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Soil greenhouse gas emissions and crop production with implementation of alley cropping in a Mediterranean citrus orchard

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ABSTRACT

The implementation of alley cropping in orchards has been suggested as a sustainable strategy to increase farmer revenues by crop diversification, enhance soil organic matter (SOM) and fertility, water retention, overall biodiversity, and contribute to climate change mitigation. Thus, the objective of this study was to assess if alley cropping with annual crops can contribute to i) mitigate soil greenhouse gas (GHG) emissions, ii) enhance C sequestration in a semiarid Mediterranean irrigated citrus orchard, and iii) increase land productivity. For this, two different treatments were established: i) conventional mandarin monoculture (MC) with no alley cropping; and ii) mandarin diversified with alley cropping of barley/vetch and fava bean (DIV). Measurements of soil CO₂ and N₂O emissions were periodically performed (every 7-20 days) during two years. Soil CO₂ emission rates followed the soil moisture trend, and showed no significant differences between treatments. As an average, soil CO_2 emission rates were 147 mg m⁻² h⁻¹ in MC and 196 mg m⁻² h⁻¹ in DIV. Soil N₂O emission rates were not correlated to soil moisture nor temperature, and showed average values of 0.026 mg m $^{-2}$ h $^{-1}$ in MC and - 0.002 mg m⁻² h⁻¹ in DIV. Alley cropping did not contribute to significantly increase soil organic C and total nitrogen in two years' time. With regard to production, mandarin yield showed no significant differences between treatments, but alley crops contributed to complementary commodities to the main cash crop, increasing overall land productivity. Thus, alley cropping in irrigated Mediterranean orchards has no significant effect on soil C sequestration and GHG emissions at short-term, with increased land productivity owing to new commodities grown in the alleys. These results confirm that under semiarid Mediterranean climate, long periods are needed to efficiently assess soil C sequestration potential of sustainable practices in orchards.

1. Introduction

The green revolution was a milestone in the last century since it allowed the increase of crop production worldwide, thanks to the adoption of high-yielded crop varieties, chemical fertilizers, pesticides and nd mechanization (Pingali, 2012). This new paradigm was fostered for decades leading to the current agri-business models based on monocultures and intense mechanization and dependence on external inputs (Morugán-Coronado et al., 2020). However, it is of international consensus that this production model has also led to environmental degradation, with soil and water pollution, decrease of biodiversity, loss of soil health, loss of soil by erosion, greenhouse gas (GHG) emissions, higher incidence of pests/diseases with higher resistance to pesticides, and, in the end, low resilience of the agro-ecosystems (Maleki et al., 2021; Martínez-Núñez et al., 2020; Morugán-Coronado et al., 2020; Zeng et al., 2022). This fact is even aggravated in most tree orchards under Mediterranean conditions, where trees are irrigated and alleys remain bare to avoid competition for water and nutrients with weeds or other crops. In addition, there has been a traditional social belief that maintaining vegetation cover in the alleys is associated to abandonment and dirt, and so weeds are controlled by tillage or herbicides application (Cerdà et al., 2018; Tsanis et al., 2021). This is usually associated to the

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Received 28 April 2022; Received in revised form 5 October 2022; Accepted 1 November 2022 Available online 17 November 2022 1161-0301/© 2022 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/). lack of consideration of soil as a non-renewable resource, but as a mere physical substrate (Cerdà et al., 2018). As a consequence, soil erosion rates in Mediterranean orchards are high, with high mineralization of soil organic matter (SOM) and GHG emissions, leading to a soil with poor structure and low content of SOM and nutrients, with low capacity to hold water and favor infiltration (Cerdà et al., 2021; Martínez-Mena et al., 2019; Tsanis et al., 2021).

The use of cover crops is being fostered in orchards worldwide to counteract the negative effects of keeping the soil bare, with positive effects such as enhancement of SOM, increased soil fertility, higher water retention capacity and attraction of auxiliary fauna, increasing overall biodiversity (Almagro et al., 2017; Martínez-Mena et al., 2019; Martínez-Núñez et al., 2020). However, their use is still not generalized in the Mediterranean region, and more incentives are needed to encourage farmers to adopt this practice. In this sense, alley cropping could be a suitable strategy to achieve environmental benefits but also increasing land profitability by production of complementary commodities (Swieter et al., 2021; Yang et al., 2021). Alley cropping is the agronomic strategy of growing annual or perennial crops in the alleys of tree orchards to increase farmer revenues, while increasing the land productivity by producing several commodities in the same space during the same year (Tsonkova et al., 2012; Wolz and DeLucia, 2018). If properly selected and managed, alley crops can increase soil organic matter content in the soil, contributing to soil C sequestration (Reyes et al., 2021), can increase soil fertility by stimulation of soil microorganisms that can fix atmospheric N or solubilize soil nutrients (Beule and Karlovsky, 2021) and can attract pollinators and auxiliary fauna to fight against pests (Morugán-Coronado et al., 2020; Staton et al., 2019). Under this strategy, farmers could reduce the use of external fertilizers and pesticides while increase C sequestration in soil and biomass, reducing the C footprint of the agroecosystems (Pavlidis et al., 2020). In fact, this strategy is aligned with the European Green Deal (European Commision, 2019) and the European Climate Law (European Commission, 2020), which aim to make a fair transition in the EU's economy to achieve climate neutral farms by 2050. In this sense, agricultural soils present a unique opportunity for C sequestration and compensating emissions by sustainable cropping systems (Chabbi et al., 2017).

It is essential, so, to define what strategies can lead to climate neutrality in farms by reducing GHG emissions and increasing C sequestration in soil and biomass. For this, a proper selection of species as alley cropping is vital. Legumes have been proposed as perfect candidates in crop diversification, since they can improve soil fertility by biological N fixation with their symbiosis with rhizobia, with higher efficiency in the use of N (Sánchez-Navarro et al., 2019). Besides, legumes have a very active rhizodeposition, stimulating microbial communities that can stabilize organic matter in the soil and solubilize essential nutrients to the plant (Sánchez-Navarro et al., 2020), attracting beneficiary insects into the farm (Cole et al., 2022).

