RESEARCH ARTICLE

Plant residues differing in C/N ratio in mulch and soil – the effect of the mulch on nutrient availability and microbial biomass is more pronounced with higher leaching amount

Thi Hoang Ha Truong^{1,2,*}, Petra Marschner¹

¹ School of Agriculture, Food and Wine, The University of Adelaide, South Australia, 5005, Australia

² Quang Binh University, Dong Hoi City, Quang Binh Province, Vietnam

ARTICLE INFO

Article history:

Received December 29, 2019

Revised March 21, 2020

Accepted April 19, 2020

Keywords:

Leaching amount

Mulching

Residues

C/N ratio

Microbial biomass N

N availability

ABSTRACT

This study investigated the effect of C/N ratio, placement of plant residues, and leaching amounts on soil respiration, microbial biomass, and nutrient availability within four weeks after amendment. Young faba bean shoots (FB, C/N 7) and mature wheat straw (WH, C/N 80) were used as low and high C/N residue, respectively. Soil was unamended, mulched with FB or WH only, or mulched with one residue and mixed with the other residue. Leaching with 5 or 25 mL water was carried out on days 4, 12, and 20. Cumulative respiration and microbial biomass N were higher with 25 than 5 mL only in treatments with two residues. WH under FB mulch reduced N availability compared to FB mulch alone, whereas FB under WH increased N availability compared to WH mulch alone. When soils were leached with 25 mL water, available N in FB mulch over WH was lower on day 12, but higher later, compared to WH mulch over FB. In contrast, in WH mulch over FB microbial biomass N increased over time whereas available N decreased. In conclusion, the effect of C/N ratio of the mulch on soil available and microbial biomass N was greater with the higher leaching amount.

© Higher Education Press 2020

1 Introduction

Plant residues can be incorporated (mixed) into soils or placed as mulches on the soil surface. In practice, mulch may be applied to soil in which plant residues are already present (e.g., when mulch is applied after harvest of a crop). But there are a few studies on the effect of plant residues in the soil on nutrient availability and nutrient leaching after mulching.

Organic amendments influence soil physical, chemical, and biological properties. For example, amendments increase soil water holding capacity (Farrell and Jones, 2009) and mulch can reduce raindrop impact and evaporation (Cook et al.,

2006; Mulumba and Lal, 2008). Decomposition of organic amendments and subsequent nutrient release are influenced by properties of the amendments including: nutrient and lignin concentration (Rahman et al., 2013; Hall et al., 2015), particle size (Bremer et al., 1991; Jensen, 1994), environmental conditions such as soil water content (Quemada and Cabrera, 1997; Paul et al., 2003), and temperature (Davidson and Janssens, 2006). Placement method (mulched or incorporated) can also influence decomposition rate and nutrient release of plant residues (Schomberg et al., 1994; Stemmer et al., 1999). In general, decomposition rate of mulches is lower than that of incorporated materials because mulched materials have smaller contact area to decomposers in the soil and are exposed to alternating wet and dry periods that may limit microbial activity (Coppens et al., 2006). Incorporated plant residues are decomposed faster because the water content in the soil is more stable than on the soil surface and because

* Corresponding author

E-mail address: thihoangha.truong@adelaide.edu.au (T.H.H. Truong)

they are in direct contact to soil microbes (Coppens et al., 2006; Nicolardot et al., 2007).

In areas with high precipitation or flood irrigation nutrients may be lost from soils via leaching of available and mobile nutrients (Lehmann et al., 1999). Mulching with organic materials can minimize leaching by reducing rain drop impact on the soil surface (Yang et al., 2003), increasing water retention, and increasing percentage of water-stable aggregates (Duiker and Lal, 1999). Organic mulches may also slowly release nutrients (Lehmann et al., 1999). Amendment with low carbon(C)/nitrogen(N) organic materials ($C/N < 20$) results in net N mineralization (Hadas et al., 2004) whereas high C/N amendments induce net N immobilization (Moritsuka et al., 2004). The C/N ratio of mulches influences N mineralization in a similar manner. Low C/N mulches increase N availability compared to high C/N mulches [e.g., Aulakh et al. (1991), Rosecrance et al. (2000)].

However, mulching can increase the risk of N loss via leaching compared to non-mulched soil if mulch materials have high N content (low C/N ratio) (Campiglia et al., 2011). In an 80-day incubation study, Lehmann et al. (1995) reported that both decomposition and N release of leaf mulch (C/N 14–26) were faster than those in twig mulch (C/N 52–118). The authors concluded that rapidly decomposing mulches should be combined with slowly decomposing mulches to reduce N leaching (Lehmann et al., 1995).

