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Abstract

Purpose: Cover crop nitrogen (N) cycling has an important role in agricultural production and contributes to peach (*Prunus persica* (L.) Batsch.) N nutrition. This study evaluated black oat (*Avena strigosa* Schreb) and ryegrass (*Lolium multiflorum* L.) residue decomposition dynamics, N recovery from cover crop residues, and N compartmentalization in peach tree organs.

Methods: A two-year field trial was developed with labeled (5 atom% ^{15}N excess) cover crop shoot biomass application in a 5-year old peach orchard. The region's climate is warm temperate (Cfb), and the soil is classified as a Typic Hapludalf. Litter bags with unlabeled shoot residues were also deposited in the orchard to assess biomass, carbon (C), N, lignin, cellulose, and non-structural biomass decomposition dynamics.

Results: After 13 months, the leaves, trunk, and roots showed the greatest proportion of N derived from residues (Ndfr) (35.4, 25.1, and 22.4%, respectively) while the greatest concentrations of ^{15}N and Ndfr occurred in roots <2 mm (0.0376 and 0.94%, respectively). The N derived from cover crop shoots in the second production cycle was similar among tree organs. Ryegrass residues presented the highest decomposition constant (k) values for dry matter, total organic carbon (TOC), cellulose, and lignin. Hence, black oat residues presented a higher half-life ($t^{½}$) for dry matter, TOC, total N, cellulose, and lignin.

Conclusions: The N derived from black oat and ryegrass residues in mature trees was expressively low (<1%) and similar between species. Within organs, the highest Ndfr occurred in peach leaves during the flowering stage, when the greatest residue decomposition rate also occurred. Soil N and plant internal N reserves are the major N sources for newly formed organs, but greater contributions to tree N nutrition may occur with long-term cover crop residue deposition and different plant species.

Keywords: *Avena strigosa*, *Lolium multiflorum*, N cycling; ^{15}N recovery, *Prunus persica* (L.) Batsch.

1 Introduction

Nutrient management in orchards has a great impact on plant growth, yields, and fruit quality. Sustainable soil fertility management in orchards requires not only a fine-tuning of fertilizer rates, but also a higher use efficiency of nutrients already present in soil (Tagliavini 2012). There is a multitude of factors, including the cycling of plant residues, that dictate nutrient availability in the soil system. A key step to improving nutrient use efficiency is understanding the nutrient's fate during leaf-litter decomposition on the soil surface and the ability of that nutrient to become available for plant uptake (Tagliavini and Scandellari 2013).

Nitrogen (N) is a nutrient with a great impact on growth and reproductive development in fruit trees, such as peach [*Prunus persica* (L.) Batsch.], as it directly affects the flowering quality, fruit set, fruit quality, and yield potential. However, mature peach trees tend to absorb only small amounts of the N derived from fertilization (Policarpo et al. 2002) as the majority of the N used by the new vegetative organs comes from internal N reserves. Part of the N-fertilizer applied to the orchard soils can be lost by volatilization (especially when using urea as N source) (Roccuzzo et al. 2017), leaching (greater loss potential in sandy soils with low organic matter content) (Lorensini et al. 2012), and surface runoff (especially in areas with steep grades) (Martínez et al. 2006). Moreover, greater losses can occur when the N-fertilizers are not applied at the right time and rate to meet trees' nutritional demand.

Cover crops, such as black oat (*Avena strigosa* Schreb.) and ryegrass (*Lolium multiflorum* L.), are used in the inter-row space of orchards to dissipate the kinetic energy of rainfall and reduce soil erosion. Throughout tree development, cover crop shoots are frequently cut and deposited on the soil surface contributing to increasing soil organic matter (SOM)

52 content and nutrient cycling (Brunetto et al. 2017; Ferreira et al. 2014).

53 During the decomposition of cover crop residues, part of the carbon (C) present in the
54 plant tissue may remain in the soil, especially from residues with lower lability. However, the
55 majority of the C will probably return to the atmosphere as CO_2, depending on the residue
56 composition and mineralization rate (Oliveira BS et al. 2016; Reichert et al. 2015). Part of the
57 plant tissue N can increase soil available N (nitrate or ammonium), which can be absorbed by
58 mature trees. The decomposition of plant material and the release of N from cover crop residues
59 depends on the residue biochemical composition, especially the cellulose and lignin content,
60 but also the C/N, lignin/N, and cellulose/lignin ratios (Carranca et al. 2009). Residues with low
61 cellulose content, high lignin content, and high C/N ratio typically have a low decomposition
62 rate and may even temporarily immobilize soil N (Bonanomi et al. 2013). On the other hand,
63 more labile residues with higher cellulose content, lower lignin content, and lower C/N and
64 cellulose/lignin ratios can mineralize N to the soil, increasing the plant-available N forms
65 (Cabrera et al. 2005). The residue decomposition and nutrient release is also dependent on
66 edaphoclimatic characteristics, especially soil texture, moisture, aeration, temperature, and
67 nutrient availability, which directly affect soil microbial activity (Brunetto et al. 2011; Cabrera
68 et al. 2005; Ferreira et al. 2014).

69 Studies investigating the N derived from cover crop residues can be accurately
70 performed using the ^{15}N isotope dilution technique (Brunetto et al. 2014; Neto et al. 2008;
71 Tagliavini et al. 2007). Annual tree organs such as leaves, twigs of the year, and fruits are major
72 sinks for the absorbed N derived from in-season fertilization (Brunetto et al. 2017), SOM
73 mineralization, and decomposing cover crop residues. Part of the N stored in perennial organs
74 is remobilized to the tree meristematic tissues in the subsequent growth cycle, reducing the tree
75 N needs from N fertilizers at this vegetative phase. Even though the contribution of cover crops
76 for plant nutrition has been evidenced for different crops and production systems (Amossé et

al. 2014; Brunetto et al. 2011, 2014; Oliveira RA et al. 2016; Ovalle et al. 2010), information on how the N derived from cover crop residues is redistributed in different peach organs is still scarce.

We hypothesize that a) ryegrass and black oat residues will present similar decomposition dynamics, and b) the N released will be used by trees and preferentially redistributed to newly formed organs and trunk. Thus, this study aims to evaluate a) the recovery of N derived from the decomposition of black oat and ryegrass shoot residues in mature trees, b) the N absorption dynamics and the N compartmentalization in different tree organs, and c) to assess black oat and ryegrass shoot residue decomposition dynamics in a mature peach orchard.

