REVISED

Title Page

Date of submission	November 24, 2014
Type of contribution	Regular paper
Number of text pages	16
Number of Tables	04
Number of Figures	03

Title: Impact of the addition of different plant residues on nitrogen mineralization-immobilization turnover and carbon content of a soil incubated under laboratory conditions

Names of Authors:	M. Kaleeen Munazza K		Majid	Mahmood	Tahir,	Nadia Sabir, and
Postal addresses of a	uthors	Department of Poonch, R				nces, The University mir, Pakistan

Short title:

N mineralization of plant residues

Correspondence address

M. Kaleem Abbasi Department of Soil and Environmental Sciences, The University of Poonch, Rawalakot Azad Jammu and Kashmir, Pakistan Tel.: +92 (0) 5824 960041 Fax: +92 (0) 5824 960004 E-mail: kaleemabbasi@yahoo.com

1 Abstract

Application of plant residues as soil amendment may represent a valuable recycling strategy that 2 affects on carbon (C) and nitrogen (N) cycling, soil properties improvement and plant growth 3 promotion. The amount and rate of nutrient release from plant residues depend on their quality 4 5 characteristics and biochemical composition. A laboratory incubation experiment was conducted for 120 days under controlled conditions [25°C and 58% water filled pore space (WFPS)] to quantify 6 initial biochemical composition and N mineralization of leguminous and non-leguminous plant 7 8 residues i.e. the roots, shoots and leaves of Glycine max, Trifolium repens, Zea mays, Poplus euramericana, Rubinia pseudoacacia and Elagnus umbellate incorporated into the soil at the rate of 9 200 mg residue N kg⁻¹ soil. The diverse plant residues showed wide variation in total N, carbon, lignin, 10 11 polyphenols and C/N ratio with higher polyphenol content in the leaves and higher lignin content in the roots. The shoot of G. max and the shoot and root of T. repens displayed continuous mineralization by 12 releasing a maximum of 109.8, 74.8 and 72.5 mg N kg⁻¹ and representing a 55, 37 and 36% of added N 13 14 being released from these resources. The roots of G. max and Z. mays and the shoot of Z. mays showed 15 continuous negative values throughout the incubation showing net immobilization. After an initial immobilization, leaves of P. euramericana, R. pseudoacacia and E. umbellate exhibited net 16 mineralization by releasing a maximum of 31.8, 63.1 and 65.1 mg N kg⁻¹, respectively and 17 representing a 16, 32 and 33% of added N being released. Nitrogen mineralization from all the 18 19 treatments was positively correlated with the initial residue N contents (r = 0.89; $p \le 0.01$), and negatively correlated with lignin content (r = -0.84; $p \le 0.01$), C/N ratio (r = -0.69; $p \le 0.05$), lignin/N 20 ratio (r = -0.68; $p \le 0.05$), polyphenol/N ratio (r = -0.73; $p \le 0.05$) and ligin+polyphenol/N ratio(r = -0.73; $p \le 0.05$) 21 22 0.70; $p \le 0.05$) indicating a significant role of residue chemical composition and quality in regulating N transformations and cycling in soil. The present study indicates that incorporation of plant residues 23 strongly modify the mineralization-immobilization turnover (MIT) of soil that can be taken into 24

account to develop synchronization between net N mineralization and crop demand in order to
 maximize N delivery and minimize N losses.

3 Key words: Biochemical composition, lignin, N immobilization, N mineralization,
4 polyphenols, residues quality

5 1 Introduction

6 Application of organic materials as soil amendments is an important management strategy that can improve and uplift soil quality characteristics and alter the nutrient cycling through mineralization or 7 8 immobilization turnover of added materials (Khalil et al., 2005; Baldi and Toselli, 2014). Use of local 9 organic materials derived either from livestock or plants have been attaining worldwide support to improve the fertility and productivity potential of degraded and nutrient poor soils (Huang et al., 2004). 10 Indeed, plant residues and animal manures are potentially important sources of nutrients for crop 11 production in smallholder agriculture. However, the Hindu Kash Himalayan (HKH) regions including 12 the State of Azad Jammu and Kashmir have a wide diversity of leguminous species and non-13 leguminous plants compared to the livestock production. Hence, use of plant residues as organic 14 nutrient source is relatively simple for the farmers compared to the manures application. Incorporating 15 plant residues into agricultural soils can sustain organic carbon content, improve soil physical 16 17 properties, enhance biological activities, and increase nutrient availability (Hadas et al., 2004; Cayuela et al., 2009). In the short-term, incorporation of plant residues provides the energy and nutrients for 18 microbial growth and activity, and act as a driving force for the mineralization-immobilization 19 20 processes in the soil and a source of nitrogen (N) for plants (Jansson and Persson, 1982). In the longterm, incorporation of crop residues is important for the maintenance of organic carbon (C) and N 21 stock in the nutrient pool of arable soils (Rasmussen and Parton, 1994). 22

Incorporation of crop residues provides readily available C and N to soils depending upon the decomposition rates and synchrony of nutrient mineralization (Murungu et al., 2011). The N availability from these residues depending on the amount of N mineralized or immobilized during

