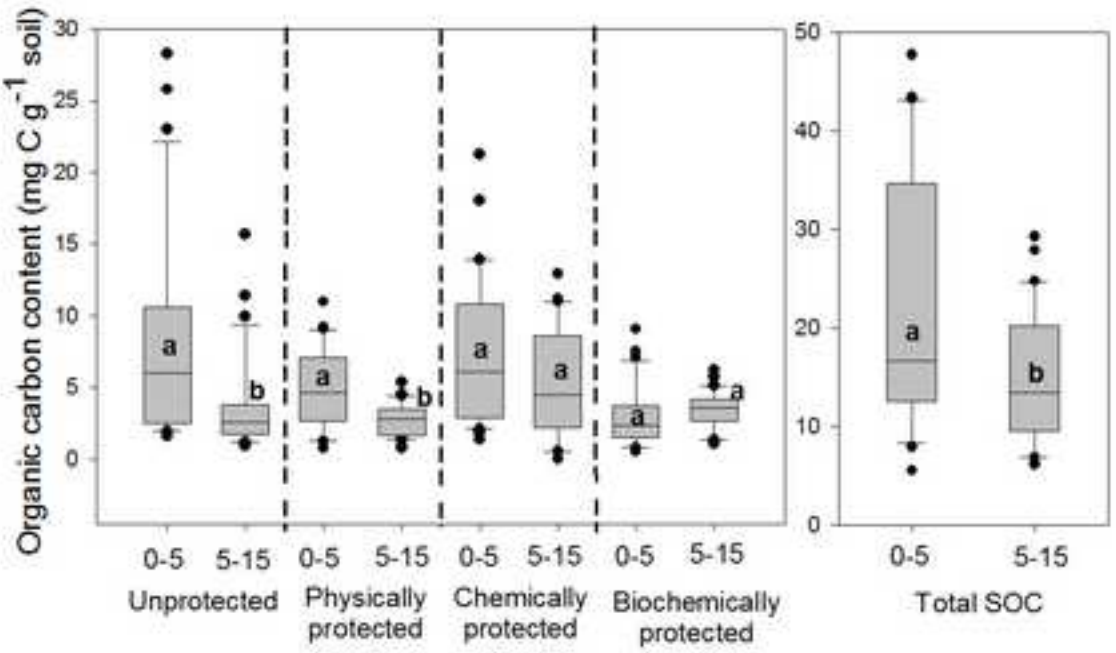

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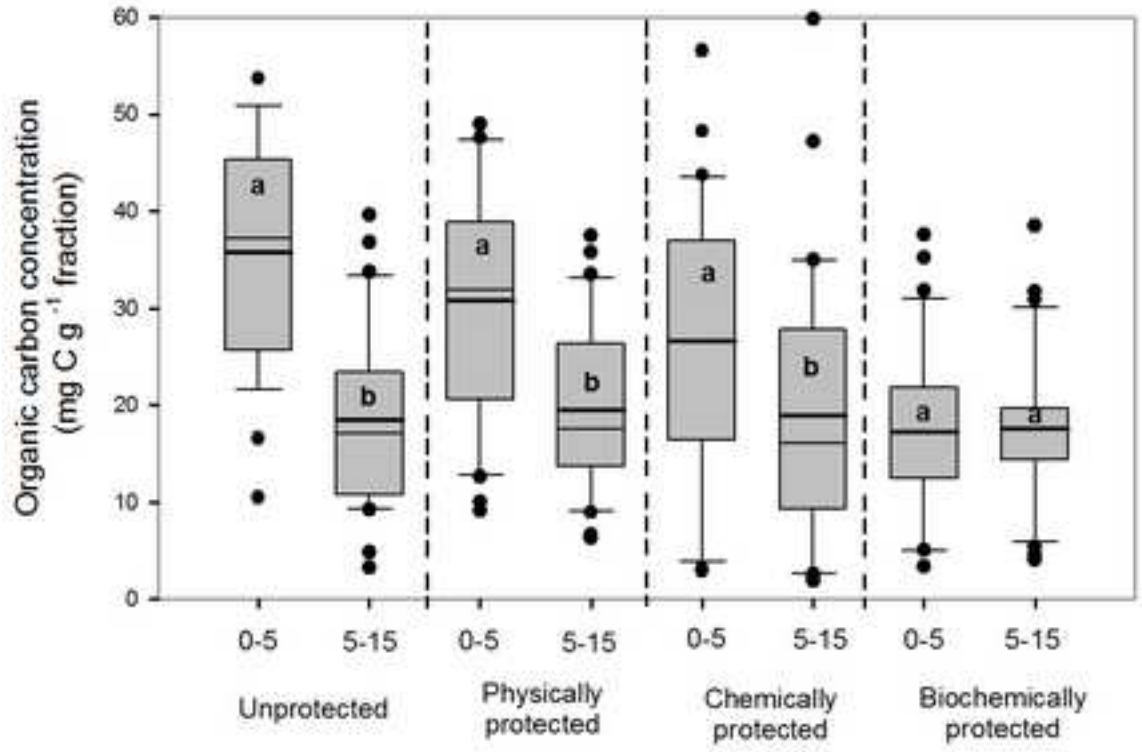

