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The importance of incorporating soil in the life cycle assessment procedure to improve the sustainability of agricultural management

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Catena

The importance of incorporating soil in the life cycle assessment procedure to improve the sustainability of agricultural management --Manuscript Draft--

Manuscript Number:	CATENA18225R1
Article Type:	Research Paper
Keywords:	Cambisols; fruit orchards; soil organic C; CO ₂ eq; LCA
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First Author:	Mauro De Feudis
Order of Authors:	Mauro De Feudis Claudio Selmi Gloria Falsone Daniele Missere Marcello Di Bonito Livia Vittori Antisari
Abstract:	<p>The formidable ability of soil to store carbon has attracted an increasing number of studies, but few of them included soil organic carbon (SOC) sequestration as part of a carbon balance assessment in the agroecosystem. This raises some interesting questions: 1) how orchards conversion increase soil capacity to mitigate the green-house gases (GHG) emissions by storing C? 2) can it be considered in life cycle assessment (LCA)? 3) can SOC pools and soil biochemical properties determination improve LCA interpretation? To answer these questions, this study selected a ten- and fifteen-years-old peach orchards, a twenty-years-old pear orchard, a thirty-years-old kiwi orchard in south-east part of Emilia-Romagna Region (Italy), and a cereals' field as reference. Soil samples were collected from 0-15 and 15-30cm depths, and the SOC pool amounts (i.e., labile and recalcitrant) determined. LCA was used to estimate the GHG emissions (CO₂eq) from the orchards. Results showed that the conversion from cereals to orchard production increased OC stock (+82% on average) suggesting that orchards cultivation systems have the capacity to enrich soil organic matter. Fertilization had the greatest impact on CO₂eq emission accounting for at least 40% of total CO₂eq emissions. Kiwi cultivation had the highest impact on GHG emissions mainly due to the high water and nutrient demand (0.045 and 0.149 kg CO₂eq kg⁻¹ fruit yr⁻¹, respectively). When taking into account the C-CO₂eq loss by fruit cultivation and C storage in soils, results would indicate that peach and pear orchard agroecosystems promote C sequestration. Conversely, kiwi cultivation showed large CO₂eq emissions only partly counterbalanced by SOC sequestration. This study highlights the importance of including soils in LCA: if made mandatory this would allow a wider, yet more detailed, picture of the impact of agricultural practices on C budget. This simple step could help optimise resource management and at the same time improve agroecosystem sustainability.</p>
Suggested Reviewers:	Claire Chenu claire.chenu@inra.fr Sonja Keel sonja.keel@agroscope.admin.ch Tommaso Chiti tommaso.chiti@unitus.it Chukwuebuka Christopher Okolo okolo.chukwuebuka@mu.edu.et Wenting Feng fengwenting@caas.cn

Bologna, July 7th, 2022

Dear Editor,

on behalf of the co-authors, I submit the revised version of the manuscript entitled “*The importance of incorporating soil in the life cycle assessment procedure to improve the sustainability of agricultural management*” (CATENA 18225) by: Mauro De Feudis, Claudio Selmi, Gloria Falsone, Daniele Missere, Marcello Di Bonito and Livia Vittori Antisari for a possible publication in Catena.

All the comments from Reviewers were addressed and the changes to the manuscript are highlighted in yellow

The article falls in the aims and scope of the Journal, and is an original work, not published or under consideration for publication elsewhere.

Sincerely yours

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Ten-years-old peach orchard



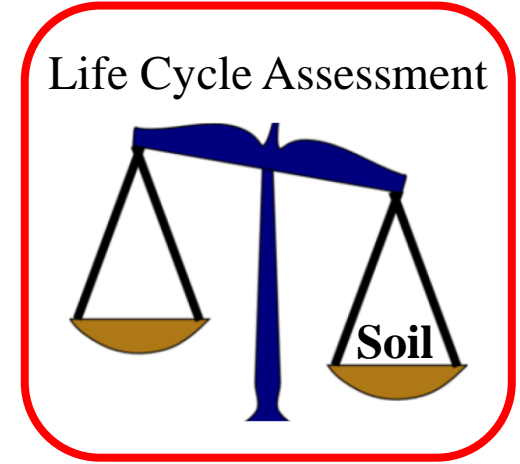
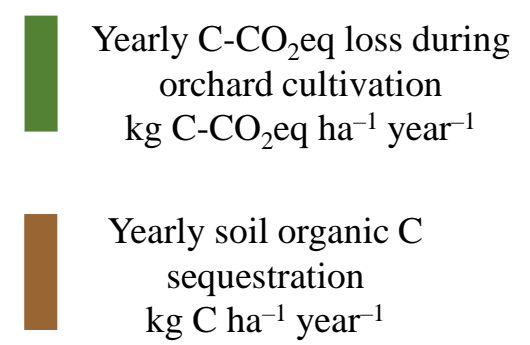
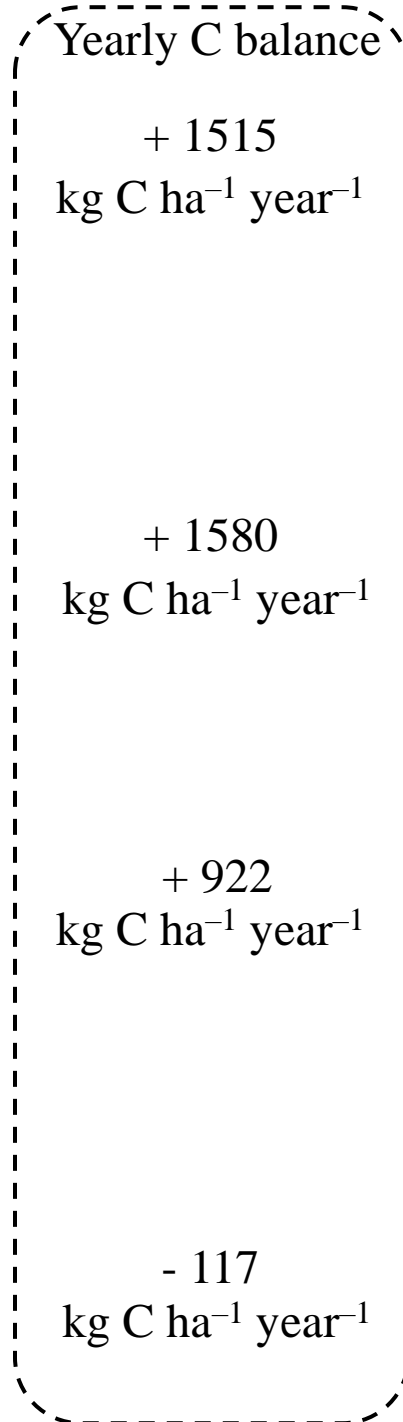
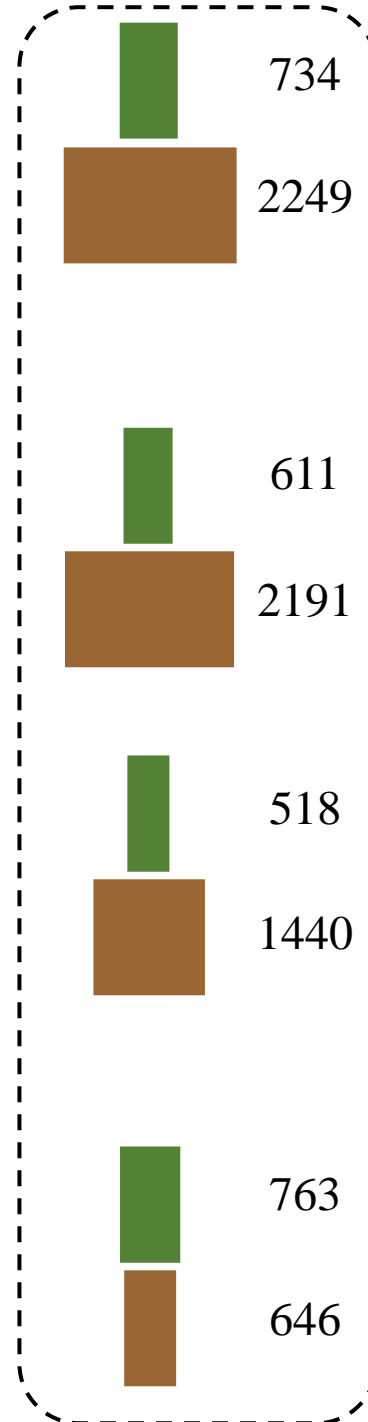
Fifteen-years-old peach orchard



Twenty-years-old pear orchard



Thirty-years-old kiwi orchard



Highlights

- C balance in peach, pear and kiwi orchard systems was investigated
- Fruit orchard cultivation promoted soil C sequestration
- Most of the C gained by soil was found in stable forms
- Soil counterbalanced the CO₂eq released in the atmosphere by agricultural practices
- LCA taking in consideration the soil resource should be promoted

Reviewer #1: This study by De Feudis et al. quantifies the impact of including changes in soil organic carbon (SOC) stocks in life cycle assessments related to greenhouse gas emissions of fruit orchards. To quantify changes in SOC they compared SOC stocks in a total of four orchards including peach, pear and kiwi with SOC stocks of a reference, which was a wheat field. A wide array of soil measurements complemented this study such as the extraction of specific carbon fractions, stable isotope analysis, and measurements of basal respiration. The SOC change rates they estimated are very high and led to positive C balances for three out of the four orchards.

For improving the sustainability of food production being able to quantify GHG balances is important. The topic of this study is therefore very relevant. Overall the experiments have been carried out with great care and the data presentation is good. The text is mostly well written, but there are many small English mistakes and I suggest to have it checked.

We thank the Reviewer 1 for the appreciation of the manuscript. The small English mistakes were corrected

My main concern are the very high SOC stock changes you report. They are much higher compared to previous studies for agroforestry systems: Cardinael et al., 2017, 2019; De Stefano and Jacobson, 2018; Shi et al., 2018 summarised in Wiesmeier et al. 2020 (Geoderma). On average a change of 0.7 Mg or t C ha⁻¹ year⁻¹ was reported. You have up to 2.2 t C ha⁻¹ year⁻¹ (Table 5)! Since these are among the most important results of your study, I think it is necessary to add more information. Please explain exactly how you calculated these values. Do you have any information related to the amount of shredded wood that was left on the field? Your estimates also strongly depend on the SOC stock at your reference cite.

The equation for calculating the yearly change of C stock into the soil was added at Line 256 of the new version of the manuscript.

At Lines 149-151 of the new version of the manuscript, the amount of pruned material was added

We agree with the reviewer that the C stock change in orchards depends on the C stock in the reference soil. In our case, CK had an average C stock of 31 Mg/ha as reported at Line 351 of the new version of the manuscript. The C stored in the investigated orchards was 57± 3 Mg/ha. These values were in agreement with previous studies conducted in Europe (Álvaro-Fuentes et al., 2012; Bateni et al., 2021;; Funes et al., 2019). We have now specified it at Lines 358-360 of the new version. However, since the soil of the orchards was kept covered by natural grasses (see Line 130 of the old version of the manuscript and Line 148 of the new version one), a relative high increase of C stock in orchards was quite expected. Indeed, the presence of a such herbaceous plants established on whole surface of the fields it is worldwide recognized to increase soil C stock (de Torres et al., 2021; Xiang et al., 2022; Novara et al., 2019) The role of the permanent grasses on soil organic C stock is now reported at Lines 361-369 of the new version of the manuscript).

How similar was the soil of the wheat field and do you have e.g. information regarding clay contents to compare?

The clay content in the investigated soils is similar (see data reported below) and now it is showed in Table S1 of the supplementary materials

	CK	Ph10	Ph15	Pr20	Ki30
Clay (g kg ⁻¹)	201 (44)	200 (28)	242 (52)	187 (20)	243 (73)

I was rather surprised (and impressed!) by the many different soil analysis you did as for a C balance it would have been sufficient to just analyse total C. You may want to add a few words in the introduction to explain what the advantage of including these analysis is.

The investigated soil parameters allowed us to understand the dynamics promoting C stabilization processes and, thus, the increase of C sequestration as stable C forms in soil. In this sense, some more words at Lines 109-116 of the new version of the manuscript and a further aim (Lines 26 and 121-122 of the new version of the manuscript) were added. Thank you for the suggestion.

Minor comments:

I honestly do not understand how you decide how many arrows to use in the graphical abstract and suggest to replace them by bars directly related to the size of the C fluxes.

The graphical abstract was modified accordingly

The first statement of the highlights is very vagues. Perhaps add "C balances"

The highlight n.1 was modified as follow: C balance in peach, pear and kiwi orchard systems was investigated

L52: I would add: "...and CH₄ emissions from enteric fermentation". Additionally you may want to explain that emissions are not only associated with the energy intensive production of fertilizers, but that the addition of N fertilizer is responsible for N₂O emissions (you mention that only in a footnote of a table, but I think it is important for the readers to know).

At Line 52 of the new version of the manuscript we specified that we referred to the agricultural crop production systems. Further, at Line 54 of the new version of the manuscript we added the role of the agricultural N inputs to N₂O emissions.

L71: C pools vary largely regarding their turnover rate and some fractions also turn over rapidly.

At Lines 75-77 of the new version of the manuscript the different turnover of soil organic carbon fractions was reported

L82: There are several reviews that show that no tillage only leads to a change in the vertical distribution of C in soil (more in topsoil, less in subsoil). The net effect on the C budget is zero. (Luo et al. 2010, Agr Eco Environ)

The sentence was modified highlighting that at global scale the no-tillage does not affect SOC content (see Lines 90-91 of the new version of the manuscript)

L137: please change to "grain production" (or cereal production)

Done, thank you

L216: perhaps repeat abbreviations here.

The explanation of the abbreviations were added (see Lines 259 of the new version of the manuscript)

L236: Please specify whether results are for 0-30 cm.

Done, thank you (Lines 284-286 of the new version of the manuscript)

L308: Please mention the SOC change rates of these studies as your values are very high (see above).

We agree with Reviewer 1 that the stock change rates are high, but the C stock values of both CK and orchards are similar to the values reported in literature. Please, see previous reply.

L313: Not only rhizodeposition, but also root turnover is an important source of C input to the soil.

Root turnover as source of C was added (see Line 367 of the new version of the manuscript)

L320: Which data are you exactly referring to? For surface soils, I see a significant difference of nearly 2 per mille between the wheat control and Pr20!

As showed in Figure 3, 0-15 cm soil depth in Pr20 had higher values compared to Ck, but it is the only one difference. The common trend in the studied sites is however to show quite homogenous values of C13. We added therefore the term “generally” (see Line 377 of the new version of the manuscript)

Cited literature

Álvaro-Fuentes, J., Easter, M., Paustian, K., 2012. Climate change effects on organic carbon storage in agricultural soils of Northeastern Spain. *Agric. Ecosyst. Environ.* 155, 87-94.

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Novara, A., Minacapilli, M., Santoro, A., Rodrigo-Comino, J., Carrubba, A., Sarno, M., Venezia, G., Gristina, L., 2019. Real cover crops contribution to soil organic carbon sequestration in sloping vineyard. *Sci. Total Environ.* 652, 300–306.

Reviewer #2: The paper has the correct ambition to include in a more stable way the SOC sequestration into the LCA of agricultural systems. The way investigated by the authors was a comparison between orchards of different age against cropland in northern Italy.

I have found the paper easy to follow in its structure. Anyway, despite the interesting topic and the ease of reading, I see some major issues in the submitted manuscript in several parts. I report my major comments here, and attached is also a pdf file with other points that should be followed to improve this piece of work.

We thank the Reviewer 2 to appreciate our manuscript and his/her comments were addressed

- A first big issue is related to the methodological approach. First of all, authors should consider that soil C stock potential depends on the soil type and its natural capacity to store C, in particular the clay content. Plenty of papers have discussed this in details, also in the Italian context of northeastern Italy. Therefore, a simple comparison between agricultural systems is excluding very important sources of variability, and I think they were not included in the analysis so far.

We agree with Reviewer 2 that soil type and clay content influence the capacity of soil to store C. All the investigated soils are Cambisols, as indicated at Line 124 in the old version of the manuscript (now Line 141), and their clay content was similar (as now reported in Table S1). We apologize that we did not report such data. In Table S1 of the Supplementary materials the clay content and the equivalent soil mass are reported which show similar values among the study sites.

Second, why not including deep SOC changes? The most recent literature is focussing on such topic, which could be very important for tree cultivation. Please provide justification about it.