1 decomposition. However, previous studies demonstrated that the decomposition and nutrient release 2 rates of residues is often regulated by environmental factors, such as temperature and soil moisture, 3 and biochemical composition of plant materials and their interaction (Abiven et al., 2005; Khalil et al. 2005). The biochemical composition or quality parameters such as total N concentration, lignin (LG), 4 5 polyphenols (PP), carbon:nitrogen (C/N) ratio, LG/N, PP/N and (LG+PP)/N ratios are considered 6 useful indicators controlled decomposition and N release of added residues (Nakhone and Tabatabai, 7 2008; Vahdat et al., 2011; Abera et al., 2012). However, it has not been clearly established that which of these variables correlate the best with N mineralization of plant residues (Nakhone and Tabatabai, 8 9 2008) as contrasting results have been reported in the literature (Nourbakhsh and Dick, 2005). It has been reported that N released from leguminous tree leaves indicated that the (lignin+polyphenol):N 10 ratio was the most important factor in predicting N mineralization (Mafongoya et al., 1998). On the 11 other hand, Frankenberger and Abdelmagid (1985) suggested that lignin content of the legumes is not a 12 good predictor of the N mineralization. Handayanto et al. (1994) suggested that the N concentration or 13 lignin:N ratio of the leaves were not good indicators of N release for agroforestry materials. Palm and 14 Sanchez (1991) attributed the differences in N mineralization rates of various tropical legumes was due 15 to polyphenols. Handayanto et al. (1994) found however, that the total N content of plant residues was 16 17 not correlated with rates of N released under non-limiting N condition.

Earlier studies clearly demonstrated the beneficial effects of plant residues to soil-plant systems 18 (Huang et al., 2004; Cayuela et al., 2009; Khalil et al., 2005; Baldi and Toselli, 2014). However, still 19 there is a scope to explore the possibilities for achieving maximum benefits in term of rate, time and 20 amount of N released. For example, the synchronization of net N mineralization with plant/crop 21 growth is desirable to maximize N delivery for the crop and minimize N losses. Abiven et al. (2005) 22 reported that one of the tools to achieve synchronization is the use of plant residues with different 23 nature and qualities. Application of residues with high C/N ratio results in immediate net N 24 immobilization while residues with low C/N ratio results in net N mineralization showing that 25

mineralization-immobilization turnover (MIT) can be influenced differently by chemical components
of added plant materials. To achieve this target, combination of legumes and non-legumes plant
materials or different plant components of the same plant species i.e. root, shoot and leaves can be
tested.

5 Keeping in view the beneficial effects of plant residues on soil-plant systems especially in the 6 mountainous upland soils vulnerable to soil (water) erosion, the present work aims to i) examine the 7 initial biochemical composition and quality characteristics of on farm available plant residues and to ii) 8 quantify the N release potential (mineralization) of these residues added to a soil incubated under 9 controlled laboratory conditions (25^oC) at Rawalakot Azad Jammu and Kashmir, Pakistan.

10 2

2 Materials and methods

11 2.1 Soil sampling

The soil used in this study was collected from an arable field located at the research farm, Faculty of 12 Agriculture, The University of Poonch, Rawalakot Azad Jammu and Kashmir, Pakistan. The study site 13 is located at latitude 33°51'32.18"N, longitude 73° 45'34.93"E and an elevation of 5374 feet above the 14 sea level. The climate of the region is sub-temperate. Mean daily maximum and minimum air 15 temperatures ranged from 27 to 29 °C (June-July) and 1.0 to -3.5 °C (January-February). The mean 16 annual rainfall ranged between 1100-1500 mm with more than 50% of the total precipitation during 17 18 monsoon each year. The soil in the study site was clay loam in texture, classified as Humic Lithic 19 Eutrudepts (Inceptosols) (Ali et al., 2006). The field was bare at the time of sampling but previously 20 maize (Zea mays L.) and wheat (Triticum aestivum L.) were cultivated. The selected field was divided 21 into10 sub-plots to ensure proper and representative soil sampling. Soil samples were collected from 0-15 cm depth at random from three points in each plot (randomly) using a 5 cm diameter soil auger. 22 The soil samples from all the selected plots were thoroughly mixed to get a composite sample. The 23 field fresh soil was passed through a 4 mm sieve to eliminate coarse rock and plant material, 24 thoroughly mixed to ensure uniformity and stored at 4°C before use (not more than 2 wk). A 25

subsample of about 0.5 kg was taken, air dried, and passed through a 2 mm sieve and used for the
 determination of physical and chemical characteristics. The original soil analysis is presented in Table
 1.

4 2.2 Collection of plant residues

Six predominant on farm available plant species were selected. These included: Glycine max shoot, 5 6 Glycine max root, Trifolium repens shoot, Trifolium repens root, Zea mays shoot, Zea mays root, 7 leaves of Poplus euramericana, Rubinia pseudoacacia and Elagnus umbellate. Plant samples/residues 8 were collected at different timings in the year 2012. Glycine max and Trifolium repens samples were 9 collected from the field before flowering (summer) while Zea mays samples were taken one week before crop harvest. The tree leaves were sampled late in fall. Plant residues were washed with running 10 tap water, rinsed three times with distilled water, dried at 65°C for 48 h, milled and passed through a 1-11 mm sieve. Triplicate samples of plant residue were taken and analyzed for their C, N, lignin and 12 polyphenol concentration. Total N contents of the residues were determined by Kjeldhal digestion, 13 distillation, and titration method (Bremner and Mulvaney, 1982). Wet digestion method was used for 14 organic C analysis (Nelson and Sommers, 1982). The lignin content was determined using Van Soest 15 methods (Van Soest et al., 1991). Soluble polyphenols were extracted in hot water (100^oC, 1h) and 16 17 determined by colorimetery using Folin-Denis reagent (Folin and Denis, 1915).