Hence, there is a need to provide scientific data about agronomic strategies than can foster climate neutrality in farms, by decreasing GHG emissions and increasing C storage. In previous research, we focused on GHG emissions on the orchard tree rows and how irrigation affects their trends (Zornoza et al., 2018, 2016); so, new information is needed on how alley management can contribute to GHG in tree orchards. Accordingly, the objectives of this study were to: i) assess if alley cropping with the multiple cropping of barley/vetch and fava bean can contribute to mitigate alley soil GHG emissions and increase soil C sequestration in a Mediterranean irrigated citrus orchard; and ii) evaluate if alley cropping can contribute to increase land productivity. Thus, we hypothesized that the growth of alley crops among the tree rows may increase soil CO2 and N2O emissions by active rhizodeposition and stimulation of soil microbial communities. However, higher organic outputs by root exudates and plant residues would foster an increase in SOM, which may counteract increased GHG emissions; so, the net balance would be C sequestration in the system. The growth of barley/vetch and fava bean in the same space than the citrus trees would increase land

productivity by the harvest of new products, with no negative effect on the main tree crop yield.

2. Materials and methods

2.1. Study site and experimental design

This experiment was performed from February 2019 to March 2021 in a commercial mandarin orchard (Citrus reticulata Blanco var. Clemenvilla), located in Murcia, SE Spain (37° 57' 31''N; 0° 56' 17'' W). The C. reticulata orchard had an extension of 2.3 ha, with 970 trees, at a spacing of 6 m between rows and 4 m between trees within the same row, planted in the year 2000. The climate is semiarid Mediterranean with mean annual temperature of 18 °C and mean annual rainfall of 280 mm. The potential evapotranspiration rate is 1300 mm year $^{-1}$. The soil is a Calcaric Regosol (IUSS Working Group WRB, 2014), with silt loam texture (7 %, 70 % and 23 % of sand, silt and clay, respectively), 55 % of CaCO₃ content, pH of 7.56, bulk density of 1.20 g cm⁻³, total organic carbon of 7.6 g kg⁻¹, total nitrogen of 1.02 g kg⁻¹ and a cation exchange capacity of 14.5 cmol₊ kg⁻¹. A drip irrigation system was installed in all tree rows, with one line per tree row and 3 pressure-compensated emitters (4 L h⁻¹) per tree. Irrigation was scheduled weekly and applied at night or early morning. The frequency of irrigation varied according to the evaporative demand which was 1-2 times per week in winter, 2-7 times per week in spring and autumn, and 7-14 times per week in summer. The EC of the irrigation water varied between 1.0 and 1.5 dS m^{-1} .

Two different treatments were established as split-plot design with three replicates in February 2018. Plots of 12 m \times 28 m were established, with the long side of each one following the direction of the maximum slope, including three rows of 6 trees. The average plot slope was 12 %. Treatments were: i) mandarin monoculture with no alley cropping (MC); and ii) mandarin diversified (DIV) with fava bean (Vicia faba L.) grown from September to December for human consumption and barley/vetch (Hordeum vulgare L./Vicia sativa L.) grown from February to June for feed as alley cropping (see photos of the plots in the Figs. S1-3 of the Supplementary material). Fava bean seeds were manually sown under drip irrigation in three rows in each alley in early September 2018, 2019 and 2020, with a spacing of 100 cm between rows and 40 cm between plants (2.5 plants m^{-2}). Crop residues were incorporated in the soil after harvest of pods. Barley/vetch seeds (1:3 ratio) were manually sown at 150 kg ha⁻¹ covering the entire alley surface in early February 2018, 2019, 2020 and 2021. Three drip irrigation lines were established for irrigation of alley crops in DIV with pressure-compensated emitters (4 L h⁻¹) every 40 cm. Tillage was performed (0-30 cm) immediately before sowing the alley crops to prepare the soil and after the harvest of aerial parts for animal feed to incorporate crop residues into the soil. For DIV, the alley crop water needs were estimated as the product of precipitation, reference crop evapotranspiration (ET_0) and the crop coefficients (between 0.25 and 0.55) proposed by the Agricultural Information System of Murcia (http://siam.imida.es) for this area. Consumption of water in the alley crops in DIV was 1770 m³ ha⁻¹ for fava bean and 1280 m³ ha⁻¹ for barley/vetch (a total of 3050 m³ ha⁻¹ for the two years of study).

Usual cultural practices (e.g. fertilization, pruning, fruit thinning and banding) were carried out by the technical department of the commercial orchard. Weed control was carried out in MC by tillage (chisel plowing 4–5 times yr⁻¹ at 20 cm depth), while weeds were not controlled in DIV. Pruning residues were chopped *in situ* and left as much on the soil surface in the monoculture. Pruning residues were not applied as mulch in DIV because at that time alley was cover with barley/vetch crop. So, pruning residues were removed from the system. Table S1 shows the C and N inputs applied to soil as pruning residues in MC and alley crop residues in DIV. Fertilizers were applied as fertigation by use of the commercial products Neptuno PK 28, Neptuno Triton and Neptuno Pandora (Medifer, Constantino Gutiérrez, SA), as a mixture of soluble N, P, K, Ca, Mg and chelated micronutrients. No irrigation was performed in the allays of MC. Table S2 shows the quantity of N, P and K added with fertilizers per treatment and crop each month.

