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Soil chemical responses to fertilization, with or without a cover crop, in an olive orchard in southwestern Buenos Aires (Argentina)

Respuestas químicas del suelo a la fertilización, con o sin un cultivo de cobertura, en una finca de olivo en el sudoeste de Buenos Aires (Argentina)

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ABSTRACT

Our objective was to study the effect of fertilization on soil chemical traits on an olive orchard (artificially irrigated), considering areas with or without a cover crop, in southwestern Buenos Aires, Argentina, during the period 2020/2021. Fertilization treatments were (1) organic manure applied to the soil near the tree trunk; inorganic fertilization applied to the (2) soil or (3) to the leaves of *Olea europaea* L. trees; and (4) unfertilized control. Seeding of *Vicia benghalensis L.* and *Avena sativa* L. around subplots (one per each of the four studied treatments) constituted the areas with a cover crop. Subplots which were not seeded corresponded to the control areas. Soil pH was lower ($p<0.05$) under organic and inorganic soil fertilization. In areas without a cover crop in April 2021, soil nitrate concentrations were greater ($p<0.05$) under organic soil fertilization than in the control. At 0-20 cm soil depth, P concentrations were greater ($p<0.05$) under organic and inorganic soil fertilizations than in the other treatments. The greatest ($p<0.05$) K

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concentrations were found in the organic fertilization treatment. Organic soil fertilization on areas without a cover crop showed greater values for the soil chemical studied traits.

Keywords: Cover crop; NPK fertilizer; olive; soil quality.

RESUMEN

Nuestro objetivo fue estudiar el efecto de la fertilización sobre las propiedades químicas del suelo en una finca de olivo (bajo riego), considerando áreas sembradas o no con un cultivo de cobertura, en el sudoeste de Buenos Aires (Argentina) durante el periodo 2020/2021. Los tratamientos de fertilización fueron (1) abono orgánico aplicado al suelo cercano al tronco del árbol; fertilización inorgánica aplicada al (2) suelo o a (3) las hojas de los árboles de *Olea europaea* L., y (4) control no fertilizado. La siembra de *Vicia benghalensis* L. y *Avena sativa* L. alrededor de subparcelas (una para cada uno de los 4 tratamientos estudiados) constituyeron las áreas con cultivo de cobertura. Las subparcelas que no fueron sembradas correspondieron a áreas control. El pH del suelo fue más bajo ($p<0.05$) cuando se fertilizó en forma orgánica e inorgánica. En áreas sin cultivo de cobertura en abril, la concentración de nitrato del suelo fue mayor ($p<0.05$) en el tratamiento con fertilización orgánica del suelo que en el control. A 0-20 cm de profundidad del suelo la concentración de P fue mayor ($p<0.05$) en las fertilizaciones orgánica e inorgánica del suelo que en los otros tratamientos. Las mayores ($p<0.05$) concentraciones de K se observaron en el tratamiento de fertilización orgánica del suelo. La fertilización orgánica del suelo en áreas sin un cultivo de cobertura mostró mayores valores para las características químicas del suelo estudiadas.

Palabras clave: Calidad de suelo; cultivo de cobertura; fertilizante NPK; olivo.

INTRODUCTION

Currently, Argentina is the main producer and exporter of olive oil of South America and the 10th at a worldwide level. The major producer Provinces are Catamarca, La Rioja, San Juan, Mendoza and Córdoba (Fig. 1). These Provinces concentrate more than 95% of the total oil country production, with more than 100,000 ha implanted with olive (International Olive Oil Council [IOC], 2015). The southwestern region of the Province of Buenos Aires, Argentina, is integrated by the semiarid, arid and subhumid-dry Pampas, with 6.5 million ha divided in 12 districts. Since the conditions of soil and climate of this region are very different from those of the northwestern Provinces, farmers of the southwestern region of Buenos Aires are very interested in developing management techniques adequate to the

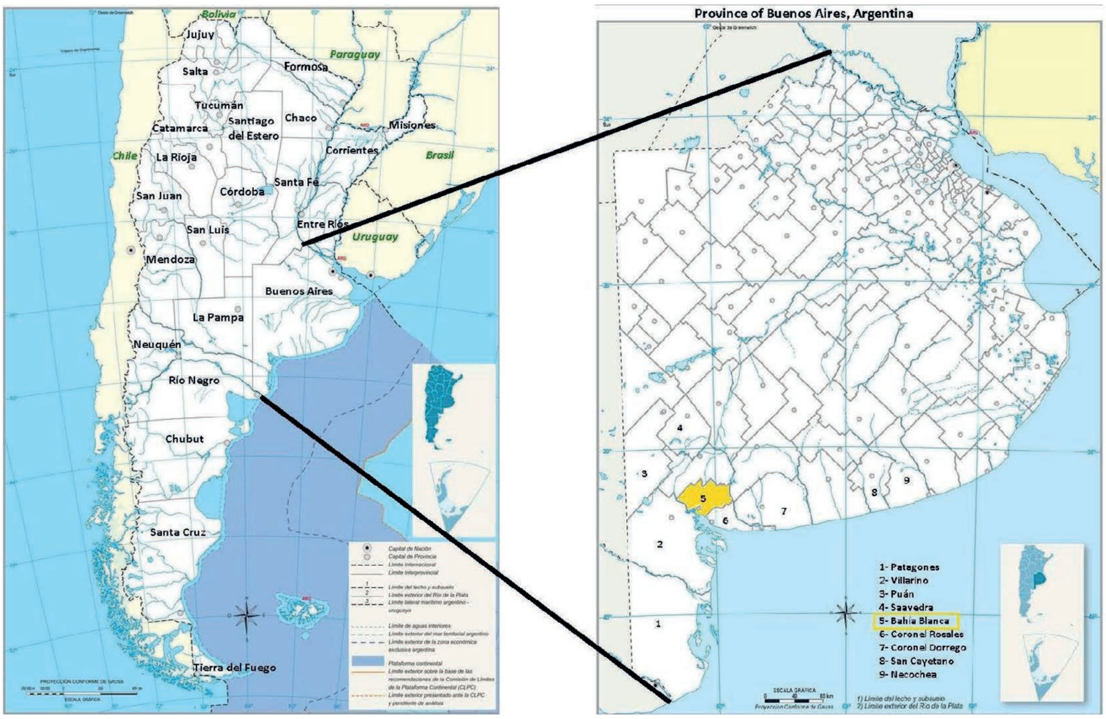

Fig. 1. Localization of the olive orchard where the study was conducted. Argentina, Province of Buenos Aires, and district of Bahía Blanca indicated with a shaded area in yellow.

Fig. 1. Localización del olivar donde se realizó el estudio. Argentina, Provincia de Buenos Aires, y partido de Bahía Blanca indicado con un área sombreada en amarillo.

region. Its agropecuarian productivity is lower than the rest of the Pampa region as a result of the prevailing agroecological conditions (Cincunegui *et al.*, 2019). Such region is ecologically suitable for the olive culture, which contributes to reduce the advancement of the desertification in the region (Elías & Barbero, 2017). It is important to highlight that the region has competitive advantages since it possess the port of greater depth in the country (Puerto Ingeniero White), adequate transporting systems, and the provision of associated services necessary for the commercialization and general development of the activity (Cincunegui *et al.*, 2019).

A large part of the producers of olive groves in the southwest of Buenos Aires use foliar fertilization with macronutrients (N, P and K) as the only method to increase the production of olive groves. However, this type of application is only recommended for micronutrients such as, B, Fe, Mn, among others. In addition, several foliar applications should be made throughout the cycle taking into account that: (1) foliar fertilization depends on the quantities in which the nutrients are necessary for the olive tree (if it is high, their absorption must be via roots), (2) the very low absorption efficiency of fertilizers through the leaves due to the presence of pubescent undersides, and (3) the large amounts of fertilizer that are not absorbed due to dripping, evaporation, drifting, etc. Many of these regional producers do not make more than two or three foliar applications annually, assuming that the crop is well nourished. In this framework, more efficient fertilization methods should be found, not only incorporating inorganic nutrients to the olive grove but also providing organic matter to the soils where this species is grown. This is important for the current fertility (inorganic nutrients, immediately available), but also for the potential fertility (inorganic nutrients slowly released by organic matter) of the soil.

