

# Adapting service crop termination strategy in viticulture to increase soil ecosystem functions and limit competition with grapevine

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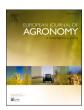


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## Adapting service crop termination strategy in viticulture to increase soil ecosystem functions and limit competition with grapevine

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#### ABSTRACT

Soil management and particularly service crops are a promising solution for addressing current challenges in viticulture as they limit the use of herbicides while increasing potential ecosystem services. Scientific literature barely considers the importance of service crop management to reach trade-offs between ecosystem services and disservices. This study evaluates during a three-year experiment, 6 service crop termination strategies for winter service crops, combining two service crops termination periods (early termination in February vs. termination at grapevine budburst) and three termination methods (mower (M), mower + tillage (T), roller-crimper (R)). Service crop (biomass, C:N ratio, weeds, and mulch following termination), soil (soil organic matter, microbial biomass, and water and nitrogen stocks), and grapevines (predawn leaf water potential, yield components,  $\delta^{13}$ C, yeast assimilable nitrogen in juice, and pruning weight) were monitored from 2019 to 2022. Allowing service crops to develop until the budburst of the vine resulted in a two to three-fold increase in biomass compared to early destruction. Termination involving soil tillage was the most effective method, as treatments destroyed early with soil tillage exhibited almost no regrowth. Soil tillage termination led to the lowest biomass of weeds at the grapevine flowering two years out of three, and prevented the regrowth of certain sown plants, especially Poaceae. The roller was less effective in destroying service crop species but was the best method for maintaining plant residues on the soil surface. A higher soil microbial biomass was observed with termination at budburst, combined with no-till termination methods. Termination involving soil tillage was notably more effective in stopping service crop transpiration, increasing soil water stocks and improving grapevine water status. In 2020 and 2022, soil inorganic nitrogen stocks were almost 4 times higher in the T treatments compared to the other two termination methods, achieving a level of approximately 61 kg ha<sup>-1</sup> that closely corresponds to the annual nitrogen requirements of grapevines. The yeast-assimilable nitrogen (YAN) content in grape juice mirrored this trend. Pruning weight varied significantly between different termination methods, with T treatments exhibiting a higher pruning weight per vine in comparison to R and M treatments. In general terms, the T treatment had a significantly higher number of bunches, the M treatment had the lowest, and the R treatment exhibited intermediate values. Overall, the average grapevine yield ranged from 7.25 to 13.7 t ha<sup>-1</sup>, corresponding to 52–98 hL  $ha^{-1}$  (with 4000 vines  $ha^{-1}$ , 140 kg  $hL^{-1}$ ). This level of production may be accepted for Protected Designations of Origin that limit grapevine yield to 40 or 60 hL ha<sup>-1</sup>, but it could be a limitation for Protected Geographical Indications, which permit 90 hL ha<sup>-1</sup>, or unlabeled productions without yield limitations. Given the Mediterranean climate context, with rising frequencies of dry winters due to climate change, termination involving soil tillage appears to be the least risky strategy to preserve grapevine vigor and production while improving soilbased ecosystem functions. However, this might be contingent on the targeted yield and wine valuation.

#### 1. Introduction

Viticulture faces major challenges including the reduction of phytosanitary products (Etienne et al., 2023; Fouillet et al., 2023, 2022), adaptation to and mitigation of climate change (Naulleau et al., 2021;

van Leeuwen et al., 2019), and the maintenance and improvement of soil quality (Coll et al., 2011; Salomé et al., 2016). Fungicides and herbicides, frequently used in viticulture (Fouillet et al., 2022; Jacquet et al., 2022; Urruty et al., 2016), are particularly under close watch due to increasing regulatory pressures from European countries, and source of

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public concern (Jacquet et al., 2022; Kudsk and Mathiassen, 2020; Lee et al., 2019). Furthermore, climate change necessitates the mobilization of various mitigation strategies such as the use of plant material adapted to changing conditions (Lamarque et al., 2023), and changes to agronomic practices like irrigation, canopy management, and soil management strategies in favor of soil carbon storage (Merot et al., 2019; Naulleau et al., 2021).

Soil management, and particularly service crops are a promising solution for addressing these challenges as they limit the use of herbicides while increasing potential ecosystem services. Service crops are herbaceous crops that are grown to provide non-marketed ecosystem services (Garcia et al., 2018), and have demonstrated the potential to mitigate climate change through the increase of soil organic matter and carbon storage (Abad et al., 2021a; Garcia et al., 2018; Griesser et al., 2022; Vicente-Vicente et al., 2016), reduce pesticide use by impacting weed suppression, pest biological control, and fungal diseases regulation (Beaumelle et al., 2021; Bernaschina et al., 2023; Jacometti et al., 2007; Tworkoski and Glenn, 2012; Valdés-Gómez et al., 2008). The role of service crops in soil water and nitrogen (N) dynamics is complex, with potential benefits such as improved water infiltration and increased nitrogen content (Capó-Baucà et al., 2019; Gaudin et al., 2010; Hartwig and Ammon, 2002; Zanzotti and Mescalchin, 2019), balanced against potential drawbacks like competition for these resources with grapevines (Abad et al., 2021b; Celette and Gary, 2013; Garcia et al., 2018; Griesser et al., 2022; Novara et al., 2018; Raffa et al., 2022).

Scientific literature shows contradictory results on the effects of service crops in vineyards without explicitly considering the importance of service crop management to reach trade-offs between ecosystem services and disservices. The management of service crops and consequently the services they provide depends on various decisions: choice of species (alone or in a mixture) and establishment (sowing date and density), spatial structure in the vineyard, duration of the service crop and termination strategy and tools (Garcia et al., 2018). For instance, the choice of service crop species impacts their ability to promote biological pest control, or affects water and nitrogen provision (Beaumelle et al., 2021; Capri et al., 2023; Garcia et al., 2020; Raffa et al., 2022; Sulas et al., 2017); favoring legume species can increase grapevine nitrogen content and yield in comparison with gramineous species (Raffa et al., 2022; Sulas et al., 2017),; non-legume species, exhibiting higher C:N ratios, are more likely to cause competition for soil resources, especially nitrogen, due to their high N depletion capacity, slower mineralization of residues or soil N immobilization (Celette et al., 2009; Justes et al., 2009; White et al., 2017). Additionally, the spatial and temporal diversity of service crops practices may influence their effects on vineyard components (Fernández-Mena et al., 2021; Griesser et al., 2022). For example, adapting soil coverage in the vineyard can mitigate yield and vigor loss due to competition between grapevines and associated vegetation (Delpuech and Metay, 2018). Moreover, the duration of service crop presence in the vineyard also affects its impacts on soil resources such as water and nitrogen, and their effect of grapevine water and nitrogen status, vigor and yield (Celette et al., 2009, 2008; Griesser et al., 2022). Thus, adapting service crop management with partial spatial or temporal disturbance of the inter-row may offer trade-offs between permanent service crops and bare soils (Griesser et al., 2022).

Residue management, interacting with the choice of service crop species and termination dynamics, also influences the services and disservices provided by service crops. For instance, incorporating service crop residues into the soil can increase their mineralization compared with mowing without incorporation, with subsequent positive impacts on grapevine nitrogen content (Coppens et al., 2006a; Raffa et al., 2022). Conversely, the use of a roller-crimper for termination can enhance weed suppression (Canali et al., 2013; Hefner et al., 2020), although its effectiveness may depend on the service crop growth stage and termination dynamics (Ashford and Reeves, 2003). The effect of the service crop termination period and method have primarily been studied separately, indicating a gap in the understanding of how these factors

interact, especially within viticulture. In a cabbage (*Brassica oleracea* L.) cropping system, Hefner et al. (2020) observed higher soil nitrogen content with the incorporation of a legume service crop, compared to using a roller-crimper, and a pure legume service crop was found to provide more soil nitrogen than a mixed crop of legumes and grasses. While research has generally focused on the effects of termination period and method in separate studies, the literature lacks comprehensive evaluations of the interaction between these two factors combined, especially in the context of viticulture. Moreover, these effects can vary greatly from one year to the next, depending on rainfall dynamics in particular and the resulting growth and biomass of the service crop (Finney et al., 2016; Haruna et al., 2020).