The studies on leaching from mulches were generally conducted over longer periods from 70 days in Rosolem et al. (2005) to 2 years in Campiglia et al. (2011). In these studies, mulches over unamended soil were leached once or several times to release nutrients. For example Rosolem et al. (2005) leached mulches five times to determine the effect of C/N ratio and N content of mulches on N release. Little is known about the effect of mulching on soil mixed with plant residues on nutrient availability, nutrient leaching, and microbial biomass shortly after mulching which may have important implications for the early stages of crop growth.

The aim of this short-term incubation study was to determine the effect of high or low C/N plant residues, the presence of plant residues in the soil, and leaching amount on respiration, microbial biomass N and P, and available N and P in soil within the first four weeks after amendment. We hypothesized with low C/N mulch and high leaching amount compared to low leaching amount, soil respiration, microbial biomass, and available N will be lower because water extractable compounds are leached from of the soil. The effect will be greater with nutrient-rich mulches that are easily decomposed with low C/N residue than with high C/N residue because the former releases more nutrients. The second hypothesis was that in soil mulched with faba bean (*Vicia faba* L.), N availability will be lower in soil that contains wheat straw (*Triticum aestivum* L.) than soil without straw because microbes decomposing wheat will immobilize N leached from faba bean mulch.

2 Materials and methods

2.1 Soil and organic materials

The sandy clay loam used in this study was collected from 0 to 10 cm depth at Waite Campus, The University of Adelaide, Australia (longitude 138°38'3.2" E, latitude 34°58'0.2" S). Collection and soil properties have been previously described in Truong and Marschner (2018). Briefly, the area is in a semi-arid region and has a Mediterranean climate. The soil is a Chromosol in Australian soil classification and a Rhodoxeralf according to US Soil Taxonomy. The soil has the following properties (for methods see below): sand 54%, silt 20% and clay 25%, pH (1:5 soil:water) 6.3, electrical conductivity (EC 1:5 soil:water) 143 $\mu\text{S cm}^{-1}$, total N 1.5 g kg^{-1} , total phosphorus(P) 371 mg kg^{-1} , total organic carbon (TOC) 17 g kg^{-1} , available N 15 mg kg^{-1} , available P 10 mg kg^{-1} , maximum water holding capacity (WHC) 378 g kg^{-1} , and bulk density 1.3 g cm^{-3} . After collection in the field, the soil was air-dried at 40°C in a fan-forced oven. After air-drying, visible plant debris was removed, and the soil was sieved to < 2 mm.

Two plant residues were used: young faba bean shoots (referred to as FB, C/N 7) as low C/N residue and mature wheat straw (referred to as WH, C/N 80) as high C/N residue (Table 1). The residues were dried at 40°C in a fan-forced oven and then ground and sieved to 0.25–2 mm. This small particle size was used to maximize contact of organic materials with the soil and thus increase their decomposition in soil and mulches. Total organic C was about 30% higher in WH than in FB. Total N and P were 8–9 times higher in FB than in WH. The C/N and C/P ratios were 11–12 times higher in WH than in FB.

Table 1 Total organic C, N, P, C/N ratio, C/P ratio, and pH of mature wheat straw (WH) and young faba bean shoots (FB) ($n = 4$).

	Wheat	Faba bean
Total organic C (g kg^{-1})	405.9 ^b	318.4 ^a
Total N (g kg^{-1})	5.1 ^a	47.1 ^b
Total P (g kg^{-1})	1.2 ^a	9.5 ^b
C/N	79.7 ^b	6.8 ^a
C/P	352.1 ^b	33.5 ^a
pH (1:10)	5.4	5.4

Within rows, means followed by different letters are significantly different ($P \leq 0.05$).

To determine nutrient leaching from mulches, 0.54 g of WH or FB was moistened and placed evenly on fabric pieces, which were placed into empty PVC cores similar to those used with soil in the experiment (see below), and were incubated moist for 4 days at room temperature ranging from 20 to 25°C. The mulches were leached on day 4 with 5 or 25 mL of reverse osmosis (RO) water. Organic C and inorganic N and P in leachates were measured as described below.

2.2 Experimental design

Before the start of experiment, the air-dried soil was incubated for 10 days at 50% of maximum WHC at 20–25°C in the dark. This water content was chosen because in previous studies using this soil, microbial activity was maximal at 50% (Marschner et al., 2015). Soil was either left unamended or amended with organic materials by only mulching or mulching and mixing.