Farmer's interest into cultivating cover crops has increased due to numerous benefits of this conservation practices (Sánchez *et al.*, 2007; Oliveira *et al.*, 2019). Cover crops are defined as non-cash crops that can be grown before or together with the main crops to keep the soil covered with vegetation for as long as possible—even all year round (Melander *et al.*, 2005) and generate numerous positive effects on the soil system (Sánchez *et al.*, 2007; Blanco-Canqui *et al.*, 2015; Cutti *et al.*, 2016; Wallace *et al.*, 2017; Rizzardi & Silva, 2006).

Cover crops can be introduced into plant production systems by cultivation during the off-season and destruction before the main crop cultivation, or by cultivation alongside the main crop, serving as living mulch for part or all of its growing season. The first is a common practice in annual growing systems, and the second in perennial growing systems (Lemessa & Wakjira, 2015). The most common cover crops used in farming belong to Fabaceae, Brassicaceae, and Poaceae families. They play a different role in the main crop due to variations in their growth rate, amount of biomass produced, uniformity of the soil coverage and C/N ratio.

Das *et al.* (2020) reported that the continuous use of green manures enhanced the organic matter content and supplemented the nutrient pool of the soil, which ultimately, improves the soil's physical, chemical and biological properties. They also emphasized that the beneficial effects of green manures on succeeding crops depends largely on residue quantity and quality, soil type, soil fertility, soil acidity, biological activity, and soil moisture and temperature. Sarwar *et al.* (2010) informed that use of green compost increased soil pH, enhanced the percentage of organic matter, N and C generating a net decrease in the C/N ratio, and increased the concentrations of P and K in the soil.

Abdennbi *et al.* (2023) showed that long-term olive mill wastewater application to the soil surface in an olive orchard at 50 m^3 ha^{-1} satisfied plant P, K and N requirements. Similarly, Busso & Busso (2022) reviewed the effects of different fertilization sources (such as organic fertilizers applied to the soil and green manures and inorganic fertilizers applied to the plant leaves and the soil) on various parameters in *Olea europaea* L, indicating that applications of olive-oil extraction by-products to soils could lead to important mid-to -long-term agro-environmental benefits.

The objective of this study was to determine the effects of the four fertilization treatments (1) organic fertilization with commercial fertilizer ("bioorganutsa") applied to the soil; inorganic fertilization with macronutrients (N, P and K) applied to the (2) leaves or (3) the soil, and (4) non-fertilization (control). on areas either with or without a nitrogen-fixing grass-legume consociated cover crop formed by *Avena sativa* L. and *Vicia benghalensis* L. on several soil chemical parameters (i.e., pH, and the concentrations of soil organic matter -SOM-, soil ammonia, nitrate, and extractable P and K) in an olive orchard at southwestern Buenos Aires province, Argentina in order to obtain alternative ways of fertilization that improve soil properties.

MATERIALS AND METHODS

Study site

The study was conducted during the growing cycle 2020/2021 in the District of Bahía Blanca, Province of Buenos Aires (38º34'S, 61º59'W; Fig. 1) in an olive orchard under irrigation pertaining to the private agropecuarian establishment of "Nobles Caciques". The mean annual precipitation (period 1896-2000) is 584.6 mm, and the mean annual temperature (period 1971-1990) is 15 ºC. The lowest mean monthly temperatures are produced in July (7.5°C) and the greatest ones in January (23.2°C). The absolute maximum and minimum registered temperatures are 43.8 and -11.8°C, respectively. Soils are classified as Petrocalcic Paleustols (Blanco *et al.*, 2003).

Drip irrigation system

A super-intensive orchard requires 1000 mm/year of water (rainfall + irrigation). Thereafter, irrigation was complementary to the precipitations. In the study region, annual precipitations are about 600 mm, so irrigation supplies 400 mm/year. Of these 400 mm, 200 mm were applied during summer (irrigation was about 3mm/day). Irrigation was suspended about 3 weeks before the beginning of autumn, at least one month before the olive harvest, to avoid the formation of watery fruits (with low oil concentration). Another 100 mm of irrigation were applied towards the end of autumn and during winter, and the remaining 100 mm were applied at the end of spring.

Experimental design

The general experimental design was subsplit plots (Fig. 2). The major experimental units were 6 large plots (48 m length x 4 m width each) where the 2 vegetation cover levels (with cover, without cover) were applied (three plots with cover crops and other three without cover crop) (Fig. 2). The secondary experimental units were 4 subplots (6 m length x 4 m width each) within each large plot where the 4 fertilization treatments were applied (1 treatment/subplot) (Fig. 2). Spacing among subplots within any large plot was 6 m length x 4 m width to avoid edge effects. The large plots, subplots and the spacing among subplots were not made at scale in Fig. 2. The

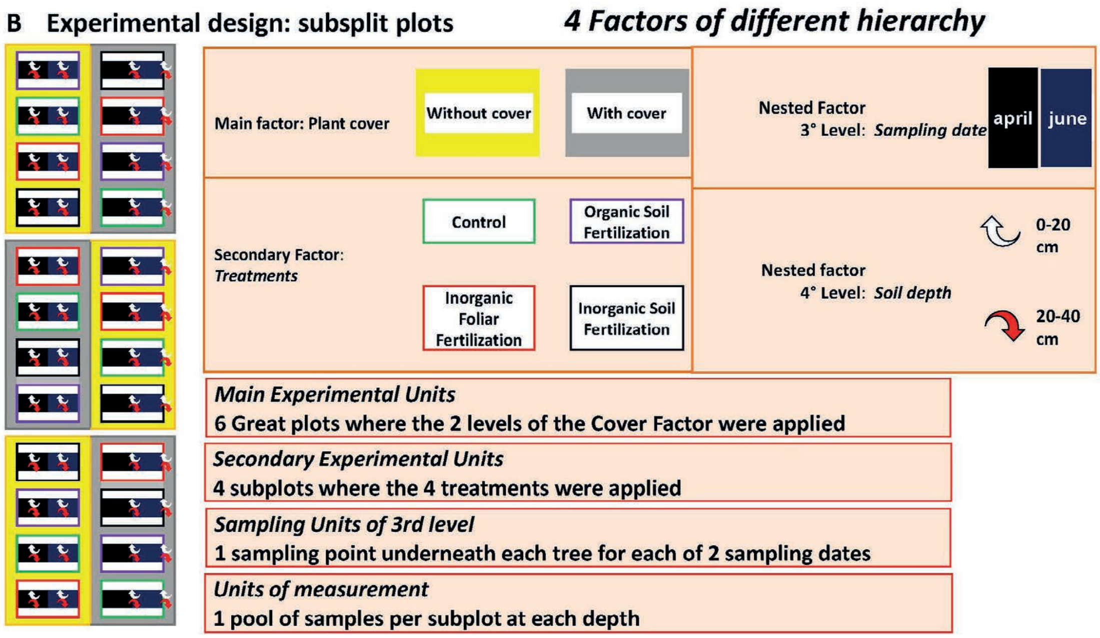

Fig. 2. General experimental design. It was subsplit plots. The major experimental units were 6 large plots with 2 vegetation cover levels applied: with cover or without cover. The secondary experimental units were 4 subplots within each large plot where 4 fertilization treatments were applied [1 treatment/subplot; the treatments where: control; a commercial organic fertilizer ''bioorganutsa'' applied to the soil, and inorganic fertilizers applied to the leaves or to the soil]. There were 8 trees within each subplot in all 6 large plots. The large plots, subplots and the spacing among subplots were not made at scale in this figure (please see text of the manuscript under Experimental Design to obtain this information). Each subplot was sampled in April and June 2021 at 0-20 and 20-40 cm of soil depth.