2.2. Soil greenhouse gas measurements

Measurements of soil CO2 and N2O were made every 7-20 days, depending on climate conditions, in all replicated treatments from 05/ 04/2019 to 04/03/2021, between 9:00 and 11:00. The basic experimental procedure used in this study was the dynamic gas chamber technique. The chamber was made of non-oxidizable steel, with a diameter of 7.5 cm and a height of 20 cm, with one inlet and one outlet connected to a photoacoustic infrared spectroscopy multi-gas analyser with ultra-sensitive cantilever pressure sensor (Gasera One, Gasera Ltd). The dynamic system with inlet and outlet in the chamber permits a continuous flow and avoids pressure fluctuations. The chambers were adjusted over a non-oxidizable steel base which was randomly inserted into the bare soil to a depth of 15 cm in the middle of the alleys in MC and in DIV when no alley crop was present and with the barley/vetch crop. Chambers were randomly inserted between two fava bean plants in the central row. Bases were kept inserted in the soil and without vegetation, except for tillage events, when they were removed and replaced. Inserts were also kept out of vegetation in a surrounding ring of 20 cm wide to reduce the effect of autotrophic respiration (Smith et al., 2010). CO_2 and N_2O were quantified every 1 min for a period of 5 min to assess the linear trend. CO2 and N2O emissions rates were expressed in mg m⁻² h⁻¹ as the difference between the quantification at the end and the beginning of the measure period divided by the time. CO2 and N2O cumulative emissions for each treatment were estimated by numerical integration (Chen et al., 2013). GHG emissions were converted into CO2equivalent (CO2e) for cumulative emission data (g m⁻²) for the experimental period. For this, N₂O emissions were converted into CO2e according to their global warming potential, which is 265 (Vasconcelos et al., 2022). Overall CO2e emission was the sum of CO2 emissions and N2O emissions converted into CO2e.

2.3. Crop production, soil sampling and analytical methods

Meteorological data were measured using an automatic weather station located in a nearby orchard (4 km). Soil temperature (T) and soil moisture (M) were measured using a ProCheck and 5TM sensors (Decagon Devices, USA) introduced at 15 cm depth adjacent to the place where GHG measures were done.

Mandarin yield was calculated by weighing all the mandarins harvested directly from the trees in each plot on 24/01/2019, 07/01/2020 and 03/02/2021. Barley/vetch yield was determined by weighing the aerial biomass cut on 13/06/2019 and 22/06/2020 (no commercial production was obtained in 2018). Fava bean was harvested by collecting all the pods in each plot when the seeds were fresh, on 09/01/2019, 23/12/2020 and 12/02/2021, and weighed as crop yield.

Two soil sampling campaigns were performed: 21/02/2019 and 17/02/2021 at two different depths (0–10 cm and 10–30 cm) with an auger. Three composite samples derived from 5 random subsamples were collected in each plot (9 composite soil samples per treatment). Soil cores using steel cylinders were taken to determine soil bulk density (BD). Soil was air-dried for one week and sieved at < 2 mm.

Particle size distribution was measured using an Mastersizer analyser 2000LF (Malvern Instruments). Soil pH and electrical conductivity (EC) were measured in deionized water (1:2.5 and 1:5 w/v, respectively). Total organic carbon (TOC), total inorganic C and total nitrogen (Nt) were determined by an elemental CHNS-O analyser (EA-1108, Carlo Erba). Soil NH⁺₄ was extracted with 2 M KCl in a 1:10 soil:extractant ratio and calorimetrically measured (Kandeler and Gerber, 1988; Keeny and Nelson, 1982). Soil NO⁻₃ was extracted with deionized water in a 1:10 soil:extractant ratio and measured by ion chromatography (Metrohm 861).

2.4. Statistical analysis

Data were checked to ensure normal distribution using the Kolmogorov-Smirnov test at P < 0.05. Homoscedasticity was checked by the Levene test. GHG data were submitted to two-way repeated measures ANOVA, with measurement date as within-subject factor, and treatment (MC and DIV) as between-subject factor. Crop yield and GHG data were submitted, independently for each date, to a t-test to compare significant differences between MC and DIV. Soil data were submitted to two-way repeated measures ANOVA, with sampling date (2019 and 2021) as within-subject factor, and treatment (MC and DIV) as between-subject factor. Relationships among properties were studied using Pearson correlations. Statistical analyses were performed with the software IBM SPSS for Windows, Version 20.

3. Results

3.1. Crop yields

Mandarin crop yield showed no significant differences between treatments in the experimental period of study (Table 1). However, during the last crop cycle, mandarin yield decreased ten times the values obtained in the first year, owing to intense affection of a *Alternaria* sp. that hindered fructification of most flowers. Fava bean and barley/vetch contributed to complementary commodities to the main cash crop, which supposed 10–15 % of overall land production during the first two years but reached 50 % of overall land production in the second year owing to the low mandarin yields. In fact, overall land productivity in DIV (8565 kg ha⁻¹) was significantly higher than in MC (3020 kg ha⁻¹) during the last year, with no significant differences the previous years.

3.2. Greenhouse gas emission rates

Soil CO₂ emission rates followed the soil moisture trend, as shown in Fig. 1, with a positive significant correlation between both properties (R = 0.41; P < 0.01). Highest CO₂ emissions were associated to highest soil moisture and high temperature values. Alley cropping contributed to significantly increase (P < 0.05) soil moisture compared to monoculture owing to irrigation (Fig. 1). As an average, soil moisture was 11.6 % in MC and 14.9 % in DIV for the entire experimental period (24 months).

Table 1

Crop yield for mandarin trees and alley crops in the two crop cycles of study (kg ha⁻¹). Values are mean \pm standard error (n = 3). Overall land productivity represents the sum of all crop yields during that cycle.

Treatment	Mandarin	Barley/ vetch	Fava bean	Overall land productivity
Cycle 2018/2019				
Diversified system	$\begin{array}{c} \textbf{18,676} \pm \\ \textbf{869} \end{array}$	-	$\begin{array}{c} 1019 \pm \\ 254 \end{array}$	$\textbf{19,695} \pm \textbf{2520}$
Monoculture	$\begin{array}{c} \textbf{20,922} \pm \\ \textbf{2509} \end{array}$	-	-	$\textbf{20,922} \pm \textbf{2509}$
t-Student	1.587 ns			0.278 ns
Cycle 2019/2020				
Diversified system	$33,107 \pm 4721$	$\begin{array}{c} 4728 \pm \\ 258 \end{array}$	1589 ± 164	$\textbf{39,421} \pm \textbf{1686}$
Monoculture	$45,436 \pm 1860$	-	-	$\textbf{45,436} \pm \textbf{4950}$
t-Student	1.281 ns			-2.25 ns
Cycle 2020/2021				
Diversified system	$\begin{array}{c} 4456 \pm \\ 1853 \end{array}$	$\begin{array}{c} 1417 \pm \\ 141 \end{array}$	$\begin{array}{c} 2691 \pm \\ 271 \end{array}$	8565 ± 1881
Monoculture t-Student	$\begin{array}{l} 3020\pm247\\ \text{-0.728 ns} \end{array}$	-	-	3020 ± 247 -2.92*

ns: not significant (P > 0.05).