There were 10 treatments differing in amendment type (no amendment, only mulching or mulching and mixing) and leaching amounts (5 and 25 mL RO water which simulated rainfalls of 4.7 and 23.3 mm) (for an overview, see Table 2). Leaching treatments with 5 and 25 mL RO water are referred to as 5-mL treatments and 25-mL treatments, respectively. Treatment names indicate soil treatment and amount of water used for leaching (5 or 25 mL). Soil was left unamended (NM 5, NM 25), only mulched with FB (FBm 5, FBm 25), mulched with FB and mixed with WH (FBm-WHs 5, FBm-WHs 25), only mulched with WH (WHm 5, WHm 25), or mulched with WH and mixed with FB (WHm-FBs 5, WHm-FBs 25). Mulching and mixing rates were 0.5 kg m⁻² and 10 g kg⁻¹ soil which is equivalent to 0.54 g and 0.3 g per core, respectively. The mulching rate of 0.5 kg m⁻² was chosen because it has been used in other studies [e.g., Halde and Entz (2016)]. Further, our preliminary experiments showed that there were little differences in N availability among four different mulching rates: 0.25, 0.5, 0.75 and 1 kg m⁻². With the mixing rate of 10 g kg⁻¹ soil, the amount of C added into the soil was about 4 g kg⁻¹, which was within the range used by previous studies [e.g., from 3 g kg⁻¹ in Shi et al. (2013) to 7 g kg⁻¹ in Nguyen and Marschner (2016)]. For treatments with both mulching and mixing the organic C addition rate was about 10 g kg⁻¹ soil, which was more than 2-fold higher than the rate added to the soil.

Soil mixed with residues or left unamended was filled into PVC cores of 3.7 cm diameter and 5 cm height with a nylon net base (7.5 µm, Australian Filter Specialist) and packed to a bulk density of 1.3 g cm⁻³ by adjusting the height of the soil in the cores. In treatments with mulch, a piece of fabric (mesh

size 0.1 mm) was placed on the soil surface to minimize movement of mulch particles into the soil. Then, residues were placed on the mesh. Residues and mesh were pressed on the soil surface to ensure good contact. The mulches were moistened by adding RO water and left undisturbed throughout the experiment.

Leaching was carried out on days 4, 12, and 20. For 5-mL and 25-mL treatments, 5 or 25 mL of RO water was added in 2.5 mL aliquots. Between additions, the water was allowed to drain from the mulch/soil surface before the next aliquot was added. In 5-mL treatments, the amount of RO water was used to leach nutrients from the mulches, but there was no leaching from the bottom of the soil-filled cores. In the 25-mL treatments sufficient water was added to generate leachate out of the lower end of the cores which was collected for measurement of inorganic N and P. After leaching, cores were dried at 30°C in a fan-forced oven for about 12 h until the soil water content was 50% WHC as determined by weight. Mulches were dried in the oven and were moistened again by spraying with RO water. Soils were destructively sampled 8 days after each leaching event (on days 12, 20, and 28) for measurement of available N and P, microbial biomass N (MBN), and microbial biomass P (MBP). Respiration was measured over 28 days except during the drying after leaching.

After amendment with plant residues and bulk density adjustment, the cores were placed individually into 1 L jars with gas-tight lids and equipped with septa to allow quantification of headspace CO₂ concentration as described below. In each 8-day period, only the cores to be sampled at the end of the period were placed back into the jars after leaching. The remaining cores were leached, dried and then incubated under the same conditions in large plastic trays covered with aluminum foil. After removing the cores to be analyzed from the jars, the cores to be harvested at the next sampling time were placed in the glass jars for respiration measurement. The jars and trays containing cores were incubated in the dark at 19–22°C. Between leaching events, soil water content was maintained at 50% of WHC and mulch kept moist by checking the water content every few days by

Table 2 Experimental design with treatment names and corresponding details about residue types, amendment rates, mulch types and rates, as well as, leaching amounts.

Treatment name	Residues in soil and amendment rates (g kg ⁻¹)	Mulch types and rates (kg m ⁻²)	Leaching amounts (mL RO water)
NM 5	Nil-0	Nil-0	5
NM 25	Nil-0	Nil-0	25
FBm 5	0	FB-0.5	5
FBm 25	0	FB-0.5	25
FBm-WHs 5	WH-10	FB-0.5	5
FBm-WHs 25	WH-10	FB-0.5	25
WHm 5	0	WH-0.5	5
WHm 25	0	WH-0.5	25
WHm-FBs 5	FB-10	WH-0.5	5
WHm-FBs 25	FB-10	WH-0.5	25

Table 3 Soil analyses from Marschner et al. (2015).

Parameter	Details	Reference
Soil texture	Hydrometer method	Gee and Or, 2002
Soil pH	1:5 soil:water ratio, 1 h shaking	Rayment and Higginson, 1992
Soil maximum water holding capacity	At matric potential –10 kPa	Wilke, 2005
Total organic C	Wet oxidation and titration	Walkley and Black, 1934
Total N	Digestion with H ₂ SO ₄ , measurement by modified Kjeldahl method	Bremner and Mulvaney, 1982
Total P	Digestion with 1:3 HNO ₃ and HCl, measurement by phosphovanado-molybdate	Hanson, 1950
Available N extraction	2 M KCl at a 1:10 soil extractant ratio, 1 h shaking	
Ammonium N		Willis et al., 1996
Nitrate N		Miranda et al., 2001
Available P extraction	Anion exchange resin	Kouno et al., 1995
Available P measurement		Murphy and Riley, 1962
Microbial biomass extraction	Chloroform fumigation and extraction with 0.5 M K ₂ SO ₄	Vance et al., 1987
Microbial biomass N	Ammonium N in extract, biomass $N = (\text{fumigated-unfumigated}) \times 0.57$	Moore et al., 2000
Microbial biomass P	Anion exchange resin with hexanol, biomass $P = \text{fumigated-unfumigated}$	Kouno et al., 1995
Soil respiration	CO ₂ concentration in headspace of jars	Setia et al., 2011