We agree with Reviewer 2 that deep soil is crucial in the C soil storage, as a great amount of organic C can be stored below to 30 cm. The choice to focus our attention on C included in 0–30 cm soil depth interval was based on the fact that this depth is still widely used (e.g., Guevara et al., 2020; Tangen and Bansal, 2020) and, therefore, can allow an easy comparison of our study sites with the other agricultural systems worldwide. This procedure is also recommended by FAO (Makipaa et al., 2012). The rationale to investigate the upper 30 cm layer was added at Lines 163-165 of the new version of the manuscript.

Furthermore, in the Discussion and Conclusion sections a reminder to the importance to consider the deep soil was added (see Lines 458-463 and Lines 481-484 of the new version of the manuscript)

Third, what about the equivalent soil mass? Authors should include it to compare the masses of soils there could be subjected to SOC stock changes, otherwise the comparison is biased unless the entire soil profile is sampled. This is particularly true for soils with very different bulk densities (arable vs. permanent) that can also be subjected to some compaction, and in cases where a global vision about C cycle is searched.

We agree with Reviewer 2 that it is important to consider the equivalent soil mass to better understand SOC stock changes. In this sense, in Table S1 of the Supplementary materials the equivalent soil mass is reported which show similar values among the study sites.

- I see some problems in results related to Figure 2, where authors compare different agricultural systems that are not comparable because can be affected by uncontrolled factors other than the agricultural system. I think this type of comparison is possible only if several orchards of the same type are included as replicates, while soil samplings within each system are sub-replicates. This highlights a very weak point of the paper, that is using single fields with peculiar properties to broaden results to general conclusions.

We agree with Reviewer 2 that it is not possible to compare different agricultural systems. However, in the present study, the general aim was to test if the inclusion of soil organic carbon stock in LCA can improve the LCA approach, in particular we tested whether soil can contribute to mitigate the GHG emissions related to the conversion of field for grain production to orchard. Our aim was not to compare the orchards one with another. In this sense, the text was revised in order to better highlight the aims of the manuscript

and any sentence referring to the comparison among orchards has been removed.

- I see a mismatch between the experimental SOC, microbial and mineralization characterization, and the LCA approach. Where is the first functional and determinant for the second? This is not reported in aims, results and discussion. A reader at the end does not understand why including these two parts in a single paper, and I have doubts that it is useful to do so.

To better understand the importance of SOC forms and soil biochemical properties for LCA, the Introduction section, aims and the Discussion section were modified, See also reply to Reviewer 1 comments.

- Methodologically, very poor information is reported about the LCA analysis, the used data and how experimental data were embedded into the LCA model.

More details about LCA analysis were added (see Lines 206-240 and Lines 276-280).

With regards to how experimental data can be embedded into the LCA, our findings showed the crucial role soil C stocks can have in the balance of CO₂-eq estimation in the agroecosystem. Thus, as reported at Lines 480-481 of the new version of the manuscript, we propose to insert the soil C storage rate as CO₂ soil uptake from atmosphere lowering the environmental impacts of orchards management.

- I think that comparing the CO₂eq impact of orchards per year is not correct, when it is likely hypothesized a "life cycle" of, let's say, 25-35 years per orchard (how many years? I am not a specialist about it). I mean, the LCA should include the entire life cycle, therefore results from orchards of 10 rather than 20 years are expected to have different impacts, but projections about the entire orchard cycle should be considered. Probably, some methodology, results and discussion should be around the average age of different orchards according to different tree species.

The LCA should compare the whole cycle, and to me is rather obvious that the impact of establishment is higher when the same orchard (here peach) is younger.

We agree with the Reviewer 2 that the removal of trees and preparing the ground for a new growing cycle stage and the mean life cycle of the orchards could be considered to assess the impact of the whole life of agricultural systems on GHG emissions. However, the LCA has been often used for the assessment of environment impact of agricultural systems over any time span (e.g., Cerutti et al., 2014; Linderholm et al., 2012; Haas et al., 2001; Paolotti et al., 2016; Tricase et al., 2018). Our aim was to assess the role of soil to mitigate the GHGs emissions due to orchard cultivation after a certain time from conversion of a cropland to orchard cultivation. Thus, the whole lifetime of orchards has not been considered in this study. The time boundaries of the LCA used for the present study were better highlighted within the subsection "Life Cycle Assessment (LCA) of peach, pear, and kiwi production" of the new version of the manuscript

- The discussion is not organized in a manner that experimental results are integrated in LCA. Moreover, major issues are related to, e.g., lack of discussion on deep SOC and layering, lack of discussion on possible mineralization or changes in SOC stock when orchards must be renovated, which contrasts with long-term duration of croplands; 3. changes in the crop/orchard management that can stringly modify the obtained results (e.g. organic production, different types of structures and actions against pests and diseases?). Regarding this last point, it could be very helpful a sensitivity analysis.

With regards to the first point, in order to better integrate the experimental results in LCA, the subsection "Carbon balance" was modified accordingly.

Concerning the crucial role of deep soil in the C soil storage, please see above reply to your comment.

With regards to the third point, we agree with Reviewer 2 that change in orchard management can modify its environment impact, but the comparison among orchards management was out of our aim.

Line 22: better "has attracted" because largely investigated

Done, thank you

Line 24: I think the question is not well posed. I think that are the soils in orchard systems that can help to mitigate GHG emissions from the agricultural systems.

We have reformulated the first question. The question now is: 1) how orchards conversion increase soil capacity to mitigate the green-house gases (GHG) emissions by storing C?

There is some repetition with concepts already reported in L33-35.

The repetitions were removed

Line 40: more?

Done, thank you

Introduction: to give emphasis to recent renovation about the role of SOC, I suggest to include recent literature about Carbon farming initiative in Europe.

We emphasized the role of SOC in carbon-farming initiative at Lines 65-67 of the new version of the manuscript

Line 70: I suggest revising up-to-date bibliography. See for instance <https://doi.org/10.1111/gcb.14066> or <https://doi.org/10.1111/gcb.14054>

We updated the cited literature (see Line 74 of the new version of the manuscript)

Line 96: this should be better defined. What is non-standardized?

We deleted it.

L98-L100. I do not understand where is the uncertainty in the approach. Maybe uncertainty in the results of the real contribution of agricultural systems to mitigate climate change? From my point of view, the focus should be given more to the limited number of studies that included SOC stock and all other management aspects in the valuation of potentials of agricultural systems to contribute to climate change mitigation. Besides: are there any justifications on why LCA was not broadly applied so far to agricultural systems?

The LCA approach was broadly applied for agricultural systems (Cerutti et al., 2014; Goossens et al., 2017; Zhao et al., 2021; Aguilera et al., 2021) but without considering soil. However, this part was modified highlighting the limited number of studies that included SOC stock in LCA. (see Lines 105-107 of the new version of the manuscript)

Line 107-108: I see this statement obvious as it is. You are saying that each management- and site-specific farming practice provides different mitigation/emission results...

This part was removed

L 137: grain

Done thank you

Line 198: Authors have not included the end-cycle of the orchard? Cropland cultivations are repeated year by year, while orchards need renovation after some time, which often include soil disturbance and tree disposal.

The aim of the present study was to assess if SOC can be included in LCA approach for agricultural ecosystems. To address this goal it was not necessary to take into account the end-cycle of the considered orchards. Further, for the LCA there are not fixed boundaries, but they can be decided according to the aims. In our case, the system boundary considered since the extraction of raw materials of inputs up to the farm gate when the fruits are harvested and the data for LCA were taken for the whole life cycle starting from the period of farm establishment till the time of performing this study

I would have expected more details about all other factors that I suppose were included in the LCA, such as fuel consumption, fertilizations, irrigation practices etc. I think they should be included and briefly explained, including the assumptions that have been made.

The LCA analysis description was improved through the addition of more details (see Lines 206-240 and Lines 276-280).

Was biomass not included because at the end of the orchard cycle what was gained becomes a disposal?

The plant biomass was not included because the end-cycle it will become a disposal that will be burned. This part was added at Lines 442-443 of the new version of the manuscript

L236-239: Please provide clarification on where it was found the concentration and stock that has been reported

In the new version of the manuscript (see Lines 284-286) we reported the values referred to 0-30 cm depth

Line 240: this suggests that in the lines before authors were dealing with topsoil. Not clear if this is correct. Please clarify

The lines before were modified

L252 and following: Where is SOC pool characterization functional to the LCA assessment that is the main aim of the paper? A reader does not understand this as much.

I see very little union between experimental data LCA analysis. This is confusing me and likely other readers, because it is not clear why experimental results about SOC mineralizations and microbial characterization can be useful to the second part that is the LCA. A strong storytelling merging everything is missing.

To clarify the importance of both SOC pools and soil biochemical properties for the LCA outputs, the introduction section, the aims and the discussion section were modified

A see a major issue in the way of presenting and discussing the CO₂-eq data. Why are agricultural practices different between 10yr and 15yr peach orchards? Shouldn't be the same per year?

Also the fertilizer emission is confusing because it is likely a site-specific aspect that does not matter with the years of orchard establishment, at least when the same fruit is cultivated.

The investigated orchards are different to each other. Our aim was not to compare orchards management, but to test if SOC stock can be included in LCA approach to emphasize the role of soil to mitigate the GHG emissions coming from the cultivation of orchards after the conversion of a field for grain production.

Cited literature

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1 **The importance of incorporating soil in the life cycle assessment procedure to improve the**
2 **sustainability of agricultural management**

3

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19

20

21 **Abstract**

22 The formidable ability of soil to store carbon **has attracted** an increasing number of studies, but few
23 of them included soil organic carbon (SOC) sequestration as part of a carbon balance assessment in
24 the agroecosystem. This raises some interesting questions: 1) how orchards conversion increase soil
25 capacity to mitigate the green-house gases (GHG) emissions by storing C? 2) can it be considered in
26 life cycle assessment (LCA)? 3) **can SOC pools and soil biochemical properties determination**
27 **improve LCA interpretation?** To answer these questions, this study selected a ten- and fifteen-years-

28 old peach orchards, a twenty–years–old pear orchard, a thirty–years–old kiwi orchard in south-east
29 part of Emilia–Romagna Region (Italy), and a cereals’ field as reference. Soil samples were collected
30 from 0–15 and 15–30cm depths, and the SOC pool amounts (i.e., labile and recalcitrant) determined.
31 LCA was used to estimate the GHG emissions (CO₂eq) from the orchards. Results showed that the
32 conversion from cereals to orchard production increased OC stock (+82% on average) suggesting that
33 orchards cultivation systems have the capacity to enrich soil organic matter. Fertilization had the
34 greatest impact on CO₂eq emission accounting for at least 40% of total CO₂eq emissions. Kiwi
35 cultivation had the highest impact on GHG emissions mainly due to the high water and nutrient
36 demand (0.045 and 0.149 kg CO₂eq kg⁻¹ fruit yr⁻¹, respectively). When taking into account the C–
37 CO₂eq loss by fruit cultivation and C storage in soils, results would indicate that peach and pear
38 orchard agroecosystems promote C sequestration. **Conversely, kiwi cultivation showed large CO₂eq**
39 **emissions only partly counterbalanced by SOC sequestration.** This study highlights the importance
40 of including soils in LCA: if made mandatory this would allow a wider, yet **more** detailed, picture of
41 the impact of agricultural practices on C budget. This simple step could help optimise resource
42 management and at the same time improve agroecosystem sustainability.

43

44 **Keywords**

45 Cambisols, fruit orchards, soil organic C, CO₂eq, LCA

46

47 **Introduction**

48 In 2020, carbon dioxide (CO₂) concentration in the atmosphere reached values greater than 410 ppm
49 due to the human activities (The World Meteorological Organization, 2020). Agriculture is
50 recognised as a significant contributor to anthropogenic emissions of CO₂ (Smith et al., 2014; Lynch
51 et al., 2021). Recent studies (Gkissakis et al., 2020; Goossens et al., 2017; Mousavi-Avval et al., 2017;
52 Pryor et al., 2017) pointed out that the GHG emissions **from agricultural crop production systems** are
53 mainly related to the fossil-fuel consuming and to the manufacturing and distribution of chemical

54 fertilizers. Noteworthy is also the production of nitrous oxide (N₂O) gas due to soil nitrogen input
55 (Lawrence et al., 2021). Consequently, a reduced utilization of both fuels and fertilizers could
56 improve the sustainability of agricultural management. For example, Aguilera et al. (2015) compared
57 the environmental impact of several conventional and organic cropping systems in Spain,
58 highlighting greater GHG emissions in the formers compared to the latter, mainly due to the use of
59 chemical fertilizers in the conventional system. Similarly, Pergola et al. (2017) found a greater impact
60 on climate change of apricot orchards under integrated system compared to those under biodynamic
61 one. In the context of the current climate change, soil plays a central role in the mitigation of GHGs
62 emission from agriculture through soil carbon sequestration, defined by Chenu et al. (2019) as “*the*
63 *process of transferring CO₂ from the atmosphere into the soil of a land unit, through plants, plant*
64 *residues and other organic solids which are stored or retained in the unit as part of the soil organic*
65 *matter*”. In this sense, worldwide there is a strong agreement to implement the carbon-farming
66 initiatives with the main aim to increase the soil organic carbon (SOC) stock which is a way to
67 mitigate the current climate change (Wiesmeier et al., 2020; Bradford et al., 2019; Chenu et al., 2019).
68 However, to reach this goal, the chemical, physical and edaphic conditions of the soil must allow the
69 humification process and the accumulation of organic C to be carried out rather than the
70 mineralization process. Soil stores three times the amount of C present in the atmosphere (Ciais et
71 al., 2013) and could potentially remove from the atmosphere between 0.79 and 1.54 Gt yr⁻¹ of C (Fuss
72 et al., 2018) if land uses and management practices increased C inputs and/or reduced C losses. In
73 this sense, promoting soil organic C (SOC) sequestration is one of the most important strategies to
74 reduce atmospheric CO₂ concentrations with a significant potential to mitigate climate change (Lal,
75 2018). Bulk SOC is composed of multiple functional pools differing in turnover, in fact it ranges from
76 the most labile form (i.e., the dissolved organic C) to the most stable one as the physically protected
77 and the chemically recalcitrance forms (De Feudis et al., 2019; Poeplau et al., 2018). The long
78 residence time associated with most of the SOC pools (e.g., De Feudis et al., 2019; Ferreira et al.,
79 2020) makes soil a major player in the global carbon budget (Martin et al., 2014). Moreover, soils

80 characterized by high SOC concentrations are recognized to be desirable because SOC improves soil
81 nutrient availability, cation exchange capacity, water retention capacity, soil aeration, soil aggregation
82 and structure, soil microbial biomass and its activity, plant yield and quality (Bationo et al., 2007;
83 Bronick and Lal, 2005; Chavarria et al., 2018; Martínez-Mena et al., 2021).

84 There is general agreement that management practices are important factors influencing SOC
85 contents in agricultural lands (Montanaro et al., 2017; Novara et al., 2019; Pardon et al., 2017). For
86 example, the cultivation of cover crops has been identified as an effective practice to increase of SOC
87 content (Poeplau and Don, 2015). Similarly, practices addressing the incorporation of the plant
88 residues into the soil could prevent SOC reduction (Keel et al., 2019). The no-tillage has been claimed
89 to be a potential option to decrease SOC loss in agricultural soils (Nath and Lal, 2017), **but at global**
90 **scale its effect on SOC content seems to be limited (Mondal and Chakraborty, 2022).** Moreover, it is
91 well known the increase of SOC content when organic fertilizers are applied (Morugán-Coronado et
92 al., 2020).

93 In this context, life cycle assessment (LCA) is a well established approach to help accounting for all
94 the various stages of any activity, including agricultural practices where it was introduced since 1990
95 (Haas et al., 2000). LCA is one of the most used standardized methodologies for estimating the
96 environmental impacts linked to the entire cycle of fruit production (Vinyes et al., 2015). Among the
97 environmental impacts, the evaluation of GHG emissions prevail compared to the other
98 environmental problems (Adewale et al., 2019; Bartzas et al., 2017; Rebolledo-Leiva et al., 2017).
99 Most of the studies concerning LCA in agroecosystems take in account yield, plant growth and all
100 the input factors related to the crop cultivation such as human labour, machinery, fertilizer
101 application, fossil fuel consuming and irrigations (e.g., Foteinis and Chatzisyneon, 2016; Kaab et al.,
102 2019). Conversely, despite its high potential to store carbon, soil is generally not included in LCA
103 approach for the evaluation of C budget (Garrigues et al., 2012). Only in few cases SOC was taken
104 into account for the LCA (Arzoumanidis et al., 2014; Brandão et al., 2013; Petersen et al., 2013).