2 Materials and methods

2.1 Cover crop cultivation

Soil for this experiment was collected from the 0-0.20 m depth of a soil classified as a Rhodic Paleudalf (Soil Survey Staff 2014) in Southern Brazil (29°42'54" S, 53°42'25" W). The soil was air-dried, ground to pass through a sieve with 2-mm openings, and reserved for analysis. Selected soil chemical and physical characteristics are shown in Table 1. Polypropylene pots (20.0 cm x 25.0 cm) with four kilograms of soil were used to cover crop cultivation.

Seeds of black oat and ryegrass were pre-germinated in plastic germination boxes (Gerbox®) by placing the seeds on paper towels and moistening them with distilled water amounting to 2.5 times the weight of the dry paper. The seed boxes were placed in a BOD (Biochemical Oxygen Demand) chamber at a constant temperature of 25°C and a positive photoperiod of 8 h. A germination test was carried out from 01 May to 13 May 2014, when 25 germinated seeds of each plant species were sown in the polypropylene pots. Seven days after

emergence, the less developed seedlings were thinned, leaving 20 black oat or 20 ryegrass plants in each pot.

Urea containing 46.6% N, labeled with 5 atom% ^{15}N, was applied to 15 pots containing black oat and 15 pots containing ryegrass at a rate of 10 g of N m^{-2}. Unlabeled urea with 45% N was applied to an additional 15 pots grown with black oat and 15 pots grown with ryegrass (control plots). Urea, with and without ^{15}N, was diluted in deionized water and applied to the soil, thereafter pots were irrigated daily with deionized water to maintain the soil at 70% field capacity (Casaroli and Jong van Lier 2008) during the experimental period. The N-fertilizer solution application was split six times throughout the cultivation period: 10, 17, 24, 31, 38, and 45 days after thinning. At 18 and 41 days after thinning, 50 mL of a nutrient solution containing 500, 502, 48, 19, 9, and 14000 mg L^{-1} of B, Mn, Zn, Cu, Mo, and KH_2PO_4, respectively, were applied to each pot. Throughout the crop cycle, weeds were removed by hand from the pots.

Sixty-three days after sowing (*i.e.*, 17 July 2014), the shoots of black oat and ryegrass grown with and without ^{15}N applications were cut close to the soil surface. The cover crop shoot residues were washed and oven-dried at 65 °C until constant weight. The cover crop ^{15}N-labeled shoot residues were reserved for Experiment 1, while the unlabeled shoot residues were reserved for Experiment 2. Subsamples of both ^{15}N-labeled and unlabeled residues were ground with a Wiley mill and separated for chemical characterization and ^{15}N analysis.

2.2 Experiment 1 - Mature trees N recovery from black oat and ryegrass shoot residue decomposition

2.2.1 Experimental layout

The experiment was carried out in a mature peach orchard of ‘Chimarrita’ [*Prunus persica* (L.) Batsch] (Raseira et al. 2014) grafted on 'Capdeboscq' [*Prunus persica* (L.) Batsch]

rootstock (Mayer et al. 2014). The orchard was established in 2009 in the experimental area of Embrapa Uva e Vinho, located in the city of Bento Gonçalves, Southern Brazil (29º09'44" S, 51º31'50" W). Before the installation of the orchard, 30 kg P_2O_5 ha^{-1} (as triple superphosphate) and 40 kg K_2O ha^{-1} (as potassium chloride) were applied to increase soil-test P and K to optimum levels. Subsequently, the orchard received annual fertilizer rates of 90 kg N ha^{-1} (as urea), 40 kg P_2O_5 ha^{-1} (as triple superphosphate), and 80 kg K_2O ha^{-1} (as potassium chloride) to maintain adequate nutrient levels according to the regional production guidelines (CQFS - RS/SC, 2004). After the installation of the experimental area, the adjacent plants received no fertilization. The spacing between tree rows and between plants within rows was 4.0 and 1.5 m, respectively, totaling 1667 plants per hectare. The Y-shaped pruning system was used for trees. The climate of the region is warm temperate (Cfb), according to the Köppen-Geiger classification system, with an average air temperature of 17.1°C and an average annual rainfall of 1755 mm (Fig. 1). The soil is classified as a Typic Hapludalf (Soil Survey Staff 2014) and its selected physical and chemical characteristics are presented in Table 1. The local landscape is gently sloping.

Before the experiment set up, the orchard included cover crops, mainly grasses (*e.g.,* black oat and ryegrass). In the evaluation area, however, all vegetation, including roots, was removed, so that no plants were competing with the trees. On 17 July 2014, ^{15}N-labeled shoot residues of black oat and ryegrass (sampled 63 days after sowing) were deposited at the soil surface to an area of 0.96 m^2 (0.8 m x 1.2 m) surrounding the tree trunk. Each plot was composed of three trees that each received 0.13 kg dry matter of either black oat or ryegrass shoot residue, corresponding to 1354 kg of dry matter per hectare. A nylon net (2 mm mesh) covered the residue and was fixed to the soil with metal clamps to prevent residue displacement by the wind, rain, and animals. For the duration of the experiment, non-residual herbicide was applied to the area of residue deposition to manage weeds and avoid ^{15}N absorption by weeds.

2.2.2 ^{15}N recovery by mature trees

To follow the pattern of ^{15}N uptake during the mature tree development, the first peach leaf sampling was carried out on 17 August 2014 (30 days after cover crop shoot residue deposition), while subsequent leaf sampling was performed monthly from September 2014 to April 2015. At each leaf sampling time, twenty mature leaves per plant were collected from the middle position on twigs of the year, surrounding the tree canopy. The leaves were oven-dried with forced air at 65 ºC until constant weight, ground, and reserved for total N and ^{15}N analysis.

On 17 December 2014,115 days after cover crop shoot residue deposition, a PVC pipe (250 mm diameter) was inserted into the soil to collect a soil core sample from the 0-0.20 m depth of each plot. Three soil cores were collected in the central position at one side of the trees row, where ^{15}N-labeled black oat and ryegrass shoot residues were deposited. Each soil core was separated into 0.05 m layers (*i.e.*, 0-0.05, 0.05-0.10, 0.10-0.15, and 0.15-0.20 m depths) and combined for a composite sample of each depth. Thereafter, the soil samples were air-dried, ground to pass a 2-mm sieve, and reserved for total N and ^{15}N analysis.