Fig. 2. Diseño experimental general. Fue parcelas subdivididas. Las unidades experimentales principales fueron 6 parcelas grandes donde se aplicaron los 2 niveles de cobertura de la vegetación: con cobertura o sin cobertura. Las unidades experimentales secundarias fueron 4 subparcelas dentro de cada parcela grande; en las mismas se aplicaron los 4 tratamientos de fertilización [1 tratamiento/subparcela; tratamientos: control; fertilizante orgánico comercial ''bioorganutsa'' aplicado al suelo; y fertilizantes inorgánicos aplicados a las hojas o el suelo.]. Hubo 8 árboles en cada subparcela de las 6 parcelas grandes. Las parcelas grandes, las subparcelas y el espaciamiento entre subparcelas no se hicieron a escala en esta figura (por favor ver el texto del manuscrito bajo Diseño Experimental para obtener esta información). Cada subparcela fue muestreada en Abril y Junio 2021 a 0-20 y 20-40 cm de profundidad del suelo.

nested experimental subunits contained 8 trees per subplot. A total of 192 trees [8 trees/subplot x 24 subplots = 192 trees] were selected randomly distributed in 2 rows of 4 trees each on each subplot. The sampling units of the 3rd level consisted of 1 sampling point underneath each tree at 2 different depths, 0.20 and 20-40 cm from the soil surface. Two sampling dates in April and June 2021 were evaluated. The unit of measurement was a compound pool of the 8 samples per subplot at each of 2 studied depths (0.20 and 20-40 cm from the soil surface) (Fig. 2). Three replicates per fertilization treatment in each plot with or without a vegetation cover were utilized (Fig. 2).

Cover crops

A mixture of *V. benghalensis* with *Avena sativa* was implanted around each subplot on 3 out of the 6 plots (Figs. 2, 3). *Vicia benghalensis* was utilized as the main culture and *A. sativa* as a support culture. *Vicia benghalensis* was seeded to a density such that it allowed to obtain 90 plants m^{-2}; while seeding of *A. sativa* only allowed to obtain 30 plants m^{-2}, avoiding the interference with *Vicia* (Renzi, 2009). Seeding was made in May of the first studied year and thereafter the reseeding was natural. A sowing machine to steady flow with mono disk ploughs in lines separated to 20 cm, locating the seeds to a soil depth between 3 and 5 cm was used. The amounts of seed utilized to obtain the proposed plant densities were 57.1 kg ha^{-1} for *V. benghalensis* and 36.95 kg ha^{-1} for *A. sativa*. The seeds of *V. benghalensis* were inoculated with a commercial starter contining *Rhizobium leguminosarum* pv *viciae*. The inoculum, on the base of sterile peat, was adhered to the seeds of *V. benghalensis* previous to the seeding through a sugar solution (Renzi, 2009).

The second cover condition consisted of leaving the soil without vegetation around each subplot on the other 3 out of the 6 plots (Figs. 2, 3) using an eccentric harrow with roll and subsequent applications of herbicides. The ridges were maintained without weeds, through chemical control with herbicides.

Soil Fertilization

Soil fertilization (Fig. 2) was made on October 28th, 2020 using ''Bioorganutsa'' or a mixture of 3 inorganic fertilizers. ''Bioorganutsa'' is a fertilizer brand developed for organic agriculture. It is composed by goat manure, mixtures of guano, blood flour, sunflower peel ashes, natural phosphorous and calcium components. Its chemical characteristics are listed in Tables 1 and 2. This amendment meets all requirements to be utilized in organic production (Daasons, 2013). It was applied at 1,778 kg ha^{-1}.

About the inorganic fertilization, urea [$CO(NH_2)_2$] was used as a source of N, diammonium phosphate [$(NH4)_2HPO_4$; PDA], as a source of N and P and potassium nitrate (KNO_3) as a source of N and K. To make the edaphic fertilization, a mixture of urea, PDA and KNO_3 (21:7:28 w/w/w, Table 2) was applied at 758 kg ha^{-1}. This fertilization was made in the soil located underneath the tree canopy, incorporating the fertilizer to the soil immediately following its application. This was to avoid mainly the volatilization of the urea (Sierra Bernal, 2009; Rosati *et al.*, 2015).

The degree of a fertilizer is the amount (gr) of the chemical element/100 gr of fertilizer (i.e., 46 gr N/100 gr urea: Table 2). The urea, diammonium phosphate and potassium nitrate already come formulated with such a degree from the factory (Table 2), and these are the ones which compound the mixture of the inorganic fertilizer applied to the soil which has a degree 3:1:4 (Table 2).

Table 1. Chemical composition of “Bioorganutsa”.
Tabla 1. Composición química de “Bioorganutsa”.

EC (dS/m)	pH	N_t (%)	P_t (%)	K (%)	C_t (%)	Ca (%)	Mg (%)	Na (%)	S (%)	Fe (%)
2.9	7.8	1.27	0.35	0.81	13.38	2.52	0.87	0.14	0.24	1.83

References: EC, Electrical conductivity; t, total.
Referencias: EC, conductividad eléctrica; t, total.

Table 2. Degree and experimental use of fertilizers employed in the mixtures. OF: Organic soil fertilization, FF: Inorganic leaf fertilization, IF: Inorganic soil fertilization.

Tabla 2. Grado y uso experimental de los fertilizantes usados en las mezclas. OF: Fertilización orgánica del suelo, FF: Fertilización inorgánica foliar, IF: Fertilización inorgánica del suelo.

Fertilizers	Degree In the relationship between N, P and K			Experimental use
	N	P (P_2O_5)	K (K_2O)	
Urea	46	0	0	IF, FF
Diammonium phosphate	18	46	0	IF
Potassium nitrate	13	0	45	IF, FF
“Bioorganutsa”	9	3	12	OF

Foliar Fertilization

Foliar fertilization required the mixture of urea, phosphoric acid (concentrated at 85%), potassium nitrate, and a non-ionic surfactant at 0.1% (v/v) to promote foliar absorption (Table 2). Preparation of the fertilizer for foliar application consisted in dissolving each fertilizer separately in water according to its solubility (Guerrero Riascos, 2004). Thereafter, each solution was filtered to remove impurities and avoid the possible obstruction of the pulverization tablets. The three solutions were then mixed and the surfactant agent was added. Pulverizations of 11.6 L per plant of the mixture solution, fractioned on 5 applications of 2.3 L per application, were made with the objective of applying quantities equivalent to those used in edaphic fertilization. The volume of broth for application that each plant can maintain on its canopy for absorption was calculated following Hidalgo Moya *et al*. (2020). Foliar fertilizations were made at the stages of sprouting (September), pre-flowering (October), fruit growth and filling (December), hardening of the stone (January) and olive color change (February). These fertilizations were made using a pressure sprayer (9 liters capacity) with a tow bar of adjustable peak.

Soil sampling

To determine physicochemical properties of the soil under the different treatments, soil samples were take at 0.20 and 20-40 cm soil depth using a soil auger. The major part of the root system is located between 0-20 to 20-40 cm, near the wet bulb generated by the drip irrigation system (Medina Bedoya, 2013). Soil samples were taken on each treatment underneath the

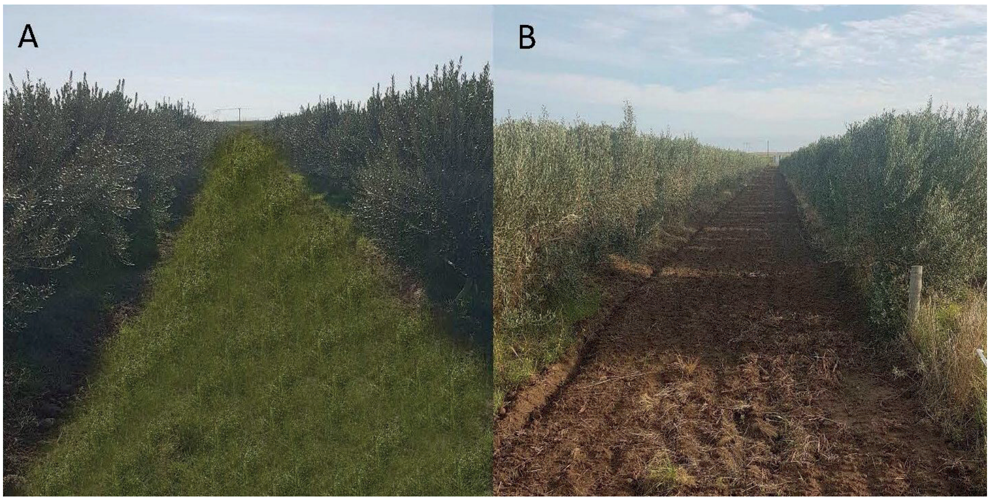

Fig. 3. Olive plots with a cover crop (A) or without a cover crop (B).
Fig. 3. Parcelas de olivos con cultivo de cobertura (A) o sin cultivo de cobertura (B).

canopy, considering as control the soil from olive trees which were neither fertilized nor seeded with a cover crop. The unit of measurement was a mixture pool of 8 samples/subplot/treatment taken on each of the three replicate points. These samples were taken on April 19th and June 28th, 2021.