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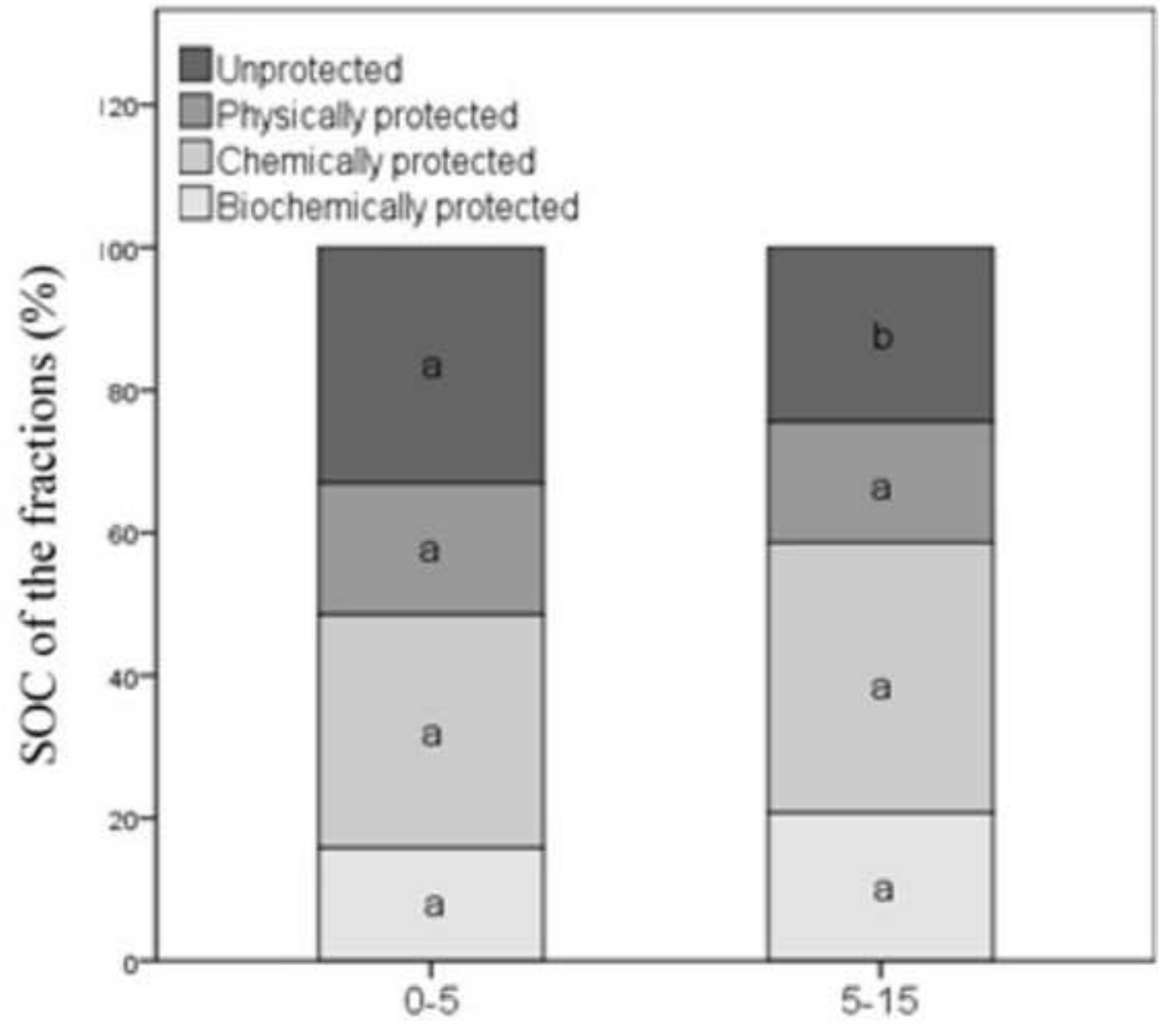