weighing cores and adding RO water if necessary. Soil respiration was measured daily from day 1 to 4, and then every two days from day 5 until day 28.

2.3 Analyses and calculations

Soil analyses were carried out as described in Marschner et al. (2015). For details, see Table 3.

2.4 Statistical analysis

There were four replicates per treatment and sampling event. Prior to the analysis, residuals were checked for normality using residual plots in Genstat 18th edition (VSN Int. Ltd., UK). Data were analyzed by two-way ANOVA in Genstat (residue treatment $\times$ leaching amount). Tukey's multiple comparison tests at a 0.05 level of significance was used to determine differences among treatments. One-way ANOVA was used to analyze properties of plant residues and nutrients leached from mulches and soil of 25-mL treatments.

3 Results

3.1 Nutrients leached from mulches in the first leaching event (day 4)

Leached organic C was lowest in WH leached with 5 mL RO water (WH 5), which was about 6-fold lower than in FB leached with 5 mL (FB 5) and 12-fold lower than in FB (FB 25) and WH (WH 25) leached with 25 mL (Table 4). Leached inorganic N was highest in FB 25 which was about 0.4, 4, and 100 times higher than in FB 5, WH 25, and WH 5, respectively. Leached inorganic P was highest in FB 25, which was about

25% higher than in FB 5 and 9–47 times higher than in WH 5 and WH 25.

3.2 Cumulative respiration

Total cumulative respiration did not differ between 5-mL and 25-mL treatments with only FB or WH mulch. However, in treatments that contained both FB and WH cumulative respiration was higher in the 25-mL treatment (Fig. 1). Among treatments with the same leaching amount, total cumulative respiration was highest in FBm-WHs, which was about 20% higher than in WHm-FBs, 30% higher than FBm, 2-fold higher than WHm, and 900% higher than the control. In treatments with FB mulch, cumulative respiration was highest in the first 12 days, but in treatments with WH mulch, it was higher from day 13 to 20.

3.3 Microbial biomass N and P

Throughout the experiment, there were no differences in MBN

Table 4 Inorganic N and P and water extractable organic carbon (WEOC) (mg kg⁻¹ soil) leached from WH and FB mulches in the first leaching event (day 4) ($n = 4$).

Mulch	Leaching amount (mL)	WEOC (mg kg ⁻¹)	Inorganic N (mg kg ⁻¹)	Inorganic P (mg kg ⁻¹)
FB	5	2122 ^b	1142 ^c	225 ^b
WH	5	345 ^a	17 ^a	6 ^a
FB	25	3932 ^c	1633 ^d	280 ^c
WH	25	4022 ^c	428 ^b	30 ^a

Within columns, means followed by different letters are significantly different ($P \leq 0.05$).

Fig. 1 Cumulative respiration in periods: days 1–12, 13–20 and 21–28 in soil alone, or soil either amended or mulched with young faba bean shoot (FB) or mature wheat straw (WH) and then leached with different leaching amounts. For treatment details see Table 2. Different lower case letters indicate significant differences in cumulative respiration among treatments for a given interval. Different upper case letters indicate significant differences in total cumulative respiration.

between the two leaching amounts in NM and WHm, few differences in FBm and FBm-WHs, and always differences in WHm-FBs (Fig. 2A). On day 12, MBN was highest in FBm-WHs 25, where it was 0.4- to 5-fold higher than the other treatments. On day 20, MBN was about 3-fold higher in WHm-FBs 25 than WHm-FBs 5, but it differed little among the other treatments. On day 28 MBN differed little among soil treatments and between leaching amounts, except for treatments with mulch and mixed residues where it was 3- to 4-fold lower in 5-mL than 25-mL treatments.

In most treatments, MBP remained stable throughout the experiment and did not differ between the two leaching amounts (Fig. 2B). Compared to the control, MBP was about 2-fold higher treatments with both FB and WH on days 12 and 20, but only 0.4-fold higher on day 28. The smaller difference compared to the control on day 28 was mainly due to higher MBP in the control than on the previous sampling dates.