Fig. 1. Environmental conditions during the duration of the experiment (top), soil CO_2 emission rates (center) and soil N_2O emission rates (bottom) from the mandarin monoculture and diversified orchard. Values are mean \pm standard error (n = 3). P: precipitation; T: soil temperature. For repeated measures ANOVA data: significant at *** P < 0.001; ns: not significant (P > 0.05).

Soil CO₂ emission rates were, in general, not significantly different between MC and DIV (diversification factor) for the complete experimental period (Fig. 1). However, 11 out of the 60 CO₂ emission rate measures were significantly higher in DIV than in MC, mostly associated to the fava bean growth; these significant increases also appeared in the fallow periods and in one occasion in the 2020 barley/vetch cycle (Fig. 1). There were two occasions when MC had significantly higher emissions than DIV: i) in the fallow period after barley/vetch 2020, and ii) in the fava bean cycle of 2020, both associated to an increase in soil moisture owing to rainfall events. As an average, CO₂ emission rates were 152 \pm 114 mg m⁻² h⁻¹ in MC and 196 \pm 109 mg m⁻² h⁻¹ in DIV for the entire experimental period.

Soil N₂O emission rates were not correlated to soil moisture nor temperature, with a flat trend with small oscillations up and down of 0 mg m⁻² h⁻¹ (Fig. 1). Soil N₂O emission rates were not significantly different between MC and DIV for the complete experimental period.

However, four out of the 60 N_2O emission rate measures were significantly higher in MC, with two episodes in autumn 2019, one episode in late spring 2020 and the last episode in winter 2021. There was only once, on 14/01/2021, when N_2O emission was higher in DIV, when fava bean was growing. As an average, N_2O emission rates were 0.026 \pm 0.114 mg m $^{-2}$ h^{-1} in MC and - 0.002 \pm 0.118 mg m $^{-2}$ h^{-1} in DIV for the entire experimental period.

There was no clear evidence about the direct effect of tillage on changes in soil CO_2 or N_2O emissions (Fig. 1). The incorporation of pruning residues in the alleys of MC on 25/05/2020 was led by a peak in soil CO_2 emissions. However, this peak was also observed in DIV, and so it may be likely due to high soil temperatures during those days, associated to high soil moisture in all plots.

3.3. Overall cumulative emissions

The estimation of cumulative CO_2 , N_2O and CO_2e released during the experimental period confirmed the lack of significant differences between treatments owing to the high temporal variability of these fluxes (Table 2). Nonetheless, DIV tended to decrease the cumulative N_2O emission and to increase the CO_2 emission. Cumulative CO_2 emission was not correlated with any soil property measured (clay, sand, silt, pH, EC, TOC, soil inorganic carbon, Nt, NH_4^+ , NO_3), while cumulative N_2O emission was significantly correlated to soil NO_3 (R = 0.48; P < 0.01), but not with Nt neither NH_4^+ .

3.4. Soil organic carbon and total nitrogen contents

TOC and Nt was initially lower in DIV than in MC at 0–10 cm depth, but alley cropping did not contribute to significantly increase these properties in any of the soil depths in two years' time (Fig. 2).

4. Discussion

Alley cropping was successfully established in the study site, with increased land productivity, achieving several crops. Nonetheless, to ensure alley crop production, irrigation was needed, and thus, soil from the alleys in DIV received more water and nutrients than the alley soil in MC. Root exudates by alley crops may have stimulated and activated soil microbial communities, as previously reported (D'Hervilly et al., 2021; Wachendorf et al., 2020). An evidence of that is that fava bean showed no nodules during the first cycle but developed a lot during the last crop cycle (Supplementary Table S3). This is a sign of improvement in soil health by increases in the abundance of beneficial microorganisms (Bertola et al., 2021). Thus, factors introduced in the alleys of DIV and not present in those of MC such as: i) irrigation; ii) fertilization and iii) vegetation growth and inputs of organic compounds by rhizodeposition and crop residues, have likely stimulated microbial communities and so microbial activity should be higher, associated with the metabolic activity of plant roots (root respiration). In this line, D'Hervilly et al. (2021) observed that alley cropping with herbaceous strips showed higher microbial biomass and a higher density of earthworms than the uncultivated alleys. Guillot et al. (2021) also reported that compared to tree monocultures, microbial basal respiration and glucose-induced respiration increased with alley cropping. However, we did not find significant differences in terms of soil CO₂ emissions between the bare alley soil in MC and the plant covered alley soil in DIV in the entire experimental period of 24 months. Thus, despite some isolated episodes with highest CO₂ emissions in DIV, the overall system is not significantly contributing to increase GHG emissions compared to a bare alley, typical of orchards in the Mediterranean regions. Similarly, Almagro et al. (2016) showed no significant differences in soil CO₂ emissions between rainfed almond orchards with bare soil and covered with cover crops. However, it was expected, as initial hypothesis, that higher soil moisture would contribute to even higher CO₂ emissions from alley soil in DIV, as reported by Zornoza et al. (2018). These authors concluded that increases in soil moisture were associated to significant increases in CO₂ emissions in irrigated orchards. On the contrary, the lack of differences

Table 2

Cumulative values of soil CO_2 , N_2O and CO_2 equivalent emissions released from alley soil in the monoculture and diversified treatments during the entire experimental period (11/04/2019 to 04-03-2021).

	Cumulative CO ₂	Cumulative N ₂ O	Cumulative CO ₂ e
	g m ⁻²		
Diversified system Monoculture t-Student	$\begin{array}{l} 3330 \pm 318 \\ 2575 \pm 64 \\ 2.292 \ \text{ns} \end{array}$	$\begin{array}{l} \text{-0.044} \pm 0.199 \\ \text{0.329} \pm 0.176 \\ \text{-1.446 ns} \end{array}$	3316 ± 369 2673 ± 115 1.649 ns

ns: not significant (P > 0.05).