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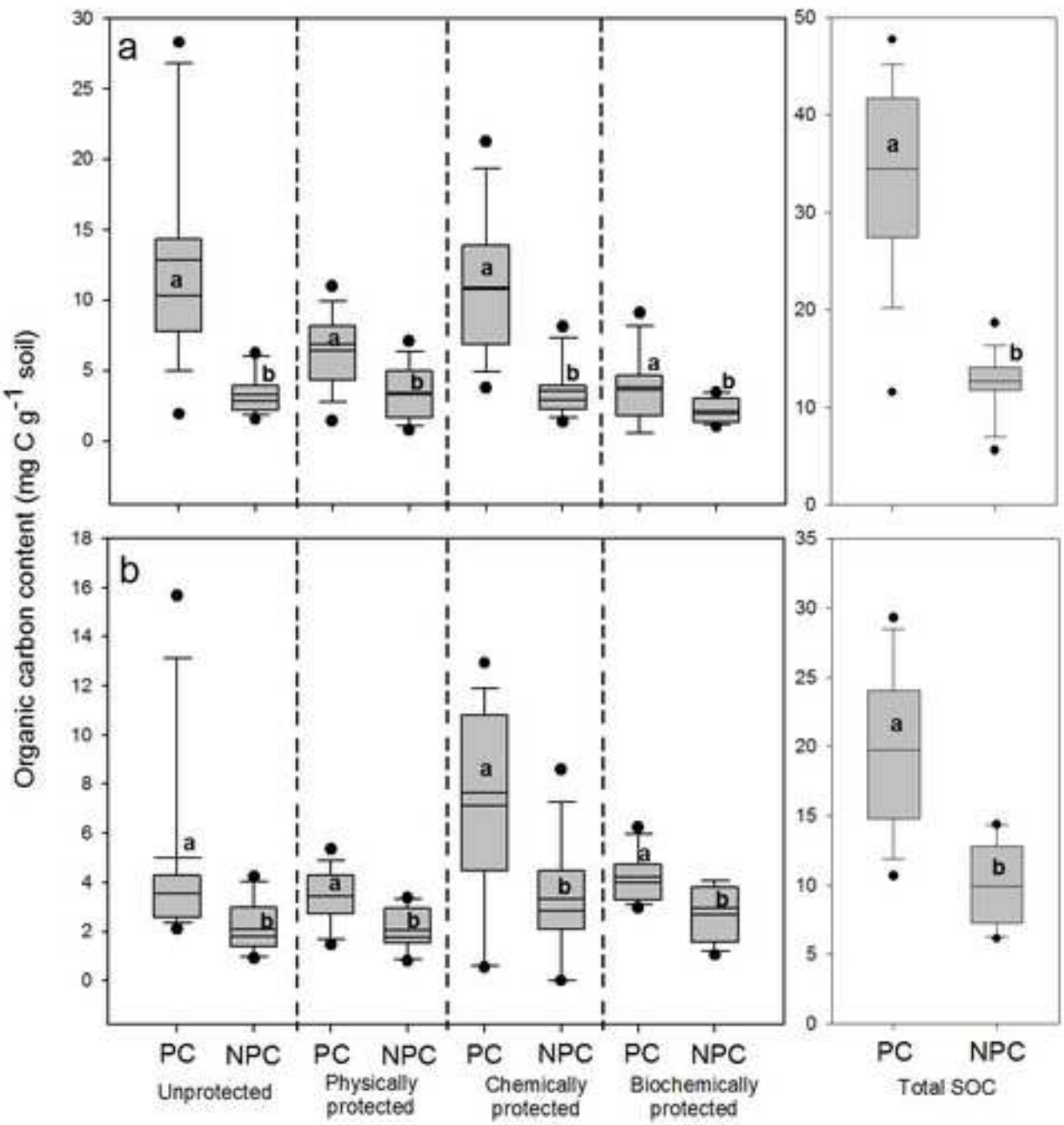