105 **Hence, although SOC is essential if LCA is to be applied in case studies where carbon balances must**

106 be calculated, the limited number of LCA studies that took into consideration SOC would highlight
107 how soil is generally the forgotten part of the agro-ecosystems. In addition, although the estimation
108 of the bulk SOC stock could be sufficient for C balance in LCA approach, the knowledge of SOC
109 pools and their dynamics are necessary for improving the interpretation of LCA outputs. Specifically,
110 since the important role of LCA to improve the management of agricultural systems for preventing
111 environmental hazards (e.g., the GHGs emissions) in the long-term, the agricultural managements
112 and/or systems able to promote the storage of the most stable SOC forms should be promoted.
113 Therefore, for a reliable C balance through the LCA procedure, it is important that soil C is stored in
114 the most stable forms. Further, because of the key role of soil microbial community to transform and
115 stabilize SOC (Angst et al., 2021; Domeignoz-Horta et al., 2021), the evaluation of their properties
116 (e.g., amount and activity) could be of interest in LCA to understand whether (or not) soil stabilize
117 C.

118 This study tries to address this gap in the literature and provide a justification for a more widely
119 accepted introduction of soil in agroecosystems LCA. In particular, the study will focus on *i*) how
120 orchards conversion increase soil capacity to mitigate the green-house gases (GHG) emissions by
121 storing C; *ii*) how soil C stock can therefore be included in LCA approach; and *iii*) if SOC pools and
122 soil biochemical properties determination can improve LCA interpretation. In order to address these
123 aims, the following hypotheses were set: 1) orchards increase soil C stock compared to grain fields;
124 2) and soil C storage capacity can mitigate the GHG emissions related to the fruit orchard agricultural
125 practices.

126

127 **Materials and Methods**

128 *Study sites description*

129 The present study was conducted in the south-east part of Emilia Romagna Region, Italy. This area
130 had a mean cumulative annual precipitation of 763 mm and a mean annual air temperature of 14.2 °C
131 for the period 1986 – 2015. The study was conducted in 2017, and the specific study site selected

132 included a ten- and fifteen-years-old peach orchards (Ph10 and Ph15, respectively) with a tree
133 density of 1,300 plants ha⁻¹; a twenty-years-old pear orchard (Pr20) with a tree density of 820 plants
134 ha⁻¹, and a thirty-years-old kiwi orchard (Ki30) with a tree density of 710 plants ha⁻¹ (see Fig. 1).
135 Some more details of study sites are reported in Vittori Antisari et al. (2021). The choice of the
136 selected tree species was based on their wide distribution in Italy. The mean yields of the selected
137 orchards were 48, 35, 30 and 28 × 10³ kg (fresh weight) ha⁻¹ for Ph10, Ph15, Pr20, Ki30, respectively.
138 According to the farmers, such yields were reached within the fifth year after the orchard
139 establishment. However, because of the missing data about yield during first years of orchard
140 cultivation, in the present study we arbitrarily considered the aforementioned yields also for the first
141 years of cultivation after orchard establishment. All the soils were classified as Cambisols with a
142 texture from silty clay loam to loam, a slight alkaline reaction (pH = 7.7 on average) and bulk density
143 ranging from 1.14 to 1.59 g cm⁻³, with lower values in 0–15 cm compared to 15–30 cm soil layer. On
144 average, cation exchange capacity of the soils studied was 24.9 cmol(+) kg⁻¹, the exchangeable Ca²⁺,
145 Mg²⁺ and K⁺ concentrations were 15.7, 1.9 and 0.60 cmol(+) kg⁻¹, respectively, and the base
146 saturation was of 75.4%. Details about clay content and equivalent soil mass of the study sites are
147 reported in Table S1 of the Supplementary materials.

148 In the orchards, soil was kept covered by natural grasses which were periodically cut (4–5 times per
149 year). Pruned wood materials were shredded and left on the soil surface. According to the farmers,
150 the average amount of pruned materials for Ph10, Ph15, Pr20 and Ki30 were 3.0, 3.0, 2.5 and 3.5 Mg
151 dry matter ha⁻¹. Some differences occurred for fertilization treatments (Table 1). In Ph10, no chemical
152 fertilization was performed, but exhausted substrate for mushroom cultivation at a rate of 7 Mg ha⁻¹
153 was spread on soil surface every year. In Ph15, Pr20 and Ki30, fertilization was carried out both by
154 fertigation, through drip irrigation lines (one line per plant row), and foliar spray. The amounts of
155 elements applied by fertilization is reported in Table 1.

156 To estimate C accumulation/loss of fruit orchard soils, a field for grain production (wheat) was used
157 as reference (CK). The rationale to use a field for grains production as reference soil was based both

158 on the widespread cultivation of such crops in the northern Italy and because the considered fruit
159 orchards were formerly wheat fields for at least 5 years.

160

161 *Soil sampling and analyses*

162 Within each field, three 30 cm depth soil pits were dug, and soil samples were collected from 0–15
163 cm (hereafter, surface soil) and 15–30 cm depths (hereafter, subsurface soil). This study used the
164 convention to investigate the 0–30 cm soil depth interval because such interval is worldwide used for
165 the SOC stock evaluation (Makipaa et al., 2012; Guevara et al., 2020; Tangen and Bansal, 2020). The
166 surface and subsurface soil samples were air-dried, passed through a 2-mm sieve and then an aliquot
167 was finely ground for SOC and total nitrogen (TN) concentrations determination.

168 SOC and TN were determined by a CHN elemental analyser (EA 1110 Thermo Fisher, USA) after
169 addition of hydrochloric acid to remove carbonates. The relative abundance of C and N stable isotopes
170 were determined by continuous flow- isotope ratio mass spectrometry (CF-IRMS) using an isotopic
171 mass spectrometer Delta V advantage (Thermo- Finnigan, DE). Measurements were expressed in
172 standard δ ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) notation (‰) relative to Vienna Pee Dee Belemnite and air, respectively.

173 Different SOM fractions, like particulate organic matter (POM), fulvic-like and humic-like
174 substances, and non-extractable organic matter (NEOM), were chemically extracted (Agnelli et al.,
175 2014). A volume of 100 mL of distilled water were added to 10 g of soil and shaken on a horizontal
176 shaker for 16 h at 25 °C, centrifuged and the supernatant was separated from the precipitate. The
177 supernatant was passed through a 53 μm sieve and the particles $>53 \mu\text{m}$ represented the POM. The
178 precipitate remaining into the centrifugation tubes was re-suspended in 100 mL 0.1 M NaOH + 0.1
179 M $\text{Na}_4\text{P}_2\text{O}_7$ solution and the samples were shaken for 24 h at 25 °C and then again centrifuged. The
180 NaOH extract was passed through a 0.45 μm polycarbonate filter, while the remaining precipitate,
181 containing NEOM was washed using deionized water to remove the excess of Na until the pH of the
182 rinsed solution was ≤ 7 . The 0.45 μm filtered NaOH extract was acidified to about pH 1.5 with 6 M
183 HCl and allowed to settle overnight to separate fulvic-like and humic-like substances and

184 centrifuged. To remove the excess of Na from the obtained fractions, the supernatant (fulvic-like
185 substances) was dialyzed through 1000 Da cut-off membranes (Spectra/Por® Dialysis membrane)
186 against distilled water, while the residual (humic-like substances) was washed with 0.002 M HCl.
187 Both purified fractions were freeze-dried. The POM and NEOM fractions were dried at 40 °C. The
188 organic C (OC) and N contents of POM, fulvic-like, humic-like substances and NEOM were
189 determined by a CHN elemental analyser (EA 1110 Thermo Fisher, USA).

190 Soil microbial respiration was determined according to Falsone et al. (2015). Soil samples were
191 adjusted to 60% of water holding capacity and incubated for 28 days at 25 °C. The CO₂ emitted from
192 incubated soils was measured through alkali (0.5 M NaOH solution) absorption of the produced CO₂
193 from each sample. Then, the titration of the rest of NaOH solution was carried out using 0.05 M HCl
194 in presence of 0.75 M BaCl₂. The soil basal respiration (SBR) of each soil sample was computed as
195 the hourly flux of CO₂ per gram of soil, while the cumulative soil basal respiration (RCUM) was
196 expressed as the total amount of CO₂ evolved during the 28 days of incubation.

197 Soil microbial biomass C (C_{mic}) was measured on soil samples at 60% of WHC using chloroform
198 fumigation extraction method with 0.5 M K₂SO₄ solution (Vance et al., 1987). Both fumigated and
199 non-fumigated extracts were analysed using a TOC-V CPN total organic carbon analyser (Shimadzu,
200 Japan). C_{mic} was calculated as EC×2.64, where EC was the difference between organic C extracted
201 from fumigated soils and organic C extracted from non-fumigated soils (Vance et al., 1987). The
202 organic C inside the filtered solution obtained from non-fumigated soil samples were considered as
203 water-extractable organic C (WEOC) (Chantigny et al., 2007).

204

205 *Life Cycle Assessment (LCA) of peach, pear, and kiwi production*

206 The LCA methodology used in the present study aimed to assess the annual impact on global warming
207 potential of fruit production expressed as kg equivalent CO₂ kg fruit⁻¹ yr⁻¹ (ISO14040, 2006 and
208 ISO14044, 2006). The following assumptions were made for this LCA:

209 - The system boundary of this study is considered from the extraction of raw materials of inputs up
210 to the farm gate when the fruits are harvested.

211 - Data for LCA were taken for the whole life cycle starting from the period of farm establishment till
212 the time of performing this study. Specifically, the LCA was carried out taking in account orchard
213 establishment, cultivation, harvesting and final disposal stages. The nursery stage was excluded,
214 mainly due to the lack of reliable data regarding this phase. The orchard establishment stage included
215 soil preparation, the construction of the fixed structures (irrigation system and supporting structures)
216 and trees plantation. During this stage, the fuel consumption was 430 kg ha^{-1} for peach and pear
217 orchards, and 1117 kg ha^{-1} for Ki30. The cultivation stage included production of fertilisers and their
218 application to the field, pest and weed management substances manufacture and their application,
219 irrigation, pruning, energy use for irrigation and fuel consumption, and machinery use. The mean
220 yearly consumption of electricity, fuel and agrochemicals for the considered orchards are reported in
221 Table 2. The electricity was used for irrigation purposes. In particular, the average water use was
222 $2400, 3240, 2300$ and $4130 \text{ m}^3 \text{ ha}^{-1}$ for Ph10, Ph15, Pr20 and Ki30, respectively. The plants were
223 watered through drip irrigation system. The disposal stage considered the disposing of wastes
224 collected during orchard establishment and cultivation stages to thermal–power plants or to landfills.
225 During the period going from orchards establishment until 2017, the waste production was on average
226 $5.3, 12.5, 15.8$ and $25.1 \text{ kg ha}^{-1} \text{ year}^{-1}$ for Ph10, Ph15, Pr20 and Ki30, respectively.

227 - The LCA took into account the production of the materials (e.g., concrete poles, iron wires and
228 irrigation tubes) used for the construction of the fixed structures in the orchards.

229 - For fertilizers and agrochemicals production, LCA includes the transport of primary and secondary
230 materials to the production plants, the synthesis of the chemical components and the waste treatment
231 or disposal.

232 - The LCA included emissions to air of nitrous oxide (N_2O) coming from soil after fertilizations were
233 calculated according to Stehfest and Bouwman (2006).

234 - For machinery, the performed LCA did include the manufacture, transport, maintenance, repair, and
235 waste management of the machinery used for field operations.

236 - LCA did not include the transport of raw materials (pesticides, fertilisers, plantlets, poles, etc.) from
237 the local storehouse to farms as well as the production of the packaging used for such raw materials.

238 - LCA did not include the human labour.

239 The data used for the life cycle inventory (e.g, fuel consumption, used fertilizers and irrigation) were
240 retrieved from the farmers.

241

242 *Calculations and statistical analyses*

243 For the investigated study sites, various calculations were performed, encompassing: soil C stock,
244 expressed as Mg ha^{-1} ; the yearly soil C stock gain or loss rate (C_{soil}) in 0–30 cm depth since the
245 conversion of CK up today, expressed as $\text{Mg ha}^{-1} \text{ yr}^{-1}$; C balance (C_{bal}), expressed as $\text{Mg ha}^{-1} \text{ yr}^{-1}$,
246 which is the yearly loss or gain of C of the fruit orchards (with exclusion of plant biomass); the
247 metabolic quotient ($q\text{CO}_2$), expressed as $\text{mg C-CO}_2 \text{ h}^{-1} \text{ mg C}_{\text{mic}}^{-1}$, which is an indicator of stress in
248 soils (Anderson and Domsch, 1993) and describes the efficiency of the microbial biomass in C use
249 (Pinzari et al., 2017); the microbial quotient ($q\text{MIC}$), expressed as $\text{mg C}_{\text{mic}} \text{ g SOC}^{-1}$, which represents
250 the microbial ability to assimilate soil C (Sun et al., 2020); and the Dilly index which relates soil
251 quality to microbial biomass and respiration (Dilly, 2005) as follows:

$$252 \quad C_{\text{stock}} = \text{SOC} \times th \times BD \times (1 - \%gravel) \times 0.1 \quad (1)$$

253 where th is the considered soil thickness and $\%gravel$ is the gravel amount in the considered soil
254 thickness;

255

$$256 \quad C_{\text{soil}} = \frac{C_{\text{stock in orchard}} - C_{\text{stock in CK}}}{\text{orchard age}} \quad (2)$$

257

$$258 \quad C_{\text{bal}} = \frac{C_{\text{stock in orchard}} - C_{\text{stock in CK}}}{\text{orchard age}} - \text{orchard mean annual age} \times \text{CLCA} \quad (3)$$

259 where C_{bal} is the carbon balance, CK is the reference field and CLCA is the C–CO₂eq
260 Within the C balance, the C of plant biomass was not considered because it was burned at the end of
261 plants' life.

$$262 \quad qCO_2 = \frac{100 \times SBR}{C_{mic}} \quad (4)$$

263

$$264 \quad qMIC = \frac{C_{mic}}{SOC} \quad (5)$$

265

$$266 \quad Dilly \ index = \frac{qCO_2 \times 1000}{SOC} \quad (6)$$

267

268 Two-way analysis of variance was performed to assess the effect of both orchard crop type and soil
269 depth on the selected soil physical, chemical and biochemical parameters. Because of the absence of
270 orchard crop type \times soil depth interaction ($P > 0.05$), the effects of both main factors were evaluated
271 through one-way analysis of variance. Prior analysis of variance, the normality and homoscedasticity
272 of residuals were evaluated through graphical analysis and the data were transformed if necessary.
273 To identify statistically significant differences among the means the Tukey's honest significant
274 difference test was conducted as multi-comparison test ($P < 0.05$). The results presented are based
275 on mean values and their standard error. The data were analysed using R software 4.0.3.

276 Concerning to LCA, SimaPro 8.5.0 software was used to analyze the life cycle inventory data.
277 SimaPro 8.5.0 is an LCA tool that can be used to monitor the performance of the sustainability of a
278 product or service. This software can analyse a complex life cycle systematically and can evaluate
279 the environmental impact of a product or service at each stage of the life cycle. Ecoinvent 3.4 was
280 chosen as background data sources (Weidema et al., 2013).

281

282 **Results**

283 *Soil physical, chemical and biochemical properties*

284 The SOC concentration and stocks in the 0 – 30 cm depth ranged from 8.02 in CK to 15.36 g kg⁻¹ in
285 Ph15 and from 31.6 in CK to 64.4 Mg ha⁻¹ in Ph15 (Figure 2a, b). The TN concentrations varied from
286 0.96 in CK to 2.03 g kg⁻¹ in Pr20 (Figure 2c).

287 Comparing the surface layer of the selected orchard crop types, CK had the lowest value of SOC and
288 TN concentration and C stock, while Pr20 had the highest ones. In subsurface soil layer, instead, only
289 the peach orchards showed higher SOC and TN concentrations than CK (Figure 2a, c), and no
290 differences in C stock occurred among orchard crop types (Figure 2b).