Fig. 2. Evolution of soil total organic carbon (A) and total nitrogen (B) from 2019 to 2021 in the two different systems at the two sampling depths (0–10 cm and 10–30 cm). Vertical bars indicate standard error. MC: monoculture; DIV: diversified orchard. (0–10) and (10–30) in the X file denotes soil depth. No significant differences between sampling dates for each treatment and depth.

in emissions between DIV and MC may indicate that current soil management in MC with frequent tillage and incorporation of pruning residues (Table S1) also contributes to high soil GHG emissions, as reported in literature (García-Orenes et al., 2010; Morell et al., 2010; Pramanik and Phukan, 2020). The lack of correlations between CO_2 emission and soil properties such as texture, pH, EC, TOC, Nt, nitrates and ammonium may suggest that climatic conditions and management are controlling CO_2 emissions by soil microbial activation through the presence of alley crops, tillage or pruning residues additions rather than inherent soil characteristics.

Contrarily to our initial hypothesis, alley cropping, incorporation of fava bean crop residues (with barley/vetch only roots remain in soil) and the likely activation of microbial communities did not lead to increases in TOC neither Nt in the DIV soil at the short term (2 years). This confirms the high organic matter mineralization rates under Mediterranean climate and the long time needed to achieve C storage in these soils, despite the high content of clay. In fact, alley soil in MC also received pruning residues, but this strategy has not contributed in two years' term to evidence an increase in C storage in the soil. This is contrary to a metaanalysis performed on Mediterranean orchards highlighting the positive effect of the implementation of alley cropping or cover crops, minimum or no tillage and on-farm organic amendments to increase TOC, with an average increase of $3.8 \text{ t C ha}^{-1} \text{ yr}^{-1}$ (Vicente-Vicente et al., 2016). However, these authors also indicated that Mediterranean sub-climates of low annual precipitation and high temperatures had very low values of soil C sequestration rate ($< 1 \text{ t C ha}^{-1} \text{ yr}^{-1}$). This is the situation of our study site, with semiarid Mediterranean conditions and 18 °C of mean annual temperature. These results are in agreement with Martínez-Mena et al. (2013), who reported that TOC did not change after three years of implementation of reduced tillage and cover crops in an almond orchard from SE Spain. However, an increase in TOC was observed after 6 years of implementation of these practices compared to

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the intensive tilled almond orchard in the same study site (Martínez-Mena et al., 2021), confirming that under semiarid conditions, C sequestration is a slow and long-term process.

With regard to soil N_2O emissions, it has been reported that the use of legumes can contribute to reduce N_2O emissions by higher efficiency in the use of N (Rochette and Janzen, 2005; Zhong et al., 2009). In fact, soil nitrate content was higher in the alley soil from MC compared to DIV (Supplementary Table S4). This may be due to its uptake by alley crops in DIV, which seems to contribute to decrease N_2O emissions owing to lower nitrate content in the soil, confirmed with the positive correlation between cumulative N_2O emissions and soil nitrate content. This fact may also contribute to sequester mobile N forms by biomass that otherwise can leach and pollute groundwater. In this line, alley cropping and cover crops have been proposed to foster nutrient cycling and avoid nutrient losses by leaching (Bergeron et al., 2011; Wolz et al., 2018).

Hence, results confirm that most variation in GHG emission rates was provided by climatic conditions, being soil moisture the main factor controlling emissions, followed by soil temperature. With this regard, Zornoza et al. (2018, 2016) reported that CO₂ emissions were more influenced by temperature than by moisture when soil water was not limited in Mediterranean irrigated orchards; however, when water content became a limiting factor, CO₂ emissions were more related to soil moisture than to temperature. Thus, since soil heterotrophic activity is regulated by soil water content, CO2 emissions followed the soil water trend, confirming the water deficit in this system. Thus, the higher the soil water content, the higher soil microbial activity and so, the CO₂ emissions (Diao et al., 2022; Song et al., 2018). Higher differences among MC and DIV with regard to soil moisture were found with fava bean crop, mostly during the last cycle 2020, associated with the highest differences in CO2 emissions between MC and DIV. This should be associated to higher microbial activity with high soil moisture. Thereby, although as a general pattern, the DIV system did not significantly contribute to increase CO₂ emissions compared to MC, when fava bean grew, CO2 emissions increased likely due to high soil moisture that enhanced rhizodeposition that stimulated microbial activity.

Alley cropping, when citrus production was high, counted only 15 % of the 2019 annual production, confirming that main revenues will come from the cash crop (mandarin). However, alley cropping reached 50 % of the 2020 annual production owing to a high incidence of a fungal disease that affected mandarin fructification. This confirms the benefits of alley cropping not only from an environmental point of view, but also as an economic benefit since diversified production can make farmers more resilient to negative effects of pests/diseases on one crop. Accordingly, Kurdyś-Kujawska et al. (2021) proved higher economic efficiency in diversified farms compared to monocultural farms in Poland. John et al. (2021) demonstrated that diversifying maize with legumes increased yield stability, nutritional gains, and profitability in central Africa. Di Falco and Zoupanidou (2016) also evidenced the important role of agrobiodiversity in the resilience of agroecosystems, proposing crop diversification as a strategy to support productivity and farmers revenues when soils are less fertile and monocultures cannot provide high yields.