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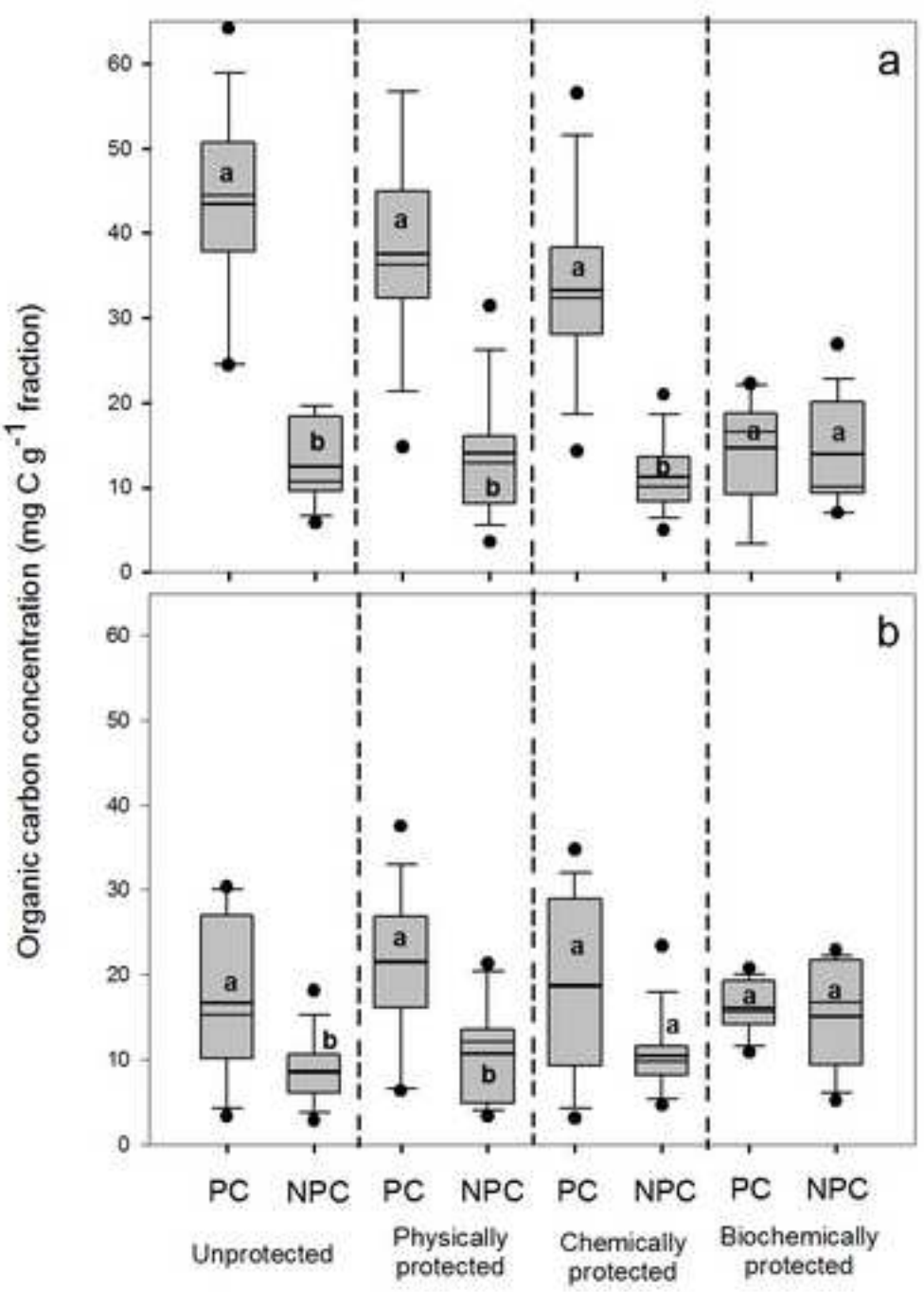