291 Between soil layers (0-15 and 15-30 cm), CK soils did not show differences in SOC and TN
292 concentrations, and C stock. Some differences instead occurred in orchards: Ph10, Pr20 and Ki30
293 showed higher SOC and TN concentrations in surface than in subsurface layer (Figure 2a, c); Ph15,
294 Pr20 and Ki30 showed higher C stock in surface soil layer than in subsurface one (Figure 2b).

295 The water-extractable organic C varied from 112 to 294 mg kg⁻¹, and no differences were found,
296 neither between soil depth nor among orchard crop types (Figure 2d).

297 The $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values ranged from -25.20 to -27.29 and from 2.06 to 9.59 ‰ (Figure 3a and b),
298 respectively. Soils under Pr20 showed less negative value of both $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of organic matter
299 compared to CK (Figure 3) and this was more pronounced for N where $\delta^{15}\text{N}$ in surface soil layer was
300 the highest value (Figure 3b).

301 The SOC pools obtained through chemical fractionation showed the major differences only for the
302 more chemically stable ones (i.e., humic-like C and non-extractable organic C; Figure 4).
303 Specifically, no humic-like C was found in subsurface soil layers of CK and Pr20, moreover only in
304 Ki30 the surface layer showed higher content of humic-like C compared to subsurface one (Figure
305 4c). In the surface layer, the C content associated to NEOM (NEOC) assumed the lowest value in CK
306 (7.35 g kg⁻¹) and it was lower in Ph10 compared to Ph15 and Pr20 (Figure 4d). Furthermore, NEOC
307 concentration decreased with soil depth in Pr20 and Ki30.

308 Both soil microbial respiration and C_{mic} content did not differ among the selected fields in surface
309 soil, while some differences occurred for the subsurface soil (Figure 5a, c). SBR showed higher values

310 in Ph10 than in Pr20 (Figure 5a) and C_{mic} content showed the lowest value in Pr20 and a higher value
311 in Ph10 than in CK (Figure 5c). Taking in consideration the soil depth, soil microbial respiration and
312 C_{mic} generally were higher in surface compared to subsurface soil of Ph15, Pr20 and Ki30.
313 Like microbial respiration and C_{mic} content, no differences of qCO_2 and $qMIC$ occurred among the
314 selected fields in surface soil (Table 3). For the subsurface soil, instead, the Pr20 showed the highest
315 qCO_2 and the lowest $qMIC$. Moreover, some differences occurred between the two soil depths in
316 Pr20 and Ki30. Specifically, while qCO_2 increased with depth in Pr20 and decreased in Ki30, the
317 opposite occurred for $qMIC$. The Dilly index showed similar values among the fields in the surface
318 soil ranging from 170 to 570 (Table 3). In the subsurface soil, the Dilly index showed the highest
319 value under Pr20 (2083) and the lowest ones under Ph10 and Ph15 (236 and 331, respectively).
320 Generally, the Dilly index did not change with soil depth with the exception of Pr20 where the
321 subsurface soil had a higher value compared to surface soil.

322

323 *CO₂ loss estimation from orchards through Life Cycle Assessment and carbon balance*

324 When looking to the overall impact of the considered orchards on CO_{2eq} emission, kiwi production
325 presented the greatest impact (Table 4). In all orchards, the main source of CO_{2eq} is attributed to
326 fertilizers. Specifically, in the investigated orchards the contribution of fertilizers' manufacturing
327 ranged from 21.97 to 33.91% of the total CO_{2eq} emissions while the GHGs emission developed after
328 the fertilizers' distribution ranged between 16.47 and 18.12% of the total CO_{2eq} emissions.
329 Comparing the considered orchards, Ki30 showed the highest CO_{2eq} emission from fertilizers use.
330 The lowest CO_{2eq} emissions related to fertilizers production were observed in Pr20 (0.042 kg CO_{2eq}
331 kg^{-1} fruit), while the lowest CO_{2eq} emissions related to fertilizers emissions were observed in Ph10
332 (0.029 kg CO_{2eq} kg^{-1} fruit). The agricultural practices during the cultivation period showed to be the
333 second greatest source of GHG, with the exception of Ph10 where the use of agrochemicals accounted
334 for the 22.4% of total CO_{2eq} emissions followed by agricultural practices with 21.4% (Table 4).
335 Unlike fertilizers use, the agricultural practices showed the highest CO_{2eq} emission value in Pr20. It

336 is interesting to observe the high relevance of orchard establishment on CO₂eq emission ranging from
337 5.8% of Ph10 to 21.7% for Ph15. Because of the scarcity of precipitations during the summer period,
338 irrigation too showed a significant impact on CO₂eq emission, with the highest value in Ki30 (0.045
339 kg CO₂eq kg⁻¹ fruit) and the lowest one in Ph10 (0.0081 kg CO₂eq kg⁻¹ fruit).

340 In the selected orchards, soils showed a yearly increase of organic C stock (C_{soil}) in the 0 – 30 cm
341 depth (Table 5). The highest soil organic C accumulation rate was observed in Ph10 (2294 kg C ha⁻¹
342 year⁻¹), while the lowest one was found in Ki30 (646 kg C ha⁻¹ year⁻¹).

343 The conversion of a field for grains production to peach and pear orchards had a positive effect on C
344 immobilization (Table 5). Conversely, kiwi cultivation seemed to be an agroecosystem that promotes
345 C release to the atmosphere. Specifically, the highest C storage rates (C_{bal}) were observed in peach
346 orchards (1515 and 1580 kg C ha⁻¹ year⁻¹ in Ph10 and Ph15, respectively), while Ki30 showed a C
347 loss of 117 Mg ha⁻¹ year⁻¹.

348

349 **Discussion**

350 *Soil chemical properties*

351 SOC content and C stock of the CK plot (8 g kg⁻¹ and 31 Mg ha⁻¹, respectively) were similar to that
352 found in Cambisols of croplands in the Emilia-Romagna region and in the plain of northern Italy
353 (Vittori Antisari et al., 2021a; Brombin et al., 2020; Dal Ferro et al., 2020; Lugato et al., 2007)
354 suggesting its representativeness as reference soil.

355 The increased SOC concentration and C stock in soils due to the land use change from wheat
356 production to orchard would suggest the capacity of orchards cultivation systems to enrich soil of
357 organic matter. Several studies (e.g., Massaccesi et al., 2018; Neilsen et al., 2014) found an increase
358 in organic carbon amount after orchards establishment. Specifically, a mean C stock of 57 Mg ha⁻¹
359 in 0–30 cm depth was observed which was similar to the values reported by previous studies
360 conducted in Europe (e.g., Álvaro-Fuentes et al., 2012; Bateni et al., 2021; Funes et al., 2019). The
361 increased C stock could be mainly attributed to the presence of a permanent herbaceous plants

362 established on whole surface of the fields which is worldwide recognized to increase soil C stock (de
363 Torres et al., 2021; Xiang et al., 2022; Novara et al., 2019). In fact, the conversion of cropland to
364 grassland promotes SOC storage (Auerswald and Fiener, 2019) due to the higher root turnover in
365 grasslands compared to cropland and due to the harvest of the whole aboveground biomass in
366 cropland (Poeplau and Don, 2013). Since root derived C through rhizodeposition processes and root
367 turnover (De Feudis et al., 2016; Douglas et al., 2020) has been identified as the major source of SOC
368 (Rasse et al., 2005), the presence of trees and perennial grasses may explain higher SOC accumulation
369 in orchards compared to CK. Such differences were marked in surface soil mainly due the generally
370 larger distribution of roots in the surface soil (Forey et al., 2017; Ruiz-Sánchez et al., 2005; Sokalska
371 et al., 2009) and to the degradation of the chopped pruning residues left on soil surface (Massacesi
372 et al., 2018; Zhao et al., 2017). The greater influence of fruit orchards on surface soil compared to
373 subsurface soil can be confirmed by the higher SOC content and C stock in the former in Ph15, Pr20
374 and Ki30. Because of the role of SOC on soil microbial activity (e.g., Martínez-García et al., 2018),
375 the higher amount of organic matter in the surface soil might explain the generally higher soil
376 microbial respiration and biomass in the superficial soil layer.

377 The generally homogeneous $\delta^{13}\text{C}$ values would indicate that orchard cultivation did not affect the
378 organic matter decomposition (Blagodatskaya et al., 2011; Solomon et al., 2002). The unchanged
379 SOC decomposition rate could be confirmed by the negligible differences between CK and the
380 considered orchards of those biochemical indicators (i.e., SBR, RCUM, C_{mic} , $q\text{CO}_2$ and $q\text{MIC}$) related
381 to C cycle. The similar SOC degradation combined with the high organic material input due to the
382 shredded pruning residues might have promoted an accumulation of NEOC in the surface soil of the
383 orchards. The plant residues could release water-insoluble compounds (e.g., lignin and waxes) and
384 labile substances readily available to microorganisms whose cell residues could bind to soil minerals
385 increasing the NEOM fraction (Hayes et al., 2017; Wang et al., 2021).

386 Like SOC content and C stock, the cultivation of fruit orchards increased the TN content in surface
387 soil. This can be attributed to the addition of N by amendment (i.e., in Ph10) and chemical fertilizers.

388 The higher $\delta^{15}\text{N}$ values in orchards compared to the wheat field might be attributed both to the
389 contribution of N-enriched fertilizers to $\delta^{15}\text{N}$ values and to the preferential microbial utilization of
390 ^{14}N compounds (Boström et al., 2007; Lobe et al., 2005). The latter maybe limited under Ki30.
391 It was interesting to note that for the subsurface soil, among the selected orchards, Pr20 showed the
392 lowest humic-like C content which would cause a limited SOC stabilization (Martins Gomes et al.,
393 2018). The limited SOC stabilization might be due to the less suitable conditions for the soil microbial
394 community which did not allow the transformation of the soil organic matter (Liebich et al., 2007).
395 In fact, the subsurface soil of Pr20 also showed the lowest C_{mic} , q_{Mic} and the highest $q\text{CO}_2$ indicating
396 a lower C use efficiency by the microbial community (Anderson, 2003; Anderson and Domsch, 1989;
397 Okolo et al., 2020) compared to other fields and, therefore, the occurrence of poor conditions (Vittori
398 Antisari et al., 2021). Such unfavourable conditions in subsurface soil for Pr20 was confirmed by the
399 very high Dilly index value, which would suggest the worsening of the energy use efficiency by the
400 microbial community, in turn not promoting organic C accumulation (Dilly, 2005).

401

402 *Life Cycle Assessment*

403 In agreement with previous studies (e.g., Romero-Gómez et al., 2017; Vinyes et al., 2017), this study
404 found that fertilization was the procedure that had the greatest impact on CO_2eq emission from the
405 orchards, accounting for at least 40% of total CO_2eq emission. In this context, it was interesting to
406 observe that, although in Ph10 no chemical fertilizers were applied, the use of organic amendment
407 had a great impact on CO_2eq emissions. In fact, organic amendment production is both an energy-
408 intensive process and a source of methane and nitrous oxide while its application causes N_2O emission
409 (Bacenetti et al., 2016; Galgani et al., 2014). However, because of the greatest use of N and P
410 fertilizers, the highest CO_2eq emission related to fertilizers was observed in Ki30. Indeed, N and P
411 fertilizers are considered highly impacting on climate change, fossil fuel depletion, acidification,
412 eutrophication, and resources depletion (Hasler et al., 2015). This result, together with the highest
413 CO_2eq emission related to the irrigation, would indicate the higher demands of nutrients and water of

414 kiwi plants compared to peach and pear trees (Allen et al., 1998; Carranca et al., 2018; Peticila et al.,
415 2015).

416 The consume of fuel related to agricultural practices as tillage, weed control and pruning showed to
417 be the second most important CO₂eq source. In this sense, Milà I Canals et al. (2006) suggested the
418 use of biofuel in order to limit the impact of the agricultural practices on CO₂ emission.

419 Several studies (e.g., Martin-Gorriz et al., 2020; Vinyes et al., 2017) reported the high impact of
420 agrochemicals on CO₂eq released into the atmosphere. However, in this study the contribution of
421 agrochemicals on CO₂eq emission in Ph15, Pr20 and Ki30 resulted low due to the sustainable
422 approach used on the studied farms. In this context, it was important to highlight the greater
423 contribution of agrochemicals on CO₂eq emissions for Ph10. In this case, the amounts of
424 agrochemicals used was 10 times higher than those used in the other orchards, and they were mainly
425 sulphur based. This higher amounts of agrochemicals can be attributed to the types of agrochemicals
426 generally used in organic farming. These findings are in agreement with the work of Longo et al.
427 (2017) which observed a larger use of pesticides to produce organic apples compared to those
428 produced with conventional approaches.

429 Overall, this study clearly showed how kiwifruit cultivation had the highest impact on GHG emissions
430 mainly due to the high water and nutrient demand, suggesting that such tree species is less suitable
431 than peach and pear for the considered study area.

432

433 *Carbon balance*

434 When taking in account the C–CO₂eq loss by fruit cultivation and C gained and stored into the soil,
435 results from this study would indicate that peach and pear orchard ecosystems promote C
436 sequestration. The capability of the studied orchards to sequester C was mainly attributed to the soil
437 on which they grow. In fact, the investigated soil was able to store each year a large amount of organic
438 C. Notably, such C was stored in the most stable form preventing C to go back to the atmosphere as
439 CO₂ in the short– or mid–term. It is important to note that in the present study we did not consider C

440 fixed in plant biomass because it is not a long-living component. In fact, orchards for fruit production
441 generally have a lifetime of few decades. Also, at the end of the cultivation period the plant biomass
442 is removed and burnt on the field or in thermal power plants or processed for pellet production which
443 are common practices for fruit orchards (Brand and Jacinto, 2020; Giuntoli et al., 2016). Conversely,
444 the organic carbon stored as fulvic-like C, humic-like C and NEOC could have a mean residence time
445 which spans from centuries to thousands of years (Certini et al., 2004; Piccolo, 2002).

446 The generally similar soil microbial efficiency to use C and, therefore, to transform C in stable forms,
447 together with similar $\delta^{13}\text{C}$ values and soil characteristics (e.g., clay content) between the orchards and
448 the reference field would indicate that C sequestration was mainly related to the management
449 practices carried-out in each orchard.

450 Taking in consideration each orchard type, it is important to mention the negative C balance (-117 kg
451 $\text{C ha}^{-1} \text{ year}^{-1}$) of Ki30. The negative value can be mainly attributed to the high inputs (fertilizers and
452 irrigation) requested by the kiwi plants which caused large CO_2eq emission just partly
453 counterbalanced by soil carbon storage processes. Indeed, when taking in consideration the soil
454 environment, generally no differences in SOC content and its chemical forms were found among the
455 selected orchards. Unlike Ki30, Pr20 showed similar values of CO_2eq emissions of peach orchards
456 (Table 5 and Table 6) but a lower mean annual C storage increase (Table 6). The weak mean annual
457 C storage increase in Pr20 could be attributed to the more stressful conditions for the microbial
458 biomass in subsurface soils. Overall, the C balance performed in this study by taking in consideration
459 the topsoil highlighted the importance of SOC sequestration into the LCA of agricultural systems.
460 However, because of its pivotal role on C storage (Guillaume et al., 2022; Antony et al., 2022) and
461 its greater influence on the agricultural managements compared to topsoil (Samson et al., 2021;
462 Osanai et al., 2020), future LCA studies should take into consideration the subsoil and its key role in
463 the overall C cycle. .

464

465 **Conclusions**

466 The results from the present study suggest that the conversion of a field from grains production to the
467 fruit orchards cultivation promoted soil carbon gain. The majority of the gained C was found in the
468 most chemically recalcitrant form suggesting that in the selected fruit orchards the C stabilization
469 processes were promoted. The organic C increase in orchards could be mainly attributed to the
470 permanent grasses covering such fields. However, such increase could be also promoted both by the
471 direct release from plant residues of chemically recalcitrant compounds and by the release of readily
472 available C for microorganisms whose necromass could bind to soil mineral particles. However, the
473 C gain rate is not unlimited as it depends on soil properties (e.g., clay content) as well as on orchard
474 management. For example, in Ki30, soil stored C, but it was not able to counterbalance the GHG
475 emissions coming from the cultivation of kiwi though it had similar clay content and similar
476 biochemical properties of the reference field. A key tool in this sense may therefore be LCA as it
477 allows us to take into consideration soil resources and their contribution. The systematic inclusion of
478 soil in LCA would allow to enhance agroecosystems sustainability and give soil resources their
479 rightful place in the quest to tackle sustainable development goals and combat climate change.
480 Therefore, we propose to insert the soil C storage rate as CO₂ soil uptake from atmosphere lowering
481 the environmental impacts of orchards management. Finally, although the present study only
482 considered topsoil (0–30 cm depth), in future LCA procedures that also considered deep soil would
483 provide an important additions to give a more realistic view of the role of soil on the mitigation of
484 the GHG emissions coming from the cultivation practices.