18 2.3 Laboratory incubation

The incubation methods used in this study was followed by the methods used in our previous studies (Abbasi et al., 2011; Abbasi and Khizar, 2012). Briefly, about hundred grams of soil already stored in the refrigerator at 4^oC was weighed and transferred into 200 ml glass jars. The initial moisture content of soil was 28% (w/w) that was increased by adding distilled water to achieve a final water filled pore space (WFPS) of 58%. The treatments were comprised of a control (no N) and nine plant residues sources, i.e., *Glycine max* shoot, *Glycine max* root, *Trifolium repens* shoot, *Trifolium repens* root, *Zea mays* shoot, *Zea mays* root, leaves of *Poplus euramericana, Rubinia pseudoacacia* and *Elagnus* *umbellate*; ten incubation timings, i.e., 0, 7, 14, 21, 28, 42, 60, 80, 100 and 120 days and three replications. Altogether, a total of 300 jars (10 treatments × 10 incubation timings × 3 replications) were arranged in a completely randomized design. Plant residues were weighed and added into the jars at the rate equivalent to 200 mg N kg⁻¹. After adding residues, all the jars were weighed and their weight was recorded. The soil was then incubated under controlled conditions at 25°C. Soil moisture was checked/adjusted after every two days by weighing the glass jars and adding the required amount of distilled water when the loss was greater than 0.05 g.

8 2.4 Soil extraction and analysis

Samples of all ten treatments were analyzed for total mineral nitrogen (TMN) as described earlier 9 (Abbasi and Khizar, 2012). Initial concentration of TMN (NH_4^+ –N+ NO_3^- –N) at day 0 was determined 10 by extracting soil samples with 200 ml of 1 M KCl added directly to the flask immediately after 11 incorporation of each N source. Thereafter, triplicate samples from each treatment were removed 12 randomly from the incubator at different incubation timings and extracted by shaking for one hour with 13 200 ml of 1 M KCl followed by filtration. The total mineral N of the extract were determined by using 14 the steam distillation and titration method (Keeney and Nelson, 1982). Net cumulative N mineralized 15 (NCNM) from different plant residue treatments was calculated following the method described earlier 16 17 (Sistani et al., 2008).

18 **2.5** Statistical analysis

All data were statistically analyzed by multifactorial analysis of variance (ANOVA) using the software package MSTATC Version 3.1 (1990). Least significant differences (LSD) was used as posthoc test to indicate significant variations within the values of either treatments or time intervals. Correlation coefficient used between the studied variables was calculated using SPSS 20 software. A probability level of $p \le 0.05$ was considered significant (Steel and Torrie, 1980).

24 **3 Results and discussion**

25 3.1 Chemical composition of the residues – residues quality

A significant difference ($p \le 0.05$) in N content among different residues was observed and *Glycine max* 1 2 shoot, Rubinia pseudoacacia and Elagnus umbellate leaves exhibited significantly ($p \le 0.05$) higher N compared to the remaining plant amendments (Table 2). The total C contents varied from 397 g kg⁻¹ 3 for the Trifolium repens shoot to 486 g kg-1 for the Zea mays root and Zea mays shoot and root 4 displayed significantly ($p \le 0.05$) higher C contents than the remaining plant residues. Similar trend was 5 noticed for C:N. The lignin content varied from a minimum of 11 g kg⁻¹ in the *Glycine max* shoot to a 6 maximum of 48 g kg⁻¹ in the Zea mays roots. A minimum polyphenol contents were recorded in 7 8 Glycine max shoot while a maximum polyphenol was found in the Poplus euramericana leaves. The 9 LG/N, PP/N and LG+PP/N ratios were highest in the Zea mays root while the lowest values were recorded for the *Glycine max* shoot. Generally, total N contents of the legume residues were higher 10 compared to the non-legumes. Similarities could be observed between the same organs of the different 11 species, i.e., all the roots were characterized by high C, LG and PP contents and lower N 12 concentration. Leaves were particularly rich in PP and total N. The differences in the concentration of 13 quality characteristics of residues according to plant components i.e. shoot, root and leaves had been 14 reported earlier (Abiven et al., 2005; Nourbakhsh and Dick, 2005). It has been reported that high lignin 15 content in root was due to presence of suberin in the roots and its ability to form complex barriers 16 when associated with lignin (Abiven et al., 2005). Plant residues used in this study provided a wide 17 range of contrasted chemical composition and significant variation in quality characteristics because of 18 the difference in: i) type of species i.e. leguminous and non-leguminous, trees and crops; and ii) plant 19 components/organs i.e. shoot, root and leaves. 20