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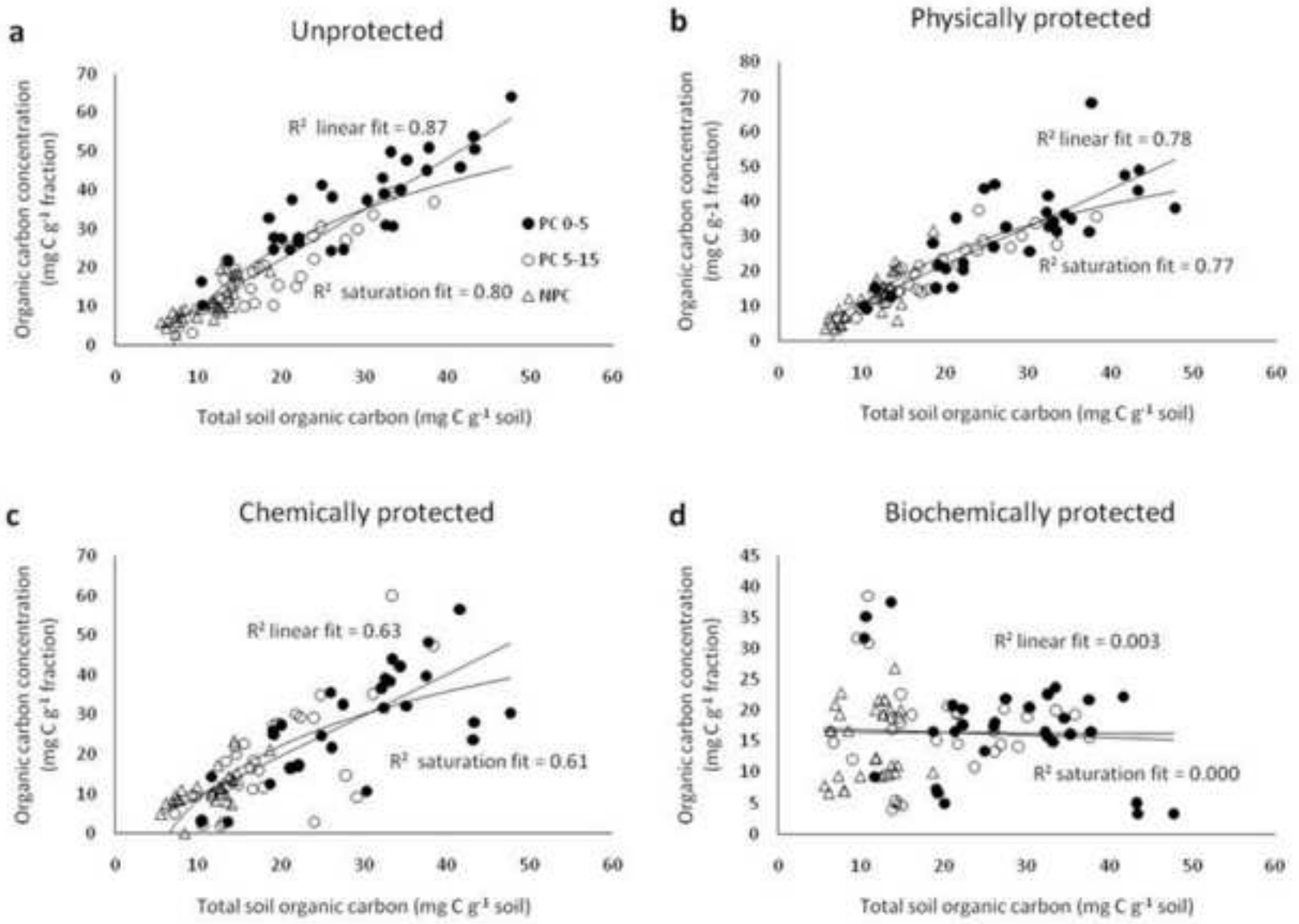