485

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489

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850

1 **The importance of incorporating soil in the life cycle assessment procedure to improve the**
2 **sustainability of agricultural management**

3

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20

21 **Abstract**

22 The formidable ability of soil to store carbon has attracted an increasing number of studies, but few
23 of them included soil organic carbon (SOC) sequestration as part of a carbon balance assessment in
24 the agroecosystem. This raises some interesting questions: 1) how orchards conversion increase soil
25 capacity to mitigate the green-house gases (GHG) emissions by storing C? 2) can it be considered in
26 life cycle assessment (LCA)? 3) can SOC pools and soil biochemical properties determination
27 improve LCA interpretation? To answer these questions, this study selected a ten- and fifteen-years-

28 old peach orchards, a twenty–years–old pear orchard, a thirty–years–old kiwi orchard in south-east
29 part of Emilia–Romagna Region (Italy), and a cereals’ field as reference. Soil samples were collected
30 from 0–15 and 15–30cm depths, and the SOC pool amounts (i.e., labile and recalcitrant) determined.
31 LCA was used to estimate the GHG emissions (CO₂eq) from the orchards. Results showed that the
32 conversion from cereals to orchard production increased OC stock (+82% on average) suggesting that
33 orchards cultivation systems have the capacity to enrich soil organic matter. Fertilization had the
34 greatest impact on CO₂eq emission accounting for at least 40% of total CO₂eq emissions. Kiwi
35 cultivation had the highest impact on GHG emissions mainly due to the high water and nutrient
36 demand (0.045 and 0.149 kg CO₂eq kg⁻¹ fruit yr⁻¹, respectively). When taking into account the C–
37 CO₂eq loss by fruit cultivation and C storage in soils, results would indicate that peach and pear
38 orchard agroecosystems promote C sequestration. Conversely, kiwi cultivation showed large CO₂eq
39 emissions only partly counterbalanced by SOC sequestration. This study highlights the importance
40 of including soils in LCA: if made mandatory this would allow a wider, yet more detailed, picture of
41 the impact of agricultural practices on C budget. This simple step could help optimise resource
42 management and at the same time improve agroecosystem sustainability.

43

44 **Keywords**

45 Cambisols, fruit orchards, soil organic C, CO₂eq, LCA

46

47 **Introduction**

48 In 2020, carbon dioxide (CO₂) concentration in the atmosphere reached values greater than 410 ppm
49 due to the human activities (The World Meteorological Organization, 2020). Agriculture is
50 recognised as a significant contributor to anthropogenic emissions of CO₂ (Smith et al., 2014; Lynch
51 et al., 2021). Recent studies (Gkissakis et al., 2020; Goossens et al., 2017; Mousavi-Avval et al., 2017;
52 Pryor et al., 2017) pointed out that the GHG emissions from agricultural crop production systems are
53 mainly related to the fossil-fuel consuming and to the manufacturing and distribution of chemical

54 fertilizers. Noteworthy is also the production of nitrous oxide (N₂O) gas due to soil nitrogen input
55 (Lawrence et al., 2021). Consequently, a reduced utilization of both fuels and fertilizers could
56 improve the sustainability of agricultural management. For example, Aguilera et al. (2015) compared
57 the environmental impact of several conventional and organic cropping systems in Spain,
58 highlighting greater GHG emissions in the formers compared to the latter, mainly due to the use of
59 chemical fertilizers in the conventional system. Similarly, Pergola et al. (2017) found a greater impact
60 on climate change of apricot orchards under integrated system compared to those under biodynamic
61 one. In the context of the current climate change, soil plays a central role in the mitigation of GHGs
62 emission from agriculture through soil carbon sequestration, defined by Chenu et al. (2019) as “*the*
63 *process of transferring CO₂ from the atmosphere into the soil of a land unit, through plants, plant*
64 *residues and other organic solids which are stored or retained in the unit as part of the soil organic*
65 *matter*”. In this sense, worldwide there is a strong agreement to implement the carbon–farming
66 initiatives with the main aim to increase the soil organic carbon (SOC) stock which is a way to
67 mitigate the current climate change (Wiesmeier et al., 2020; Bradford et al., 2019; Chenu et al., 2019).
68 However, to reach this goal, the chemical, physical and edaphic conditions of the soil must allow the
69 humification process and the accumulation of organic C to be carried out rather than the
70 mineralization process. Soil stores three times the amount of C present in the atmosphere (Ciais et
71 al., 2013) and could potentially remove from the atmosphere between 0.79 and 1.54 Gt yr⁻¹ of C (Fuss
72 et al., 2018) if land uses and management practices increased C inputs and/or reduced C losses. In
73 this sense, promoting soil organic C (SOC) sequestration is one of the most important strategies to
74 reduce atmospheric CO₂ concentrations with a significant potential to mitigate climate change (Lal,
75 2018). Bulk SOC is composed of multiple functional pools differing in turnover, in fact it ranges from
76 the most labile form (i.e., the dissolved organic C) to the most stable one as the physically protected
77 and the chemically recalcitrance forms (De Feudis et al., 2019; Poeplau et al., 2018). Tthe long
78 residence time associated with most of the SOC pools (e.g., De Feudis et al., 2019; Ferreira et al.,
79 2020) makes soil a major player in the global carbon budget (Martin et al., 2014). Moreover, soils

80 characterized by high SOC concentrations are recognized to be desirable because SOC improves soil
81 nutrient availability, cation exchange capacity, water retention capacity, soil aeration, soil aggregation
82 and structure, soil microbial biomass and its activity, plant yield and quality (Bationo et al., 2007;
83 Bronick and Lal, 2005; Chavarria et al., 2018; Martínez-Mena et al., 2021).

84 There is general agreement that management practices are important factors influencing SOC
85 contents in agricultural lands (Montanaro et al., 2017; Novara et al., 2019; Pardon et al., 2017). For
86 example, the cultivation of cover crops has been identified as an effective practice to increase of SOC
87 content (Poeplau and Don, 2015). Similarly, practices addressing the incorporation of the plant
88 residues into the soil could prevent SOC reduction (Keel et al., 2019). The no-tillage has been claimed
89 to be a potential option to decrease SOC loss in agricultural soils (Nath and Lal, 2017), but at global
90 scale its effect on SOC content seems to be limited (Mondal and Chakraborty, 2022). Moreover, it is
91 well known the increase of SOC content when organic fertilizers are applied (Morugán-Coronado et
92 al., 2020).

93 In this context, life cycle assessment (LCA) is a well established approach to help accounting for all
94 the various stages of any activity, including agricultural practices where it was introduced since 1990
95 (Haas et al., 2000). LCA is one of the most used standardized methodologies for estimating the
96 environmental impacts linked to the entire cycle of fruit production (Vinyes et al., 2015). Among the
97 environmental impacts, the evaluation of GHG emissions prevail compared to the other
98 environmental problems (Adewale et al., 2019; Bartzas et al., 2017; Rebolledo-Leiva et al., 2017).

99 Most of the studies concerning LCA in agroecosystems take in account yield, plant growth and all
100 the input factors related to the crop cultivation such as human labour, machinery, fertilizer
101 application, fossil fuel consuming and irrigations (e.g., Foteinis and Chatzisyneon, 2016; Kaab et al.,
102 2019). Conversely, despite its high potential to store carbon, soil is generally not included in LCA
103 approach for the evaluation of C budget (Garrigues et al., 2012). Only in few cases SOC was taken
104 into account for the LCA (Arzoumanidis et al., 2014; Brandão et al., 2013; Petersen et al., 2013).

105 Hence, although SOC is essential if LCA is to be applied in case studies where carbon balances must

106 be calculated, the limited number of LCA studies that took into consideration SOC would highlight
107 how soil is generally the forgotten part of the agro-ecosystems. In addition, although the estimation
108 of the bulk SOC stock could be sufficient for C balance in LCA approach, the knowledge of SOC
109 pools and their dynamics are necessary for improving the interpretation of LCA outputs. Specifically,
110 since the important role of LCA to improve the management of agricultural systems for preventing
111 environmental hazards (e.g., the GHGs emissions) in the long-term, the agricultural managements
112 and/or systems able to promote the storage of the most stable SOC forms should be promoted.
113 Therefore, for a reliable C balance through the LCA procedure, it is important that soil C is stored in
114 the most stable forms. Further, because of the key role of soil microbial community to transform and
115 stabilize SOC (Angst et al., 2021; Domeignoz-Horta et al., 2021), the evaluation of their properties
116 (e.g., amount and activity) could be of interest in LCA to understand whether (or not) soil stabilize
117 C.

118 This study tries to address this gap in the literature and provide a justification for a more widely
119 accepted introduction of soil in agroecosystems LCA. In particular, the study will focus on *i*) how
120 orchards conversion increase soil capacity to mitigate the green-house gases (GHG) emissions by
121 storing C; *ii*) how soil C stock can therefore be included in LCA approach; and *iii*) if SOC pools and
122 soil biochemical properties determination can improve LCA interpretation. In order to address these
123 aims, the following hypotheses were set: 1) orchards increase soil C stock compared to grain fields;
124 2) and soil C storage capacity can mitigate the GHG emissions related to the fruit orchard agricultural
125 practices.

126

127 **Materials and Methods**

128 *Study sites description*

129 The present study was conducted in the south-east part of Emilia Romagna Region, Italy. This area
130 had a mean cumulative annual precipitation of 763 mm and a mean annual air temperature of 14.2 °C
131 for the period 1986 – 2015. The study was conducted in 2017, and the specific study site selected

132 included a ten- and fifteen-years-old peach orchards (Ph10 and Ph15, respectively) with a tree
133 density of 1,300 plants ha⁻¹; a twenty-years-old pear orchard (Pr20) with a tree density of 820 plants
134 ha⁻¹, and a thirty-years-old kiwi orchard (Ki30) with a tree density of 710 plants ha⁻¹ (see Fig. 1).
135 Some more details of study sites are reported in Vittori Antisari et al. (2021). The choice of the
136 selected tree species was based on their wide distribution in Italy. The mean yields of the selected
137 orchards were 48, 35, 30 and 28 × 10³ kg (fresh weight) ha⁻¹ for Ph10, Ph15, Pr20, Ki30, respectively.
138 According to the farmers, such yields were reached within the fifth year after the orchard
139 establishment. However, because of the missing data about yield during first years of orchard
140 cultivation, in the present study we arbitrarily considered the aforementioned yields also for the first
141 years of cultivation after orchard establishment. All the soils were classified as Cambisols with a
142 texture from silty clay loam to loam, a slight alkaline reaction (pH = 7.7 on average) and bulk density
143 ranging from 1.14 to 1.59 g cm⁻³, with lower values in 0–15 cm compared to 15–30 cm soil layer. On
144 average, cation exchange capacity of the soils studied was 24.9 cmol(+) kg⁻¹, the exchangeable Ca²⁺,
145 Mg²⁺ and K⁺ concentrations were 15.7, 1.9 and 0.60 cmol(+) kg⁻¹, respectively, and the base
146 saturation was of 75.4%. Details about clay content and equivalent soil mass of the study sites are
147 reported in Table S1 of the Supplementary materials.

148 In the orchards, soil was kept covered by natural grasses which were periodically cut (4–5 times per
149 year). Pruned wood materials were shredded and left on the soil surface. According to the farmers,
150 the average amount of pruned materials for Ph10, Ph15, Pr20 and Ki30 were 3.0, 3.0, 2.5 and 3.5 Mg
151 dry matter ha⁻¹. Some differences occurred for fertilization treatments (Table 1). In Ph10, no chemical
152 fertilization was performed, but exhausted substrate for mushroom cultivation at a rate of 7 Mg ha⁻¹
153 was spread on soil surface every year. In Ph15, Pr20 and Ki30, fertilization was carried out both by
154 fertigation, through drip irrigation lines (one line per plant row), and foliar spray. The amounts of
155 elements applied by fertilization is reported in Table 1.

156 To estimate C accumulation/loss of fruit orchard soils, a field for grain production (wheat) was used
157 as reference (CK). The rationale to use a field for grains production as reference soil was based both

158 on the widespread cultivation of such crops in the northern Italy and because the considered fruit
159 orchards were formerly wheat fields for at least 5 years.

160

161 *Soil sampling and analyses*

162 Within each field, three 30 cm depth soil pits were dug, and soil samples were collected from 0–15
163 cm (hereafter, surface soil) and 15–30 cm depths (hereafter, subsurface soil). This study used the
164 convention to investigate the 0–30 cm soil depth interval because such interval is worldwide used for
165 the SOC stock evaluation (Makipaa et al., 2012; Guevara et al., 2020; Tangen and Bansal, 2020). The
166 surface and subsurface soil samples were air-dried, passed through a 2-mm sieve and then an aliquot
167 was finely ground for SOC and total nitrogen (TN) concentrations determination.

168 SOC and TN were determined by a CHN elemental analyser (EA 1110 Thermo Fisher, USA) after
169 addition of hydrochloric acid to remove carbonates. The relative abundance of C and N stable isotopes
170 were determined by continuous flow- isotope ratio mass spectrometry (CF-IRMS) using an isotopic
171 mass spectrometer Delta V advantage (Thermo- Finnigam, DE). Measurements were expressed in
172 standard δ ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) notation (‰) relative to Vienna Pee Dee Belemnite and air, respectively.

173 Different SOM fractions, like particulate organic matter (POM), fulvic-like and humic-like
174 substances, and non-extractable organic matter (NEOM), were chemically extracted (Agnelli et al.,
175 2014). A volume of 100 mL of distilled water were added to 10 g of soil and shaken on a horizontal
176 shaker for 16 h at 25 °C, centrifuged and the supernatant was separated from the precipitate. The
177 supernatant was passed through a 53 μm sieve and the particles $>53 \mu\text{m}$ represented the POM. The
178 precipitate remaining into the centrifugation tubes was re-suspended in 100 mL 0.1 M NaOH + 0.1
179 M $\text{Na}_4\text{P}_2\text{O}_7$ solution and the samples were shaken for 24 h at 25 °C and then again centrifuged. The
180 NaOH extract was passed through a 0.45 μm polycarbonate filter, while the remaining precipitate,
181 containing NEOM was washed using deionized water to remove the excess of Na until the pH of the
182 rinsed solution was ≤ 7 . The 0.45 μm filtered NaOH extract was acidified to about pH 1.5 with 6 M
183 HCl and allowed to settle overnight to separate fulvic-like and humic-like substances and

184 centrifuged. To remove the excess of Na from the obtained fractions, the supernatant (fulvic-like
185 substances) was dialyzed through 1000 Da cut-off membranes (Spectra/Por® Dialysis membrane)
186 against distilled water, while the residual (humic-like substances) was washed with 0.002 M HCl.
187 Both purified fractions were freeze-dried. The POM and NEOM fractions were dried at 40 °C. The
188 organic C (OC) and N contents of POM, fulvic-like, humic-like substances and NEOM were
189 determined by a CHN elemental analyser (EA 1110 Thermo Fisher, USA).

190 Soil microbial respiration was determined according to Falsone et al. (2015). Soil samples were
191 adjusted to 60% of water holding capacity and incubated for 28 days at 25 °C. The CO₂ emitted from
192 incubated soils was measured through alkali (0.5 M NaOH solution) absorption of the produced CO₂
193 from each sample. Then, the titration of the rest of NaOH solution was carried out using 0.05 M HCl
194 in presence of 0.75 M BaCl₂. The soil basal respiration (SBR) of each soil sample was computed as
195 the hourly flux of CO₂ per gram of soil, while the cumulative soil basal respiration (RCUM) was
196 expressed as the total amount of CO₂ evolved during the 28 days of incubation.

197 Soil microbial biomass C (C_{mic}) was measured on soil samples at 60% of WHC using chloroform
198 fumigation extraction method with 0.5 M K₂SO₄ solution (Vance et al., 1987). Both fumigated and
199 non-fumigated extracts were analysed using a TOC-V CPN total organic carbon analyser (Shimadzu,
200 Japan). C_{mic} was calculated as $EC \times 2.64$, where EC was the difference between organic C extracted
201 from fumigated soils and organic C extracted from non-fumigated soils (Vance et al., 1987). The
202 organic C inside the filtered solution obtained from non-fumigated soil samples were considered as
203 water-extractable organic C (WEOC) (Chantigny et al., 2007).