In the laboratory, soil samples were air dried and then screened through a 2 mm screen mesh. The following edaphic properties of fertility were analyzed: pH [relationship soil: water (1:2,5)]; organic matter following Davies (1974); ammonium and nitrate following the Kjeldahl method (Bremner & Mulvaney, 1982); extractable P (Bray & Kurtz, 1945) and exchangeable K through the extraction with ammonium acetate 1N pH 7 (Jackson, 1964).

Statistical analysis

A FOUR -WAY ANOVA table (2 green cover types x 4 fertilization treatments x 2 soil depths x 2 dates) was conducted for each factor. Two-way interactions were open whenever they were significant. When F tests were significant, means were compared using the LSD test of Fisher at a significant level of 5%. See the Appendix for more details.

RESULTS

pH

There were interactions that involved the different factors: fertilization treatment x soil depth, and cover x fertilization treatment x date. However, the effects of treatments were very important, about 60% of the model fixed by the researcher. This percentage is called Eta squared and it is similar to

R^2 when using regression analysis. It is the sum of squares of any variation source (i.e., treatments in this case) divided by the sum of all sum squares in the ANOVA Table, including all major effects and the interactions (the error sum of squares is not used when making this ratio). This meant that the results on average were not highly affected by those interactions. Because of this, the treatment means were compared using LSD. The highest ($p<0.05$) pHs were found in the control and foliar fertilization treatments, and the lowest ($p<0.05$) pHs were shown by the organic and inorganic soil fertilizations (Fig. 4).

Due to the existing interactions that involve all factors, fertilization treatments were compared for each combination of the levels of the different factors, including, with or without cover crops, different sampling dates and soil depths, separately (Table 3). In 6 out of 8 comparisons, the highest ($p<0.05$) pH was found in the control with soil cover (Table 3). Also, in 5 of 8 comparisons, the lowest ($p<0.05$) pH was found in the treatment with organic soil fertilization.

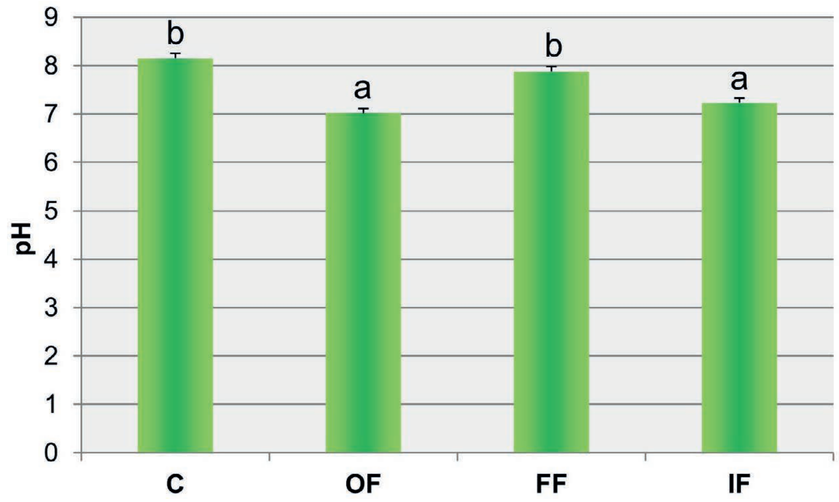

Fig. 4. Soil pH in each of the treatments as follows: C, control without fertilization; OF, organic soil fertilization; FF, inorganic foliar fertilization; IF, inorganic soil fertilization. Histograms followed by different letters are significantly different at $p <0.05$. Each histogram is the mean+1 S.E. of 24 replicates.

Fig. 4. pH del suelo en cada uno de los tratamientos como sigue: C, Control; OF, Fertilización del suelo orgánica; FF, Fertilización foliar inorgánica; IF, Fertilización del suelo inorgánica. Los histogramas seguidos por letras diferentes son significativamente diferentes a $p <0.05$. Cada histograma es el promedio +1E.E. de 24 réplicas.

Table 3. pH-comparison for each fertilization treatment combined with the other levels of the factors under analysis (sampling date, soil depth, plant cover). Different letters within the same column indicate significant differences ($p<0.05$) among them. Each value of soil pH is the average of n=3.

Tabla 3. Comparación de pH para cada tratamiento de fertilización, en combinación con diferentes niveles de los otros factores bajo análisis (fecha de muestreo, profundidad del suelo, cobertura vegetal). Letras diferentes dentro de una misma columna indican diferencias significativas ($p<0.05$) entre ellas. Cada valor de pH del suelo es el promedio de n=3.

Treatment/ Date	April				June			
Depth	0-20 cm		20-40 cm		0-20 cm		20-40 cm	
Soil cover	With	Without	With	Without	With	Without	With	Without
Control	8.60c	8.01b	8.04bc	7.41a	8.36c	8.14b	8.0b	8.10b
OF	7.46b	6.73a	7.09 a	6.88a	6.70a	7.49b	6.71a	7.02a
FF	8.11bc	7.54b	8.37c	7.67ab	7.59bc	7.94b	7.85b	7.94b
IF	6.59a	7.53ab	7.50ab	7.86b	7.39ab	6.33a	7.86b	6.75a

Organic matter

No interactions ($p>0.05$) were found in the new ANOVA table. Because of this the factors can be tested on average. There were significant differences between dates ($p=0.0000$) and soil depths ($p=0.0002$) in the new ANOVA Table. The organic matter percentage was greater ($p<0.05$) in June than in April, and at 0-20 cm than 20-40 cm soil depth (Fig. 5). The mean values (n=12) of organic matter percentage in the different treatments in the areas with or without green cover ranged between 2.70% (with green cover, inorganic foliar fertilization) and 2.87% (without green cover, organic soil fertilization).

Ammonium

There were neither significant interactions nor significant differences between any of the major factors considered in the study regarding ammonium content in the soil. As a result, the mean values (n=12) of ammonium in the different treatments in the areas with or without green cover ranged between 3.53 ‰ (with green cover, control) and 10.94 ‰ (without green cover, inorganic foliar fertilization).