3.4 Available N and P

Available N was 0.2- to 10-fold higher in 5-mL treatments than in corresponding 25-mL treatments, except in FBm-WHs 5 and FBm-WHs 25 on day 12 where it was similar (Fig. 3A). On day 12 in 5-mL treatments available N was about 2-fold higher in FBm and WHm-FBs compared to the control. Available N was about 50% lower in FBm-WHs 25 than the other 25-mL treatments. Differences among treatments were generally similar on days 20 and 28. Among 5-mL treatments, available N was 70% higher than the control in FBm, but lower in WHm.

With 25 mL leaching, available N was low in the control and WHm, but 6- to 8-fold higher than the control in treatments with FB mulch. Presence of WH in soil reduced available N on days 20 and 28 compared to FB mulch alone by 30%–50%, but available N was 2- to 3-fold higher in WHm-FBs than WHm.

In most treatments, available P changed little throughout experiment and did not differ between 5-mL treatments and corresponding 25-mL treatments. The exception was on day 28 when available P was 2-fold higher in NM 5 than in NM 25 (Fig. 3B). Available P was about 2-fold higher in FBm and WHm-FBs than the control, but available P differed little between control and WHm and FBm-WHs.

3.5 Leached N and P

Only 25-mL treatments generated leachate from the soil. Leached N was highest on day 12 (Fig. 4A). On day 12, leached N was about 50% lower in FBm-WHs than the other treatments. Leached N on day 20 was 50% lower in NM and WHm than the other treatments. On day 28, leached N in FBm was 2.5- to 10-fold higher than in the other treatments. At all sampling times, presence of WH in soil reduced leached N compared to FBm, but leached N differed between WHm and WHm-FBs only on day 20 when it was higher in WHm-FBs.

Leached P was lowest on day 12 (Fig. 4 B). On day 12, leached P differed compared to the control only in FBm where it was about 2-fold higher. Leached P was about 4-fold higher in FBm than FBm-WHs, and 4-fold higher in WHm-FBs than

Fig. 2 Microbial biomass N (A) and P (B) on days 12, 20 and 28 in soil alone, or soil either amended or mulched with young faba bean shoot (FB) or mature wheat straw (WH) and then leached with different leaching amounts. For treatment details see Table 2. At a given sampling date, different letters indicate significant differences among treatments.

WHm. On days 20 and 28, leached P was lowest in WHm, where it was 2- to 4-fold higher than the other treatments. On day 20, leached P was highest in treatments with FB mulch, but these treatments did not differ from the control on day 28. On days 20 and 28, presence of WH in the soil did not affect leached P with FB mulch. But leached P was 2- to 4-fold higher in WHm-FBs than WHm.

4 Discussion

This experiment showed that the effect of the leaching amount on respiration, available N and P, and microbial biomass N and P varied with time and amendment treatment. Presence of high C/N residue under low C/N mulch reduced available N

compared to low C/N mulch alone, but only temporarily increased MBN. In contrast, low C/N in soil under high C/N mulch resulted in higher available N than with high C/N mulch alone.

As expected from previous studies (Partey et al., 2014; Marschner et al., 2015), mulching with low C/N faba bean resulted in higher respiration than high C/N wheat straw. The initial high MBN with faba bean mulch suggests that faba bean was rapidly decomposed in the first 12 days (as indicated by the high cumulative respiration) and mineralized N taken up by microbes. However later, easily decomposable compounds were depleted resulting in biomass turnover and high available N. Wheat straw mulch provided a longer lasting C source which lead to N immobilization in the later stages of the experiment.

Fig. 3 Available N (A) and P (B) on days 12, 20 and 28 in soil alone, or soil either amended or mulched with young faba bean shoot (FB) or mature wheat straw (WH) and then leached with different leaching amounts. For treatment details see Table 2. At a given sampling date, different letters indicate significant differences among treatments.

4.1 Leaching amount

The first hypothesis (with low C/N mulch and high leaching amount compared to low leaching amount, soil respiration, microbial biomass, and available N will be lower) can be only be confirmed for available N. Total cumulative respiration per g soil differed between leaching amounts only in treatments with mulch and residues mixed into the soil where it was higher in 25-mL than 5-mL treatments. This is likely due to the greater movement of decomposable compounds from the mulch into the soil in 25-mL treatments which complemented the nutrients from the decomposing residue in the soil. The greater leaching amount may have also released nutrients from the residues through action of water or by softening the residue particles which may have made them more decom-

posable by microbes (Swift et al., 1979). In the 5-mL treatments, there was less transfer of nutrients from the mulch into the soil. MBN also differed between leaching amounts only in treatments with mulch and residues mixed into the soil. In FBm-WHs, the higher MBN on day 12 in 25-mL than 5-mL treatments indicates that in the first 12 days more N was leached into the soil and taken up by microbes with the higher leaching amount. Later, the greater leaching amount resulted in higher N loss out of the soil as indicated by the lower available N than with 5 mL. After the second leaching event, it is likely sufficient N for microbial N uptake had been released even with the smaller leaching amount. In WHm-FBs, MBN was lower with 25 mL than 5 mL on day 12, but higher at the later sampling times. A possible explanation is that C and N released during faba bean decomposition in the