204

205 *Life Cycle Assessment (LCA) of peach, pear, and kiwi production*

206 The LCA methodology used in the present study aimed to assess the annual impact on global warming
207 potential of fruit production expressed as kg equivalent CO₂ kg fruit⁻¹ yr⁻¹ (ISO14040, 2006 and
208 ISO14044, 2006). The following assumptions were made for this LCA:

209 - The system boundary of this study is considered from the extraction of raw materials of inputs up
210 to the farm gate when the fruits are harvested.

211 - Data for LCA were taken for the whole life cycle starting from the period of farm establishment till
212 the time of performing this study. Specifically, the LCA was carried out taking in account orchard
213 establishment, cultivation, harvesting and final disposal stages. The nursery stage was excluded,
214 mainly due to the lack of reliable data regarding this phase. The orchard establishment stage included
215 soil preparation, the construction of the fixed structures (irrigation system and supporting structures)
216 and trees plantation. During this stage, the fuel consumption was 430 kg ha⁻¹ for peach and pear
217 orchards, and 1117 kg ha⁻¹ for Ki30. The cultivation stage included production of fertilisers and their
218 application to the field, pest and weed management substances manufacture and their application,
219 irrigation, pruning, energy use for irrigation and fuel consumption, and machinery use. The mean
220 yearly consumption of electricity, fuel and agrochemicals for the considered orchards are reported in
221 Table 2. The electricity was used for irrigation purposes. In particular, the average water use was
222 2400, 3240, 2300 and 4130 m³ ha⁻¹ for Ph10, Ph15, Pr20 and Ki30, respectively. The plants were
223 watered through drip irrigation system. The disposal stage considered the disposing of wastes
224 collected during orchard establishment and cultivation stages to thermal–power plants or to landfills.
225 During the period going from orchards establishment until 2017, the waste production was on average
226 5.3, 12.5, 15.8 and 25.1 kg ha⁻¹ year⁻¹ for Ph10, Ph15, Pr20 and Ki30, respectively.

227 - The LCA took into account the production of the materials (e.g., concrete poles, iron wires and
228 irrigation tubes) used for the construction of the fixed structures in the orchards.

229 - For fertilizers and agrochemicals production, LCA includes the transport of primary and secondary
230 materials to the production plants, the synthesis of the chemical components and the waste treatment
231 or disposal.

232 - The LCA included emissions to air of nitrous oxide (N₂O) coming from soil after fertilizations were
233 calculated according to Stehfest and Bouwman (2006).

234 - For machinery, the performed LCA did include the manufacture, transport, maintenance, repair, and
235 waste management of the machinery used for field operations.

236 - LCA did not include the transport of raw materials (pesticides, fertilisers, plantlets, poles, etc.) from
237 the local storehouse to farms as well as the production of the packaging used for such raw materials.

238 - LCA did not include the human labour.

239 The data used for the life cycle inventory (e.g, fuel consumption, used fertilizers and irrigation) were
240 retrieved from the farmers.

241

242 *Calculations and statistical analyses*

243 For the investigated study sites, various calculations were performed, encompassing: soil C stock,
244 expressed as Mg ha⁻¹; the yearly soil C stock gain or loss rate (C_{soil}) in 0–30 cm depth since the
245 conversion of CK up today, expressed as Mg ha⁻¹ yr⁻¹; C balance (C_{bal}), expressed as Mg ha⁻¹ yr⁻¹,
246 which is the yearly loss or gain of C of the fruit orchards (with exclusion of plant biomass); the
247 metabolic quotient (qCO₂), expressed as mg C-CO₂ h⁻¹ mg C_{mic}⁻¹, which is an indicator of stress in
248 soils (Anderson and Domsch, 1993) and describes the efficiency of the microbial biomass in C use
249 (Pinzari et al., 2017); the microbial quotient (qMIC), expressed as mg C_{mic} g SOC⁻¹, which represents
250 the microbial ability to assimilate soil C (Sun et al., 2020); and the Dilly index which relates soil
251 quality to microbial biomass and respiration (Dilly, 2005) as follows:

$$252 \quad C_{stock} = SOC \times th \times BD \times (1 - \%gravel) \times 0.1 \quad (1)$$

253 where th is the considered soil thickness and %gravel is the gravel amount in the considered soil
254 thickness;

255

$$256 \quad C_{soil} = \frac{C_{stock\ in\ orchard} - C_{stock\ in\ CK}}{orchard\ age} \quad (2)$$

257

$$258 \quad C_{bal} = \frac{C_{stock\ in\ orchard} - C_{stock\ in\ CK}}{orchard\ age} - orchard\ mean\ annual\ age \times CLCA \quad (3)$$

259 where C_{bal} is the carbon balance, CK is the reference field and CLCA is the C–CO₂eq
260 Within the C balance, the C of plant biomass was not considered because it was burned at the end of
261 plants' life.

$$262 \quad qCO_2 = \frac{100 \times SBR}{C_{mic}} \quad (4)$$

263

$$264 \quad qMIC = \frac{C_{mic}}{SOC} \quad (5)$$

265

$$266 \quad \text{Dilly index} = \frac{qCO_2 \times 1000}{SOC} \quad (6)$$

267

268 Two-way analysis of variance was performed to assess the effect of both orchard crop type and soil
269 depth on the selected soil physical, chemical and biochemical parameters. Because of the absence of
270 orchard crop type × soil depth interaction ($P > 0.05$), the effects of both main factors were evaluated
271 through one-way analysis of variance. Prior analysis of variance, the normality and homoscedasticity
272 of residuals were evaluated through graphical analysis and the data were transformed if necessary.
273 To identify statistically significant differences among the means the Tukey's honest significant
274 difference test was conducted as multi-comparison test ($P < 0.05$). The results presented are based
275 on mean values and their standard error. The data were analysed using R software 4.0.3.

276 Concerning to LCA, SimaPro 8.5.0 software was used to analyze the life cycle inventory data.
277 SimaPro 8.5.0 is an LCA tool that can be used to monitor the performance of the sustainability of a
278 product or service. This software can analyse a complex life cycle systematically and can evaluate
279 the environmental impact of a product or service at each stage of the life cycle. Ecoinvent 3.4 was
280 chosen as background data sources (Weidema et al., 2013).

281

282 **Results**

283 *Soil physical, chemical and biochemical properties*

284 The SOC concentration and stocks in the 0 – 30 cm depth ranged from 8.02 in CK to 15.36 g kg⁻¹ in
285 Ph15 and from 31.6 in CK to 64.4 Mg ha⁻¹ in Ph15 (Figure 2a, b). The TN concentrations varied from
286 0.96 in CK to 2.03 g kg⁻¹ in Pr20 (Figure 2c).

287 Comparing the surface layer of the selected orchard crop types, CK had the lowest value of SOC and
288 TN concentration and C stock, while Pr20 had the highest ones. In subsurface soil layer, instead, only
289 the peach orchards showed higher SOC and TN concentrations than CK (Figure 2a, c), and no
290 differences in C stock occurred among orchard crop types (Figure 2b).

291 Between soil layers (0-15 and 15-30 cm), CK soils did not show differences in SOC and TN
292 concentrations, and C stock. Some differences instead occurred in orchards: Ph10, Pr20 and Ki30
293 showed higher SOC and TN concentrations in surface than in subsurface layer (Figure 2a, c); Ph15,
294 Pr20 and Ki30 showed higher C stock in surface soil layer than in subsurface one (Figure 2b).

295 The water-extractable organic C varied from 112 to 294 mg kg⁻¹, and no differences were found,
296 neither between soil depth nor among orchard crop types (Figure 2d).

297 The $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values ranged from -25.20 to -27.29 and from 2.06 to 9.59 ‰ (Figure 3a and b),
298 respectively. Soils under Pr20 showed less negative value of both $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of organic matter
299 compared to CK (Figure 3) and this was more pronounced for N where $\delta^{15}\text{N}$ in surface soil layer was
300 the highest value (Figure 3b).

301 The SOC pools obtained through chemical fractionation showed the major differences only for the
302 more chemically stable ones (i.e., humic-like C and non-extractable organic C; Figure 4).
303 Specifically, no humic-like C was found in subsurface soil layers of CK and Pr20, moreover only in
304 Ki30 the surface layer showed higher content of humic-like C compared to subsurface one (Figure
305 4c). In the surface layer, the C content associated to NEOM (NEOC) assumed the lowest value in CK
306 (7.35 g kg⁻¹) and it was lower in Ph10 compared to Ph15 and Pr20 (Figure 4d). Furthermore, NEOC
307 concentration decreased with soil depth in Pr20 and Ki30.

308 Both soil microbial respiration and C_{mic} content did not differ among the selected fields in surface
309 soil, while some differences occurred for the subsurface soil (Figure 5a, c). SBR showed higher values

310 in Ph10 than in Pr20 (Figure 5a) and C_{mic} content showed the lowest value in Pr20 and a higher value
311 in Ph10 than in CK (Figure 5c). Taking in consideration the soil depth, soil microbial respiration and
312 C_{mic} generally were higher in surface compared to subsurface soil of Ph15, Pr20 and Ki30.
313 Like microbial respiration and C_{mic} content, no differences of qCO_2 and $qMIC$ occurred among the
314 selected fields in surface soil (Table 3). For the subsurface soil, instead, the Pr20 showed the highest
315 qCO_2 and the lowest $qMIC$. Moreover, some differences occurred between the two soil depths in
316 Pr20 and Ki30. Specifically, while qCO_2 increased with depth in Pr20 and decreased in Ki30, the
317 opposite occurred for $qMIC$. The Dilly index showed similar values among the fields in the surface
318 soil ranging from 170 to 570 (Table 3). In the subsurface soil, the Dilly index showed the highest
319 value under Pr20 (2083) and the lowest ones under Ph10 and Ph15 (236 and 331, respectively).
320 Generally, the Dilly index did not change with soil depth with the exception of Pr20 where the
321 subsurface soil had a higher value compared to surface soil.

322

323 *CO₂ loss estimation from orchards through Life Cycle Assessment and carbon balance*

324 When looking to the overall impact of the considered orchards on CO_{2eq} emission, kiwi production
325 presented the greatest impact (Table 4). In all orchards, the main source of CO_{2eq} is attributed to
326 fertilizers. Specifically, in the investigated orchards the contribution of fertilizers' manufacturing
327 ranged from 21.97 to 33.91% of the total CO_{2eq} emissions while the GHGs emission developed after
328 the fertilizers' distribution ranged between 16.47 and 18.12% of the total CO_{2eq} emissions.
329 Comparing the considered orchards, Ki30 showed the highest CO_{2eq} emission from fertilizers use.
330 The lowest CO_{2eq} emissions related to fertilizers production were observed in Pr20 (0.042 kg CO_{2eq}
331 kg^{-1} fruit), while the lowest CO_{2eq} emissions related to fertilizers emissions were observed in Ph10
332 (0.029 kg CO_{2eq} kg^{-1} fruit). The agricultural practices during the cultivation period showed to be the
333 second greatest source of GHG, with the exception of Ph10 where the use of agrochemicals accounted
334 for the 22.4% of total CO_{2eq} emissions followed by agricultural practices with 21.4% (Table 4).
335 Unlike fertilizers use, the agricultural practices showed the highest CO_{2eq} emission value in Pr20. It

336 is interesting to observe the high relevance of orchard establishment on CO₂eq emission ranging from
337 5.8% of Ph10 to 21.7% for Ph15. Because of the scarcity of precipitations during the summer period,
338 irrigation too showed a significant impact on CO₂eq emission, with the highest value in Ki30 (0.045
339 kg CO₂eq kg⁻¹ fruit) and the lowest one in Ph10 (0.0081 kg CO₂eq kg⁻¹ fruit).

340 In the selected orchards, soils showed a yearly increase of organic C stock (C_{soil}) in the 0 – 30 cm
341 depth (Table 5). The highest soil organic C accumulation rate was observed in Ph10 (2294 kg C ha⁻¹
342 year⁻¹), while the lowest one was found in Ki30 (646 kg C ha⁻¹ year⁻¹).

343 The conversion of a field for grains production to peach and pear orchards had a positive effect on C
344 immobilization (Table 5). Conversely, kiwi cultivation seemed to be an agroecosystem that promotes
345 C release to the atmosphere. Specifically, the highest C storage rates (C_{bal}) were observed in peach
346 orchards (1515 and 1580 kg C ha⁻¹ year⁻¹ in Ph10 and Ph15, respectively), while Ki30 showed a C
347 loss of 117 Mg ha⁻¹ year⁻¹.

348

349 **Discussion**

350 *Soil chemical properties*

351 SOC content and C stock of the CK plot (8 g kg⁻¹ and 31 Mg ha⁻¹, respectively) were similar to that
352 found in Cambisols of croplands in the Emilia-Romagna region and in the plain of northern Italy
353 (Vittori Antisari et al., 2021a; Brombin et al., 2020; Dal Ferro et al., 2020; Lugato et al., 2007)
354 suggesting its representativeness as reference soil.

355 The increased SOC concentration and C stock in soils due to the land use change from wheat
356 production to orchard would suggest the capacity of orchards cultivation systems to enrich soil of
357 organic matter. Several studies (e.g., Massaccesi et al., 2018; Neilsen et al., 2014) found an increase
358 in organic carbon amount after orchards establishment. Specifically, a mean C stock of 57 Mg ha⁻¹
359 in 0–30 cm depth was observed which was similar to the values reported by previous studies
360 conducted in Europe (e.g., Álvaro-Fuentes et al., 2012; Bateni et al., 2021; Funes et al., 2019). The
361 increased C stock could be mainly attributed to the presence of a permanent herbaceous plants

362 established on whole surface of the fields which is worldwide recognized to increase soil C stock (de
363 Torres et al., 2021; Xiang et al., 2022; Novara et al., 2019). In fact, the conversion of cropland to
364 grassland promotes SOC storage (Auerswald and Fiener, 2019) due to the higher root turnover in
365 grasslands compared to cropland and due to the harvest of the whole aboveground biomass in
366 cropland (Poeplau and Don, 2013). Since root derived C through rhizodeposition processes and root
367 turnover (De Feudis et al., 2016; Douglas et al., 2020) has been identified as the major source of SOC
368 (Rasse et al., 2005), the presence of trees and perennial grasses may explain higher SOC accumulation
369 in orchards compared to CK. Such differences were marked in surface soil mainly due the generally
370 larger distribution of roots in the surface soil (Forey et al., 2017; Ruiz-Sánchez et al., 2005; Sokalska
371 et al., 2009) and to the degradation of the chopped pruning residues left on soil surface (Massaccesi
372 et al., 2018; Zhao et al., 2017). The greater influence of fruit orchards on surface soil compared to
373 subsurface soil can be confirmed by the higher SOC content and C stock in the former in Ph15, Pr20
374 and Ki30. Because of the role of SOC on soil microbial activity (e.g., Martínez-García et al., 2018),
375 the higher amount of organic matter in the surface soil might explain the generally higher soil
376 microbial respiration and biomass in the superficial soil layer.

377 The generally homogeneous $\delta^{13}\text{C}$ values would indicate that orchard cultivation did not affect the
378 organic matter decomposition (Blagodatskaya et al., 2011; Solomon et al., 2002). The unchanged
379 SOC decomposition rate could be confirmed by the negligible differences between CK and the
380 considered orchards of those biochemical indicators (i.e., SBR, RCUM, C_{mic} , $q\text{CO}_2$ and $q\text{MIC}$) related
381 to C cycle. The similar SOC degradation combined with the high organic material input due to the
382 shredded pruning residues might have promoted an accumulation of NEOC in the surface soil of the
383 orchards. The plant residues could release water-insoluble compounds (e.g., lignin and waxes) and
384 labile substances readily available to microorganisms whose cell residues could bind to soil minerals
385 increasing the NEOM fraction (Hayes et al., 2017; Wang et al., 2021).

386 Like SOC content and C stock, the cultivation of fruit orchards increased the TN content in surface
387 soil. This can be attributed to the addition of N by amendment (i.e., in Ph10) and chemical fertilizers.