Figure captions

Figure 1. Frequency distribution of the annual production of aboveground biomass (a) and organic carbon (b) in the PC olive oil orchards.

Figure 2. Box-plot representation of whole SOC and unprotected, and physically, chemically and biochemically protected organic carbon of soils of 0 – 5 and 5 – 15 cm soils depth of PC olive oil farms. Boundaries of the boxes closest to, and furthest from zero indicate the 25th and 75th percentiles, respectively. The thin lines within the box mark the average. Bars above and below the box indicate the 90th and 10th percentiles, respectively. Outliers are represented as black dots. Average values with the same letter indicate no significant differences between depths ($p < 0.05$).

Figure 3. Box-plot representation of soil organic carbon concentration (mg C g^{-1} fraction) in the unprotected, and physically, chemically and biochemically protected organic carbon fractions of soils (0 – 5 and 5 –15 cm) of PC olive oil farms. Boundaries of the boxes closest to, and furthest from zero indicate the 25th and 75th percentiles, respectively. The thin lines within the box mark the average. Bars above and below the box indicate the 90th and 10th percentiles, respectively. Outliers are represented as black dots. Average values with the same letter indicate no significant differences between depths ($p < 0.05$).

Figure 4. Percentage contribution (on average) of soil organic carbon fractions to the whole SOC of soils (0 – 5 and 5 –15 cm) of PC olive oil farms. Average values with the same letter indicate no significant differences between management types ($p < 0.05$).

Figure 5. Box-plot representation of whole SOC and in the unprotected, and physically, chemically and biochemically protected organic carbon fractions of soils 0 – 5 cm (a) and 5 – 15 cm (b) of PC and comparable NPC olive oil farms. Boundaries of the boxes closest to, and furthest from zero indicate the 25th and 75th percentiles, respectively. The thin lines within the box mark the average. Bars above and below the box indicate the 90th and 10th percentiles, respectively. Outliers are represented as black dots. Average values with the same letter indicate no significant differences between management types ($p < 0.05$).

Figure 6. Box-plot representation of soil organic carbon concentration (mg C g^{-1} fraction) in the unprotected, and physically, chemically and biochemically protected organic carbon fractions of soils of 0 – 5 cm (a) and 5 – 15 cm (b)of PC and comparable NPC olive oil farms. Boundaries of the boxes closest to, and furthest from zero indicate the 25th and 75th percentiles, respectively. The thin lines within the box mark the average. Bars above and below the box indicate the 90th and 10th percentiles, respectively. Outliers are represented as black dots. Average values with the same letter indicate no significant differences between management types ($p < 0.05$).

Figure 7. Relationship between whole SOC (mg C g^{-1} soil) and soil organic carbon concentration (mg C g^{-1} fraction) of the (a) unprotected, and (b) physically, (c) chemically and (d) biochemically protected organic carbon fractions of top 5 cm (full circle) and 5 – 15 cm (empty circle) of soils of the PC olive oil farms, and 0 – 5 and 5 – 15 cm soils of NPC farms. Linear and saturation functions and R^2 coefficients are

included for each soil organic carbon fraction. All regressions are significant at $p < 0.05$ except those of the biochemically protected pool.