Fig. 4 Leached inorganic N (A) and P (B) from soil leached with 25 mL RO water in the first (day 4), second (day 12) and third (day 20) leaching events. For treatment details see Table 2. At a given sampling date, different letters indicate significant differences among treatments.

first 12 days were leached from the soil with 25 mL, but retained in the soil with 5 mL. Later, the greater leaching of C from wheat mulch with 25 mL led to higher supply of C, which, together with N from faba bean, resulted in higher MBN than with 5 mL where less C from wheat mulch entered the soil.

4.2 Presence of residue in the soil

Compared to mulch only treatments, presence of residues in the soil increased cumulative respiration likely because of the greater amount of plant residues. The hypothesis that in soil mulched with faba bean, N availability will be lower in soil that contains wheat straw than without straw can be confirmed for available N.

With wheat mulch alone, MBN changed little during the experiment with both leaching amounts. But in WHm-FBs, MBN decreased over time with 5 mL whereas it increased with 25 mL. The decrease in MBN with 5 mL occurred despite high available N which indicates that the reduction of microbial N uptake was due to lack of easily decomposable C as it is often the most limiting factor for microbes in soil (Lynch, 1988; Smith and Paul, 1990). With 25 mL on the other hand, the increase in MBN over time was accompanied by a decrease in available N. This indicates greater N immobilization in 25-mL treatments likely due to higher supply of C from wheat mulch compared to 5 mL.

In FBm-WHs, MBN was higher than with faba bean mulch alone only on day 12 with 25 mL. Therefore the lower available

N cannot be explained by greater N immobilization in the later stages of the experiment. After day 12, it is likely that less N was released from faba bean mulch. This limited microbial N uptake particularly with 25 mL due to the greater loss of C compared to 5 mL. The lower available N in FBM-WHs compared to faba bean mulch alone may also be due to denitrification in anaerobic microsites caused by high respiration (Munch and Velthof, 2007).

Compared to WHm-FBs, FBM-WHs had higher cumulative respiration which was mainly due to the high initial respiration of FB mulch. In WHm-FBs, decomposition may have been delayed due to N limitation of microbes decomposing wheat mulch. The two treatments differed strongly in MBN and available N only with 25 mL leaching, probably due to the greater movement of nutrients in the soil than with 5 mL leaching. With 25 mL, MBN was higher in FBM-WHs than WHm-FBs on day 12, but lower on day 20. Available N, on the other hand, was lower in FBM-WHs on day 12 whereas it was higher at the later sampling times. This suggests that wheat under faba bean mulch initially induced net N immobilization (Tian et al., 1992; Bending and Turner, 1999). But the subsequent increase in available N suggests that more N was released by FB mulch than could be immobilized by microbes in the soil which were possibly limited by decomposable C (Lynch, 1988; Smith and Paul, 1990). In WHm-FBs with 25 mL on the other hand, available N decreased over time whereas MBN increased which indicates continuous supply of C from wheat. Thus, the interaction between available N and MBN over time was influenced by which residue was in the soil and which was mulched.

Both MBP and available P were higher in WHm-FBs than in WHm. Thus, faba bean, which has low C/P ratio, apparently released sufficient P to increase microbial P uptake and available P. Unlike MBN, which changed over time, MBP remained stable and was not affected by leaching amount. Microbes can accumulate P in storage compounds (Kulakovskaya et al., 2012; Cédric et al., 2016) and therefore retain P even when microbial activity is low. In contrast, microbial N is closely related to growth rate as N containing cell compounds are rapidly turned over (Schmidt et al., 2007).

5 Conclusion

This study showed that particularly N availability and microbial biomass N were influenced by leaching amount and by the arrangement of residues. The effect of the C/N ratio of the residue in the soil and that of the mulch on available N and microbial biomass N was greater with the higher leaching amount (25 mL), likely because nutrients were leached from the mulch reached greater depth in the cores than with 5 mL leaching. With 25 mL leaching, changes of available N and microbial biomass N differed between faba bean mulch over wheat and wheat mulch over faba bean. Compared to wheat mulch over faba bean, net N immobilization occurred initially in faba bean mulch over wheat, but later more N was released than could be taken up by the microbial biomass which was

limited by C availability. Compared to faba bean mulch alone, faba bean mulch over wheat in soil will minimize N leaching and should still provide available N to plants. In wheat mulch over faba bean, on the other hand, C supply from wheat stimulated microbial N uptake leading to net immobilization. N immobilized in the microbial biomass in this treatment may provide a source of N for plants after biomass turnover. Thus, this residue management could be suitable for slow growing plants. Experiments with plants are required to elucidate the effects of high or low C/N residues in mulched soil on nutrient uptake and availability.