388 The higher $\delta^{15}\text{N}$ values in orchards compared to the wheat field might be attributed both to the
389 contribution of N-enriched fertilizers to $\delta^{15}\text{N}$ values and to the preferential microbial utilization of
390 ^{14}N compounds (Boström et al., 2007; Lobe et al., 2005). The latter maybe limited under Ki30.
391 It was interesting to note that for the subsurface soil, among the selected orchards, Pr20 showed the
392 lowest humic-like C content which would cause a limited SOC stabilization (Martins Gomes et al.,
393 2018). The limited SOC stabilization might be due to the less suitable conditions for the soil microbial
394 community which did not allow the transformation of the soil organic matter (Liebich et al., 2007).
395 In fact, the subsurface soil of Pr20 also showed the lowest C_{mic} , q_{Mic} and the highest $q\text{CO}_2$ indicating
396 a lower C use efficiency by the microbial community (Anderson, 2003; Anderson and Domsch, 1989;
397 Okolo et al., 2020) compared to other fields and, therefore, the occurrence of poor conditions (Vittori
398 Antisari et al., 2021). Such unfavourable conditions in subsurface soil for Pr20 was confirmed by the
399 very high Dilly index value, which would suggest the worsening of the energy use efficiency by the
400 microbial community, in turn not promoting organic C accumulation (Dilly, 2005).

401

402 *Life Cycle Assessment*

403 In agreement with previous studies (e.g., Romero-Gómez et al., 2017; Vinyes et al., 2017), this study
404 found that fertilization was the procedure that had the greatest impact on CO_2eq emission from the
405 orchards, accounting for at least 40% of total CO_2eq emission. In this context, it was interesting to
406 observe that, although in Ph10 no chemical fertilizers were applied, the use of organic amendment
407 had a great impact on CO_2eq emissions. In fact, organic amendment production is both an energy-
408 intensive process and a source of methane and nitrous oxide while its application causes N_2O emission
409 (Bacenetti et al., 2016; Galgani et al., 2014). However, because of the greatest use of N and P
410 fertilizers, the highest CO_2eq emission related to fertilizers was observed in Ki30. Indeed, N and P
411 fertilizers are considered highly impacting on climate change, fossil fuel depletion, acidification,
412 eutrophication, and resources depletion (Hasler et al., 2015). This result, together with the highest
413 CO_2eq emission related to the irrigation, would indicate the higher demands of nutrients and water of

414 kiwi plants compared to peach and pear trees (Allen et al., 1998; Carranca et al., 2018; Peticila et al.,
415 2015).

416 The consume of fuel related to agricultural practices as tillage, weed control and pruning showed to
417 be the second most important CO₂eq source. In this sense, Milà I Canals et al. (2006) suggested the
418 use of biofuel in order to limit the impact of the agricultural practices on CO₂ emission.

419 Several studies (e.g., Martin-Gorriz et al., 2020; Vinyes et al., 2017) reported the high impact of
420 agrochemicals on CO₂eq released into the atmosphere. However, in this study the contribution of
421 agrochemicals on CO₂eq emission in Ph15, Pr20 and Ki30 resulted low due to the sustainable
422 approach used on the studied farms. In this context, it was important to highlight the greater
423 contribution of agrochemicals on CO₂eq emissions for Ph10. In this case, the amounts of
424 agrochemicals used was 10 times higher than those used in the other orchards, and they were mainly
425 sulphur based. This higher amounts of agrochemicals can be attributed to the types of agrochemicals
426 generally used in organic farming. These findings are in agreement with the work of Longo et al.
427 (2017) which observed a larger use of pesticides to produce organic apples compared to those
428 produced with conventional approaches.

429 Overall, this study clearly showed how kiwifruit cultivation had the highest impact on GHG emissions
430 mainly due to the high water and nutrient demand, suggesting that such tree species is less suitable
431 than peach and pear for the considered study area.

432

433 *Carbon balance*

434 When taking in account the C–CO₂eq loss by fruit cultivation and C gained and stored into the soil,
435 results from this study would indicate that peach and pear orchard ecosystems promote C
436 sequestration. The capability of the studied orchards to sequester C was mainly attributed to the soil
437 on which they grow. In fact, the investigated soil was able to store each year a large amount of organic
438 C. Notably, such C was stored in the most stable form preventing C to go back to the atmosphere as
439 CO₂ in the short– or mid–term. It is important to note that in the present study we did not consider C

440 fixed in plant biomass because it is not a long-living component. In fact, orchards for fruit production
441 generally have a lifetime of few decades. Also, at the end of the cultivation period the plant biomass
442 is removed and burnt on the field or in thermal power plants or processed for pellet production which
443 are common practices for fruit orchards (Brand and Jacinto, 2020; Giuntoli et al., 2016). Conversely,
444 the organic carbon stored as fulvic-like C, humic-like C and NEOC could have a mean residence time
445 which spans from centuries to thousands of years (Certini et al., 2004; Piccolo, 2002).

446 The generally similar soil microbial efficiency to use C and, therefore, to transform C in stable forms,
447 together with similar $\delta^{13}\text{C}$ values and soil characteristics (e.g., clay content) between the orchards and
448 the reference field would indicate that C sequestration was mainly related to the management
449 practices carried-out in each orchard.

450 Taking in consideration each orchard type, it is important to mention the negative C balance (-117 kg
451 $\text{C ha}^{-1} \text{ year}^{-1}$) of Ki30. The negative value can be mainly attributed to the high inputs (fertilizers and
452 irrigation) requested by the kiwi plants which caused large CO_2eq emission just partly
453 counterbalanced by soil carbon storage processes. Indeed, when taking in consideration the soil
454 environment, generally no differences in SOC content and its chemical forms were found among the
455 selected orchards. Unlike Ki30, Pr20 showed similar values of CO_2eq emissions of peach orchards
456 (Table 5 and Table 6) but a lower mean annual C storage increase (Table 6). The weak mean annual
457 C storage increase in Pr20 could be attributed to the more stressful conditions for the microbial
458 biomass in subsurface soils. Overall, the C balance performed in this study by taking in consideration
459 the topsoil highlighted the importance of SOC sequestration into the LCA of agricultural systems.

460 However, because of its pivotal role on C storage (Guillaume et al., 2022; Antony et al., 2022) and
461 its greater influence on the agricultural managements compared to topsoil (Samson et al., 2021;
462 Osanai et al., 2020), future LCA studies should take into consideration the subsoil and its key role in
463 the overall C cycle. .

464

465 **Conclusions**

466 The results from the present study suggest that the conversion of a field from grains production to the
467 fruit orchards cultivation promoted soil carbon gain. The majority of the gained C was found in the
468 most chemically recalcitrant form suggesting that in the selected fruit orchards the C stabilization
469 processes were promoted. The organic C increase in orchards could be mainly attributed to the
470 permanent grasses covering such fields. However, such increase could be also promoted both by the
471 direct release from plant residues of chemically recalcitrant compounds and by the release of readily
472 available C for microorganisms whose necromass could bind to soil mineral particles. However, the
473 C gain rate is not unlimited as it depends on soil properties (e.g., clay content) as well as on orchard
474 management. For example, in Ki30, soil stored C, but it was not able to counterbalance the GHG
475 emissions coming from the cultivation of kiwi though it had similar clay content and similar
476 biochemical properties of the reference field. A key tool in this sense may therefore be LCA as it
477 allows us to take into consideration soil resources and their contribution. The systematic inclusion of
478 soil in LCA would allow to enhance agroecosystems sustainability and give soil resources their
479 rightful place in the quest to tackle sustainable development goals and combat climate change.
480 Therefore, we propose to insert the soil C storage rate as CO₂ soil uptake from atmosphere lowering
481 the environmental impacts of orchards management. Finally, although the present study only
482 considered topsoil (0–30 cm depth), in future LCA procedures that also considered deep soil would
483 provide an important additions to give a more realistic view of the role of soil on the mitigation of
484 the GHG emissions coming from the cultivation practices.

485

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489

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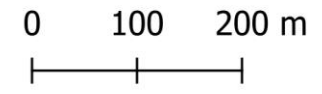
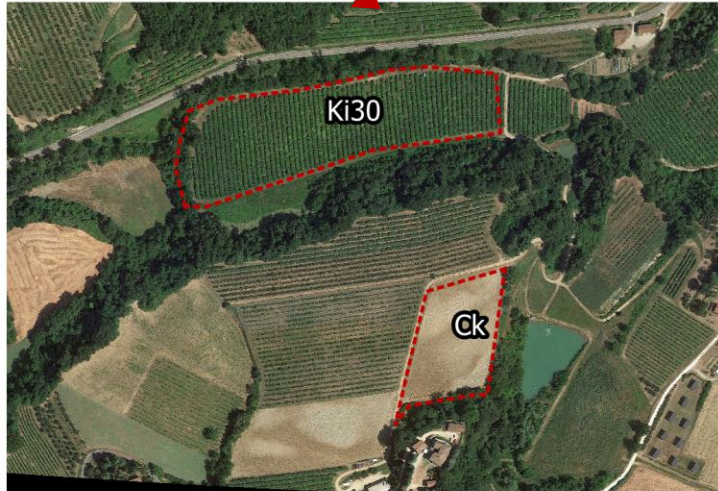
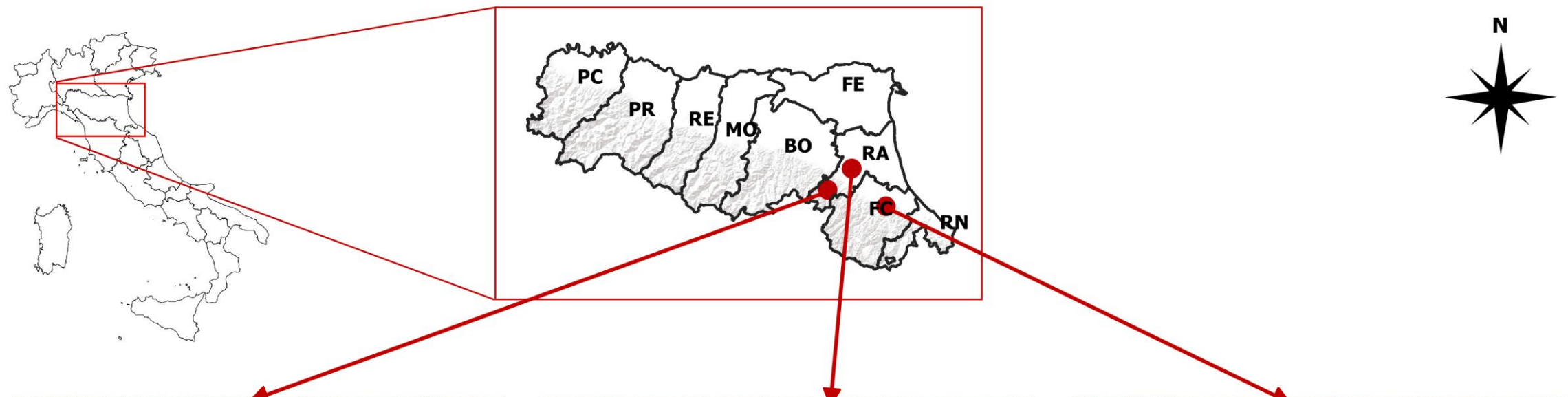
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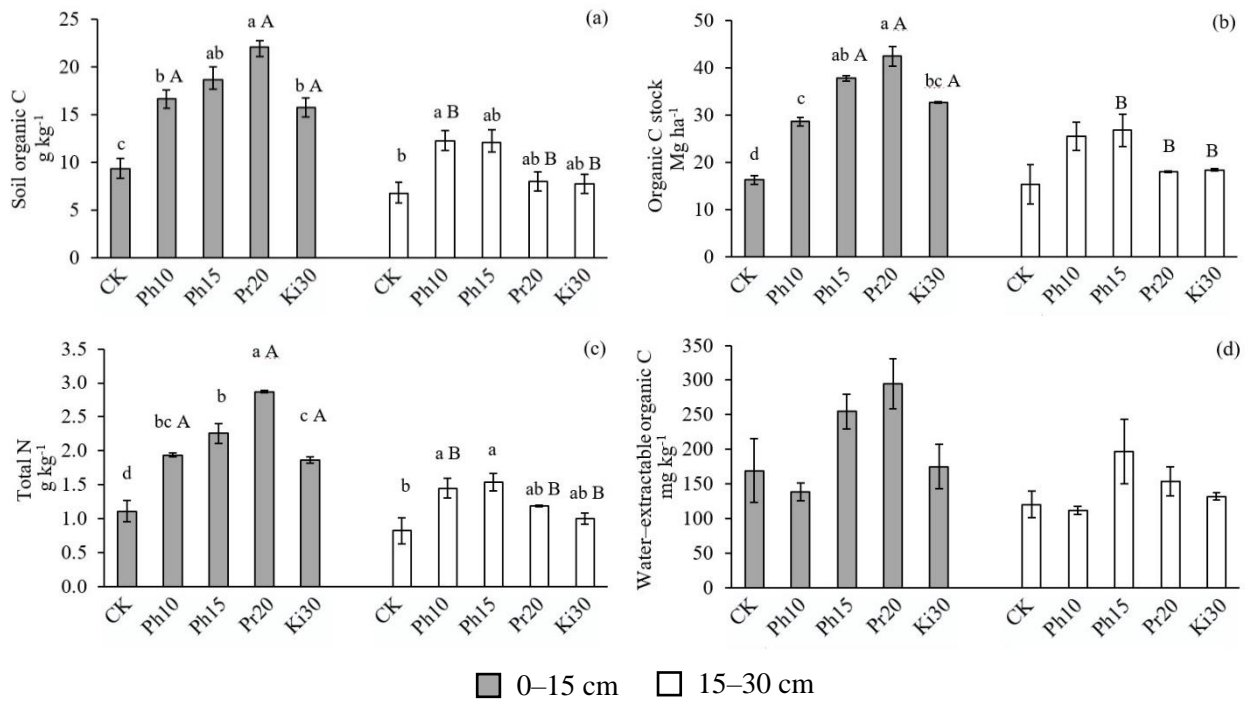
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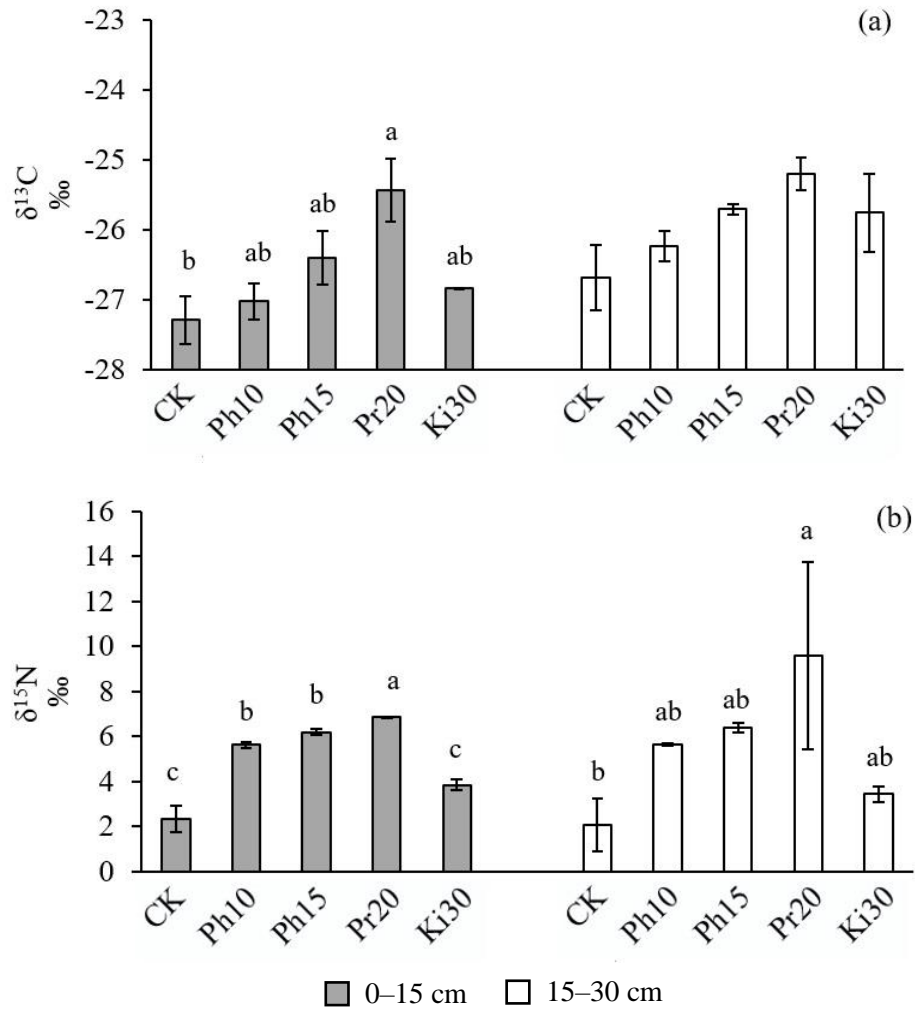
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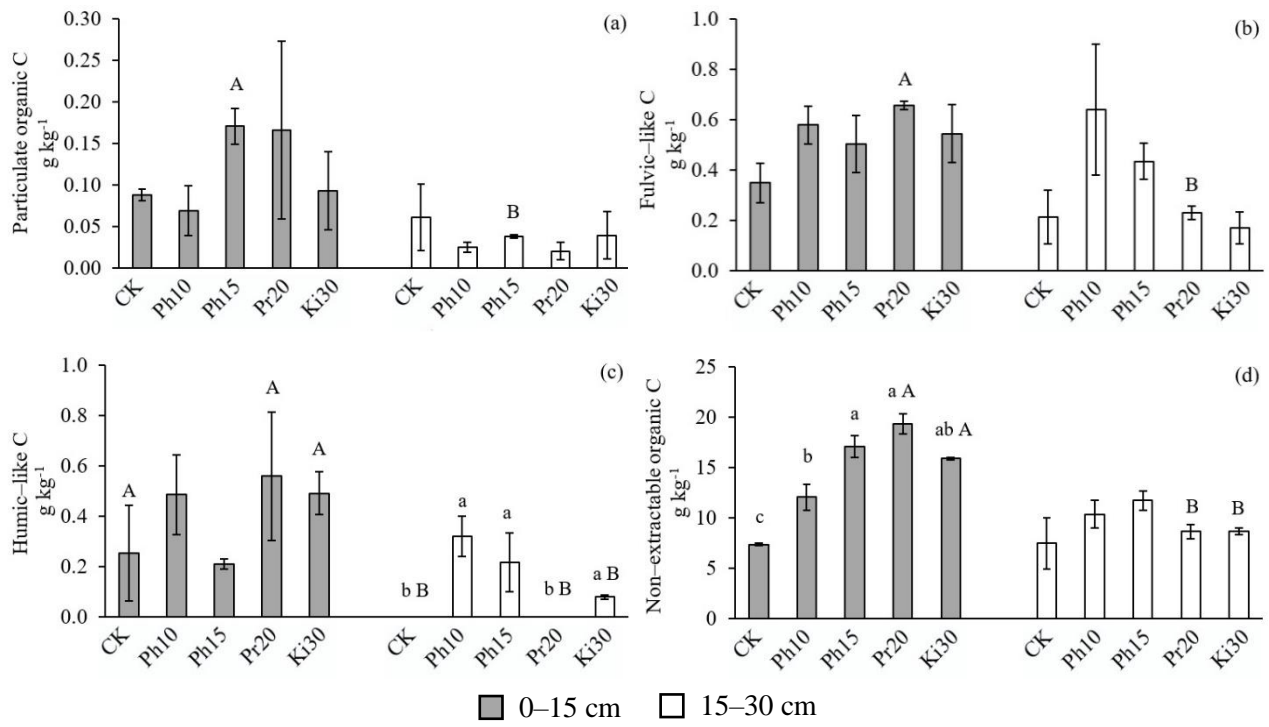
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Figure 1