5. Conclusion

Alley cropping with barley/vetch and fava bean contributed to increase land productivity in a mandarin orchard from the Mediterranean region. Alley cropping did not contribute to higher overall GHG emissions, with no differences between bare and plant covered alleys. Soil CO₂ was regulated by soil moisture in the area, with higher emissions during those days with higher soil moisture owing to rainfall events or irrigation in the diversified system. Furthermore, management (alley crops, tillage, incorporation of pruning residues) had higher influence on CO₂ emissions than inherent soil characteristics. The growth of fava bean and barley/vetch seems to contribute to decrease N₂O emissions. After two years, the addition of crop residues in the alley soil was not

enough to increase soil organic C content in the soil, confirming the trend to organic matter mineralization under these Mediterranean climatic conditions. More time is so needed to demonstrate if this practice is actually efficient for soil C sequestration and storage in irrigated orchards from the Mediterranean basin. These findings can encourage farmers, land managers and decision-makers to implement and foster the adoption of alley cropping as a sustainable practice to enhance land production with no negative effect on the environment, with potential positive impact long-term. However, long-term monitoring programs are needed to provide robust data about the impact of alley cropping on the delivery of ecosystem services, since changes in soil are a long process.

CRediT authorship contribution statement

Virginia Sánchez-Navarro: Conceptualization, Investigation, Writing – original draft. Silvia Martínez-Martínez: Conceptualization, Investigation, Writing – review & editing. Jose A. Acosta: Conceptualization, Investigation, Writing – review & editing. María Almagro: Investigation, Writing – review & editing. María Martínez-Mena: Conceptualization, Investigation, Writing – review & editing. Carolina Boix-Fayos: Conceptualization, Investigation, Writing – review & editing. Elvira Díaz-Pereira: Conceptualization, Investigation, Writing – review & editing. Abdelmalek Temnani: Investigation. Pablo Berrios: Investigation. Alejandro Pérez-Pastor: Conceptualization, Writing – review & editing. Raúl Zornoza: Conceptualization, Investigation, Writing – original draft, Data curation, Project administration, Funding acquisition.

Declaration of Competing Interest

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.

Data Availability

Data will be made available on request.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.eja.2022.126684.

References

- Almagro, M., de Vente, J., Boix-Fayos, C., García-Franco, N., Melgares de Aguilar, J., González, D., Solé-Benet, A., Martínez-Mena, M., 2016. Sustainable land management practices as providers of several ecosystem services under rainfed Mediterranean agroecosystems. Mitig. Adapt. Strateg. Glob. Change, vol. 21, pp. 1029–43. (https://doi.org/10.1007/s11027-013-9535-2).
- Almagro, M., Garcia-Franco, N., Martínez-Mena, M., 2017. The potential of reducing tillage frequency and incorporating plant residues as a strategy for climate change mitigation in semiarid Mediterranean agroecosystems. Agric. Ecosyst. Environ. 246, 210–220. https://doi.org/10.1016/j.agee.2017.05.016.
- Bergeron, M., Lacombe, S., Bradley, R.L., Whalen, J., Cogliastro, A., Jutras, M.-F., Arp, P., 2011. Reduced soil nutrient leaching following the establishment of tree-based intercropping systems in eastern Canada. Agrofor. Syst. 833 (83), 321–330. https:// doi.org/10.1007/S10457-011-9402-7.
- Bertola, M., Ferrarini, A., Visioli, G., 2021. Improvement of soil microbial diversity through sustainable agricultural practices and its evaluation by -omics approaches: a

perspective for the environment, food quality and human safety. Microorganisms, vol. 9, Page 1400. (https://doi.org/10.3390/MICROORGANISMS9071400).

Beule, L., Karlovsky, P., 2021. Tree rows in temperate agroforestry croplands alter the composition of soil bacterial communities. PLoS One 16, e0246919. https://doi.org/ 10.1371/JOURNAL.PONE.0246919.