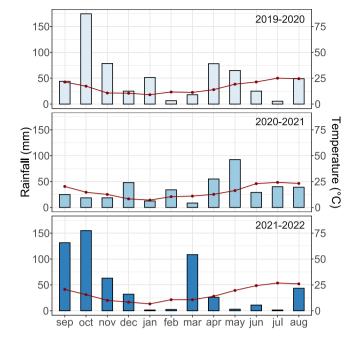
This study aimed at evaluating different service crop termination strategies for a winter mixture of species from the three commonly used families (i.e. *Fabaceae, Poaceae, Brassicaceae*), combining two service crop termination periods (early termination vs. budburst termination) and three termination methods (mower, mower + tillage, roller-crimper). For three years, we conducted an experiment in a vineyard situated in a Mediterranean climate region in the south of France. During this period, we monitored a variety of indicators in the service crop (biomass, C:N ratio, weeds, and mulch following termination), the soil (such as soil organic matter, microbial biomass, and water and nitrogen stocks), and the grapevine (predawn leaf water potential, yield components,  $\delta^{13}\mathrm{C}$ , yeast assimilable nitrogen in juice, and pruning weight). We hypothesized that:

- Terminating the service crop early (i.e. before grapevine budburst)
  reduces competition for water and nutrients, thus promoting
  grapevine yield. However, it offers fewer benefits for the soil's
  organic matter and microbial biomass due to less biomass input to
  the soil.
- 2. Termination methods involving tillage enhance the effectiveness of service crop termination and increase service crop mineralization rate and consequently grapevine yield and berry quality, but these methods provide less weed suppression compared to the rollercrimper, due to the absence of mulch at the soil surface and weed regrowth after tillage.
- 3. No-till termination methods, such as using a mower or roller-crimper, benefit soil organic matter and microbial biomass. However, these methods increase competition with the grapevine by decreasing nitrogen inputs, leading to extended depletion of water and nutrients due to less effective termination practices.
- 4. Interactions between the termination period and method exist, providing opportunities to identify trade-offs. By adjusting the combination of these two factors, vine growers can align their strategies with their objectives.

#### 2. Material and methods

#### 2.1. Experimental site and design

The experiment was conducted in a conventional vineyard located in Villeneuve lès Maguelone in southern France (43°32.5243'N, 3°50.8240'E), spanning from fall 2019 to fall 2022. The region features a Mediterranean climate, with rainfall accumulation reaching 621, 421, and 579 mm in each year of the experiment respectively (Fig. 1). The vineyard, planted with *Vitis vinifera* L. variety 'Syrah' grafted onto an SO4 rootstock in 2003, has a density of 4000 vines per hectare (2.5 m  $\times$  1 m), and had an average slope of 2% along the direction of the rows. Vines were trained using a midwire bilateral cordon system, and spur pruned to 12 nodes per vine (6 spurs and 2 nodes per spurs). Throughout the duration of the experiment, fruit thinning, green pruning, and the application of fertilizers were not conducted, and the row was mechanically weeded. The soil composition averages 35% clay, 42% silt, and 23% sand, with a 30% proportion of stoniness and 3.7% of total organic matter. Soil pH (water method) was 8.3, with 1.8 g kg $^{-1}$  of total



**Fig. 1.** Cumulated rainfall per month (Rainfall bars, mm) and mean air temperature per month (Temperature points and lines, °C) for the 3 years of the experiment.

nitrogen, and  $1.3\,\mathrm{g\,kg}^{-1}$  of P2O5 (Joret-Hébert method).

Before initiating the experiment, no cover crops were established, and the inter-rows were consistently cultivated. During the three years of experimentation, a mixture of species from three families (legumes, grasses and crucifers, Table 1) was sown in all inter-rows (1.7 m width) with a direct seeding machine (AURENSAN SV-200), without prior seedbed preparation. The seeds used were sourced from a local cooperative producing population varieties (SCIC Graines Equitables). We included Vicia faba and Sinapis alba in the mixture each year, but the second year (2020) we replaced Avena sativa with Hordeum vulgare due to seed availability. Moreover, the last year we added *Pisum sativum* in the mixture to lower the risk of a weak emergence of *Fabaceae* species as experienced in the second year (Fig. 2). Consequently, the composition of the seeded mixture slightly differed across years, but remained the same in all inter-rows. We designed 6 experimental treatments as a combination of two cover crop termination periods - early termination (E) and budburst termination (B), and three termination methods mower and tillage (T), mower only (M), and roller (R). Each treatment was applied to plots covering a vine row and its two adjacent inter-rows of 45 m length. Treatments were randomly situated and replicated thrice across three blocks in the vineyard (Figure A1). The early termination occurred between mid-February and early March depending on the year, on E treatments, while budburst termination occurred between late March and mid-April (Table 1), on both E and B treatments. When

the service crops were not completely terminated, a supplementary pass was done later in the season (Table 1). Mowing was performed using a rotary mower for viticulture (PERFECT T-series), the tillage was performed using a toothed frame soil cultivator (PHILIPAGRI Polyculteur 2000), and the roller was applied using a roller with separated, notched, and cast iron discs (ROLL'N'SEM RWS200).

#### 2.2. Service crops, weeds and mulch measurements

#### 2.2.1. Aboveground service crop sampling

Aboveground service crop biomass was sampled twice, before each termination date. At grapevine flowering we collected remaining sown species, residues (i.e. dead plant material), and living weeds. In each treatment and block, three quadrats measuring  $0.5 \times 0.5$  m were used for sampling. All sown species and weeds were separated, dried at  $60^{\circ}$ C over 72 hours, and then weighed. Due to the restrictions imposed during the COVID-19 pandemic, specifically during the lockdown, the aboveground biomass for E treatments was not recorded at budburst in 2020

#### 2.2.2. C:N ratio determination

At both termination periods, samples from each sown species were sent to a laboratory (Celesta Lab, Mauguio, France) for carbon (C) and nitrogen (N) content analysis (determined by dry combustion elemental analysis). For each quadrat, the C:N ratio was calculated by dividing the total C content by the total N content.

#### 2.3. Soil functions indicators

#### 2.3.1. Soil organic matter and microbial biomass

At the beginning of the experiment (October 2019), three composite soil samples per block were collected and sent to a laboratory (Celesta Lab) for determination of particulate and dissolved soil organic matter (SOM, determined by dry combustion elemental analysis after particle size fractionation with a 50  $\mu m$  threshold). To build one soil composite we collected three soil samples on one inter-row per treatment (0–20 cm soil layer) and mixed the samples between two adjacent treatments (total 6 samples per soil composite).

By the end of the experiment in October 2022, we collected three more soil samples from the 0–20 cm layer in one inter-row of each treatment (54 in total). These samples were sent to the same laboratory for particulate and dissolved SOM determination (%), providing three replicates per treatment and per block. We also used the samples collected in Block S1 (Figure A1) for the measurement of soil microbial biomass (MB, determined by fumigation-extraction, mg kg $^{-1}$ ) in addition to SOM.

#### 2.3.2. Soil water and inorganic nitrogen stocks

Soil water and inorganic nitrogen stocks were measured on one out of the three blocks (Block S1, Figure A1). Each year, shortly after grapevine flowering, we extracted three soil cores from the same location where aboveground sampling had occurred (i.e. areas with weeds, residues, and remaining sown species; for details, refer to Section 2.3.1). These cores, extracted down to a depth of  $1\,\mathrm{m}$ , were then divided into

Table 1 Service crop composition, sowing and termination dates for the 3 years of the experiment. Growing Degree-Day (calculated with a base temperature of  $0^{\circ}$ C) between sowing and termination dates are indicated in italic for early and budburst termination.

	Composition	Sowing density	Sowing date	Early termination	<b>Budburst termination</b>	Supplementary passes
Year 1	Vicia faba Sinapis alba Avena sativa	$100 \text{ kg ha}^{-1}$ $5 \text{ kg ha}^{-1}$ $60 \text{ kg ha}^{-1}$	2019–10–03	2020-02-14 1591 GDD	2020–03–24 2038 GDD	Roller on 2020–05–28
Year 2	Vicia faba Sinapis alba Hordeum vulgare	100 kg ha <sup>-1</sup> 5 kg ha <sup>-1</sup> 60 kg ha <sup>-1</sup>	2020-09-30	2021–03–03 1641 GDD	2021–04–12 2096 GDD	Roller on 2021–06–08
Year 3	Vicia faba Sinapis alba Avena sativa Pisum sativum	100 kg ha <sup>-1</sup> 5 kg ha <sup>-1</sup> 60 kg ha <sup>-1</sup> 50 kg ha <sup>-1</sup>	2021–10–07	2022–02–17 1327 GDD	2022-04-13 1923 GDD	Roller + mower on 2022–05–16 Tillage on 2022–06–06

four distinct layers (0–20 cm, 20–40 cm, 40–60 cm, and 60–100 cm). For each layer, we weighed a subsample of the soil to determine its fresh mass. The samples were then oven dried at 103 °C for 72 hours, and reweighed to measure their dry mass. Following this, the samples were submerged in water inside a 2 mm sieve to separate the soil from the gravel. The gravel was oven-dried at 103 °C for 24 hours and then weighed. We calculated the gravimetric water content for each soil layer, and calculated the soil water stock by applying the equation:

$$WS = WC \times D \times (100 - GC) \times BD \times 10^{-4}$$

with WS the water stock (mm), WC the gravimetric soil water content (%), D the thickness of the soil layer (mm), GC the gravel content (%, in mass) and BD the bulk density of the soil layer (g cm $^{-3}$ ). Total soil water stock at 1 m depth was calculated by aggregating the water stock of each soil layer.

From the 0–20 cm soil layer, another subsample was sent to a laboratory to determine its inorganic nitrogen content (NO3- and NH4+, determined by KCl extraction and colorimetry, mg kg $^{-1}$ ). While it might not encompass the entire pool of inorganic N available to grapevines, the 0–20 cm soil layer is the zone affected by soil tillage, which is important for the mineralization of service crop residues in our study. The 0–20 cm nitrogen stock was calculated with the same formula as for water stock, by replacing the soil water content with the inorganic nitrogen content, and used as an indicator of N provision.