Nitrate

There was a three-way interaction (cover x treatments x dates; $p=0.0113$) affecting nitrate concentrations. There was an interaction ($p=0.0062$) between cover crops x fertilization treatments. There were no interactions ($p>0.05$) involving soil depths; therefore, these data were averaged to analyze the remaining factors. The fertilization treatments were compared within with cover crops, on average for both sampling dates. Within the areas with plant cover, the lowest ($p<0.05$) nitrate concentrations were found in the soil cover control without fertilizers, inorganic foliar fertiliza-

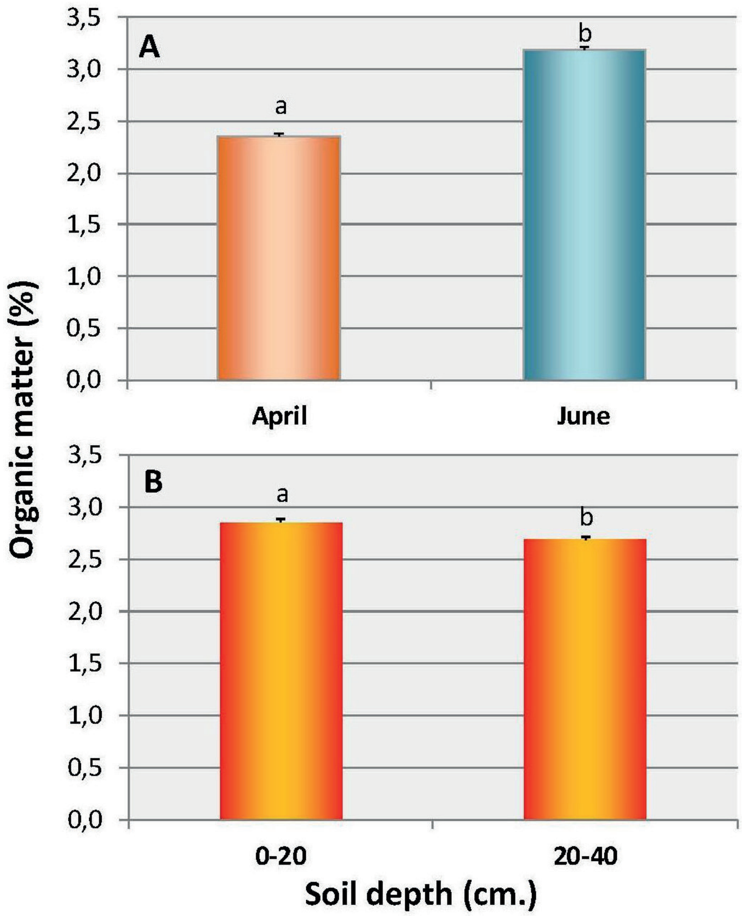

Fig. 5. Organic matter (%) in April and June, and at 0-20 and 20-40 cm soil depth. Different lowercase letters above histograms for sampling dates or soil depths indicate significant differences at $p<0.05$. Each histogram is the mean + 1 S.E. of n= 48 for the two sampling dates and soil depths.

Fig. 5. Materia orgánica (%) en Abril y Junio, y a 0-20 y 20-40 cm de profundidad del suelo. Letras minúsculas diferentes encima de los histogramas para las fechas de muestreo o profundidades del suelo indican diferencias significativas a $p<0.05$. Cada histograma es el promedio + 1 E.E. de n= 48 para las dos fechas de muestreo y profundidades del suelo.

tion and organic soil fertilization (Fig. 6); the highest ($p<0.05$) value was shown with inorganic soil fertilization, which was similar ($p>0.05$) to the organic soil fertilization (Fig. 6).

For the subplots without cover crop, treatments were compared for each date separately. In April 2021, the lowest ($p<0.05$) values of nitrate concentrations were found in the control without fertilizer and without cover crop, and in the inorganic foliar and inorganic soil fertilization treatments, while the highest ($p<0.05$) values were determined in the organic soil fertilization treatments (Fig. 6). In June 2021, the lowest ($p<0.05$) values were shown in the organic soil and inorganic soil fertilization treatments, and the highest ($p<0.05$) values in the control and inorganic foliar fertilization treatments (Fig. 6).

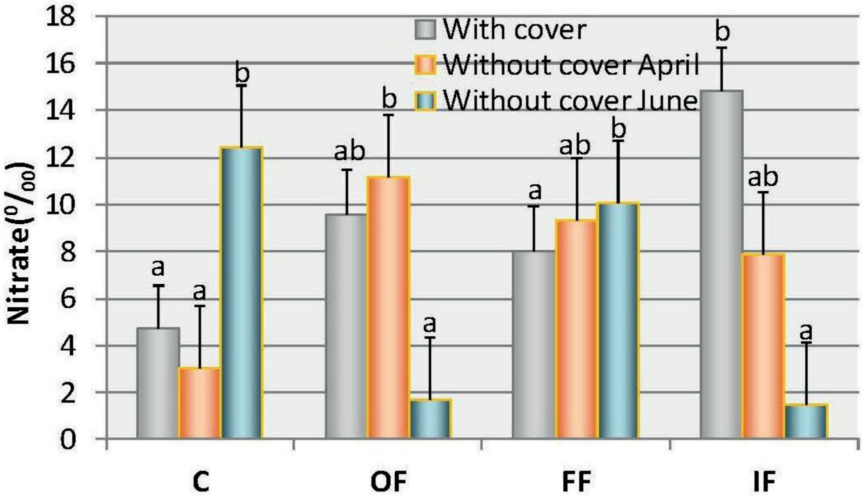

Fig. 6. Soil nitrate concentrations (‰) on each of the studied treatments. Treatments were as follows: C, control without fertilization; OF, organic soil fertilization; FF, inorganic foliar fertilization; IF, inorganic soil fertilization. Each histogram is the mean + 1 S.E. Different letters above histograms on the figure legends either With (on average for both dates; n=12) or Without cover in April or June 2021 (n=6) indicate significant differences ($p<0.05$).

Fig. 6. Concentraciones de nitrato del suelo (‰) en cada uno de los tratamientos estudiados en Abril y Junio 2021, con cultivos de cobertura (promedio para ambas fechas; n=12) y sin cultivo de cobertura (n=6, respectivamente para cada fecha). Los tratamientos fueron: C, control sin fertilización; OF, fertilización orgánica del suelo; FF, fertilización inorgánica foliar; IF, fertilización inorgánica del suelo. Cada histograma es el promedio + 1 E.E. Letras diferentes sobre los histogramas en las leyendas indican diferencias significativas ($p<0.05$).

Phosphorus

In terms of P concentration, there were interactions which involved the fertilization treatments, dates and soil depths ($p= 0.0183$). The effect of the depths was quite important, and that of the dates was not negligible. Because of this the average of each of these factors was graphed (Fig. 7).

It appeared to exist a greater (although not significant) average concentration of P in June than in April 2021 (Fig. 7). It also appeared to exist a greater (although not significant) P concentration at 0-20 than at 20-40 cm soil depth (Fig. 7).

There was also a two-way significant interaction (treatment x depth, $p=0.0011$). Within 0-20 cm soil depth, the greater ($p<0.05$) P concentrations were found in the organic and inorganic soil fertilization treatments (Fig. 8). At this soil depth, the lower ($p<0.05$) P concentrations were determined in the control and inorganic foliar fertilization treatments (Fig. 8). Within 20-40 cm soil depth, the greatest and lowest ($p<0.05$) P concentrations were found in the control without fertilization and the inorganic soil fertilization treatments, respectively (Fig. 8).

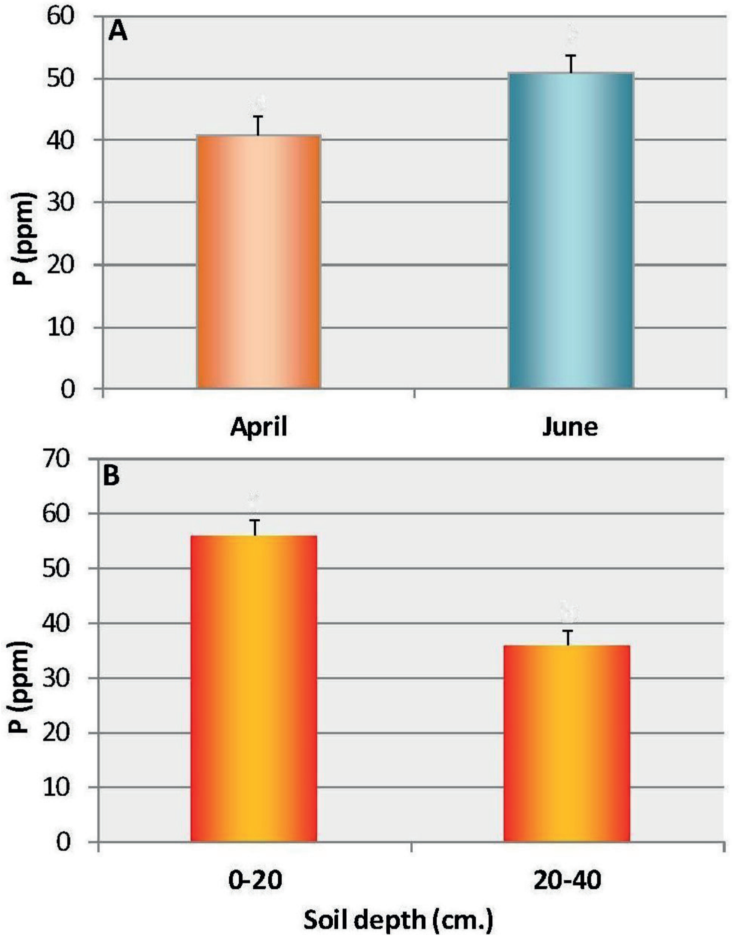

Fig. 7. Phosphorous concentration in April and June (A) and at 0-20 and 20-40 cm soil depth (B). Each histogram is the mean + 1 S.E. of n=48.