On 23 November 2015 (15 months after cover crop shoot residue deposition), the trunks of trees that received cover crop residue were cut close to the soil surface and separated into the stem, twigs of the year, 2-year-old branches, and leaves. The fruits were collected from each tree and weighed to determine fruit yield. Thereafter, ten fruits per tree were randomly selected to determine total N content and ^{15}N enrichment in the pulp (*i.e.,* mesocarp plus exocarp). The tree roots were removed from the soil, washed with distilled water, and separated into three diameter classes (< 2 mm, 2 - 5 mm, > 5 mm). Root material was reserved for analysis of total N and ^{15}N enrichment. The same stratified soil sampling procedure and analysis, described above, was repeated before peach roots were removed from the soil.

2.3 Experiment 2 - Black oat and ryegrass shoot residue decomposition and N release in

a mature peach orchard

2.3.1 Experimental layout

Experiment 2 was laid out in the same orchard used for Experiment 1. Unlabeled black oat and ryegrass shoot residues were placed in nylon litter bags (2 mm mesh) covering an area of 0.16 m^2 (0.4 m x 0.4 m) and deposited on 22 July 2014 on the soil surface of planting rows. Each plot was composed of four trees that received either black oat or ryegrass shoot residue. Each tree received 64 g of dry matter from black oat or ryegrass shoot residue, corresponding to 4,000 kg dry matter per hectare. The bags were fixed to the soil with a metal clamp to increase the area of contact with the soil surface and prevent displacement by the wind. The litter bags were collected at 30-day intervals (*i.e.,* 0, 30, 60, 90, 120, 150, and 180 days after deposition), and at each sampling time, four litter bags of each cover crop were removed from the field. In the laboratory, the residues were washed to remove the adhering soil particles and then oven-dried at 65 ºC, ground, sieved through 2-mm openings, and reserved for further analysis.

2.4 Plant and soil analysis

Before cover crop residue deposition in the experiments, a subsample was subjected to sulfuric acid digestion to determine total N, phosphorus (P), potassium (K), calcium (Ca), and magnesium (Mg) contents (Tedesco et al. 1995). Total N was determined by steam distillation (TE-0364, Tecnal, Brazil). Total P was determined in a spectrophotometer (Bell Photonics, 1105, Brazil) at 882 nm (Murphy and Riley 1962). Total K was determined in a flame photometer (Digimed, BM-62, Brazil). Calcium and Mg were determined in an atomic absorption spectrophotometer (PerkinElmer, AAnalyst 200, USA). Total organic carbon (TOC) was determined by wet combustion (Yeomans and Bremner 1988). The determination of lignin, cellulose, and non-structural biomass was performed according to the methodology described by Aber and Martin (1999). Total N and ^{15}N were analyzed in an elemental analyzer (Thermo

Scientific, Flash EA 1112, Milan, Italy) and by isotope-ratio mass spectrometry (Thermo Scientific, Delta V Advantage, Bremen, Germany), respectively. Cover crop shoot residue chemical characteristics are presented in Table 2.

The tree organs' tissue and soil samples from Experiment 1 were also analyzed for total N and $\%^{15}N$ following the above-mentioned procedures. The remaining cover crop residues from litter bags in Experiment 2 were also analyzed for C content, lignin, cellulose, and non-structural biomass following the above-mentioned methodologies. The decomposition of residue dry matter and the release of C, N, cellulose, lignin, and non-structural biomass were estimated by subtracting the initial content from the amount determined after each sampling time.

2.5 Calculations and statistical analysis

Atom% ^{15}N excess in soil and plant tissue samples was calculated based on the natural ^{15}N abundance (Mariotti 1983). The N derived from residue (Ndfr) and the N derived from soil (Ndfs) was calculated by the following equations:

$$^{15}N\ excess\ in\ sample\ (\%) = \%\ ^{15}N\ in\ sample - 0.3663\% \quad \text{(Eq. 1)}$$

$$Ndfr\ (\%) = (\%\ ^{15}N\ excess\ in\ sample\ /\ \%\ ^{15}N\ excess\ in\ residue)\ x\ 100 \quad \text{(Eq. 2)}$$

$$Ndfs(\%) = 100 - Ndfr \quad \text{(Eq. 3)}$$

The residual percentage of each variable (total C and N, cellulose, lignin, and non-structural biomass) were adjusted by the exponential mathematical model described by Wieder and Lang (1982):

$$X = X_0^{(-kt)} \quad \text{(Eq. 4)}$$

Where: X is the amount of dry matter or nutrient remaining in the residue after a period t (days); X_0 is the initial amount of dry matter or nutrient in the residue; k is the decomposition constant.

The half-life ($t^{½}$) was calculated with the value of k (Paul and Clark 1996) (Equation 5). The $t^{½}$ expresses the time required for half of the residue to decompose and half of the nutrients contained in the residue to be released.

$$t^{1/2} = 0.693/k \quad \text{(Eq. 5)}$$

All data were submitted to normality and homogeneity of variance by the Lilliefors and Shapiro-Wilk tests prior to the analysis of variance (ANOVA). The dry matter, atom% ^{15}N, total N (mg tree^{-1}), and total soil N (Experiment 1) variables were transformed [log10(x)] to fit a normal distribution before running the ANOVA. Experiment 1 had a completely randomized block design with five replicates. ANOVA was performed to determine the influence of black oat and ryegrass shoot residues on leaf total N, atom% ^{15}N excess, and Ndfr for each leaf's sampling time. ANOVA was also conducted to determine the influence of black oat and ryegrass shoot residue, tree organs, and their interaction on dry matter, total N, atom% ^{15}N excess, Ndfr, and Ndfs. Experiment 2 had a completely randomized block design with five replicates. ANOVA was performed to determine the influence of black oat and ryegrass shoot residues on the remaining dry matter, TOC, total N, cellulose, lignin, and non-structural biomass decomposition constant rate (k) and half-life (t½). Means were compared by the Tukey-test ($p < 0.05$). When no difference was observed among cover crop species, the mean