#### 2.4. Grapevine performances

#### 2.4.1. Sampling strategy

At the experiment's onset, we selected 10 vines from each treatment and block to monitor pruning weight, predawn leaf water potential, and yield components over the experiment's duration. The selection process deliberately avoided vines that lacked neighboring plants or displayed signs of disease (such as dead wood or severely diminished vigor). Regrettably, a portion of the initially chosen vines either perished or manifested wood diseases as the experiment progressed, necessitating their replacement, either at the grapevine harvest or pruning periods.

#### 2.4.2. Grapevine water status during reproductive phase

Shortly after grapevine flowering (between one and two weeks before flowering started), fruit set and veraison, the grapevine water status was monitored by measuring predawn leaf water potential ( $\psi$ , MPa). From each treatment and block, two fully expanded leaves located on two primary shoots were collected from six separate vines and their  $\psi$  values were determined using two respective pressure chambers (Soil Moisture Equipment Corp., Santa Barbara, California, USA). The recorded leaf water potential for each vine, used for subsequent analysis, was computed as the mean of the readings from the two pressure chambers.

#### 2.4.3. Number of bunches, yield and berry measurements

At the time of grapevine harvest, the count of bunches per vine was recorded for 10 vines in each treatment and block. The bunches from each vine were gathered to weigh the overall fruit biomass per vine. Subsequently, a composite sample for each treatment and block was constituted by collecting a single bunch from each of the 10 monitored vines within their respective treatment and block. Each composite sample was then manually pressed to extract the juice, yielding one juice sample per treatment and block. Three juice samples were taken from each composite for the determination of the carbon isotope ratio ( $\delta^{13}$ C), used as an integrated indicator of grapevine water status (Gaudillère et al., 2002), and another three samples were drawn for the measurement of yeast assimilable nitrogen (YAN, mg  $\mbox{L}^{-1}\mbox{)}.$  The  $\delta^{13}\mbox{C}$  analyses were carried out by the INRAE laboratory IPSiM (Montpellier, France), using a continuous-flow isotope ratio mass spectrometer (EA-IRMS) using a VarioPyrocube elemental analyser (Elementar, UK) coupled with Isoprime Precision mass spectrometer, while the YAN analyses were

performed by the INRAE Experimental Unit Pech Rouge (Gruissan, France) using enzymatic and NOPA methods for ammoniacal and amine nitrogen, respectively, followed by spectrophotometry.

#### 2.4.4. Pruning weight

Following the grapevine leaves falling in November 2019, we chose 10 vines for monitoring throughout the experiment in each treatment and block (refer to Section 2.4.1 for details). Each of these selected vines was pruned to a total of 12 nodes per vine (with 6 spurs and 2 nodes per spur), and the biomass of the pruned canes was recorded (kg vine<sup>-1</sup>). The pruning weight was subsequently recorded in the winters (November-December) of 2020, 2021, and 2022, adhering to the same sampling process. Pruning woods were let on the soil surface and managed following the service crop termination strategies (i.e. shred in the M treatments, incorporated in the T treatments and let at the surface in R treatments).

#### 2.5. Data analysis

Data were analyzed using R software (R Core Team, 2023, version 4.2.3). When data were available for Block S1 only, we analyzed the factor Year, Termination period and Termination method with fixed effects models and post hoc analyses using the HSD.test function from the agricolae package (Mendiburu, 2021). When data were available for the three blocks, mixed effects models were implemented using the *lme4* (Bates et al., 2015) and *lmerTest* (Kuznetsova et al., 2017) packages, with the factor Block as random effect. Both fixed effects and mixed effects models were subjected to ANOVAs to test the significance of variables, with non-significant interactions removed using the drop1 function. For models that included random effects, Satterthwaite's method was employed as facilitated in the *lmerTest* package. The emmeans package (Lenth, 2022) was used for post hoc analyses and multiple comparisons of mixed effects models, applying the Bonferroni adjustment.

To analyze aboveground service crop biomass, we utilized mixed effects models separately for each year and sampling date, with aboveground biomass as the dependent variable, and Termination period, Termination method, and their interaction as fixed effects (including all double interactions). The factor Block was considered a random effect. *Post hoc* tests were conducted on the interaction between Termination period and Termination method when effects were significant (Fig. 2, Table 2). For the evaluation of living weeds, residues, and remaining sown species at grapevine flowering, we combined the factors 'Termination period' and 'Termination method' into a single factor. This was due to the presence of zero values, which made it impossible to test interactions. Table 3

To analyze soil water stock, inorganic nitrogen stock, and microbial biomass, we utilized fixed effects models as these variables were sampled in Block S1 only (no random Block effect). Soil water stock, inorganic nitrogen stock, and microbial biomass were the dependent variables in the models. Termination period, Termination method, and Year were the fixed effects, including their double interactions (triple interactions were excluded). ANOVAs were executed on the models after the removal of non-significant interactions. Post-hoc tests were conducted on significant factors or their interactions (Table 4).

To analyze final SOM, predawn leaf water potential, number of bunches, yield per vine,  $\delta^{13}$ C, YAN, and pruning weight data, we used mixed effects models with Termination period, Termination method, and Year as fixed effects, including their double interactions (triple interactions were excluded). The factor Block was considered a random effect. ANOVAs were performed on the models after the removal of nonsignificant interactions. Post-hoc tests were performed on significant factors or their interactions (see Table 5).

#### 3. Results

#### 3.1. Service crops biomass and C:N ratio

The biomass of service crops varied depending on the year: in 2020,

it was around 5 t ha $^{-1}$  at early termination and between 10 and 15 t ha $^{-1}$  at late termination (i.e. budburst); in 2021, it ranged between 1 t ha $^{-1}$  and 2 t ha $^{-1}$  before early termination and approximately 2–3 t ha $^{-1}$  at budburst; in 2022, it was about 1.3 t ha $^{-1}$  before early

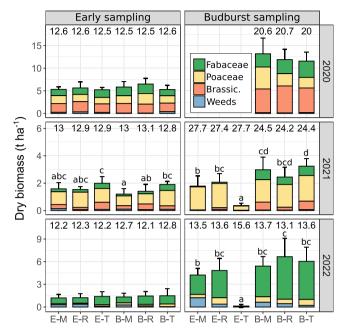


Fig. 2. Dry matter of service crops and weeds measured at the two termination dates in each treatment, and C:N ratio of the sown species. Colors indicate the mean biomass per plant family and weeds in the mixture at each measurement date. The top of the barplots indicates the mean (and standard deviation) of the total dry matter of the mixtures in each treatment. C:N ratios of the sown species are indicated for each treatment at the top of each sub-plot. Dry matter was not measured in the E treatments at budburst 2020 due to lockdown. The biomass sampled before the termination of E treatments are shown in the left part of the plot (Early), the biomass sampled before the termination of B treatments are shown in the right part of the plot (Budburst). For each year and measurement period, different letters indicate significant differences between treatments. E: early termination; B: termination at budburst; M: mowing machine; R: roller; T: tillage.

 $0.35\pm0.37$ 

 $1.75\pm0.61$  ab

 $0.12\pm0.1~\text{ab}$ 

Weeds dry biomass (t ha-1)

Residues dry biomass (t ha-1)

Living SC dry biomass (t ha-1)

termination and around 5–6 t ha<sup>-1</sup> at budburst (Fig. 2). Service crops terminated at budburst (B-M, B-R, and B-T treatments) produced an additional biomass of approximately 5–7 t ha<sup>-1</sup> in 2020, 1–2 t ha<sup>-1</sup> in 2021, and 4–5 t ha<sup>-1</sup> in 2022, between the two termination periods (Fig. 2). In 2021 and 2022, early destruction with soil tillage (i.e. E-T treatment) resulted in almost negligible biomass during measurements at budburst in the same treatment, while in the two other treatments (E-M and E-R), the biomass was similar to those terminated only at budburst (Fig. 2). The proportion of species present in the mix also varied from year to year. In 2021 the mixture was dominated by *Hordeum vulgare*, with a low presence of *Vicia faba*; in 2022, the mixture was dominated by *Vicia faba* and *Pisum sativum*, while *Sinapis alba* did not emerge (Fig. 2).

C:N ratio values were similar each year at early termination for all treatments (C:N ratio between 12.1 and 13.1 in each treatment), but varied between years for the second sampling period (i.e. before budburst termination): between 20 and 20.7 in 2020, between 24.4 and 27.7 in 2021, and between 13.1 and 15.6 in 2022 (Fig. 2).

At grapevine flowering, treatments terminated with tillage (i.e. E-T and B-T) exhibited lower plant residue biomass on average (0.01–1.52 t ha $^{-1}$ ) compared to M and R treatments (0.35–6.12 t ha $^{-1}$ , Table 2). R treatments exhibited the highest residue biomass on average in 2021 and 2022 but not in 2020. In 2020 and 2021, the T treatments exhibited the lowest living weed biomass, M treatments the highest, R treatments exhibiting intermediate values. In 2022, there were no differences between treatments.