Acknowledgment

Thi Hoang Ha Truong receives a postgraduate scholarship from Vietnam International Education Development.

References

- Aulakh, M.S., Doran, J.W., Walters, D.T., Mosier, A.R., Francis, D.D., 1991. Crop residue type and placement effects on denitrification and mineralization. *Soil Science Society of America Journal* 55, 1020–1025.
- Bending, G.D., Turner, M.K., 1999. Interaction of biochemical quality and particle size of crop residues and its effect on the microbial biomass and nitrogen dynamics following incorporation into soil. *Biology and Fertility of Soils* 29, 319–327.
- Bremer, E., Houtum, W., Kessel, C., 1991. Carbon dioxide evolution from wheat and lentil residues as affected by grinding, added nitrogen, and the absence of soil. *Biology and Fertility of Soils* 11, 221–227.
- Campiglia, E., Mancinelli, R., Radicetti, E., Marinari, S., 2011. Legume cover crops and mulches: effects on nitrate leaching and nitrogen input in a pepper crop (*Capsicum annuum* L.). *Nutrient Cycling in Agroecosystems* 89, 399–412.
- Cédric, T., Huu-Thanh, N., Alison, B., Anissa, D., Lies De, C., Raphaëlle, C., Evi, M., Erik, M., Frank, D., 2016. Characterisation of phosphate accumulating organisms and techniques for polyphosphate detection: A review. *Sensors (Basel)* 16, 797.
- Cook, H.F., Valdes, G.S.B., Lee, H.C., 2006. Mulch effects on rainfall interception, soil physical characteristics and temperature under *Zea mays* L. *Soil & Tillage Research* 91, 227–235.
- Coppens, F., Garnier, P., De Gryze, S., Merckx, R., Recous, S., 2006. Soil moisture, carbon and nitrogen dynamics following incorporation and surface application of labelled crop residues in soil columns. *European Journal of Soil Science* 57, 894–905.
- Davidson, E.A., Janssens, I.A., 2006. Temperature sensitivity of soil carbon decomposition and feedbacks to climate change. *Nature* 440, 165–173.
- Duiker, S.W., Lal, R., 1999. Crop residue and tillage effects on carbon sequestration in a Luvisol in central Ohio. *Soil & Tillage Research* 52, 73–81.
- Farrell, M., Jones, D.L., 2009. Critical evaluation of municipal solid waste composting and potential compost markets. *Bioresource Technology* 100, 4301–4310.