21 **3.2** Nitrogen mineralization

Results indicated that the control soil without any amendment released a maximum of 77.7 mg N kg⁻¹ at day 100 compared to 13.7 mg kg⁻¹ at the start showing a substantial release of N into mineral N pool (Table 3). Expressed as the total N initially present, the net N mineralized during the incubation was 14 %. The mineralization of native soil N observed here was in accordance with our previous study where

a maximum of 90 mg kg⁻¹ mineral N was released from the control soil representing 16% of the initial 1 N of the soil (Abbasi and Khizar, 2012). Among different plant materials added, the legumes, i.e., 2 3 shoot of *Glycine max* and shoot and root of *Trifolium repenes* exhibited significantly higher TMN compared to the non-legumes. The maximum TMN released from these amendments varied between 4 150 mg kg⁻¹ to 189 mg kg⁻¹. The mean values indicated that these legumes were collectively able to 5 release 85 mg N kg⁻¹ compared to 20 mg kg⁻¹ by maize and 58 mg N kg⁻¹ by leaves of the non-legumes 6 7 trees. As expected, the plant organs also affected N mineralization and in general roots displayed 8 significantly lower TMN compared to the shoot and leaves. Incorporation of Glycine max root and Zea 9 mays shoot and root resulted in a constant decrease in TMN and the maximum values ranged between 32 to 49 mg kg⁻¹ compared to 78 mg kg⁻¹ in the control treatment. On the other hand, after initial 10 negative values till day 14 and 21, leaves of Poplus euramericana, Rubinia pseudoacacia and Elagnus 11 umbellate continuously increased TMN till the end ranged between 107 to 140 mg kg⁻¹ (highest 12 values). 13

14

3.3. Net cumulative N mineralization

Nitrogen mineralization of added plant residues was determined on the basis of net cumulative N 15 mineralized (NCNM). The N mineralization from Glycine max and Trifolium repenes shoot showed 16 positive values throughout the incubation ranged between 24 to 110 mg kg⁻¹ for *Glycine max* and 5 to 17 75 mg kg⁻¹ for *Trifolium repenes* (Figure 1). Considering the NCNM at the end day 120, the net N 18 mineralized as percentage of total N applied from *Glycine max* and *Trifolium repenes* shoot was 54% 19 20 and 21%, respectively. The percent of N mineralized from Glycine max added shoot had been reported previously ranged between 39 to 43% of applied N residues (Nakhone and Tabatabai, 2008). On the 21 other hand, the NCNM from *Glycine max* roots, Zea mays shoot and Zea mays roots exhibited negative 22 values throughout the incubation indicating net immobilization. Among the three residues, Zea mays 23 roots displayed higher negative values leading to higher immobilization. Roots of *Glycine max*, and 24 leaves of *Poplus euramericana*, *Rubinia pseudoacacia* and *Elagnus umbellate* showed four phases of 25

mineralization-immobilization turnover (MIT): initial negative values from day 7 to 21, slow 1 2 mineralization from day 21 to 60, a rapid mineralization during 60 and 80 days, declining in net during 3 100 and 120 days. The net N mineralized as percentage of total N applied from roots of Glycine max, and leaves of Poplus euramericana, Rubinia pseudoacacia and Elagnus umbellate was 16, 8, 21, 21% 4 5 respectively. Net nitrogen mineralization (% of added N) from different organic materials during 110 6 days of incubation was in the range between -35% in Triticum aestivum (wheat) residues to 81% in 7 Trifolium repen (white clover) residues (Kumar and Goh, 2003). Similarly, a 44, 38 and 35% of N 8 added had been released from the leaves of peanut, pigeonpea and hairy indigo, respectively 9 (Thippayarugs et al., 2008).

All legumes (except *Glycine max* root) exhibited the highest NCNM (average 30% of added plant N residues) compared to non-legumes (17%). Similarly, the cereal crop *Zea mayz* shoot and root exhibited net immobilization compared to net mineralization observed in the legumes and tree leaves. The plant components also showed variation in NCNM. For example, shoot of *Glycine max* and *Trifolium repens* mineralized an average of 74 mg N kg⁻¹ compared to 4 mg N kg⁻¹ from the roots. Likewise, leaves of forest trees showed higher NCNM compared to the roots of legumes and nonlegumes crop.

17 The shoot of *Glycine max* and *Trifolium repens* exhibited the highest NCNM without any negative value during incubation because of high N concentration and low C/N ratio. However, it is 18 interesting to note that the total N concentration of the leaves of *Rubinia pseudoacacia* and *Elagnus* 19 20 umbellate was higher and C/N ratio was lower compared to the Trifolium repens shoot but the net mineralization (averaged) of Trifolium repens shoot was higher (47 and 58%) compared to the leaves 21 of *Rubinia pseudoacacia* and *Elagnus*, respectively. The low mineralization in leaves in spite of high 22 N content and low C/N ratio was attributed to higher concentration of LG, PP, LG/N, PP/N and 23 LG+PP/N. These results demonstrated the role of other factors in addition to total N and C/N ratio 24 affecting plant residues decomposition and N mineralization kinetics. As indicated in a previous study 25

(Trinsoutrot et al., 2000) the net accumulation (whether positive or negative) of mineral N in soil 1 2 during decomposition of organic residues is directly related to the residue N content. However, our 3 results clearly indicated that N was not the only factor affecting the mineralization of added residues but some additional quality characteristics also influenced MIT of plant residues. Likewise, the total N 4 5 content and C/N ratio of the leaves of Rubinia pseudoacacia and Elagnus umbellate were at par with 6 Glycine max shoot but the net mineralization of G. max shoot was 3-fold higher. It had been reported that organic materials with similar C/N ratios may mineralize different amounts of N because of 7 8 differences in composition that are not reflected by the C/N ratio (e.g. different lignin contents) 9 (Mohanty et al., 2011).