#### 3.2. Soil variables

#### 3.2.1. Soil water and inorganic nitrogen provision

 $0.09 \pm 0.16$ 

 $\textbf{2.28} \pm \textbf{0.42}~\textbf{b}$ 

 $0.29 \pm 0.16$  bc

The water stock around grapevine flowering varied from 171 to 247 mm in average. The factors "Year" and "Termination method" were significant over the whole dataset but not their interaction (Table A1, Table 4). Year 2022 showed lower stocks compared to years 2020 and 2021 (Fig. 3, Table 4). The ranking between the three termination methods remained constant each year: T > M > R from the highest to the lowest soil water stock. Soil water stock in the T treatments was significantly higher than water stock in the R treatments whatever the year or termination period (i.e. early vs. budburst, Table 4). Differences between T and R treatments reached 41 mm, 27.5 mm, and 21.7 mm in

**Budburst termination** 

 $0.15\pm0.24$ 

 $5.59 \pm 2.93 c$ 

 $0.43\pm0.22~\mathbf{c}$ 

 $0.05 \pm 0.07$ 

 $1.26\pm0.59~\text{ab}$ 

 $0\pm0.01~\text{a}$ 

Table 2 Death plant residue and living weeds and service crops (SC) dry matter measured at grapevine flowering. M: mowing machine; R: roller, T: tillage. For each variable, the letters indicate significant differences from the *post hoc* test performed after ANOVA ( $\alpha < 0.05$ ).

Early termination

 $0.39\pm0.55$ 

 $5.82 \pm 2.46 c$ 

 $0.33\pm0.25~\textbf{bc}$ 

	Mowing	Roller	Tillage	Mowing	Roller	Tillage
Weeds dry biomass (t ha <sup>-1</sup> )	$1.43\pm0.26~\textbf{d}$	$0.51 \pm 0.55 \text{ ab}$	$0.07 \pm 0.06 \; \mathbf{a}$	$1.09 \pm 0.51$ <b>cd</b>	$0.64 \pm 0.21$ bc	$0.1\pm0.12~\mathbf{a}$
Residues dry biomass (t ha <sup>-1</sup> )	$3.79\pm1.79~\textbf{b}$	$3.9\pm1.49~\mathbf{b}$	$0.91\pm0.5~\mathbf{a}$	$6.12\pm1.86~\mathbf{c}$	5.47 ± 1.67 <b>bc</b>	$1.52\pm0.38~\textbf{a}$
			20	021		
		Early termination			Budburst termination	
	Mowing	Roller	Tillage	Mowing	Roller	Tillage
Weeds dry biomass (t ha <sup>-1</sup> )	$0.36\pm0.25~\text{ab}$	$0.17 \pm 0.22~\textbf{ab}$	$0.14\pm0.05~\text{ab}$	$\textbf{0.54} \pm \textbf{0.62}~\textbf{b}$	$0.12\pm0.12~\textbf{a}$	$0.09 \pm 0.06 \; \mathbf{a}$
Residues dry biomass (t ha <sup>-1</sup> )	$\textbf{0.35} \pm \textbf{0.24 ab}$	$1.96\pm0.57~\textbf{d}$	$0.01\pm0.03~\textbf{a}$	$\textbf{0.46} \pm \textbf{0.24} \; \textbf{b}$	$1.45\pm0.42~\text{c}$	0
Living SC dry biomass (t ha <sup>-1</sup> )	$1.71\pm0.44~\textbf{bc}$	$2.21\pm0.69~\text{cd}$	$0.01\pm0.01~\textbf{a}$	$\textbf{1.54} \pm \textbf{0.49}~\textbf{b}$	$\textbf{2.64} \pm \textbf{0.37} \; \textbf{d}$	$0.11\pm0.05~\textbf{a}$
			20	022		
		Early termination			Budburst termination	
	Mowing	Roller	Tillage	Mowing	Roller	Tillage

 $0.2\pm0.24$ 

 $0.03 \pm 0.06$  a

 $0.11\pm0.33~\text{ab}$ 

#### Table 3

 $\delta^{13}$ C and YAN measured on berries in 2020, 2021 and 2022 in all treatments. E: early termination; B: budburst termination; M: mowing machine; R: roller; T: tillage. For  $\delta^{13}$ C, the factors Termination method, Year, and the interaction Termination method:Termination period were significant; for YAN, the factors Termination method, Year, and the interaction Termination period:Year were significant (Table A1). For  $\delta^{13}$ C letters indicate significant differences from the post hoc test performed on the interaction Termination method:Termination period; for YAN letters indicate significant differences from the post hoc test performed on the factor Termination method (Table 5, Table A1).

			2	020		
		Early termination			Budburst termination	
	Mowing	Roller	Tillage	Mowing	Roller	Tillage
$\delta^{13}$ C	-24,1 $\pm$ 1,5 <b>bc</b>	-24 $\pm$ 1,9 $\mathbf{c}$	-25 $\pm$ 1,3 $\mathbf a$	-23,6 $\pm$ 1,3 <b>c</b>	-24 $\pm$ 1,4 bc	-24,8 $\pm$ 1,8 <b>ab</b>
YAN (mg L <sup>-1</sup> )	191 ± 6 <b>a</b>	$204 \pm 34~\textbf{a}$	$230\pm11~\textbf{b}$	$195\pm18~\textbf{a}$	$188\pm24~\textbf{a}$	$211\pm23~\textbf{b}$
			2	021		
		Early termination			Budburst termination	
	Mowing	Roller	Tillage	Mowing	Roller	Tillage
$\delta^{13}C$	-22,5 $\pm$ 0,4 <b>bc</b>	-22,4 $\pm$ 1 $\mathbf{c}$	-23 $\pm$ 0,4 a	-22,5 $\pm$ 0,3 $\mathbf{c}$	-22,8 $\pm$ 0,3 <b>bc</b>	$-22,9 \pm 0,5 \text{ ab}$
YAN (mg L <sup>-1</sup> )	$145\pm29~\textbf{a}$	$160 \pm 42~\textbf{a}$	$192 \pm 31~\textbf{b}$	$160 \pm 43~\textbf{a}$	$171 \pm 42~\mathbf{a}$	$206 \pm 39~\textbf{b}$
			2	022		
		Early termination			Budburst termination	
	Mowing	Roller	Tillage	Mowing	Roller	Tillage
$\delta^{13}$ C	$-23,4 \pm 1,1$ bc	-23,4 $\pm$ 1,2 $\mathbf{c}$	-24,3 $\pm$ 0,8 <b>a</b>	-23,1 ± 0,8 <b>c</b>	-23,7 $\pm$ 0,7 <b>bc</b>	$-23,8 \pm 0,8 \text{ ab}$
VAN (mg L <sup>-1</sup> )	156 ± 43 a	156 ± 68 a	215 + 29 <b>h</b>	156 ± 46 a	149 ± 45 a	189 ± 54 <b>h</b>

2020, 2021, and 2022, respectively (Fig. 3).

The soil inorganic nitrogen content varied from 5 to 67 kg ha $^{-1}$  in average. Only the interaction between factors "Year" and "Termination method" was significant. Indeed, the treatment T showed the higher soil inorganic nitrogen values for years 2020 and 2022 (61 kg ha $^{-1}$  on average vs. 16.25 kg ha $^{-1}$  on average for M and R treatments), but not in 2021 (Fig. 3). Moreover, differences between the "mowing" and "roller" termination methods were not significant whatever the year.

#### 3.2.2. Soil organic matter and microbial biomass

Particulate and dissolved SOM did not significantly vary between treatments after three years of treatments (Fig. 4, Table 5). The soil microbial biomass measured at the end of the experiment was significantly modified by both factors "Termination period" and "Termination method" (Table A1). Indeed, the treatments B (termination at budburst) exhibited significantly higher soil MB than treatments E. On average, the treatment R exhibited higher soil MB in comparison with the treatments M and T, but differences were not significant in the post hoc test (Table 4).

#### 3.3. Grapevine growth and yield

#### 3.3.1. Grapevine water and nitrogen status

Overall, service crop termination by soil tillage (i.e. treatments T) decreased grapevine water constraint compared to other termination methods (Fig. 5, Table 3, Table 5). Regarding to the predawn leaf water potential, only the termination method had a significant effect (Table A1, Table 5). Leaf water potential in the "Till" treatments was significantly different from leaf water potential in the M and R treatments which shared similar values (Table 5). Year 2020 showed a lower water stress than 2021 and 2022, and there was no interaction between the factors Termination method and Year, showing that the same trend was observed each year. Results were similar regarding to the measurements of the  $\delta^{13}$ C performed on grape berries, with the treatments T exhibiting the lowest water stress, especially when service crop termination was performed early (Table 3, Table 5).

The factors Termination method and the interaction between Termination period and Year significantly explained YAN variations

between treatments (Table 3, Table A1). Indeed, the treatments T exhibited higher YAN content in berries compared to the treatments M and R, regardless of the year, and the treatments M and R were not significantly different (Table 5). Regarding to the effect of the factor Year, YAN was higher in 2020 compared to 2021 and 2022.