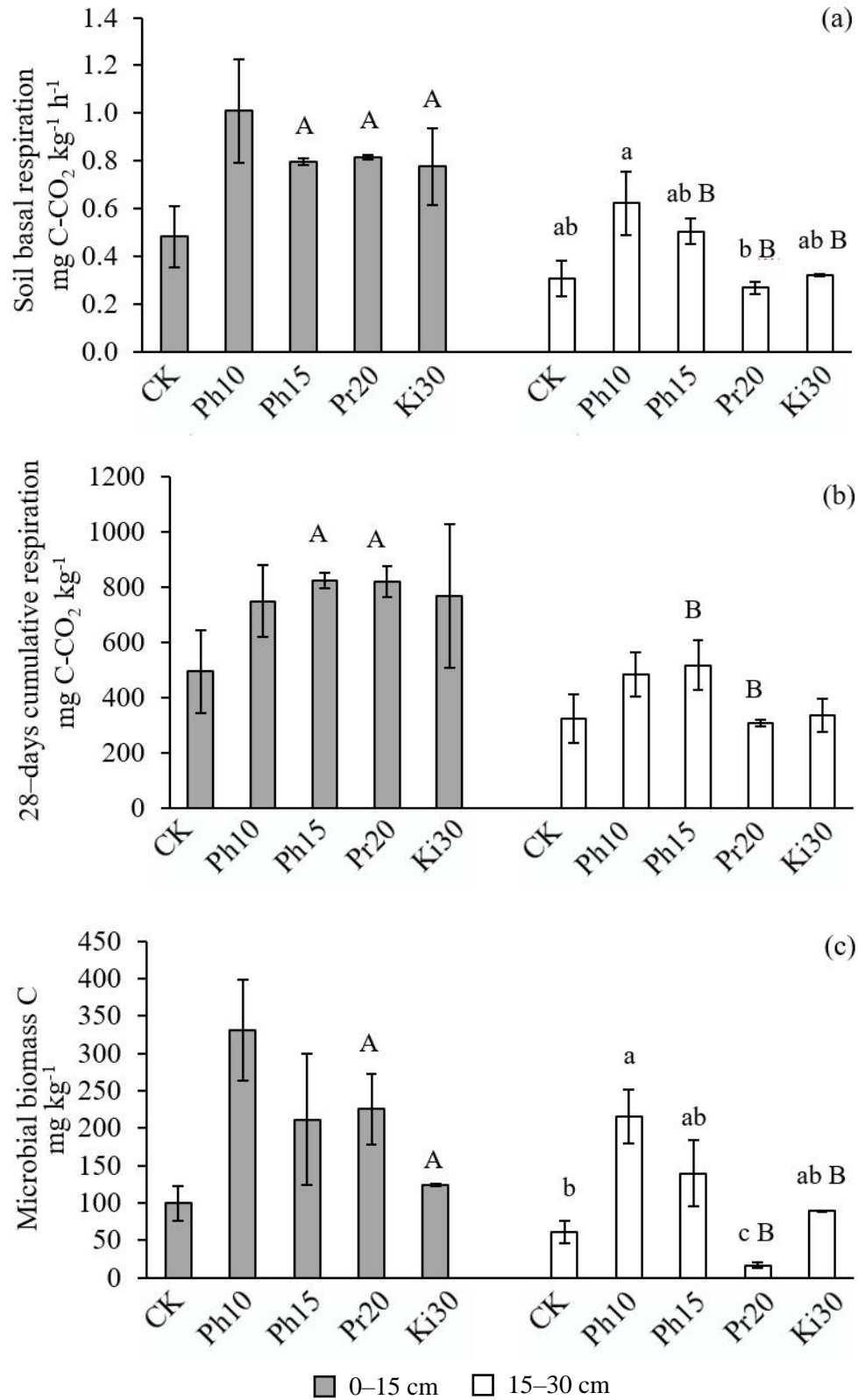


Figure captions

Figure 1. Study site locations. CK: field for grains production; Ph10: 10-years-old peach orchard; Ph15: 15-years-old peach orchard; Pr20: 20-years-old pear orchard; Ki30: 30-years-old kiwi orchard.

Figure 2. Soil organic C content (a), organic C stock (b), total N content (c) and water-extractable organic C content (d) in 0–15 (grey bars) and 15–30 cm (white bars) soil depths of a field for grains production (CK), a 10-years-old peach orchard (Ph10), a 15-years-old peach orchard (Ph15), a 20-years-old pear orchard (Pr20) and a 30-years-old kiwi orchard (Ki30). Error bars represent standard errors. Within the same soil depth, different lowercase letters indicate significant differences among the fields ($P < 0.05$). Within the same field, different uppercase letters indicate significant differences between 0–15 and 15–30 cm soil depths ($P < 0.05$).

Figure 3. $\delta^{13}\text{C}$ (a) and $\delta^{15}\text{N}$ (b) values in 0–15 (grey bars) and 15–30 cm (white bars) soil layers of a field for grains production (CK), a 10-years-old peach orchard (Ph10), a 15-years-old peach orchard (Ph15), a 20-years-old pear orchard (Pr20) and a 30-years-old kiwi orchard (Ki30). Error bars represent standard errors. Within the same soil layer, different lowercase letters indicate significant differences among the fields ($P < 0.05$).

Figure 4. Concentrations of particulate organic C (a), fulvic-like C (b), humic-like C (c) and non-extractable organic C (d) in 0–15 (grey bars) and 15–30 cm (white bars) soil depths of a field for grains production (CK), a 10-years-old peach orchard (Ph10), a 15-years-old peach orchard (Ph15), a 20-years-old pear orchard (Pr20) and a 30-years-old kiwi orchard (Ki30). Error bars represent standard errors. Within the same soil layer, different lowercase letters indicate significant differences among the fields ($P < 0.05$). Within the same field, different uppercase letters indicate significant differences between 0–15 and 15–30 cm soil depths ($P < 0.05$).

Figure 5. Soil basal respiration (a), 28-days cumulative respiration (b) and microbial biomass C content (c) in 0–15 (grey bars) and 15–30 cm (white bars) soil depths of a field for grains production (CK), a 10-years-old peach orchard (Ph10), a 15-years-old peach orchard (Ph15), a 20-years-old

pear orchard (Pr20) and a 30–years–old kiwi orchard (Ki30). Error bars represent standard errors. Within the same soil layer, different lowercase letters indicate significant differences among the fields ($P < 0.05$). Within the same field, different uppercase letters indicate significant differences between 0–15 and 15–30 cm soil depths ($P < 0.05$).

Table 1. Amounts of C, N, P₂O₅ and K₂O applied by soil fertilization (Soil), fertigation (Fert) and by foliar spray (Leaf) application to a 10–years–old peach orchard (Ph10), a 15–years–old peach orchard (Ph15), 20–years–old pear orchard (Pr20) and a 30–years–old kiwi orchard (Ki30) through organic or synthesized fertilizers.

Nutrient	Ph10	Ph15	Pr20	Ki30
	Organic	Synthesized	Synthesized	Synthesized
C (kg ha ⁻¹)	Soil = 3990	Soil = 0	Soil = 0	Soil = 0
	Fert = 0	Fert = 0	Fert = 0	Fert = 0
	Leaf = 0	Leaf = 0	Leaf = 0	Leaf = 0
N (kg ha ⁻¹)	Soil = 140	Soil = 0	Soil = 0	Soil = 54.0
	Fert = 0	Fert = 117.8	Fert = 79.8	Fert = 69.5
	Leaf = 0	Leaf = 1.4	Leaf = 5.2	Leaf = 0
P ₂ O ₅ (kg ha ⁻¹)	Soil = 80	Soil = 0	Soil = 0	Soil = 0
	Fert = 0	Fert = 36.1	Fert = 38.5	Fert = 54.3
	Leaf = 0	Leaf = 3.3	Leaf = 1.2	Leaf = 1.7
K ₂ O (kg ha ⁻¹)	Soil = 153	Soil = 0	Soil = 0	Soil = 0
	Fert = 0	Fert = 47.0	Fert = 148.5	Soil = 1.2
	Leaf = 0	Leaf = 2.6	Leaf = 1.2	Fert = 115.9

Table 2. Amounts of fuel, electricity and agrochemicals consumed in a 10–years–old peach orchard (Ph10), a 15–years–old peach orchard (Ph15), 20–years–old pear orchard (Pr20) and a 30–years–old kiwi orchard (Ki30).

Input	Unit	Ph10	Ph15	Pr20	Ki30
Fuel consumption	kg ha ⁻¹	414	405	528	484
Electricity	kwh ha ⁻¹	600	810	575	1944
Agrochemicals	kg ha ⁻¹	223	21	51	29

Table 3. Metabolic quotient (qCO₂), microbial quotient (qMIC) and Dilly index in 0–15 and 15–30 cm depth intervals in a reference field (CK), a 10–years–old peach orchard (Ph10), a 15–years–old peach orchard (Ph15), 20–years–old pear orchard (Pr20) and a 30–years–old kiwi orchard (Ki30). Standard error is reported in brackets. Different uppercase letters indicate significant differences between 0–15 and 15–30 cm soil depth intervals, different lowercase letters indicate significant differences within the same soil depth interval ($P < 0.05$).

Soil indicator	Soil depth	CK	Ph10	Ph15	Pr20	Ki30
qCO ₂ mg C-CO ₂ h ⁻¹ mg Cmic ⁻¹	0-15	5.20 (1.28)	3.42 (1.25)	4.59 (1.98)	3.77 B (0.74)	6.24 A (1.39)
	15-30	5.07 ab (1.00)	2.89 b (0.34)	4.14 b (1.69)	16.73 a A (4.58)	3.59 b B (0.02)
qMIC mg Cmic g SOC ⁻¹	0-15	10.7 (2.2)	20.0 (4.5)	11.1 (3.9)	10.3 A (2.4)	7.9 B (0.1)
	15-30	9.2 a (1.2)	18.0 a (3.4)	12.2 a (5.1)	2.1 b B (0.44)	11.6 a A (0.6)
Dilly index qCO ₂ /SOC	0-15	570 (141)	203 (71)	256 (125)	170 B (29)	397 (88)
	15-30	791 ab (242)	236 c (10)	331 c (99)	2083 aA (527)	466 bc (25)

SOC = soil organic carbon content

Table 4. Amounts and percentage distribution of carbon dioxide equivalent emitted from the establishment, cultivation and disposal stages of a 10–years–old peach orchard (Ph10), a 15–years–old peach orchard (Ph15), 20–years–old pear orchard (Pr20) and a 30–years–old kiwi orchard (Ki30).

Site	Unit	Establishment stage	Cultivation stage					Disposal stage	Total
			Agricultural practices	Irrigation	Fertilizer production	Fertilizer emissions	Agrochemicals		
Ph10	kg CO ₂ eq kg ⁻¹ fruit yr ⁻¹	0.010	0.037	0.0081	0.049	0.029	0.039	0.00081	0.17
	%	5.84	21.42	4.67	28.40	16.78	22.43	0.47	
Ph15	kg CO ₂ eq kg ⁻¹ fruit yr ⁻¹	0.041	0.043	0.015	0.053	0.034	0.0015	0.0012	0.19
	%	21.72	23.02	7.80	27.95	18.12	0.78	0.62	
Pr20	kg CO ₂ eq kg ⁻¹ fruit yr ⁻¹	0.026	0.066	0.012	0.042	0.034	0.0094	0.0021	0.19
	%	13.26	34.52	6.39	21.97	17.88	4.90	1.07	
Ki30	kg CO ₂ eq kg ⁻¹ fruit yr ⁻¹	0.033	0.067	0.045	0.100	0.049	0.0024	0.0026	0.30
	%	10.85	22.23	14.87	33.91	16.47	0.80	0.88	

The establishment stage included soil preparation, the construction of the fixed structures (irrigation system and supporting structures) and trees plantation. Agricultural practices included fuel consumption, machinery use, pruning, pest and weed control, fertilizers distribution. Fertilizer production equates to the kg CO₂eq emission related to the industrial production phase of fertilizers. Fertilizer emissions equates to the kg CO₂eq of green–house gas emissions from soil (e.g., N₂O) once the fertilizers were distributed. Agrochemicals equates to the kg CO₂eq emission related to the industrial production phase of them. Wastes equates to the disposing of wastes collected during orchard establishment and cultivation stages to thermal–power plants or to landfills.

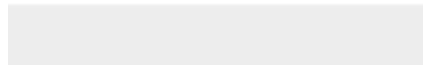
Table 5. Yearly C loss from fruit production practices (C-LCA), yearly C stock change (C soil) and C soil - C-LCA (Cbal) of a 10-years-old peach orchard (Ph10), a 15-years-old peach orchard (Ph15), 20-years-old pear orchard (Pr20) and a 30-years-old kiwi orchard (Ki30).

	Unit	Ph10	Ph15	Pr20	Ki30
C-LCA	kg C-CO ₂ eq ha ⁻¹ year ⁻¹	734	611	518	763
C soil	kg C ha ⁻¹ year ⁻¹	2249	2191	1440	646
Cbal	kg C ha ⁻¹ year ⁻¹	1515	1580	922	-117



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The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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