10 Similarly, roots of *Glycine max* and *Zea mays* showed net immobilization while roots of 11 *Trifolium repens* displayed fast decomposition and net N release pattern. This discrepancy in root MIT 12 was mainly due to high N concentration, low C/N ratio; low LG and PP contents of the roots of 13 *Trifolium repens*. The N turnover shown by *Trifolium repens* roots confirmed the strong below-ground 14 N dynamics and residual effect of *Trifolium repens* if grown in the soil.

Among the leaves of different trees tested, leaves of Rubinia pseudoacacia and Eumbellate 15 released a substantial amount of N into mineral N pool. Leaf residues have been described as high 16 17 quality litter materials in terms of high N and low-lignin contents (Thippayarugs et al., 2008), have been found to decompose easily and release mineral N substantially (Mtambanengwe and Kirchmann, 18 1995) as observed in our study. However, Poplus euramericana leaves exhibited higher net 19 20 immobilization (for a longer period) and lower net mineralization. The variation was again due to disparity in the biochemical composition. The low N content, high C/N ratio and high PP content may 21 have been largely responsible for the slow decomposition and low net mineralization of Poplus 22 euramericana leaves. These results inferred that the same plant components may not necessarily be 23 shown similar decomposition and mineralization turnover because of variation in biochemical 24 composition. 25

Pattern and trend of N mineralization 3.4.

2 The patterns of N mineralization varied among plant residues and plant components. After 3 incorporation into soil and during incubation, the added residues exhibited three main patterns of cumulative net mineralization (Figure 2): i) a pattern of the continuous and rapid release of net N 4 5 throughout the incubation without showing any negative value indicating net mineralization. This 6 pattern of mineralization was shown by the *Glycine max* shoot and *Trifolium repens* shoot; ii) a pattern shown by the Trifolium repens roots, and leaves of Poplus euramericana, Rubinia pseudoacacia and 7 8 Elagnus umbellate indicated initial negative values of net cumulative immobilization for variable 9 periods followed by slow and then a rapid release of N indicating immobilization-mineralization turnover; iii) a pattern showed continuous negative values throughout the incubation indicating net N 10 immobilization as seen in case of the Glycine max root and the Zea mays shoot and the root. The MIT 11 and N released pattern by plant residues observed here was in accordance with that reported earlier in 12 both leguminous and non-leguminous plant residues (Kumar and Goh, 2003). 13

14 The N mineralization trend over time showed wide variation (Figure 3a). These results highlighted the time taken for releasing N into mineral N pool by the added plant residues. Results showed initial lag 15 phase where most of the applied residues endured immobilization with little mineralization only from 16 17 Glycine max and Trifolium repenes shoot was shown during 0 to 21 days of incubation. The rapid mineralization phase occurred from day 28 to day 80. Thereafter, a declining phase of mineralization 18 started in the later part of the incubation at day 100 to day 120. 19

20 3.5

Changes in soil organic matter

21 In order to examine the changes in soil organic matter (SOM) in response to added plant residues, comparison between the SOM at the start at day 0 with those recorded at the end of incubation on day 22 120 had been shown (Figure 3b). Soil organic matter contents of all the treatments recorded at day 120 23 was lower than that recorded at day 0. The unaccounted SOM ranged between 32 to 67% compared to 24 that recorded at day 0. The decreasing trend of SOM was substantially higher for the treatments 25

showing mineralization (54–67%) compared to those showing immobilization (32–38%). By the end 1 2 at day 120, the loss of SOM was in the order: T. repens shoot > E. umbellate leaves > T. repens root = 3 R. pseudoacacia leaves > P. euramericana > G. max shoot > Z. mays shoot > Z.mays root = G. max root. The SOM turnover observed here was coincided with net mineralization. In the initial lag phase 4 5 when mineralization was either very low or displayed negative values, on average only 8% of the 6 initial SOM had been utilized (7-21 days). The SOM utilization during 28-80 days when 7 mineralization was rapid was 31% of the initial amount while 43% of initial SOM was utilized in the 8 later part of incubation (100 and 120 days) when mineralization start showing declining trend.