**Table 4** *Post hoc* tests performed after ANOVAs on linear models for soil variables measured only in Block S1. The multiple comparisons were only applied to significant factors, or their interaction ( $\alpha=0.05$ ). Letters indicate significant differences between treatments. Early: early termination; Bud: budburst termination; M: mowing machine; R: roller; T: tillage.

Term	Mean	Min	Max	group
Water stock at grapevine flowering				
Termination method				
M	207.6	166.0	238.7	ab
R	197.8	160.3	236.3	b
T	218.9	163.4	264.5	a
Year				
2020	224.4	191.9	264.5	a
2021	218.7	185.6	251.8	a
2022	181.2	160.3	219.8	b
Nitrogen stock at grapevine flowering				
Termination method:Year				
M:2020	17.3	6.3	28.0	b
M:2021	7.5	3.3	12.6	b
M:2022	12.1	4.5	28.8	b
R:2020	15.5	7.5	29.4	b
R:2021	10.6	4.3	21.4	b
R:2022	19.6	11.2	28.0	b
T:2020	62.6	53.3	78.9	a
T:2021	18.6	6.8	35.5	b
T:2022	61.7	18.7	104.9	a
Soil microbial biomass				
Termination period				
Early	419.6	347	481	ь
Bud	490.3	368	619	a
Termination method				
M	428.3	347	534	a
R	511.7	409	619	a
T	424.8	368	484	a

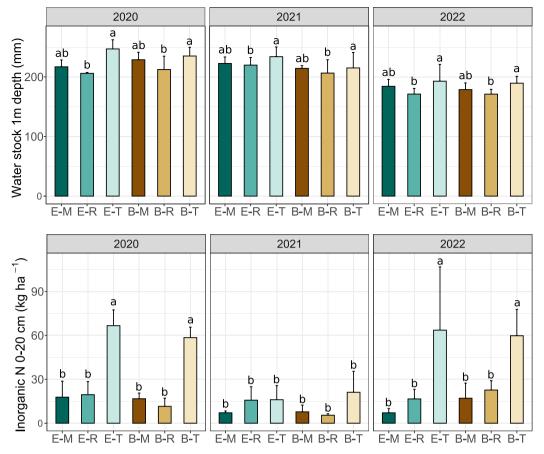


Fig. 3. Soil water stock in mm and soil inorganic nitrogen measured around grapevine flowering on 1 m and 20 cm depth, respectively. E: early termination; B: budburst termination; M: mowing machine; R: roller, T: tillage. For water stock, the factors Termination method and Year were significant; for nitrogen (N) stock, the factors Termination method, Year, and the interaction Termination method: Year were significant (Table A1). For the water stock, letters indicate significant differences from the post hoc test performed on the factor Termination method; for the nitrogen stock, letters indicate significant differences from the post hoc test performed on the interaction Termination method: Year (Table 4, Table A1).

#### 3.3.2. Yield components and grapevine vegetative growth

Only the termination method significantly explained grapevine number of bunches (Table A1). T treatments exhibited the highest number of bunches in average, M treatments exhibited the lowest number of bunches, R treatments being intermediate (Fig 6, Table 5). Moreover, the difference between the T treatment and the M treatment was significant (16.1 vs. 14.1 bunches on average, respectively), but the treatment R exhibited no significant differences from treatments M and T (15.4 bunches on average, Table 5).

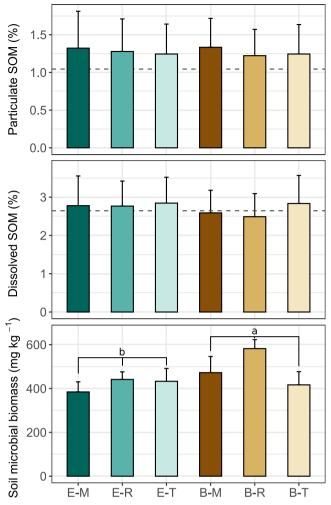
Grapevine yields ranged from 7.25 to 13.7 t ha<sup>-1</sup> over the course of the experiment (Fig 6). The factor Termination period and the interaction between Termination method and Year were significant in the analysis (Table A1). The treatment B (i.e. termination at budburst) exhibited higher yields compared to the treatments E (i.e. early termination). Moreover, differences between treatments were not stable through the three years: in 2020, there was no significant difference in yield between the three termination methods; in 2021, the ranking followed that of the number of bunches ( $M \le R \le T$ ; in 2022, the T treatment exhibited a significantly higher yield compared to the M and R treatments (Fig 6, Table 5).

The factors Year and Termination method significantly explained pruning weight, with no interaction between both factors (Table A1). Indeed, the treatments T exhibited significantly higher pruning weights than the treatments M and R regardless of the year, whereas the differences between treatments were negligible before the implementation of the experiment (Fig. 7, Table 5). Regarding to the factor Year, grapevine pruning weights in 2021 were significantly lower than grapevine pruning weights in 2020 and 2022.

#### 4. Discussion

#### 4.1. Experimental design: evaluating service crop termination practices

We opted to compare two periods for service crop termination combined with three termination methods, forgoing any comparison with a control treatment of maintained bare soil. Indeed, the use of service crops is on the rise, even in Mediterranean viticulture areas where rainfall is scarce (Fernández-Mena et al., 2021). While the differences between maintaining service crops and bare soil are well-researched and documented in scientific and technical literature (Abad et al., 2021a, 2021b; Griesser et al., 2022; Winter et al., 2018), data concerning the management of service crops in viticulture is limited. The inquiries we address in this paper arise directly from winegrowers. Thus, our experimental design enables us to examine closely the issues associated with the termination period and method by comparing six service crop management strategies, assuming service crops are established annually. While our experimental design does not facilitate a comparison of each termination strategy with a bare soil control, we assume it aligns more closely with winegrowers' concerns about managing service crops. Another limitation of our study lies in the sampling strategy for soil measurements: indeed, soil measurements were conducted only in Block S1 (Figure A1). However, we assume the results observed in the soil at the scale of one block (such as a higher soil microbial biomass for budburst termination, improved soil water and inorganic nitrogen stock with tillage) are consistent with the indicators measured at the entire experiment scale, and give some insights to interpret some of the results observed on the grapevine.



**Fig. 4.** Particulate SOM, dissolved SOM and microbial biomass measured at the end of the experiment in each treatment (0–20 cm). Dashed lines represent SOM measurements at the beginning of the experiment, averaged between the three blocks. E: early termination; B: budburst termination; M: mowing machine; R: roller; T: tillage. For particulate and dissolved SOM, no significant differences were observed between treatments. For microbial biomass, both factors Termination method and Termination period were significant (Table A1). Letters indicate significant differences from the *post hoc* test performed on the factor Termination period (Table 4, Table A1).

In addition, the mixture of sown species was not exactly the same from one year to another, potentially leading to confounding effects between the type of mixture and the climatic characteristics of the year. However, the same botanical families of species were sown each year, and the same mixture was used for all treatments annually. Moreover, we hypothesize that even by sowing the exact same mixture each year, the sowing method (direct seeding) and the highly variable precipitation from year to year around the sowing date would not have allowed us to precisely control the composition of the emerged vegetative cover, likely resulting in different mixes between years in terms of species composition and biomass. Except for the service crops' dry biomass, the year factor was included in all statistical models to account for these interannual variations when assessing service crops termination strategies. Furthermore, the potential effects of these changes in climatic conditions and species mix from one year to the next are discussed in the following sections, particularly regarding variables related to water and nitrogen stress in grapevines.

Table 5

Post hoc tests performed after ANOVAs on linear mixed models. The multiple comparisons were only applied to significant factors, or their interaction ( $\alpha = 0.05$ ). Letters indicate significant differences between treatments. Early: early termination; Bud: budburst termination; M: mowing machine; R: roller; T:

Term	emmean	lower. CL	upper. CL	group
Predawn leaf water potential				
Termination method				
T	-0.45	-0.82	-0.07	a
M	-0.49	-0.87	-0.11	b
R	-0.49	-0.87	-0.11	b
Year				
2020	-0.39	-0.77	-0.01	a
2021	-0.51	-0.89	-0.13	b
2022	-0.53	-0.91	-0.14	b
$\delta^{13}C$				
Termination period:Method				
Early:T	-24.1	-26.2	-22.0	a
Bud:T	-23.8	-25.9	-21.7	ab
Bud:R	-23.5	-25.7	-21.4	bc
Early:M	-23.3	-25.5	-21.2	bc
Early:R	-23.3	-25.4	-21.1	c
Bud:M	-23.1	-25.2	-20.9	c
Year				
2020	-24.2	-26.8	-21.7	a
2022	-23.6	-26.1	-21.1	b
2021	-22.7	-25.2	-20.1	c
Yeast Assimilable Nitrogen				
(YAN)				
Termination method				
M	167	79.1	255	а
R	171	83.1	260	a
T	207	118.8	295	b
Termination period:Year	207	110.0	250	
Bud:2022	164	78.4	250	а
Early:2021	166	79.7	252	a
Early:2022	176	89.9	262	a
Bud:2021	179	93.1	265	a
Bud:2020	198	112.0	284	b
Early:2020	208	122.4	294	b
-				
Number of bunches				
Termination method		100	15.0	
M	14.1	13.2	15.0	a .
R	15.4	14.4	16.3	ab
T	16.1	15.2	17.0	b
Yield				
Termination period				
Early	2.54	2.36	2.73	a
Bud	2.76	2.57	2.94	b
Termination method:Year				
M:2021	1.89	1.59	2.18	a
R:2021	2.40	2.10	2.69	ab
M:2022	2.46	2.17	2.76	ab
R:2022	2.54	2.24	2.84	b
T:2021	2.61	2.32	2.90	b
M:2020	2.75	2.45	3.04	bc
T:2020	2.91	2.61	3.20	bc
R:2020	2.98	2.68	3.28	bc
T:2022	3.32	3.03	3.62	c
Pruning weights				
Termination method				
M	0.53	0.43	0.62	a
R	0.56	0.46	0.65	a
T	0.80	0.71	0.90	b
Year	5.50	0.71	5.70	,
2021	0.48	0.39	0.58	a
2022	0.68	0.59	0.77	ь