Fig. 7. Concentración de P en Abril y Junio (A) y a 0-20 y 20-40 cm de profundidad del suelo (B). Cada histograma es el promedio + 1 E.E. de n=48.

Potassium

For K concentrations, the effect of the fertilization treatments was quite important (32,86 % of the model fixed by the researcher) (Fig. 9A). There was a greatest ($p<0.05$) K concentration in the organic soil fertilization than in the control and the other inorganic fertilizers (Fig. 9A). No important interactions were found involving soil depths. However, the main factor soil depth was highly significant ($p= 0.0001$) in the new general ANOVA Table (see Appendix). Potassium concentration was greater ($p<0.05$) at 0-20 than at 20-40 cm soil depth (Fig. 9B).

There was an interaction between crop cover x fertilization treatments ($p=0.0206$). In the subplots with cover crop, the greater ($p<0.05$) K concentrations were found in the treatments with organic and inorganic soil fertilizations (Fig. 10). While the lowest ($p<0.05$) K concentrations were shown in the control with cover crop and with inorganic foliar fertilization (Fig. 10). In the areas without a cover crop, the greatest ($p<0.05$) K

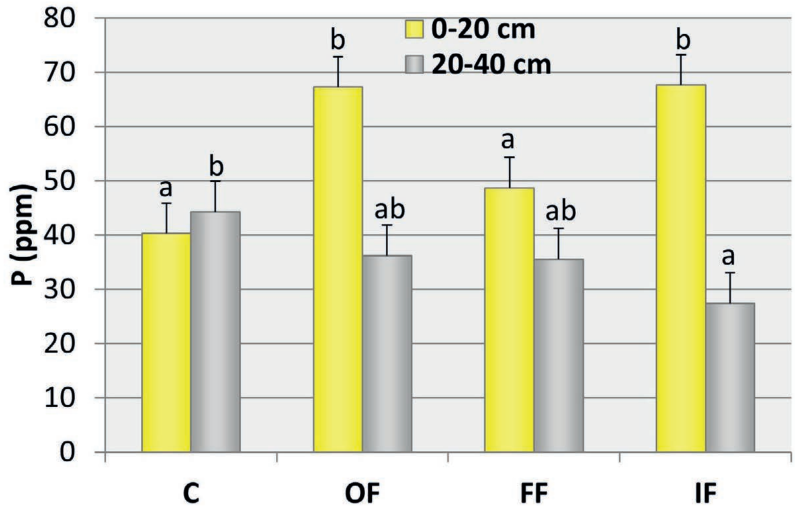

Fig. 8. Phosphorous concentration (ppm) at each studied soil depth (0-20 and 20-40 cm) in the different fertilization treatments. Treatments were as follows: C, Control; OF, organic soil fertilization; FF, inorganic foliar fertilization; IF, inorganic soil fertilization. Within each soil depth, different letters indicate significant differences (p<0.05) among treatments. Each histogram is the mean + 1 S.E. of n=12.

Fig. 8. Concentración de P (ppm) en cada profundidad del suelo estudiada (0-20 y 20-40 cm) en los distintos tratamientos. Los tratamientos fueron: C, control; OF, fertilización orgánica del suelo; FF, fertilización inorgánica foliar; IF, fertilización inorgánica del suelo. Dentro de cada profundidad del suelo, letras diferentes indican diferencias significativas (p<0.05) entre tratamientos. Cada histograma es el promedio + 1 E.E. de n=12.

concentration was observed in the subplots with organic soil fertilization, meanwhile, the lowest ($P<0.05$) K concentrations were observed in the control, and in the treatments with inorganic fertilization, considering both, foliar and soil application (Fig. 10).

DISCUSSION AND CONCLUSION

Cover cropping is a soil conservationist practice that can improve soil conditions between tree rows in orchards (Koudahe *et al.*, 2022). The positive effects of cover crops on agroecosystems include reductions in soil erosion and nitrate leaching, increases of water infiltration into the soil and maintenance of soil moisture. They also suppress weeds and reduce the occurrence of pests, nematodes and various soil pathogens, and improvements in soil quality and health by increasing the content of organic matter and the availability of nutrients (Sánchez *et al.*, 2007; Blanco-Canqui *et al.*, 2015; Duan *et al.*, 2020; Silva *et al.*, 2021). In agreement with this, we found that the use of cover crops increased the soil nitrate concentrations when the inorganic soil fertilization treatment was applied. Other studies have informed that elimination of spontaneous vegetation in olive orchards is an undesirable management system (Keesstra *et al.*, 2016). These authors showed that usage of spontaneous vegetation as cover crops is a good alternative to improve soil properties. Also, use of organic manures and cover

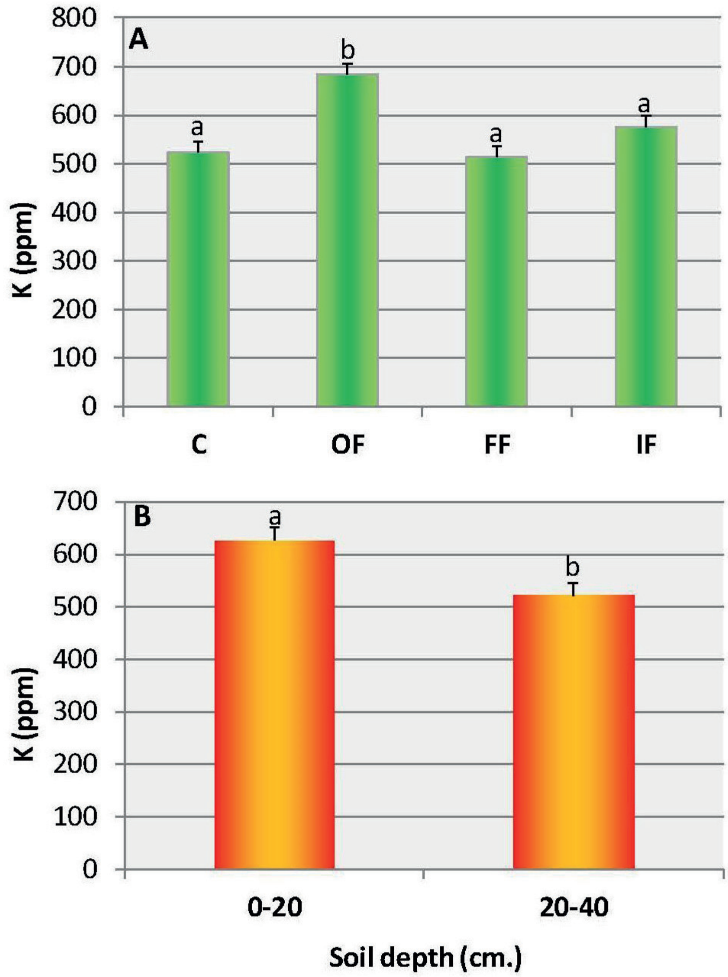

Fig. 9. Potassium concentration (ppm) in the different treatments (A; n=24) and in the two studied soil depths (B; n=48). Treatments were as follows: C, Control; OF, organic soil fertilization; FF, inorganic foliar fertilization; IF, inorganic soil fertilization. The two studied soil depths were 0-20 and 20-40 cm below the soil surface. Different lowercase letters above histograms on (A) and (B) indicate significant differences ($p<0.05$) among treatments or soil depths, respectively. Each histogram is the mean + 1 S.E.