252 value of the treatments was compared. ANOVA was also conducted to determine the influence
253 of black oat and ryegrass shoot residues, residue deposition time, and their interaction on C/N
254 ratio, dry matter, TOC, and total N. When the interaction was significant, two times the standard
255 error of means (SEM) was used as the minimum difference between means statistically different
256 for $p \leq 0.05$.
257
258 **3 Results**
259 **3.1 Experiment 1 - Mature trees N recovery from black oat and ryegrass shoot residue**
260 **decomposition**
261 Following the pattern of leaf response of mature trees cultivated in soils with black oat
262 and ryegrass ^{15}N-labeled shoot residue deposition, the highest N concentration was found in
263 trees cultivated with ryegrass shoot residue, especially in November and December 2014 (Fig.
264 2a). However, the highest leaf N concentration occurred in August 2014, shortly after the
265 deposition of both cover crop residues on the soil surface. Thereafter, leaf N concentration
266 decreased over time in all trees. The highest atom% ^{15}N and the Ndfr were observed in
267 September in leaves of trees that received the deposition of black oat residue (Fig. 2b and 2c).
268 In subsequent evaluations, there were no differences for leaf concentration of total N, atom %
269 ^{15}N, and % Ndfr between trees cultivated under both cover crop treatments.
270 After 75 weeks (*i.e.*, December 2015), the dry matter, atom % ^{15}N, and Ndfr of mature
271 peach organs did not differ between the two treatments, but significant differences were
272 observed within tree organs (Table 3). The highest total N concentration occurred in mature
273 tree leaves. The highest dry matter yield was observed in the peach trunk, followed by roots >
274 5 mm and leaves (7677.6, 2243.1, and 1212.2 g tree^{-1}, respectively). Mature trees leaves and
275 trunk presented the highest Ndfr (mg tree^{-1}) and Ndfs (mg tree^{-1}) while the highest atom % ^{15}N
276 excess and % Ndfr were observed in roots < 2 mm.

277 Soil characteristics were not affected by the type of cover crop residue, but differences
278 were observed among the soil layers (Table 4). For the mean effect of crop residues, the soil
279 samples collected at 23 and 75 weeks (*i.e.*, 115 and 375 days, respectively) after cover crop
280 residue deposition showed the highest atom% ^{15}N and Ndfr at the 0-0.05 m depth (Table 4),
281 with little variation among other soil depths. It should be noted that 23 weeks after the
282 deposition of black oat and ryegrass shoot residues, atom% ^{15}N and Ndfr values were higher
283 than the values observed 75 weeks after the residue deposition. Overall, the percentage of soil
284 Ndfr below the 0.05-m depth was low.
285
286 **3.2 Experiment 2 - Black oat and ryegrass shoot residue decomposition and N release in**
287 **a mature peach orchard**
288 The temporal dynamics of shoot residue dry matter, TOC, N, lignin, cellulose, and
289 non-structural biomass contents were explained by the exponential decay model (Eq. 4). Cover
290 crop residue dry matter and TOC, N, cellulose, lignin, and non-structural biomass contents
291 decreased rapidly during the experimental period, with slight differences between cover crop
292 species (Fig. 3). At the end of the trial (*i.e.*, 150 days after the residue deposition), only
293 approximately 15% of the cover crop residue dry matter and TOC remained in the soil surface
294 (Fig. 3a and 3b, respectively). There was an 82 and 79% decrease in the N content of black oat
295 and ryegrass remaining residues, respectively (Fig. 3c). Lignin and cellulose presented
296 comparable decomposition dynamics, with remaining percentages close to zero after 150 days
297 of residue decomposition. The remaining non-structural biomass followed a similar trend as
298 dry matter, TOC, and total N, with slightly higher values (20 and 30% for black oat and ryegrass
299 shoot residues, respectively) at the end of the experiment (Fig. 3f). The highest *k* values for dry
300 matter, TOC, cellulose, and lignin were observed for ryegrass shoot residue (Table 5). Black
301 oat residue presented the highest $t^{½}$ values for dry matter, TOC, total N, cellulose, and lignin

(Table 5). There was no statistical difference for *k* and $t^{½}$ values of the non-structural biomass in both cover crop residues. The C/N ratio of litter bag residues over time was greater in black oat than in ryegrass, except at 90 and 180 days after deposition, when the two cover crops presented similar values (Table 6). The C/N ratio constantly decreased until 90 days of deposition, then remained stable for a month and slightly increased at the end of the trial. Black oat shoot residue showed greater dry matter than ryegrass, with values decreasing over time in both cover crops. Total organic C decreased across time and was different between cover crop species. Total organic C of black oat shoot residue was higher than ryegrass from 30 to 120 days after deposition; at the beginning of the experiment, ryegrass showed higher values than black oat, while at the end of the experiment (150 and 180 days after deposition), the values were similar between the two cover crops species. Black oat total N content was higher than ryegrass at 0, 30, 90, and 120 days after deposition; in other sampling times, the situation was inverted. The remaining residue N content decreased over time, the only exception was black oat that showed a slight increase 90 days after deposition and then again decreased until the end of the trial (Table 6).

4 Discussion

Different from annual crops, perennial plants, such as fruit trees, use two main sources of N for their vegetative growth and reproduction: the root N uptake and the internal N cycling (Carranca et al. 2018). This behavior can be observed in our study by the highest total N concentration in mature tree leaves at the first sampling time, which is probably related to the plant's internal N reserves. Likewise, the highest N derived from cover crop residues measured in leaves at 30 and 60 days after black oats and ryegrass shoot deposition, respectively, indicates that the cover crop residues were rapidly decomposing and releasing N to the soil, which was taken up by trees during an active absorption phase. This timing coincided with the flowering

stage when plants increase young root growth, which is responsible for the absorption of water and nutrients, including soil N (Bravo et al. 2012). The trees' superficial roots, which were in proximity with the cover crops' decomposing residues, may have contributed to a greater extent to the N absorption at this stage. Furthermore, this behavior indicates that synchronizing cover crop termination (*e.g.,* mowing) so that a greater decomposition rate occurs in a period of high tree N demand can contribute to enhancing N recovery.

A higher soil N availability favors plant dry matter production by increasing the formation and growth of new vegetative organs such as leaves and twigs, which have a high nutritional N demand due to intense cell division and elongation (Nario et al. 2003; Roccuzzo et al. 2017). However, the highest leaf N content in mature trees at different phenological stages was mostly derived from other N forms than cover crop shoot residue decomposition. This is because the mineral N fertilization that the orchard received over the years may have had a residual effect on our study. The low N recovery from the cover crop shoot residue decomposition by the peach organs might also be related to N losses from the plant-soil system, especially by denitrification, runoff, and leaching (Carranca et al. 2018; Roccuzzo et al. 2017) as only a small proportion of Ndfr (below 0.8%) remained in the soil. On the other hand, the high N content in newly formed organs may also be a response to internal N remobilization.