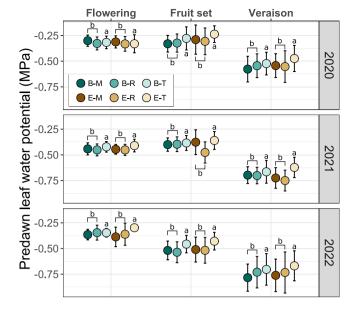
0.73

0.63

0.82

ь

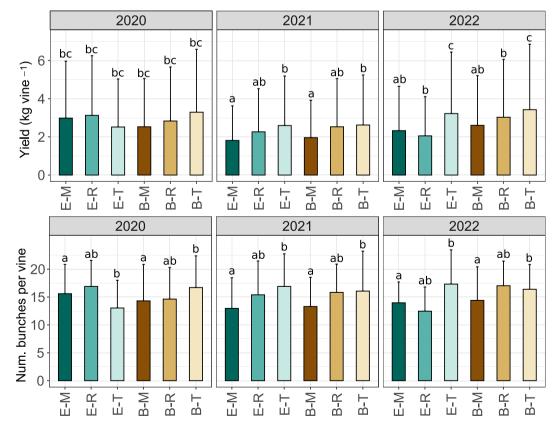
2020



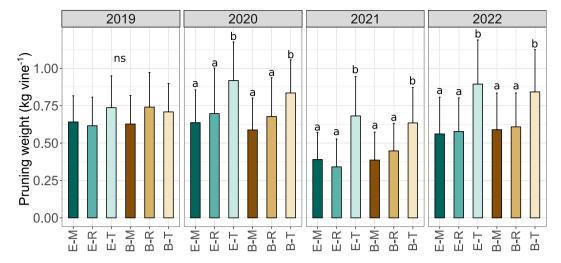
**Fig. 5.** Predawn leaf water potential measured in all treatments just after grapevine flowering, and at nouaison and veraison stages. E: early termination; B: budburst termination; M: mowing machine; R: roller; T: tillage. Both factors Termination method and Year significantly explained predawn leaf water potential variations. Letters indicate significant differences from the *post hoc* test performed on the factor Termination method (Table 5, Table A1).

#### 4.2. Termination strategies effectiveness to control service crops

Biomass production and mixture composition varied greatly from one year to another (Fig. 2), probably as a consequence of the weather conditions, particularly precipitation and temperature patterns (Fig. 1). The year 2019-2020 combined high fall and winter precipitations, that contrasted with the lower winter rainfall in year 2021-2022 and the drought experienced in year 2020–2021 (Fig. 1). Temperature was also higher in December and January of year 2019-2020 compared to 2020-2021 and 2021-2022. Agricultural practices and their interaction with climate also affected biomass production. For instance, decisions about species selection (two legume species were sown in 2022 but not in 2020 and 2021) or the timing of sowing and its alignment with rainfall patterns are key factors influencing biomass production (Anugroho et al., 2009; Constantin et al., 2015). In our study, both termination methods and timings modified service crops control: allowing service crops to develop until the budburst of the vine resulted in a two to three-fold increase in biomass compared to early destruction (Fig. 2). The longer duration of the growing season, and the rise of mean temperature the month before grapevine budburst may explain these observations (Table 1, Ruis et al., 2019). Termination involving soil tillage was the most effective method, as treatments destroyed early with soil tillage exhibited almost no regrowth, unlike treatments destroyed with the mowing machine or roller (Fig. 2). The use of a mowing machine led to the regrowth of the service crop, but the biomass of treatments destroyed only at the budburst of the grapevine (B-M, B-R and B-T) remained superior on average, even though the differences were not always significant between treatments sampled at budburst (Fig. 2). Thus, using a mowing machine resulted in a double biomass production and organic restitution to the soil after mowing. In the case of the roller,



**Fig 6.** Grapevine yield and number of bunches measured at harvest in 2020, 2021 and 2022. E: early termination; B: budburst termination; M: mowing machine; R: roller; T: tillage. For grapevine yield, the factors Termination method, Termination period, Year, and the interaction Termination method: Year were significant; for the number of bunches, only the factor Termination method was significant (Table A1). For grapevine yield, letters indicate significant differences from the *post hoc* test performed on the interaction Termination method: Year; for grapevine number of bunches, letters indicate significant differences from the *post hoc* test performed on the factor Termination method (Table 5, Table A1).



**Fig. 7.** Grapevine pruning weights measured at the beginning of the experiment (2019) and during the 3 years of the experiment. ns: non-significant differences between treatments. E: early termination; B: budburst termination; M: mowing machine; R: roller; T: tillage. Both factors Termination method and Year significantly explained pruning weight variations (Table A1). Letters indicate significant differences from the *post hoc* test performed on the factor Termination method (Table 5, Table A1).

the double production was more uncertain as some plants were not completely destroyed following early destruction, leading to a mixture of regrowth and continued growth. The low height of the service crops at early termination period, combined with a termination occurring during species vegetative growth may explain the lower efficiency of the roller (Alonso-Ayuso et al., 2020; Hefner et al., 2020; Kornecki and Kichler, 2022).

Destruction involving soil tillage led to the lowest biomass of weeds at the grapevine flowering two years out of three (Table 2). This is important as grapevine flowering is a key period for the grapevine yield elaboration (Guilpart et al., 2014), and a period when water competition can be intense in a Mediterranean context. Additionally, the termination method involving soil tillage prevented the regrowth of certain sown plants, especially Poaceae, as opposed to the roller, which was less effective in destroying this service crop species (*Avena sativa* and *Hordeum vulgare*). However, the roller produced the largest amount of dead plant residues measured at the soil surface at grapevine flowering (Table 2), and seemed to be the best method for *in situ* mulching practices (Table 2).

## 4.3. Impacts of termination strategies on soil organic matter and microbial biomass

Changes in soil particulate and dissolved organic matter did not exhibit any significant variations across treatments by the end of the experiment (Fig. 4). An upward trend in particulate SOM seemed to manifest in all treatments from the start to the end of the study, but a similar variation was not discernible for dissolved SOM (Fig. 4). Additionally, soil SOM content was not sampled in each treatment at the beginning of the experiment, but rather via a less precise sampling method at the block scale. This made it impossible to verify the Year effect in the evolution of SOM. The accumulation of carbon in the soil is largely determined by the biomass produced by the service crops, but also by its characteristics (C:N ratio). Moreover, years 2021 and 2022 exhibited low rainfall patterns during the service crop growth, and arid conditions are unfavorable for biomass production and the subsequent inputs of carbon into the soils (Blanco-Canqui et al., 2022). Indeed, it is often highlighted in the literature that an increase in SOM might be observable only over an extended period under a permanent cover cropping approach (Celette et al., 2009; Morlat and Jacquet, 2003). Notably, a minimum of three years seems to be the briefest duration within which to observe an increase in SOM (Belmonte et al., 2016;

Marques et al., 2010), even though we found few examples in literature showing short-term effects of cover crops on soil organic carbon in Mediterranean olive grove and vineyard, for soil with low organic matter content (Belmonte et al., 2016; González-Rosado et al., 2020).

Regarding to the microbial biomass measured at the conclusion of the experiment in Block S1, both Termination period and Termination method factors significantly influenced the results. Termination at budburst demonstrated a higher soil MB than early termination (Fig. 4, Table 4), likely a result of the higher biomass returned to the soil and the continued root activity within the soil. In a recent literature review, Kim et al. (2020) demonstrated that the presence of service crops can significantly increase soil microbial abundance, activity, and diversity parameters by 27%, 22%, and 2.5% respectively, compared to bare soil. However, the extent of the effects of service crops seems to vary depending on their management or tillage practices: the effects of service crops appear less pronounced after soil tillage. On average, the B-R and B-M treatments recorded the highest soil MB values, with these findings potentially explained by the combination of late service crop termination and no-till practices (Sun et al., 2020; Zuber and Villamil, 2016).