9 **3.6** Relationship between cumulative N mineralization and residues quality characteristics

Results of the study showed highly significant positive correlation between N mineralization, i.e., and 10 plant residue N concentrations (r = 0.89; $P \le 0.01$) (Table 4). In contrast, a negative significant 11 correlations existed between net cumulative N mineralized (NCNM) and LG (r = -0.84; $P \le 0.01$), 12 NCNM and C/N ratio (r = -0.69; $P \le 0.05$), NCNM and LG/N ratio (r = -0.68; $P \le 0.05$), NCNM and 13 PP:N ratio (r = -0.73; P \le 0.05) and NCNM and LG+PP/N ratio (r = -0.70; P \le 0.05). The correlation 14 between N mineralization and PP was non-significant at a p<0.05. The significant positive correlation 15 between net rates of N mineralization and residues N concentration observed is consistent with other 16 studies (Nourbakhsh and Dick, 2005; Vahdat et al., 2011). It had been reported that N availability may 17 control the decomposition of plant residues, particularly those with low N content such as cereals when 18 the N requirements of the soil decomposers are not met by the residue or soil N contents (Vahdat et al., 19 20 2011). A negative correlation was also observed between net N mineralization and C/N ratio of the plant materials. Previously total N contents and C/N ratio were considered adequate for predicting the 21 net N mineralization of crop residue. However, the latest studies including the present work 22 highlighted the role of other quality characteristics including LG and PP affecting net mineralization of 23 plant residues. The closer relationship between net mineralization with residue lignin contents (r = -24 0.84; $P \le 0.01$) than that of C/N ratio (r = -0.69; $P \le 0.05$) recorded in this study was in accordance with 25

previous findings (Müller et al., 1988; Vahdat et al., 2011). The highly significant positive correlation
 between net N mineralization and the residue N content (*r* = 0.89; *P*≤0.01) confirm the previous results
 (Nourbakhsh and Dick, 2005; Vahdat et al., 2011) indicating that residue N concentration can be
 considered a better tool to predict mineralization of added organic residues compared to the C/N ratio.

5 4 Conclusions

The experiment showed that soil amended with plant residues displayed wide variation of N 6 mineralization depended upon the plant species and plant components/organs. The decomposition and 7 8 N released potential of added materials were largely related to their biochemical composition. Overall, in addition to residues N concentration and C/N ratio, LG contents of plant residues also appeared to 9 be an important factor predicting the net N mineralization of plant residues. Shoots of Glycine max and 10 11 Trifolium repens and leaves of Rubinia pseudoacacia and Elagnus umbellate exhibited a substantial mineralization potential demonstrating that legumes and trees of these two plant species can produce 12 high quality residues and thus have the potential to promote N cycling in agro-ecosystems. This study 13 suggested that plant residues showing rapid mineralization can be used for early N demands of a crop 14 while residues with high C:N and LG contents immobilize N thus can help to counter the N loss 15 generally observed due to rapid ammonification- nitrification turn over. Use of such plant materials in 16 our cropping systems especially in the regions subjected to land degradation may be a useful 17 management strategy to restore these soils for agriculture production. 18

Acknowledgements. The authors express their appreciation to Nuclear Institute for Food and
Agriculture (NIFA), KPK Peshawar, Pakistan for providing lab facilities to analyze biochemical
characteristics of the plant materials used in the study.

22 **References**

Abbasi, M. K., and Khizar, A.: Microbial biomass carbon and nitrogen transformations in a loam soil
 amended with organic-inorganic N sources and their effect on growth and N-uptake in maize,
 Ecol. Eng., 39, 23–132, 2012.

Abbasi, M. K., Hina, M., and Tahir, M. M.: Effect of Azadirachta indica (neem), sodium thiosulphate 1 and calcium chloride on changes in nitrogen transformations and inhibition of nitrification in 2 soil incubated under laboratory conditions, Chemospher, 82, 1629-1635, 2011. 3 Abera. G., Wolde-meskel, E., and Bakken, L. R.: Carbon and nitrogen mineralization dynamics in 4 5 different soils of the tropics amended with legume residues and contrasting soil moisture contents. Biol. Fertil. Soils, 48, 51-66, 2012. 6 Abiven, S., Recous, S., Reyes, V., and Oliver, R.: Mineralisation of C and N from root, stem and leaf 7 residues in soil and role of their biochemical quality, Biol. Fertil. Soils, 42, 119–128, 2005. 8 Ali, B., Mohmand, H., and Muhammad, F. 2006. Integrated land resource survey and evaluation of 9 10 Azad Jammu & Kashmir area 2004. Soil Survey of Pakistan, Government of Pakistan, Ministry of Food, Agriculture & Livestock, pp. 156-157. 11 Baldi, E., and Toselli, M.: Mineralization dynamics of different commercial organic fertilizers from 12 agro-industry organic waste recycling: an incubation experiment, Plant Soil Environ., 60, 93-13 99, 2014. 14 Bremner. J. M., and Mulvaney C.S.: Nitrogen-total, in: Methods of Soil Analysis Part 2 Chemical and 15 Microbiological Properties, edited by: Page, A. L., Miller, R. H., and Keeney, D. R., SSSA 16 17 Madison, WI, 595-624, 1982. Cayuela, M. L., Sinicco, T., and Mondini, V.: Mineralization dynamics and biochemical properties 18 during initial decomposition of plant and animal residues in soil, Appl. Soil Ecol., 48, 118–127, 19 2009. 20 Folin, O., and Denis W.: A colorimetric estimation of phenol and phenol and derivatives in urine, J. 21 22 Biol. Chem., 22; 305-308, 1915. Frankenberger Jr., W.T., and H. M. Abdelmagid, H.M.: Kinetic parameters of nitrogen mineralization 23 rates of leguminous crops incorporated into soil. Plant Soil, 87, 257-271, 1985. 24 Hadas, A., Kautsky, L., Goek, M., and Kara, E. E.: Rates of decomposition of plant residues and 25 available nitrogen in soil, related to residue composition through simulation of carbon and 26 nitrogen turnover, Soil Biol. Biochem., 36, 255-266, 2004. 27 28 29 Handayanto, E., Cadish, G., and Giller, K. E.: Nitrogen release from prunings of hedgerow trees in 30 relation to the quality of the pruning and incubation method. Plant Soil 160, 237–248, 1994. Huang, Y., Zou, J., Zheng, X., Wang, Y., and Xu, X.: Nitrous oxide emissions as influenced by 31 amendment of plant residues with different C:N ratios, Soil Biol. Biochem., 36, 973-981, 2004. 32 33 Jansson, S. L., and Persson J.: Mineralization and immobilization of soil nitrogen, in: Nitrogen in Agricultural Soils, edited by Stevenson, F. J., ASA, SSSA Special Publication No. 22, 34 Madison WI, 229–252, 1982. 35 Keeny, D. R., and Nelson, D. W.: Nitrogen – inorganic forms, in: Methods of Soil Analysis Part 2 36 37 Chemical and Microbiological Properties, eddied by Page, A. L., Miller, R. H., and Keeney D. R., SSSA Madison WI, 643-693, 1982. 38