- Hadas, A., Kautsky, L., Goek, M., Kara, E.E., 2004. Rates of decomposition of plant residues and available nitrogen in soil, related to residue composition through simulation of carbon and nitrogen turnover. *Soil Biology & Biochemistry* 36, 255–266.
- Halde, C., Entz, M.H., 2016. Plant species and mulch application rate affected decomposition of cover crop mulches used in organic rotational no-till systems. *Canadian Journal of Plant Science* 96, 59–71.
- Hall, S.J., Silver, W.L., Timokhin, V.I., Hammel, K.E., 2015. Lignin decomposition is sustained under fluctuating redox conditions in humid tropical forest soils. *Global Change Biology* 21, 2818–2828.
- Jensen, E.S., 1994. Mineralization-immobilization of nitrogen in soil amended with low C:N ratio plant residues with different particle sizes. *Soil Biology & Biochemistry* 26, 519–521.
- Kulakovskaya, T.V., Vagabov, V.M., Kulaev, I.S., 2012. Inorganic polyphosphate in industry, agriculture and medicine: Modern state and outlook. *Process Biochemistry* 47, 1–10.
- Lehmann, J., Feilner, T., Gebauer, G., Zech, W., 1999. Nitrogen uptake of sorghum (*Sorghum bicolor* L.) from tree mulch and mineral fertilizer under high leaching conditions estimated by nitrogen-15 enrichment. *Biology and Fertility of Soils* 30, 90–95.
- Lehmann, J., Schroth, G., Zech, W., 1995. Decomposition and nutrient release from leaves, twigs and roots of three alley-cropped tree legumes in central Togo. *Agroforestry Systems* 29, 21–36.
- Lynch, J.M., 1988. Microorganisms in their natural environments: the terrestrial environment, In: Lynch, J.M., Hobbie, J.E., eds. *Microorganisms in action: concepts and applications in microbial ecology*. London, England: Blackwell Scientific Publications, 103–131.
- Marschner, P., Hatam, Z., Cavagnaro, T.R., 2015. Soil respiration, microbial biomass and nutrient availability after the second amendment are influenced by legacy effects of prior residue addition. *Soil Biology & Biochemistry* 88, 169–177.
- Moritsuka, N., Yanai, J., Mori, K., Kosaki, T., 2004. Biotic and abiotic processes of nitrogen immobilization in the soil-residue interface. *Soil Biology & Biochemistry* 36, 1141–1148.
- Mulumba, L.N., Lal, R., 2008. Mulching effects on selected soil physical properties. *Soil & Tillage Research* 98, 106–111.
- Munch, J.C., Velthof, G.L., 2007. Denitrification and Agriculture—Chapter 21, In: Bothe, H., Ferguson, S.J., Newton, W.E., eds. *Biology of the Nitrogen Cycle* 1st ed. Amsterdam: Elsevier Science, 331–341.
- Nguyen, T.T., Marschner, P., 2016. Soil respiration, microbial biomass and nutrient availability in soil after repeated addition of low and high C/N plant residues. *Biology and Fertility of Soils* 52, 165–176.
- Nicolardot, B., Bouziri, L., Bastian, F., Ranjard, L., 2007. A microcosm experiment to evaluate the influence of location and quality of plant residues on residue decomposition and genetic structure of soil microbial communities. *Soil Biology & Biochemistry* 39, 1631–1644.
- Partey, S.T., Preziosi, R.F., Robson, G.D., 2014. Improving maize residue use in soil fertility restoration by mixing with residues of low C-to-N ratio: effects on C and N mineralization and soil microbial biomass. *Journal of Soil Science and Plant Nutrition* 14, 518–531.
- Paul, K., Polglase, P., O'Connell, A., Carlyle, J., Smethurst, P., Khanna, P., 2003. Defining the relation between soil water content and net nitrogen mineralization. *European Journal of Soil Science* 54, 39–48.
- Quemada, M., Cabrera, M., 1997. Temperature and moisture effects on C and N mineralization from surface applied clover residue. *Plant and Soil* 189, 127–137.
- Rahman, M.M., Tsukamoto, J., Rahman, M.M., Yoneyama, A., Mostafa, K.M., 2013. Lignin and its effects on litter decomposition in forest ecosystems. *Chemistry and Ecology* 29, 540–553.
- Rosecrance, R.C., McCarty, G.W., Shelton, D.R., Teasdale, J.R., 2000. Denitrification and N mineralization from hairy vetch (*Vicia villosa* Roth) and rye (*Secale cereale* L.) cover crop monocultures and bicultures. *Plant and Soil* 227, 283–290.
- Rosolem, C.A., Calonego, J.C., Foloni, J.S.S., 2005. Leaching of nitrate and ammonium from cover crop straws as affected by rainfall. *Communications in Soil Science and Plant Analysis* 36, 819–831.
- Schmidt, S.K., Costello, E.K., Nemergut, D.R., Cleveland, C.C., Reed, S.C., Weintraub, M.N., Meyer, A.F., Martin, A.M., 2007. Biogeochemical consequences of rapid microbial turnover and seasonal succession in soil. *Ecology* 88, 1379–1385.
- Schomberg, H.H., Steiner, J.L., Unger, P.W., 1994. Decomposition and nitrogen dynamics of crop residues: residue quality and water effects. *Soil Science Society of America Journal* 58, 372–381.
- Shi, A., Penfold, C., Marschner, P., 2013. Decomposition of roots and shoots of perennial grasses and annual barley—separately or in two residue mixes. *Biology and Fertility of Soils* 49, 673–680.
- Smith, J.L., Paul, E.A., 1990. The significance of soil microbial biomass estimation, In: Bollag, J.M., Stotzky, G., eds. *Soil biochemistry*. New York: Marcel Dekker, Inc., 357–395.
- Stemmer, M., Lutzow, M., Kandeler, E., Pichlmayer, F., Gerzabek, M. H., 1999. The effect of maize straw placement on mineralization of C and N in soil particle size fractions. *European Journal of Soil Science* 50, 73–85.
- Swift, M.J., Heal, O.W., Anderson, J.M., 1979. *Decomposition in terrestrial ecosystems*. Oxford: Blackwell.
- Tian, G., Kang, B.T., Brussaard, L., 1992. Biological effects of plant residues with contrasting chemical compositions under humid tropical conditions—Decomposition and nutrient release. *Soil Biology & Biochemistry* 24, 1051–1060.
- Truong, T.H.H., Marschner, P., 2018. Respiration, available N and microbial biomass N in soil amended with mixes of organic materials differing in C/N ratio and decomposition stage. *Geoderma* 319, 167–174.
- Willis, R.B., Montgomery, M.E., Allen, P.R., 1996. Improved method for manual, colorimetric determination of total Kjeldahl nitrogen using salicylate. *Journal of Agricultural and Food Chemistry* 44, 1804–1807.
- Yang, Y.J., Dungan, R., Ibekwe, A., Valenzuela-Solano, C., Crohn, D., Crowley, D., 2003. Effect of organic mulches on soil bacterial communities one year after application. *Biology and Fertility of Soils* 38, 273–281.