### 4.4. Impacts of termination strategies on soil water stock and grapevine water status

Establishing service crops represents an effective lever for improving soil infiltration capacity and replenishing plant-available water in soils (Blanco-Canqui and Ruis, 2020). In our study, the Termination period factor was not significant in explaining both the soil water stock at grapevine flowering and the predawn leaf water potential measured between grapevine flowering and veraison. However, the Termination method was a significant factor influencing these two variable variations (Fig. 3, Fig. 5, Table 4, Table A1). Termination involving soil tillage was notably more effective to stop service crop transpiration, unlike mowing and rolling termination methods (Fig. 2). Thus, soil water stocks were highest in the T treatments, lowest in the R treatments, and intermediate in the M treatments (Fig. 4). This could be explained by the reduced efficacy of the roller compared to other methods, and the non-instantaneous halt of transpiration in R treatments. In M treatments, service crop regrowth occurred, but mowing greatly diminished aerial biomass and destroyed plant leaves, hence curtailing transpiration. More contrasting results were observed in predawn leaf water potential, where T treatments stood distinct from M and R treatments (Fig. 5,

Table 3), showing a lower water stress experienced by the grapevine in T treatments. These observations were confirmed by the  $\delta^{13}C$  analysis in grape berries, with T treatments showing less water constraint than M and R treatments (Table 3, Table 5). Notably, the E-T treatment (early termination involving soil tillage) showed a significant difference from all no-till termination methods, but had similar  $\delta^{13}C$  values to the E-B treatment. However, the efficiency of service crops termination method is still debated in literature: for example, Alonso-Ayuso et al., (2020), showed that roller-crimper enhanced the soil water conservation and decreased soil temperature compared to residue incorporation, and decreased energy cost of service crops termination suggesting a higher environmental sustainability of the roller-crimper in Mediterranean regions.

Regarding to the termination period, the most delayed termination strategy we analyzed in this study was at the grapevine budburst, when the grapevine growth was just beginning. This is a common practice in the region mainly explained by concerns about grapevine water stress, but some winegrowers extend the growth of service crops to a later period (e.g. grapevine flowering), or even permanently (Celette and Gary, 2013; Fernández-Mena et al., 2021). The two termination periods tested were relatively close in time (delayed by about 1.5 months), and the grapevine was still dormant, which resulted in no discernible impact of the termination period on soil water stocks and predawn leaf water potential.

The factor Year also significantly affected soil water stocks, with lower stocks in 2022 compared to 2020 and 2021 (Fig. 3, Table A1, Table 4). This observation is likely a consequence of the precipitation pattern in the spring of 2022 rather than the composition and biomass of the mix, as soil water stocks in 2020 were the highest, despite having a service crop biomass approximately twice as high as in 2022 (Fig. 2, Fig. 3). The factor Year was also significant in explaining predawn leaf water potentials (2020 > 2021 = 2022) and  $\delta^{13}$ C values (2020 < 2022 < 2021), but did not interact with the factors Termination period or Termination method to explain variations in these indicators (Table A1, Table 5).

## 4.5. Impacts of termination strategies on soil inorganic nitrogen stock and YAN in berries

In 2020 and 2022, soil inorganic nitrogen stocks were almost 4 times higher in the T treatments compared to the other two termination methods (Fig. 4, Table 3, Table 5). Furthermore, the soil inorganic nitrogen stocks in the T treatments achieved a level of approximately  $61 \text{ kg ha}^{-1}$  (16.25 kg ha<sup>-1</sup> in R and T treatments). This level is notable as it closely corresponds to the annual nitrogen requirements of grapevines (Conradie, 1980; Verdenal et al., 2021). Additionally, this occurred during a period of high nutrient absorption by the plant. Therefore, these treatments present a compelling alternative to external nitrogen inputs. The stock of mineral nitrogen in the soil is the result of all the fluxes in the nitrogen balance, possibly modified by the presence of the service crop. These fluxes are largely determined by the biomass produced by the service cover and the soil cover. Some authors stress the importance of mixing species in order to maximize these two variables (Wortman et al., 2012; Blanco-Canqui et al., 2011). The incorporation of plant residues in T treatments, which speeds up the mineralization of the reintroduced organic matter, as opposed to residues left on the soil surface (Coppens et al., 2006a, 2006b) may explain the differences between termination treatments. For years 2020 and 2022, the R and M treatments exhibited less mineralization, and the regrowth or sustained growth of service crops presumably resulted in the absorption of the soil's inorganic nitrogen (Celette and Gary, 2013).

The interaction between factors Year and Termination method significantly explained soil inorganic nitrogen stocks, likely a consequence of the service crop biomass and species composition. Indeed, the lower proportion of legumes that sprouted in the mixture in 2021, combined with a reduced biomass production compared to other years,

likely accounted for the low soil inorganic nitrogen stock measured across all treatments post-flowering (Fig. 4, e.g. Silvestri et al., 2021). Furthermore, the C:N ratio of the service crops in 2021 was higher compared to 2020 and 2022, particularly at budburst, which may have delayed their mineralization after termination (Justes et al., 2009; Nicolardot et al., 2001).

The yeast-assimilable nitrogen (YAN) content in grape juice mirrored this trend (Table 3), being significantly higher in the T treatments compared to the M and R treatments. Consequently, in the T treatments, the vine was able to utilize the enhanced mineralization resulting from soil tillage, effectively providing a green manure service (Zanzotti and Mescalchin, 2019). However, none of the treatments displayed an average YAN content lower than 140 mg  $\rm L^{-1}$ , a level below which there may be risks for fermentation and wine production (Bell and Henschke, 2005; Bely et al., 1990; Verdenal et al., 2021).

## 4.6. Managing termination strategies to achieve grapevine growth and yield objectives

Pruning weight, as per the entire data set, ranged between 0.34 and 0.92 kg per vine. These values align with the range (0.3-0.6 kg per meter of canopy) suggested by Keller (2015), and found in other studies (e.g. Guilpart et al., 2017; Rives, 2000). However, particularly in 2021, the pruning weight in M and R treatments were at the lower end of this range. Some vines sampled displayed pruning weights under 0.3 (Fig. 7), potentially risking sustainability in the event of recurring drought, like to the conditions experienced in 2021 (Fig. 1, Fig. 7). Regarding the acceptability of the strategy by the winegrowers: as far as the grapevine growth is concerned, we showed, over the three years of the experiment, that the pruning weight displayed significant variation between different termination methods, with T treatments exhibiting a higher pruning weight per vine in comparison to R and M treatments (Fig. 7, Table 5). Moreover, in T treatments, the average pruning weight showed an increase in 2020 and 2022 compared to 2019. This suggests that the tilled service crops increased grapevine vigor these years, compared to the period before the experiment.

In general, the average grapevine yield ranged from 7.25 to 13.7  $\ensuremath{t}$ ha<sup>-1</sup>, corresponding to 52–98 hL ha<sup>-1</sup> (with 4000 vines ha<sup>-1</sup>, 140 kg hL<sup>-1</sup>). This level of production may be considered acceptable for Protected Designations of Origin that limit grapevine yield to 40 or 60 hL ha<sup>-1</sup>, but it could be a limitation for Protected Geographical Indications, which permit 90 hL ha<sup>-1</sup>, or unlabeled productions without yield limitations. The Termination period and the interaction between the Termination method and Year factors significantly influenced the results (Table A1). Furthermore, on average, the grapevine yield decreased for the M and R treatments between 2020 and 2022, while it increased for the T treatment (Fig 6, Table 5), although these differences were not significant (Table 5). Much like the number of bunches, the grapevine yield is also impacted by the abiotic stresses experienced in the preceding year (Guilpart et al., 2017), which could explain the progressive ranking between the three termination methods over the course of the experiment. Prior to the experiment, no cover crops had been sown in the vineyard, which likely explains the lack of significant differences between treatments in 2020. Nevertheless, the stresses endured in 2020 could have significantly impacted the grapevine yield in 2021, and the more severe water stress experienced by the grapevine in 2021 likely had a significantly greater effect on the grapevine yield in 2022 (Fig 6, Table 5).

Only the termination method significantly controlled the number of bunches, a major grapevine yield component (Fig 6, Table A1, Table 5). In general terms, the T treatment had a significantly higher number of bunches, the M treatment had the lowest, and the R treatment exhibited intermediate values (Table 5). This pattern was particularly noticeable in 2021 and 2022 (Fig 6). The number of bunches is related to the level of water and nitrogen stress experienced by the vineyard in the previous year (Guilpart et al., 2014), which likely explains this trend. Despite

these findings, our analysis did not identify a significant Year effect. This outcome might be attributable to the variability of the data relative to the mean effects (Fig 6).

## 4.7. Managing service crop strategy to increase soil functions in the short and long terms and achieve production objectives

Determining the appropriate service crop termination strategy is an annual challenge for the winegrower. Each year, in light of prior constraints and the associated potential yield, the goal is to align the water and nitrogen availability for the grapevine with its needs, typically following the total or partial destruction and/or burial of the service crop. Nevertheless, the mineralization of the organic nitrogen contributed by service crops relies heavily on soil biological activity, which is significantly influenced by climatic factors such as soil moisture and temperature - elements often beyond the control of farmers (Crews and Peoples, 2005).