- Cerdà, A., Novara, A., Moradi, E., 2021. Long-term non-sustainable soil erosion rates and soil compaction in drip-irrigated citrus plantation in Eastern Iberian Peninsula. Sci. Total Environ. 787, 147549 https://doi.org/10.1016/J.SCITOTENV.2021.147549.
- Cerdà, A., Rodrigo-Comino, J., Giménez-Morera, A., Keesstra, S.D., 2018. Hydrological and erosional impact and farmer's perception on catch crops and weeds in citrus organic farming in Canyoles river watershed, Eastern Spain. Agric. Ecosyst. Environ. 258, 49–58. https://doi.org/10.1016/J.AGEE.2018.02.015.
- Chabbi, A., Lehmann, J., Ciais, P., Loescher, H.W., Cotrufo, M.F., Don, A., SanClements, M., Schipper, L., Six, J., Smith, P., Rumpel, C., 2017. Aligning agriculture and climate policy. Nat. Clim. Change 7, 307–309. https://doi.org/ 10.1038/nclimate3286.
- Chen, W., Wang, Y., Zhao, Z., Cui, F., Gu, J., Zheng, X., 2013. The effect of planting density on carbon dioxide, methane and nitrous oxide emissions from a cold paddy field in the Sanjiang Plain, northeast China. Agric. Ecosyst. Environ. 178, 64–70. https://doi.org/10.1016/j.agee.2013.05.008.
- Cole, L.J., Baddeley, J.A., Robertson, D., Topp, C.F.E., Walker, R.L., Watson, C.A., 2022. Supporting wild pollinators in agricultural landscapes through targeted legume mixtures. Agric. Ecosyst. Environ. 323, 107648 https://doi.org/10.1016/J. AGEE.2021.107648.
- D'Hervilly, C., Marsden, C., Capowiez, Y., Béral, C., Delapré-Cosset, L., Bertrand, I., 2021. Trees and herbaceous vegetation strips both contribute to changes in soil fertility and soil organism communities in an agroforestry system. Plant Soil 4631 (463), 537–553. https://doi.org/10.1007/S11104-021-04932-X.
- Di Falco, S., Zoupanidou, E., 2016. Soil fertility, crop biodiversity, and farmers' revenues: evidence from Italy. Ambio 462 (46), 162–172. https://doi.org/10.1007/S13280-016-0812-7.
- Diao, H., Wang, A., Yuan, F., Guan, D., Wu, J., 2022. Autotrophic respiration modulates the carbon isotope composition of soil respiration in a mixed forest. Sci. Total Environ. 807. https://doi.org/10.1016/J.SCITOTENV.2021.150834.
- European Commision, 2019. COMMUNICATION FROM THE COMMISSION. The European Green Deal. COM(2019) 640 final.
- European Commission, 2020. REGULATION OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL establishing the framework for achieving climate neutrality and amending Regulation (EU) 2018/1999 (European Climate Law). COM(2020) 80 final, 2020/0036 (COD).
- García-Orenes, F., Guerrero, C., Roldán, A., Mataix-Solera, J., Cerdà, A., Campoy, M., Zornoza, R., Bárcenas, G., Caravaca, F., 2010. Soil microbial biomass and activity under different agricultural management systems in a semiarid Mediterranean agroecosystem. Soil Tillage Res. 109. https://doi.org/10.1016/j.still.2010.05.005.
- Guillot, E., Bertrand, I., Rumpel, C., Gomez, C., Arnal, D., Abadie, J., Hinsinger, P., 2021. Spatial heterogeneity of soil quality within a Mediterranean alley cropping agroforestry system: comparison with a monocropping system. Eur. J. Soil Biol. 105, 103330 https://doi.org/10.1016/J.EJSOBI.2021.103330.
- IUSS Working Group WRB, 2014. International Soil Classification System for Naming Soils and Creating Legends for Soil Maps. World Soil Resources Reports No. 106. https://doi.org/10.1017/S0014479706394902.
- John, I., Snapp, S., Nord, A., Chimonyo, V., Gwenambira, C., Chikowo, R., 2021. Marginal more than mesic sites benefit from groundnut diversification of maize: increased yield, protein, stability, and profits. Agric. Ecosyst. Environ. 320, 107585 https://doi.org/10.1016/J.AGEE.2021.107585.
- Kandeler, E., Gerber, E., 1988. Short-term assay of soil urease activity using colorimetric determination of ammonium. Biol. Fertil. Soils 6, 68–72.
- Keeny, D.R., Nelson, D.W., 1982. Nitrogen inorganic forms. In: Page, A.L. (Ed.), Methods of Soil Analysis. Agronomy Monograph 9, Part 2. Madison, pp. 643–98.
- Kurdyś-Kujawska, A., Strzelecka, A., Zawadzka, D., 2021. The impact of crop diversification on the economic efficiency of small farms in Poland. Agriculture, vol. 11, Page 250. (https://doi.org/10.3390/AGRICULTURE11030250).
- Maleki, S., Karimi, A., Zeraatpisheh, M., Poozeshi, R., Feizi, H., 2021. Long-term cultivation effects on soil properties variations in different landforms in an arid region of eastern Iran. CATENA 206, 105465. https://doi.org/10.1016/J. CATENA.2021.105465.
- Martínez-Mena, M., Carrillo-López, E., Boix-Fayos, C., Almagro, M., García Franco, N., Díaz-Pereira, E., Montoya, I., de Vente, J., 2019. Long-term effectiveness of sustainable land management practices to control runoff, soil erosion, and nutrient loss and the role of rainfall intensity in Mediterranean rainfed agroecosystems. Catena. https://doi.org/10.1016/j.catena.2019.104352.
- Martínez-Mena, M., Garcia-Franco, N., Almagro, M., Ruiz-Navarro, A., Albaladejo, J., de Aguilar, J.M., Gonzalez, D., Querejeta, J.I., 2013. Decreased foliar nitrogen and crop yield in organic rainfed almond trees during transition from reduced tillage to notillage in a dryland farming system. Eur. J. Agron. 49, 149–157. https://doi.org/ 10.1016/j.eja.2013.04.006.
- Martínez-Mena, M., Perez, M., Almagro, M., Garcia-Franco, N., Díaz-Pereira, E., 2021. Long-term effects of sustainable management practices on soil properties and crop yields in rainfed Mediterranean almond agroecosystems. Eur. J. Agron. 123, 126207 https://doi.org/10.1016/J.EJA.2020.126207.
- Martínez-Núñez, C., Manzaneda, A.J., Isla, J., Tarifa, R., Calvo, G., Molina, J.L., Salido, T., Ruiz, C., Gutiérrez, J.E., Rey, P.J., 2020. Low-intensity management benefits solitary bees in olive groves. J. Appl. Ecol. 57, 111–120. https://doi.org/ 10.1111/1365-2664.13511.
- Morell, F.J., Álvaro-Fuentes, J., Lampurlanés, J., Cantero-Martínez, C., 2010. Soil CO₂ fluxes following tillage and rainfall events in a semiarid Mediterranean

agroecosystem: effects of tillage systems and nitrogen fertilization. Agric. Ecosyst. Environ. 139, 167–173. https://doi.org/10.1016/J.AGEE.2010.07.015.