The similar behavior observed for the black oat and ryegrass shoot residue decomposition dynamics, dry matter, organic C, N, cellulose, lignin, and non-structural biomass contents is related to the residue initial lignin concentration and C/N ratio (Table 2). The decrease of dry matter, organic C, N, cellulose, lignin, and non-structural biomass that occurred over time is mediated by the activity of soil fauna and the degradation by the microbial population (Carranca et al. 2009; Nguyen and Marschner 2017; Oliveira RA et al. 2016). During the decomposition period, cover crop residues had a C/N ratio below 20 (Table 6) at all sampling times, which facilitates the colonization and mineralization by microbial population

352 (Ferreira et al. 2014; Oliveira RA et al. 2016). Consequently, the cover crop shoot residue dry
353 matter reduction over the 150 days of the trial contributed to the decrease of soil cover (this if
354 there is no deposition of residues in short intervals of time) and the potential to dissipate the
355 kinetic energy of the raindrops, leaving the soil more susceptible to wind and water erosion
356 (Ferreira et al. 2014; Oliveira RA et al. 2016), and therefore N loss by runoff. This process was
357 probably even more intense in the surrounding area of the tree, without cover crop residues.
358 Frequent rainfall and warm temperatures during spring and summer are known to increase soil
359 microbial activity and therefore intensify residue decomposition rate (Chen et al. 2020). Hence,
360 continuous cover crop residue deposition in orchards is paramount to increase both soil cover
361 and SOM content, while reducing nutrient losses over time, which ultimately will positively
362 affect N use efficiency.

363 Using cover crops in orchards is economically feasible since, in addition to the
364 protection against soil erosion, the residue deposition can contribute to the cycling of different
365 nutrients, increasing soil health and potentially reducing the costs associated with fertilizer
366 application. On average, during the experimental period, black oat and ryegrass residues
367 released an equivalent of 120 kg N ha^{-1} and 136 kg N ha^{-1}, corresponding to 261 and 296 kg
368 urea ha^{-1}, respectively. However, this is a substantial amount of N released in 150 days for the
369 present climatic conditions, and therefore a major part of the N derived from cover crop residue
370 was probably lost from the soil-plant system, which is supported by the low N recovery by
371 mature trees. The greater N recovery by mature trees during the first 60 days was related to the
372 50% N released from both cover crops during this period (see Table 6 and Fig. 3), which
373 corresponds to the flowering phase where new roots are active and are responsible for water
374 and nutrient uptake (Bravo et al. 2012). Furthermore, during this phase the trees present an
375 intensive leaf growth, which increases leaf area and consequently surface area for transpiration,
376 resulting in a greater accumulation of minerals within the plant body. These findings agree with

377 those observed in other fruit studies, such as the grapevines, in soils amended with other types
378 of residue (Brunetto et al. 2011, 2014).
379 The overall contribution of cover crop residue to peach nutrition in our study was low
380 (below 1%). However, a greater contribution might occur with different cover crop species and
381 residue management that synchronizes the residue N release with trees nutrient demand. It is
382 also important to highlight that even with a low contribution to N nutrition in a short term, cover
383 crops play an important role in cycling other nutrients such as P, K, and S, increasing SOM
384 content and reducing soil and water loss in the system, which is paramount for sustainable food
385 production.
386
387 **5 Conclusions**
388 The highest percentage of N derived from the decomposition of black oat and ryegrass
389 shoot residues occurred in leaves at the flowering stage, *i.e.,* about 30 to 60 days after the shoot
390 residues deposition on the soil surface. The majority of peach leaf N and the N in other young
391 tree organs was derived from other N sources than the cover crop residues, such as soil available
392 N, organic matter, plant internal N reserves, or even the residual effect of mineral fertilizers
393 applied to the trees adjacent to the study area. This is partially explained by the shoot residue
394 decomposition rate on the soil surface, which was reduced to 25-50% during the flowering
395 period.
396 Most of the N contained in leaves in the first evaluation cycle and annual and perennial
397 organs in the second evaluation cycle was also derived from other N sources than cover crop
398 decomposing shoot residue. It is expected that cover crop grasses with a similar C/N ratio to
399 black oat and ryegrass will have comparable residue decomposition dynamics and contribution
400 to tree N nutrition.
401 Further studies need to be performed to evaluate other cover crop species and residue

402 management to synchronize cover crop N mineralization with the stages of greater plant N
403 demand to avoid N losses and increase the N recovery and plant nutritional status.
404
405 **Conflict of Interest** The authors declare that they have no conflict of interest.
406
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525 **Table 1** Selected physical and chemical characteristics of experimental soils at the 0-0.20 m
526 depth

Soil properties	Unit	Experiment1 and 2
		0-0.20 m layer
Clay (pipette method)	g kg^{-1}	333
Silt (pipette method)	g kg^{-1}	405
Sand (pipette method)	g kg^{-1}	262
Organic C (Walkley and Black 1934)	g kg^{-1}	17.3
pH in H_2O (1:1)	-	5.2
Total N (Kjeldahl method)	g kg^{-1}	2.0
NO_3^--N (extracted by KCl 1 mol L^{-1})	mg kg^{-1}	34
NH_4^+-N (extracted by KCl 1 mol L^{-1})	mg kg^{-1}	56
Alkaline hydrolizable N (Roberts et al. 2009)	mg kg^{-1}	183
Aluminum (exchangeable) (extracted by KCl 1 mol L^{-1})	mg kg^{-1}	15
Magnesium (exchangeable) (extracted by KCl 1 mol L^{-1})	mg kg^{-1}	150
Calcium (exchangeable) (extracted by KCl 1 mol L^{-1})	mg kg^{-1}	760
Phosphorus (available) (extracted by Mehlich 1)	mg kg^{-1}	15
Potassium (available) (extracted by Mehlich 1)	mg kg^{-1}	100

527

528 **Table 2** Chemical characterization of black oat and ryegrass shoot residues at the beginning of
529 the experiment, and the amount of dry matter and nutrients added to the soil for residues
530 decomposition (0-20 cm depth).