We observed no notable effect of the termination period on all variables, with the exception of yield and microbial biomass which were higher for the budburst termination period. Terminating at budburst facilitated a greater biomass production from service crops and promoted soil microbial biomass. As early termination may not be necessary to mitigate competition between service crop and grapevine, we recommend termination at budburst to favor soil ecosystem functions linked to microbial activity.

In terms of termination methods, soil tillage seemed to be the most effective strategy to reduce water and nitrogen competition and enhance grapevine vigor and yield, especially during and after dry years. Though the particulate SOM increased across all treatments to the same degree, soil tillage did not favor soil microbial biomass and could potentially hinder other biodiversity components such as arthropods (Inveninato Carmona et al., 2021), or increase soil erosion (Prosdocimi et al., 2016).

Given the Mediterranean climate context, with rising frequencies of drought periods and especially dry winters due to climate change, termination involving soil tillage appears to be the least risky strategy to preserve grapevine vigor and production. However, this might be contingent on the targeted yield and wine valuation. As our study demonstrated, satisfactory yields could be attained with all treatments. Regarding to grapevine vigor, a balance might be achieved by adopting an adaptive termination strategy (Ripoche et al., 2011): ensuring service crop termination with soil tillage during dry years and utilizing a roller-crimper when the winter rainfall pattern is more favorable (Fig. 1, Fig. 7). Selecting species that are easily terminated with a roller crimper, or modifying fertilization practices, could also assist in using this termination method. Lastly, spatial adjustments can aid in reaching a balance between soil microbial activity and grapevine production, by modifying soil coverage at the field scale (Delpuech and Metay, 2018) or implementing different management practices on adjacent inter-rows (e. g. alternating tillage and rolling every other inter-row).

From a broader perspective, in the context of promoting the introduction of service crop, it would be important to estimate the cost of each strategy in terms of labor and energy consumption. Indeed, our study did not include recording of agricultural work times nor energy consumption indicators whereas the issue of energy consumption in systems with or without service crops is controversial, with recent contradictory results. Indeed. soil tillage is often associated with higher energy consumption (Alonso-Ayuso et al., 2020) while other studies (Navarro-Miró et al., 2019) highlighted that the highest energetic consumption in the system including service crop in comparison with organic system was mainly due to the additional operations related to the service crop sowing and management.

#### 5. Conclusion

This study focused on the effect of various termination strategies (early and budburst termination period, and tillage, rolling and mowing

as termination methods) on soil ecosystem functions, vine water and nitrogen status, grapevine vigor and yield.

Regarding soil water stock and grapevine water status, it was found that the termination method was more influential than the termination period. Termination through soil tillage proved to be more successful at halting service crop growth compared to other methods, leading to improved soil water stocks. This improvement corresponded to a lower water stress experienced by grapevines. In terms of nitrogen, the termination involving soil tillage led to a significant increase in soil inorganic nitrogen stocks at grapevine flowering, which were nearly four times higher than the other termination methods. This increase provided a sufficient level of nitrogen that matches the annual nitrogen requirements of grapevines, offering a promising alternative to external nitrogen inputs. Across all termination treatments, an average increase in particulate SOM was observed, indicating the short -term beneficial impact of service crops on enhancing soil organic matter content, irrespective of the termination strategy. Moreover, the budburst termination period combined with no-till termination methods resulted in higher soil microbial biomass, which plays a critical role in nutrient cycling, organic matter decomposition, directly influencing the soil's

Assessing the impacts of the termination strategies on grapevine growth and yield revealed significant variations between the different termination methods. The termination involving soil tillage exhibited an overall higher pruning weight per vine, and a higher grapevine yield the third year of experiment. Consequently, this study stimulates a more comprehensive approach to discerning the impacts of diverse termination strategies (termination date and tool). For instance, exploring the long-term effects of service crop management strategies on soil functions, particularly focusing on the sustainability of organic matter storage to enhance soil structural stability, is promising. Furthermore, a thorough economic analysis could assess the cost-effectiveness of termination strategies, taking into account investment, maintenance costs, and labor. As a complement to field-based analytical experiments, which are time-consuming, a novel approach centered on the development of predictive models to simulate grapevine system functioning integrating service crops is also worth considering. Subsequently, further investigations should incorporate the development of models to simulate water and nitrogen balances in viticultural systems involving service crops. These dynamic models may inform irrigation decisions or fertilization strategies and evaluate the resilience of service crop management in response to climate change. The advancement of such knowledge and methodologies holds promise for broader adoption of cover cropping practices in vineyard plots. These models can facilitate scenario analyses to support decision-making and assess broader impacts on ecosystem services, including soil erosion, biodiversity, and carbon sequestration. Finally, this study underscores the importance of adjusting termination strategies to climatic conditions, the specific needs of the grapevine, and the objectives of winegrowers (e.g. yield and net income). By finding a balance between soil health and grapevine production, sustainable viticulture practices can be promoted.

## Declaration of Generative AI and AI-assisted technologies in the writing process

During the preparation of this work the author(s) used Chat-GPT in order to translate French text into English, or detect English language errors. After using this tool/service, the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the publication.

#### CRediT authorship contribution statement

**Léo Garcia:** Writing – review & editing, Writing – original draft, Visualization, Supervision, Project administration, Methodology, Investigation, Formal analysis, Conceptualization. **Gaïa Krafft:** 

Visualization, Methodology, Investigation, Data curation. **Clément Enard:** Resources, Methodology. **Yvan Bouisson:** Supervision, Resources, Methodology. **Aurélie Metay:** Writing – review & editing, Writing – original draft, Validation, 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

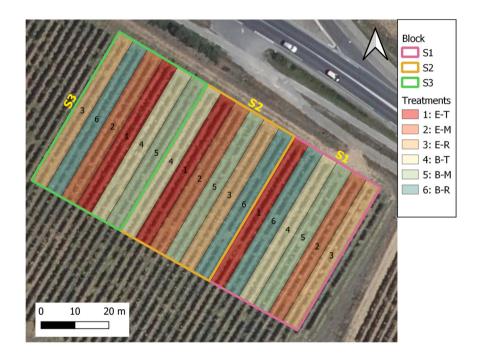


Figure A1. Design of the experiment: blocks and treatments. E: early termination; B: budburst termination; M: mower; R: roller; T: mower+tillage.

Table A1

Anova table for each variable measured during the experiment. Non-significant interactions were removed during the variable selection procedure. Satterthwaite's method was used for models including random effects. ns: non-significant effect; \* significant effect

Term	Sum Sq	DF	Stat	Pvalue	
Water stock at grapevine flowering					
Termination period	310.39	1	1.43	0.238	ns
Termination method	3996.16	2	9.19	< 0.001	***
Year	19761.09	2	45.44	< 0.001	***
Nitrogen stock at grapevine flowering					
Termination period	15.40	1	0.10	0.76	ns
Termination method	13843.19	2	43.35	< 0.001	***
Year	4448.99	2	13.93	< 0.001	***
Termination method:Year	3671.77	4	5.75	< 0.001	***
Soil Microbial biomass					

(continued on next page)

Table A1 (continued)

Term	Sum Sq	DF	Stat	Pvalue	
Termination period	22543	1	5.84	0.0299	*
Termination method	28993	2	3.75	0.0495	*
Particulate soil organic matter					
Termination period	0.003	1	0.043	0.837	ns
Termination method	0.078	2	0.563	0.573	ns
Dissolved soil organic matter					
Termination period	0.342	1	1.870	0.178	ns
Termination method	0.431	2	1.178	0.317	ns
Predawn leaf water potential					
Termination period	0.0096	1	0.845	0.358	ns
Termination method	0.4593	2	20.170	< 0.001	***
Year	3.8454	2	168.860	< 0.001	***
δ13					
Termination period	0.42	1.00	1.09	0.30	ns
Termination method	16.32	2.00	21.33	< 0.001	***
Year	67.26	2.00	87.88	< 0.001	***
Termination period:Termination method	2.44	2.00	3.19	0.0438	*
Yeast Assimilable Nitrogen					
Termination period	329.39	1	0.69	0.41	ns
Termination method	51645.72	2	53.78	< 0.001	***
Year	36845.94	2	38.37	< 0.001	***
Termination period:Year	5336.26	2	5.56	0.0047	**
Number of bunches					
Termination period	27.33	1.00	0.89	0.35	ns
Termination method	371.91	2.00	6.07	0.0025	**
Year	3.23	2.00	0.05	0.95	ns
Yield					
Termination period	6.07	1.00	4.70	0.031	*
Termination method	30.75	2.00	11.90	< 0.001	***
Year	34.79	2.00	13.46	< 0.001	***
Termination method:Year	15.14	4.00	2.93	0.021	*
Pruning weight					
Termination period	0.01	1.00	0.25	0.62	ns
Termination method	8.18	2.00	74.22	< 0.001	***
Year	6.01	2.00	55.01	< 0.001	***

#### Appendix B. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.eja.2024.127161.

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