- Morugán-Coronado, A., Linares, C., Gómez-López, M.D., Faz, Á., Zornoza, R., 2020. The impact of intercropping, tillage and fertilizer type on soil and crop yield in fruit orchards under Mediterranean conditions: a meta-analysis of field studies. Agric. Syst. https://doi.org/10.1016/j.agsy.2019.102736.
- Pavlidis, G., Karasali, H., Tsihrintzis, V.A., 2020. Pesticide and fertilizer pollution reduction in two alley cropping agroforestry cultivating systems. Water Air Soil Pollut. 2315 (231), 1–23. https://doi.org/10.1007/S11270-020-04590-2.
- Pingali, P.L., 2012. Green revolution: Impacts, limits, andthe path ahead. Proc. Natl. Acad. Sci. USA. https://doi.org/10.1073/pnas.0912953109.
- Pramanik, P., Phukan, M., 2020. Enhanced microbial respiration due to carbon sequestration in pruning litter incorporated soil reduced the net carbon dioxide flux from atmosphere to tea ecosystem. J. Sci. Food Agric. 100, 295–300. https://doi. org/10.1002/JSFA.10038.
- Reyes, F., Gosme, M., Wolz, K.J., Lecomte, I., Dupraz, C., 2021. Alley cropping mitigates the impacts of climate change on a wheat crop in a mediterranean environment: a biophysical model-based assessment. Agriculture, vol. 11, Page 356. (https://doi.or g/10.3390/AGRICULTURE11040356).
- Rochette, P., Janzen, H.H., 2005. Towards a revised coefficient for estimating N₂O emissions from legumes. Nutr. Cycl. Agroecosyst. 732 (73), 171–179. https://doi.org/10.1007/S10705-005-0357-9.
- Sánchez-Navarro, V., Zornoza, R., Faz, Á., Fernández, J.A., 2020. Comparison of soil organic carbon pools, microbial activity and crop yield and quality in two vegetable multiple cropping systems under mediterranean conditions. Sci. Hortic. 261. https:// doi.org/10.1016/j.scienta.2019.109025.
- Sánchez-Navarro, V., Zornoza, R., Faz, Á., Fernández, J.A., 2019. Comparing legumes for use in multiple cropping to enhance soil organic carbon, soil fertility, aggregates stability and vegetables yields under semi-arid conditions. Sci. Hortic. 246. https:// doi.org/10.1016/j.scienta.2018.11.065.
- Smith, P., Lanigan, G., Kutsch, W.L., Buchmann, N., Eugster, W., Aubinet, M., Ceschia, E., Béziat, P., Yeluripati, J.B., Osborne, B., Moors, E.J., Brut, A., Wattenbach, M., Saunders, M., Jones, M., 2010. Measurements necessary for assessing the net ecosystem carbon budget of croplands. Agric. Ecosyst. Environ. 139, 302–315. https://doi.org/10.1016/J.AGEE.2010.04.004.
- Song, W., Tong, X., Zhang, J., Meng, P., Li, J., 2018. How a root-microbial system regulates the response of soil respiration to temperature and moisture in a plantation. Pol. J. Environ. Stud. 27, 2749–2756. https://doi.org/10.15244/PJOES/ 81271.
- Staton, T., Walters, R.J., Smith, J., Girling, R.D., 2019. Evaluating the effects of integrating trees into temperate arable systems on pest control and pollination. Agric. Syst. 176, 102676 https://doi.org/10.1016/J.AGSY.2019.102676.
- Swieter, A., Langhof, M., Lamerre, J., 2021. Competition, stress and benefits: trees and crops in the transition zone of a temperate short rotation alley cropping agroforestry system. J. Agron. Crop Sci. 208, 209–224. https://doi.org/10.1111/JAC.12553.
- Tsanis, I.K., Seiradakis, K.D., Sarchani, S., Panagea, I.S., Alexakis, D.D., Koutroulis, A.G., 2021. The impact of soil-improving cropping practices on erosion rates: a stakeholder-oriented field experiment assessment. Land 10, 964. https://doi.org/ 10.3390/LAND10090964.
- Tsonkova, P., Böhm, C., Quinkenstein, A., Freese, D., 2012. Ecological benefits provided by alley cropping systems for production of woody biomass in the temperate region: a review. Agrofor. Syst. 851 (85), 133–152. https://doi.org/10.1007/S10457-012-9494-8.
- Vasconcelos, A.L.S., Cherubin, M.R., Cerri, C.E.P., Feigl, B.J., Borja Reis, A.F., Siqueira-Neto, M., 2022. Sugarcane residue and N-fertilization effects on soil GHG emissions in south-central, Brazil. Biomass Bioenergy 158, 106342. https://doi.org/10.1016/J. BIOMBIOE.2022.106342.
- Vicente-Vicente, J.L., García-Ruiz, R., Francaviglia, R., Aguilera, E., Smith, P., 2016. Soil carbon sequestration rates under Mediterranean woody crops using recommended management practices: a meta-analysis. Agric. Ecosyst. Environ. 235, 204–214. https://doi.org/10.1016/J.AGEE.2016.10.024.
- Wachendorf, C., Piepho, H.P., Beuschel, R., 2020. Determination of litter derived C and N in litterbags and soil using stable isotopes prevents overestimation of litter decomposition in alley cropping systems. Pedobiologia 81–82, 150651. https://doi. org/10.1016/J.PEDOBI.2020.150651.
- Wolz, K.J., Branham, B.E., DeLucia, E.H., 2018. Reduced nitrogen losses after conversion of row crop agriculture to alley cropping with mixed fruit and nut trees. Agric. Ecosyst. Environ. 258, 172–181. https://doi.org/10.1016/J.AGEE.2018.02.024.
- Wolz, K.J., DeLucia, E.H., 2018. Alley cropping: global patterns of species composition and function. Agric. Ecosyst. Environ. 252, 61–68. https://doi.org/10.1016/J. AGEE.2017.10.005.
- Yang, T., Ma, C., Lu, W., Wan, S., Li, L., Zhang, W., 2021. Microclimate, crop quality, productivity, and revenue in two types of agroforestry systems in drylands of Xinjiang, northwest China. Eur. J. Agron. 124, 126245 https://doi.org/10.1016/J. EJA.2021.126245.
- Zeng, Q., Mei, T., Wang, M., Tan, W., 2022. Intensive citrus plantations suppress the microbial profiles of the β-glucosidase gene. Agric. Ecosyst. Environ. 323, 107687 https://doi.org/10.1016/J.AGEE.2021.107687.

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- Zhong, Z., Lemke, R.L., Nelson, L.M., 2009. Nitrous oxide emissions associated with nitrogen fixation by grain legumes. Soil Biol. Biochem. 41, 2283–2291. https://doi. org/10.1016/J.SOILBIO.2009.08.009.
- Zornoza, R., Acosta, J.A., Gabarrón, M., Gómez-Garrido, M., Sánchez-Navarro, V., Terrero, A., Martínez-Martínez, S., Faz, Á., Pérez-Pastor, A., 2018. Greenhouse gas emissions and soil organic matter dynamics in woody crop orchards with different

irrigation regimes. Sci. Total Environ. 644, 1429–1438. https://doi.org/10.1016/J. SCITOTENV.2018.06.398.

Zornoza, R., Rosales, R.M., Acosta, J.A., de la Rosa, J.M., Arcenegui, V., Faz, Á., Pérez-Pastor, A., 2016. Efficient irrigation management can contribute to reduce soil CO₂ emissions in agriculture. Geoderma 263, 70–77. https://doi.org/10.1016/j. geoderma.2015.09.003.