Variable	Black oat	Ryegrass
	---------- g kg^{-1} ----------	
TOC[a]	463.2±3.5[k]	431.9±2.0
Total N[b]	42.9±07	47.1±0.6
P[c]	4.3±0.2	4.0±0.2
K[d]	24.6±0.4	27.9±1.7
Ca[e]	4.9±0.2	5.4±0.1
Mg[f]	5.8±0.1	5.6±0.1
Cel[g]	388.9±0.6	444.7±1.2
Lig[h]	141.4±0.3	140.5±1.8
Bio[i]	569.7±0.3	414.8±0.8
C/N	10.8±0.2	9.2±0.1
Lig/N	1.0±0.01	3.0±0.04
C/P	109.0±5.9	108.3±5.7
Cel/Lig	9.4±0.8	3.2±0.6
At % N^{15}	40.1±0.1	36.0±0.04
	Amount of residue and nutrients added to the soil surface (kg ha^{-1})	
DM[j]	5176.8	5607.4
TOC	220.1	228.9
N	13.6	14.9
P	2.4	2.2
K	20.1	22.0
Ca	2.0	2.5
Mg	2.1	2.5

531 [a]Total organic carbon; [b]Total nitrogen; [c]Total phosphorus; [d]Total potassium; [e]Total calcium; [f]Total magnesium;
532 [g]Cellulose; [h]Lignin; [i]Non-structural biomass; [j]Dry matter; [k]mean standard error (n = 3)

533 **Table 3** Dry matter, total N, atom ^{15}N excess, ^{15}N derived from residues (Ndfr), and N
534 derived from other sources (Ndfs) in mature peach trees organs after 375 days of black oat
535 and ryegrass shoot residues deposition on the soil surface (Experiment 1)

Cover crop	Peach tree organs								CV (%)
	Pulp	Leaves	Twigs of the year	Branches of the year	Trunk	Roots > 5 mm	Roots 2 - 5 mm	Roots < 2 mm	
					Dry matter (g tree $^{-1}$)				
Black Oat	54.43	1243.18	523.14	1163.23	6387.33	1891.52	137.58	145.95	
Ryegrass	55.33	1181.28	556.66	985.35	8967.83	2594.65	111.17	98.33	
Average	*54.88f*[(1)]	*1212.23bc*	*539.90d*	*1074.29c*	*7677.58a*	*2243.08b*	*124.37e*	*122.14e*	*6.12*[(2)]
					Total N (g kg^{-1})				
Black Oat	11.16	26.56	7.44	4.07	2.82	5.90	8.04	8.65	
Ryegrass	12.18	27.87	7.69	4.47	3.12	8.83	11.22	10.54	
Average	*11.67b*	*27.22a*	*7.60d*	*4.27e*	*2.98f*	*7.37d*	*9.63c*	*9.60c*	*7.36*
					Total N (mg tree^{-1})				
Black Oat	575.39	33012.08	3839.95	4733.8	18244.51	11110.02	1109.17	1257.05	
Ryegrass	671.81	32957.52	4288.27	4420.87	28052.09	22870.01	1228.49	1027.03	
Average	*623.60e*	*32984.80a*	*4064.11c*	*4577.33c*	*23148.30ab*	*16990.01b*	*1168.83d*	*1142.04d*	*5.5*
					atom ^{15}N excess (%)				
Black Oat	0.019	0.0138	0.0164	0.0211	0.0156	0.0216	0.0279	0.0387	
Ryegrass	0.017	0.0170	0.0154	0.0190	0.0151	0.0172	0.0266	0.0364	
Average	*0.0180c*	*0.0154c*	*0.0159c*	*0.0200bc*	*0.0153c*	*0.0194bc*	*0.0272b*	*0.0376a*	
					^{15}N derived from shoot residues (Ndfr) (% total N in the organ)				
Black Oat	0.47	0.35	0.41	0.53	0.39	0.54	0.7	0.96	
Ryegrass	0.42	0.42	0.38	0.47	0.37	0.43	0.66	0.91	
Average	*0.45 bc*	*0.38 c*	*0.39 c*	*0.50 bc*	*0.38 c*	*0.48 bc*	*0.67 ab*	*0.94 a*	
					^{15}N derived from shoot residues (Ndfr) (mg tree^{-1})				
Black Oat	2.69	110.88	15.99	25.17	73.86	60.21	7.98	12.01	
Ryegrass	2.84	140.2	16.46	21.09	104.39	98.99	8.4	8.92	
Average	*2.76e*	*125.54a*	*16.22bc*	*23.13b*	*89.12a*	*79.60a*	*8.19cd*	*10.46bc*	*21.1*
					^{15}N derived from other sources (Ndfs) (% total N in the organ)				
Black Oat	99.53	99.65	99.59	99.47	99.61	99.46	99.3	99.04	
Ryegrass	99.58	99.58	99.62	99.53	99.63	99.57	99.34	99.09	
Average	*99.56ab*	*99.62a*	*99.60a*	*99.50ab*	*99.62a*	*99.52ab*	*99.32b*	*99.06c*	
					^{15}N derived from other sources (Ndfs) (mg tree^{-1})				
Black Oat	572.71	32901.20	3823.96	4708.63	18170.65	11049.81	1101.19	1245.04	
Ryegrass	668.97	32817.32	4271.81	4399.78	27947.71	22771.01	1220.09	1018.12	
Average	*620.84e*	*32859.26a*	*4047.89c*	*4554.20c*	*23059.18ab*	*16910.41b*	*1160.64d*	*1131.58d*	*5.49*

536 [(1)]Means (n = 5) followed by the same lowercase letter in the column, for each plant organ, do not differ by the
537 Tukey-test ($p > 0.05$).[(2)]Coefficient of variation (CV) of cover crop shoot residue error 1

538 **Table 4** Soil total nitrogen (TN) concentration, atom ^{15}N excess, and ^{15}N derived from residues
539 (Ndfr) in the 0-0.20 m depth of a mature peach tree orchard with black oat and ryegrass shoot
540 residues deposition (Experiment 1)