- Khalil, M. I., Hossain, M. B., and Schmidhalte, U.: Carbon and nitrogen mineralization in different
 upland soils of the subtropics treated with organic materials, Soil Biol. Biochem. 37: 1507–
 1518, 2005.
- Kumar, K., and Goh, K. M.: Nitrogen release from crop residues and organic amendments as affected
 by biochemical composition, Commun. Soil Sci.Plant Anal., 34, 2441–2460, 2003.
- Mafongoya, P. L., Nair, P. K. R., and Dzowela, B. H.: Mineralization of nitrogen from decomposing
 leaves of multipurpose trees as affected by their chemical composition. Biol. Fertil. Soils 27,
 143–148, 1998.
- Mohanty, M., Reddy, K. S., Probert, M. E., Dalal, R. C., Subba Rao, A., and Menzie, N. W.: Modeling
 N mineralization from green manure and farmyard manure from a laboratory incubation study,
 Ecol. Model., 222, 719–726, 2011.
- MSTATC: A microcomputer program for the design, management, and analysis of agronomic research
 experiments, Michigan State University, Michigan, USA, 1990.
- Mtambanengwe, F., and Kirchmann, H.,: Litter from a tropical savanna woodland (miombo): chemical
 composition and C and N mineralization. Soil Biol. Biochem., 27, 1639–1651, 1995.
- Müller, M. M., Sundman, V., Soininvaara, O., and Meriläinen, A.: Effect of chemical composition on
 the release of nitrogen from agricultural plant materials decomposing in soil under field
 conditions, Biol. Fertil. Soils, 6, 78–83, 1988.
- Murungu, F. S., Chiduza, C., Muchaonyerwa, P., and Mnkeni, P. N. S.: Decomposition, nitrogen, and
 phosphorus mineralization from residues of summer-grown cover crops and suitability for a
 smallholder farming system in South Africa, Commun. Soil Sci. Plant Anal., 42, 2461–2472,
 2011.
- Nakhone, L. N., and Tabatabai, M. A.: Nitrogen mineralization of leguminous crops in soils, J. Plant
 Nutr. Soil Sci., 171, 231–241, 2008.
- Nelson. D. N., and Sommer, L. E.: Total carbon, organic carbon and organic matter, in: Methods of
 Soil Analysis Part 2 Chemical and Microbiological Properties, edited by Page, A. L., Miller, R.
 H., and Keeney, D. R., SSSA Madison, WI, 539–589, 1982.
- Nourbakhsh, F., and Dick, R. P.: Net nitrogen mineralization or immobilization potential in a residue amended calcareous soil, Arid Land Res. Manage., 19, 299–306, 2005.
- Palm, C. A. and Sanchez, P. A.: Nitrogen release from the leaves of some tropical legumes as affected
 by their lignin and polyphenolic contents. Soil Biol. Biochem. 23, 83–88, 1991.
- Rasmussen, P. E., and Parton, W. J.: Long term effects of residue management in wheat-fallow: I.
 Inputs, yield, and soil organic matter, Soil Sci. Soc. Am. J., 58, 523–530, 1994.
- Sistani, K. R., Adeli, A., McGowen, S. L., Tewolde, H., and Brink, G. E.: Laboratory and field
 evaluation of broiler litter nitrogen mineralization, Bioresour. Technol., 99, 2603–2611, 2008.
- Steel, R. G. D., and Torrie, J. H.: Principles and Procedure of Statistics. McGrraw Hill Book Co Inc,
 New York, 1980.

Thippayarugs, S., Toomsan, B., Vityakon, P., Limpinuntana, V., Patanothai, A., and Cadisch, G. G.:
 Interactions in decomposition and N mineralization between tropical legume residue
 components, Agroforest, Syst., 72, 137–148, 2008.