Fig. 9. Concentración de K (ppm) en los diferentes tratamientos (A; n=24) y en las dos profundidades del suelo estudiadas (B; n=48). Treatments were as follows: C, Control; OF, organic soil fertilization; FF, inorganic foliar fertilization; IF, inorganic soil fertilization. Las dos profundidades del suelo estudiadas fueron 0-20 y 0-40 cm debajo de la superficie del suelo. Letras minúsculas diferentes encima de los histogramas en (A) y (B) indican diferencias significativas ($p<0.05$) entre tratamientos o profundidades del suelo, respectivamente. Cada histograma es el promedio + 1 E.E.

crops contribute to reduce the air, water and soil pollution that the use of inorganic fertilizers may cause (Savci, 2012). An efficient tool for monitoring the soil management systems can be the evaluation of the long-term impact of cover crops and type of aggregates to the soil on its chemical properties. On this regard, Aranda *et al.* (2015) reported that after 17 years of organic management with application of olive-mill pomace co-compost, olive grove soils were of higher quality than those with conventional management where no co-compost had been applied. They found that the main studied soil chemical parameters significantly increased in soils treated with

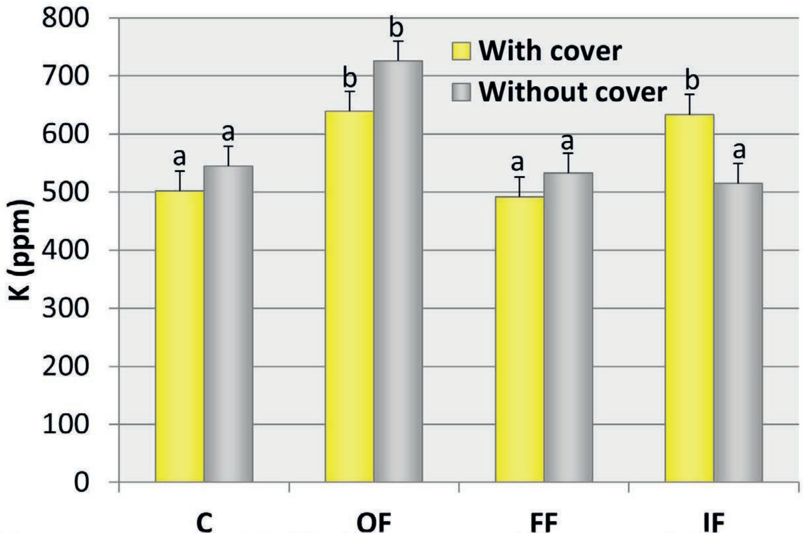

Fig. 10. Potassium concentrations (ppm) among fertilization treatments within each soil cover treatment. Treatments were as follows: C, Control; OF, organic soil fertilization; FF, inorganic foliar fertilization; IF, inorganic soil fertilization. Different lowercase letters above histograms indicate significant differences ($p<0.05$) among treatments within each type of soil cover. Each histogram is the mean + 1 S.E. of n=12.

Fig. 10. Concentraciones de K (ppm) entre tratamientos de fertilización dentro de cada tratamiento de cobertura de suelo. Los tratamientos fueron: C, Control; OF, fertilización del suelo orgánica; FF, fertilización foliar inorgánica; IF, fertilización del suelo inorgánica. Letras minúsculas diferentes encima de los histogramas indican diferencias significativas ($p<0.05$) entre los tratamientos dentro de cada tipo de cobertura del suelo. Cada histograma es el promedio + 1 E.E. de n=12.

organic amendment. Thus, application of olive-oil extraction by-products to soils could lead to important mid-to -long-term agro-environmental benefits such as the sustainability of those management practices. Use of organic manures and cover crops not only incorporate inorganic nutrients to the olive grove but also provide soil organic matter (SOM). This is important not only to have an increase of soil inorganic nutrients but also SOM that slowly releases inorganic nutrients to the soil. Zhang *et al.* (2021) demonstrated that fertilization with manure enhanced the stability of aggregates and aggregate-associated carbon by increasing the availability of soil nutrients. The stability of soil aggregates, considered to happen by cohesion and clustering of mineral particles and organic matter (Bucka *et al.*, 2019), is regarded as an indicator of aggregate formation, which is closely related to the organic carbon stability. For uniform coverage of the soil surface, it is critical that the species used as cover crops produce a substantial amount of biomass. In addition, their C/N ratio should be balanced, and they should be resistant to rapid decomposition, thus protecting the soil, even from the early stages of growth and development of the main crop (Cutti *et al.*, 2016; Wallace *et al.*, 2017). On the other hand, the exposure of the soil promotes weed infestation and increases erosion susceptibility, while high C/N can extract nitrogen from the system, reducing its availability to plants (Rizzardi & Silva, 2006).

Legume cover crops in general improve soil quality, providing more favorable conditions for the growth, development and yielding of main crops, and playing a significant role in reducing weed infestation (Blanco-Canqui *et al*., 2015; Elsalahy *et al*., 2019; Kocira *et al*., 2020). Tao *et al.* (2017) reported that four different green manure fertilization regimes with *Vicia benghalensis*, *V. sativa* L., *Astragalus* spp., and *Raphanus raphanistrum* subsp. *sativus* L. Domin, tested on Maize crops, reduced the soil pH, increased the concentrations of soil organic matter, available K and alkali solution N; and either increased or decreased the available P concentrations depending on the green manure used for fertilization. These authors also informed that soil pH, alkali solution N, and available K were the key environmental factors shaping soil bacterial communities (useful as indicators of soil quality) and determining maize yields. On the other hand, Hindersmann *et al*. (2023) reported that *Paspalum notatum* Flüggé, (Poaceae) when used as cover crop in olive orchards competed with olive trees for mineral forms of N available in the soil, resulting in decreased growth and N content in organs of the tested olive cultivars. This effect could be explained, at least in part, by the density of plants in a community, which determines the intensity of competition and the efficiency in exploiting available resources (Li *et al*., 2020). As plant density increases, competition among plants for soil water and nutrients in the belowground root systems is expected to intensify (Zhai *et al*., 2019).

The grass-legume consociated formed by *A. sativa* and *V. benghalensis* tested as cover crop, and the density selected for this work in an olive orchard, in general terms, showed interactions tending to increase the pH level and increase nitrate levels (particularly in the inorganic soil fertilization treatment -IF- with cover crop) independently of the sampling date and soil depth.

Similar to our results, Roussos *et al.* (2017) found that the application of the organic fertilizer 'Activit' significantly reduced the soil pH in comparison with the control. This reduction was probably due to the nitrification of ammonium and the production of organic acids (phenolic and carboxylic groups) during the decomposition of the organic materials (García-Ruiz *et al.*, 2012). Even more, the addition of organic matter increases the microbiological biomass and activity (Hernández *et al.*, 2014), resulting in an increased CO_2 production and consequently in soil pH reduction. Adekiya *et al.* (2019, 2020) also reported that the lower pH of organic amended soil compared with the control could be due to the fact that during microbial decomposition of the incorporated manures, organic acids may be released, neutralizing the alkalinity of the manures, thereby lowering the pH of the soil below their initial value. By the contrary, Wang *et al.* (2019) reported that long-term manure application increased soil pH and reduces soil acidification. These opposite results may be attributable to the nature of the soil in the study of Wang *et al.* (2019) which was an acidic soil, while in the present study was characterized as an alkaline soil.

Taken into account these observations an organic fertilizer may increase the pH of acidic soils and decrease the pH of alkaline soils, thus in future research adjustments of soil pH by organic materials should be paid more attention.

Fertilization with NPK exhibited the lowest soil pH as result of leaching of bases from the soil surface. Similar to our results, Adekiya *et al.* (2020) reported a reduced pH in soils fertilized with NPK inorganic fertilizer than in the control.