Cover crop	Soil depth (m)				
	0-0.05	0.05-0.10	0.10-0.15	0.15-0.20	CV (%)
	23 Weeks (i.e., 115 days)				
	Total N (g kg^{-1})				
Black Oat	4.1	3.3	2.0	1.8	
Ryegrass	4.7	2.6	2.0	1.3	
Average	*4.4 a*[(1)]	*2.9 b*	*2.0 c*	*1.6 c*	*6.22*[(2)]
	^{15}N (atom% ^{15}N excess)				
Black Oat	0.0224	0.0097	0.0056	0.0051	
Ryegrass	0.0406	0.0098	0.0057	0.0050	
Average	*0.0215 a*	*0.0097 b*	*0.0056 b*	*0.0050b*	*0.40*
	Ndfr (%)				
Black Oat	0.56	0.24	0.14	0.13	
Ryegrass	1.01	0.24	0.14	0.13	
Average	*0.79 a*	*0.24 b*	*0.13 b*	*0.13 b*	*5.92*
	75 Weeks (i.e., 375 days)				
	Total N (g kg^{-1})				
Black Oat	4.6	3.4	2.4	1.9	
Ryegrass	4.0	3.0	2.3	1.9	
Average	*4.3 a*	*3.2 b*	*2.3 c*	*1.9 c*	*5.66*
	^{15}N (atom% ^{15}N excess)				
Black Oat	0.0074	0.0041	0.0052	0.0257	
Ryegrass	0.0263	0.0058	0.0051	0.0044	
Average	*0.0169 a*	*0.0050 b*	*0.0051 b*	*0.0051 b*	*0.64*
	Ndfr (%)				
Black Oat	0.18	0.10	0.13	0.14	
Ryegrass	0.66	0.15	0.13	0.11	
Average	*0.42 a*	*0.12 b*	*0.13 b*	*0.13 b*	*9.83*

541 [(1)]Means (n = 5) followed by the same lowercase letter within columns not differ by Tukey test ($p < 0.05$).
542 [(2)]Coefficient of variation (CV) of cover crop residue error 1

Table 5 Dynamics of black oat and ryegrass shoot residues decomposition in a mature peach orchard. Remaining dry matter, total organic carbon, nitrogen, cellulose, lignin, and non-structural biomass were adjusted to the model $X = Xo^{(-kt)}$; where X is the amount of dry matter or nutrient remaining in the residue after a period t (days), X_0 is the initial amount of dry matter or nutrient in the residue, k is the decomposition constant rate and t½ is the half-life for each compartment (Experiment 2)

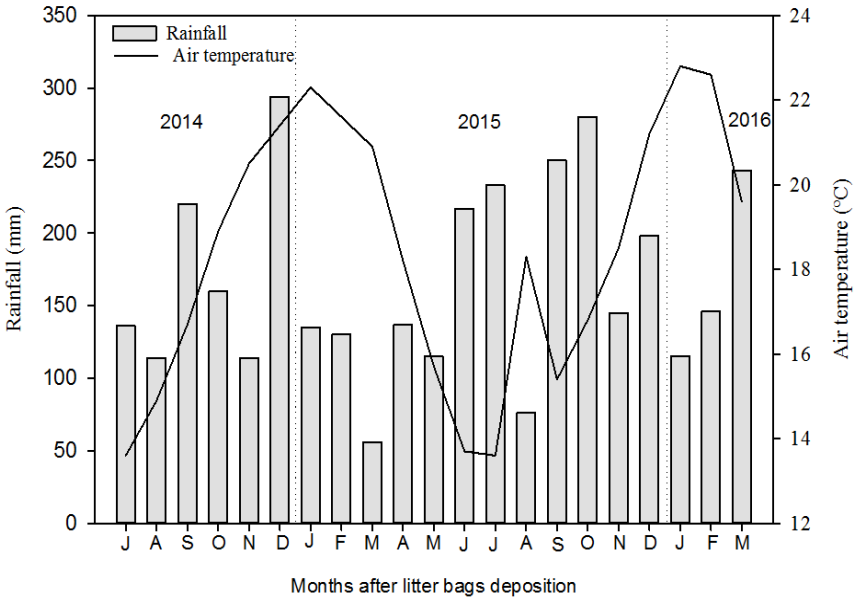
Treatment	k	t½	$R^{2(3)}$	k	t½	R^2
	g g^{-1}	days	-	g g^{-1}	days	-
	Remaining dry matter			Remaining cellulose		
Black Oat	0.0164 b(1)	42 a	0.98*	0.0278 b	25 a	0.95*
Ryegrass	0.0208 a	33 b	0.98*	0.0356 a	19 b	0.98*
CV (%)(2)	2.61	2.72		9.05	5.34	
	Remaining total organic carbon			Remaining lignin		
Black Oat	0.0170 b	41 a	0.97*	0.0325 b	23 a	0.88*
Ryegrass	0.0211 a	33 b	0.97*	0.0614 a	12 b	0.98*
CV (%)	2.26	2.33		13.62	14.16	
	Remaining nitrogen			Remaining non-structural biomass		
Black Oat	0.0118 a	54 a	0.94*	0.0117 a	59 a	0.91*
Ryegrass	0.0129 a	59 a	0.97*	0.0459 a	50 a	0.93*
CV (%)	6.06	11.38		41.36	29.48	

(1)Means (n = 5) followed by the same lowercase letter do not differ by the Tukey test ($p < 0.05$). (2) Coefficient of variation (CV) of the decomposition constant rate (k) and the half-life for each compartment (t½). (3) Coefficient of determination (R^2) of the residue's decomposition dynamics model. * significant at $p < 0.05$

552 **Table 6** Carbon/nitrogen (C/N) ratio, dry matter (DM), total organic carbon (TOC), and
553 nitrogen (N) in the remaining shoot residues of black oat and ryegrass deposited on the soil
554 surface in mature peach trees planting rows (Experiment 2)

Treatments	Day after litter bags deposition on the soil surface						
	0	30	60	90	120	150	180
				C/N ratio			
Black oat shoot	16.3	8.7	7.0	5.5	5.8	6.7	6.1
Ryegrass shoot	15.7	6.6	4.3	5.1	5.0	7.6	5.9
Significance(1)				2 SEM = 0.479			
CV (%)	8.20	15.3	23.9	24.4	23.0	40.8	10.1
				DM (kg ha^{-1})			
Black oat shoot	5177	2989	1573	1277	1120	745	161
Ryegrass shoot	5607	2702	1277	1078	838	825	142
Significance				2 SEM = 61.0			
CV (%)	3.79	7.13	16.1	9.72	12.1	20.3	26.0
				TOC (kg ha^{-1})			
Black oat shoot	2201	1126	562	512	443	290	44.7
Ryegrass shoot	2289	976	420	419	327	280	69.5
Significance				2 SEM = 35.8			
CV (%)	16.80	7.62	16.81	9.79	11.56	23.53	3.11
				N (kg ha^{-1})			
Black oat shoot	136	131	74.8	94.9	77.2	42.7	9.90
Ryegrass shoot	149	150	88.8	83.6	61.1	47.2	12.5
Significance				2 SEM = 4.12			
CV (%)	9.85	13.2	7.34	18.4	20.6	9.00	23.5