In agreement with our results, Roussos *et al.* (2017) and Gosling & Shepherd (2005) found that the application of the organic fertilizers 'Activit' and 'Agriobiosol', nor different types of fertilization respectively, did not cause significant differences in the levels of SOM in comparison with the control. Werner (1997) also found no significant differences in total organic C between two production systems: conventional and transitional organic orchard. The non remarkable changes in SOM registered by Roussos *et al.* (2017) and García-Ruiz *et al.* (2012) were attributed to the short trial period and to high summer temperatures at the studied sites which accelerated the organic matter decomposition. Similarly, Adekiya *et al.* (2020) did not find significant differences in the percentage of soil organic matter between the control and the inorganic soil fertilization treatments in their two studied years.

The upper soil layer (0–20 cm) always maintained a higher SOM than the soil layer at 20-40 cm depth under all treatment conditions, sampling dates and soil covering types. Sánchez *et al.* (2007) determined increases in SOM to the upper soil layer after six years of assays on areas seeded with various cover crops. These increases in the top soil were 31, 27.9, 23 and 18.6 g kg^{-1} in the areas covered with *Trifolium fragiferum* L, alfalfa/fescue, *Vicia sativa* and control treatments, respectively.

Regarding ammonium concentrations, no differences (overall mean = 6.82 ‰) were found neither when organic or inorganic fertilizers were applied to areas with or without a green herbaceous cover among rows of olive trees at any of the studied sampling dates or soil depths. The climatic conditions of the study site may have played a role in the mineralization of organic matter (Brunetto *et al.*, 2018) and decomposition of plant residues (Leon *et al.*, 2015), since temperature and rainfall distribution varied throughout the experimental period. The lack of significant differences among treatments in our study may also be related to the short-term duration of the study. Shelton *et al.* (2018) determined that on average, NH_4-N tended to be greater in both the inorganic (i.e., urea) and organic (a blended, pelletized, commercial animal byproduct fertilizer) fertilization treatments than in the unfertilized control of corn conservation agroecosystems in Kentucky, USA.; nonetheless, they informed that means comparisons failed to identify specific months during which soil NH_4-N significantly differed, finding only that urea N was marginally greater than the unfertilized treatment at the end of autumn.

We found that areas with a green herbaceous cover among rows of olive trees showed a significantly greater nitrate concentration when soil fertilization was inorganic than when inorganic fertilization was applied to the leaves and in the control. The concentration of soil nitrate in the organic soil fertilization, however, did not differ from the control, and in the inorganic foliar and soil fertilizations. Shelton *et al.* (2018), however, found that during the warm season, soil nitrate concentrations were greater in the organic fertilization treatment (a blended, pelletized, commercial animal byproduct fertilizer) than in the control on areas where *V. benghalensis* was used as a cover crop of corn conservation agroecosystems in Kentucky, USA. These authors also reported that soil nitrate concentrations were greater when soil N fertilization was inorganic (i.e., urea) than in the unfertilized control on areas where *Triticum aestivum* L. was used as a cover crop. Sánchez *et al.* (2007) fertilized tree trunks at 0.40 m distance with a commercially available organic N, P and K fertilizer, on areas seeded with various cover crops among tree rows and found that nitrate concentrations remained under 7.5 mg kg^{-1} during the winter months in all treatments, and increased in spring and summer. A sharp increase in soil nitrate, up to 100 ppm, was observed in late spring in areas seeded with *V. benghalensis* due to the rapid biomass decomposition after maturation of this species, concluding that even with the use of permanent cover crops, the addition of organic fertilizers is needed in order to sustain yield and tree vigor.

Similarly to our results, Beniaich *et al.* (2023) showed similar ($p>0.05$) available P concentrations at 0-5 cm soil depth when olive cultivation was intercropped with spontaneous vegetation than when olive cultivation was on bare soil or only bare soil in their first studied year. While Roussos *et al.* (2017) showed similar soil P concentrations at different sampling dates during the whole year, our results indicated not differences in P concentrations for different sampling dates.

We found that application of the organic and inorganic soil fertilizers determined a significantly increase in P concentration in relation to the control and the inorganic foliar fertilizer at 0-20 cm soil depth for both sampling dates and plant cover types. Similar results were obtained by Adekiya et. al. (2020) on okra (*Abelmoschus esculentus* L.), Roussos *et al.* (2017) and Diacono & Montemurro (2010) by the application of the organic soil fertilizer 'Activit' and manure. The application of organic material may increase P availability through the effective reduction of P sorption to the soil and increase of P solubility by the organic acids found in the organic amendment (Sanyal & De Datta, 1991).

We found that areas with a green herbaceous cover among rows of olive trees showed a significantly greater exchangeable K concentration when organic or inorganic soil fertilization was applied than when foliar fertilization was applied for both dates and soil depths. Beniaich *et al.* (2023) also found that the exchangeable K concentration at 0-5 cm soil depth was significantly greater in olive cultivation intercropped with spontaneous vegetation than olive cultivation on bare soil, and only bare soil, during two subsequent years of study.

Roussos *et al.* (2017) reported a significant increment in soil K concentration when the commercial organic fertilizer 'Activit' was use for early- and late-winter, and mid-spring. This is in agreement with the results of Adekiya *et al.* (2020) who reported higher soil K concentrations after applying various organic fertilizers. Our study areas without a cover crop showed a significant higher K concentrations in the soil when the fertilization was organic.

Iofrida *et al.* (2020) compared the socio-economic impacts of organic and conventional farming systems of the Italian olive growing. They evaluated two scenarios. The first one represented by conventional olive growing, characterized using chemical fertilizers, herbicides (glyphosate), pesticides (especially organophosphates), and mechanized soil management. The second one represented organic olive growing, which included the use of farmyard organic fertilizers, mechanical weeding, low impact pesticides and mechanized soil management. Their results showed that the organic olive growing scenario was the most economically feasible alternative because of the contribution of public subsidies and the competitive market prices for organic products that balanced the higher production costs. However, at the current market conditions and by excluding public subsidies, investments in both farming systems were not economically sustainable. According to various authors (Rodríguez Sousa *et al.*, 2019; Shackelford *et al.*, 2019; Castellano-Hinojosa & Strauss, 2020; Gómez & Soriano, 2020), the evaluation of soil indicators, the biological impact of cover crops, the effects of cover crops on multiple ecosystem services and the integration of socio-economical assessments must be assessed in future studies. It is also important to highlight the role of both innovation and collaboration on the implementation of sustainable practices.

Finally, a parallel study in the same orchard (Busso *et al.*, unpublished) evaluated the effects of the same fertilization treatments on areas with or without a cover crop on the olive oil quality (polyphenols, extinction coefficient UV_{270}, free fatty acids and concentrations of various fatty acids), not finding significant differences ($p>0.05$) neither among fertilization treatments, in areas with or without a plant cover, nor different sampling dates.

In conclusion, this study, though limited to a single growing season, demonstrated that soil nutrient concentrations were significantly higher in organic fertilization treatments compared to inorganic ones, and in areas with cover crops, including legumes. The immediate advantage for farmers using organic manures and cover crops lies in their dual role: they supply essential inorganic nutrients and enhance soil organic matter content, which gradually releases additional inorganic nutrients. Future long-term studies are necessary to evaluate whether continuous application of organic amendments and cover crops will lead to sustained increases in soil organic matter and nutrient concentrations compared to inorganic fertilization in this region.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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APPENDIX

Statistical analysis

In the general table of variance (FOUR-WAY ANOVA= 2 green cover types x 4 treatments x 2 sampling dates x 2 soil depths) for soil pH, organic matter (%), ammonium (‰), nitrate (‰), and P and K (ppm), the tests for each factor (plant cover, treatment, date and soil depth) and those of the interactions should be tested with their corresponding error mean squares (MS). However, because all error mean squares resulted very similar in the general table of variance of each of the studied factors, it was tested if those error mean squares were statistically equal using the test of Bartlett. Since the error mean squares for all factors were statistically equal ($p>0.05$), a weighed mean square error was used for all factors with the total amounts of degree of freedom (=64). This weighed mean square error with their degree of freedom was used to make a new FOUR -WAY ANOVA table (2 green cover types x 4 treatments x 2 soil depths x 2 dates) for each factor.