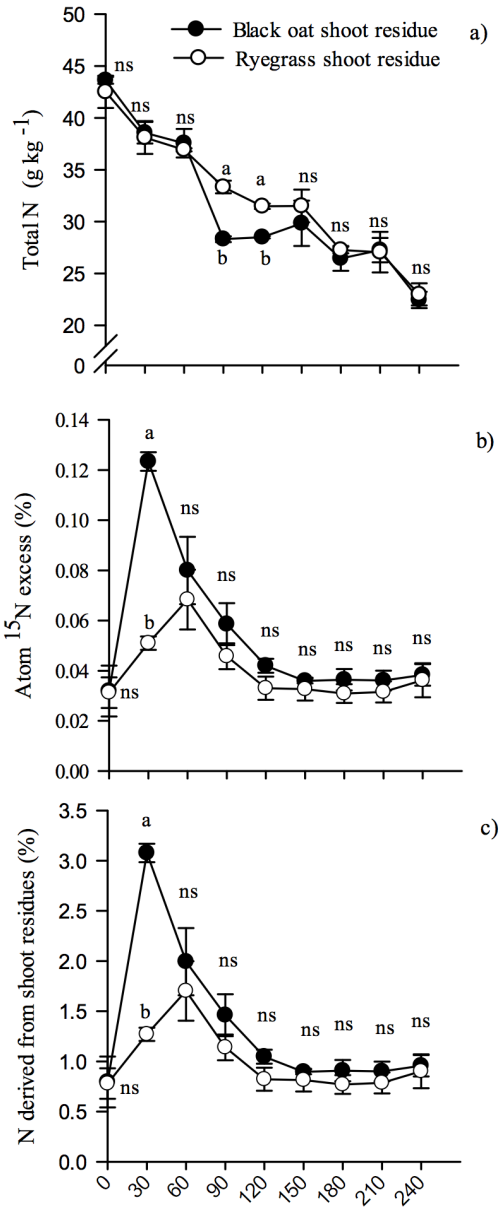
555 (1) Values differing by 2 standard error of means (SEM) are statistically different



556

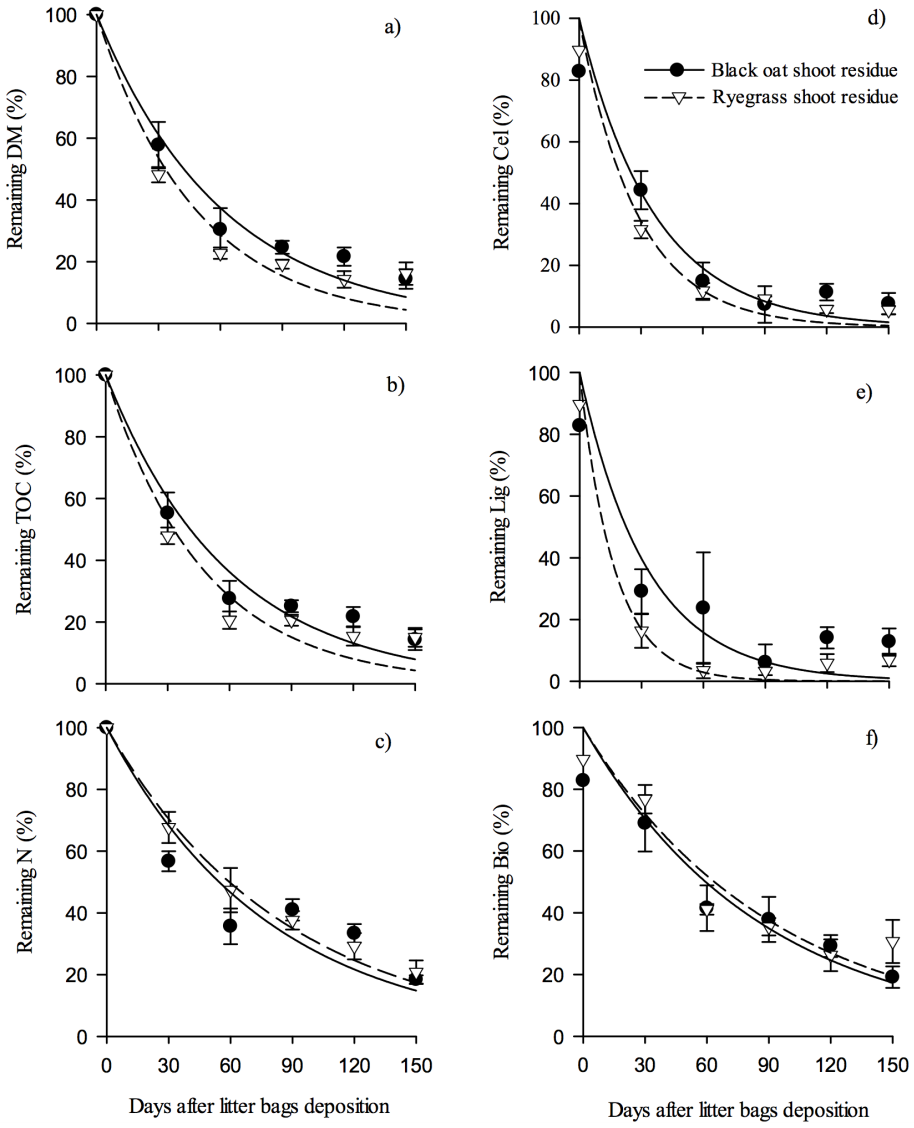
557 **Fig. 1** Average air temperature and cumulative monthly rainfall after the deposition of litter

558 bags in the experimental area



559

560 **Fig. 2** Total N a), atom% ^{15}N excess b), and ^{15}N derived from shoot residues c) in mature peach
561 trees leaves grown with black oat and ryegrass shoot residues deposition on the soil surface
562 (Experiment 1). Vertical bars indicate the standard error of the mean (n = 3). Lowercase letters
563 compare black oat and ryegrass residues within each sampling time by the Tukey-test ($p <$
564 0.05), ns = not significant



565

566 **Fig. 3** Remaining percentage of dry matter (DM) a), total organic carbon (TOC) b), nitrogen
567 (N) c), cellulose (Cel) d), lignin (Lig) e), and non-structural biomass (Bio) f) in black oat and
568 ryegrass shoot residues deposited on the planting rows of a mature peach tree orchard
569 (Experiment 2)