- Trinsoutrot, I., Recous, S., Mary, B., and Nicolardot, B.: C and N flux of decomposing ¹³C and ¹⁵N
 Brassica napus L.: effect of residue composition and N content, Soil Biol. Biochem., 32, 1717–
 1730, 2000.
- Vahdat, E., Nourbakhsh, F., and Basiri, M.: Lignin content of range plant residues controls N
 mineralization in soil, Eur. J. Soil Biol., 47, 243–246, 2011.
- 9 Van Soest, P. J., Robertson, J. B., and Lewis, B.,A.: Methods for dietary fiber, neutral detergent fiber
 10 and non-starch polysaccharides in relation to animal nutrition, J. Dairy Sci., 74, 3584–3597,
 11 1991.
- 12

Soil properties	Values
Bulk density (Mg m ⁻³)	1.20
Particle density (Mg m ⁻³)	2.48
Porosity (%)	48.3
Sand (g kg ⁻¹)	241
Silt (g kg ⁻¹)	394
Clay (g kg ⁻¹)	365
Texture class	clay loam
рН	7.2
CEC (cmol kg ⁻¹)	7.3
Organic matter (g kg ⁻¹)	10.4
Organic C (g kg ⁻¹)	6.03
Total N (g kg ⁻¹)	0.58
C:N ratio	10:1
Total mineral N (mg kg ⁻¹)	8.7
Total organic N (mg kg ⁻¹)	591.0
P (mg kg ⁻¹)	3.4
K (mg kg ⁻¹)	88.0
$Fe (mg kg^{-1})$	15.7
Mn (mg kg ⁻¹)	17.0
Cu (mg kg ⁻¹)	1.02
$Zn (mg kg^{-1})$	1.16

1	
_	
2	

Tables 2. Biochemical composition of the plant residues used in the experiment (each value is the mean of three replications)

Plant residues	Plant organs	Total N	Total C	Lignin (LG)	Polyphenols (PP)	C/N	LG/N	PP/N	LG+PP/N
	8		g	kg ⁻¹					
Glycine max	Shoot	35.2a	447c	11f	13.1f	12.7	0.3	0.4	0.7
Glycine max	Root	12.8e	466b	29d	26.9d	36.4	2.3	2.1	4.4
Zea Mays	Shoot	9.6f	472ab	41b	29.5cd	49.2	4.3	3.1	7.3
Zea Mays	Root	4.0g	486a	48a	31.4c	121.5	12.0	7.9	19.9
Trifolium repenes.	Shoot	27.4b	397g	13f	18.0e	14.4	0.4	0.6	1.1
Trifolium repenes	Root	16.0d	423de	21e	20.2e	26.4	1.3	1.2	2.5
Poplus euramericana	Leaves	20.8c	435cd	34c	53.8a	20.9	1.6	2.6	4.2
Rubinia pseudoacacia.	Leaves	33.3a	404fg	28d	32.3c	12.1	0.8	1.0	1.8
Elagnus umbellate	Leaves	34.7a	418ef	32cd	38.7b	12.1	0.9	1.1	2.0
LSD(0.05)		3.14	14.16	4.53	3.77				

5 * Different letters in each column show significant differences among different treatments at a p < 0.05

Treatments				Da	ys after plan	t residues add	lition			-	LSD
	0	7	14	21	28	42	60	80	100	120	(p≤0.05)
					mg N kg	⁻¹ soil					
T ₀	13.7	13.9	12.9	17.1	30.9	65.9	63.1	75.6	77.7	51.7	2.88
T_1	14.8	39.2	49.2	76.8	96.7	158.1	165.2	174.1	188.7	160.9	7.90
T ₂	13.7	8.1	5.2	8.3	11.8	13.8	28.4	50.4	49.4	27.7	8.15
T ₃	13.7	7.4	6.2	6.9	10.5	23.1	21.2	36.1	46.7	21.0	5.34
T ₄	14.3	7.4	9.4	7.7	8.8	15.3	22.2	21.4	32.4	26.4	4.30
T ₅	14.1	19.0	21.6	55.5	62.5	86.8	127.6	150.8	145.8	93.3	7.31
T ₆	15.5	8.2	5.2	23.9	34.0	85.3	98.0	149.9	130.2	85.8	9.46
T ₇	13.0	5.7	4.1	8.6	22.6	55.5	73.1	106.8	87.3	66.9	8.39
T ₈	13.9	7.4	9.2	23.6	46.6	91.3	111.0	138.9	127.8	93.7	7.83
T ₉	12.9	9.4	14.5	25.3	51.1	80.1	92.7	140.0	116.4	93.5	6.88
LSD (n<0.05)	2.43	4.77	3.12	5.11	7.63	8.23	6.87	9.23	8.27	7.34	

Table 3. Changes in the concentration of total mineral N (TMN) of a soil amended with different plant residues and incubated at 25°C under controlled laboratory conditions

 $\frac{(p \le 0.05)}{T_0 = \text{ control}; T_1 = Glycine \text{ max shoot}, T_2 = G. \text{ max root}; T_3 = Zea \text{ mays shoot}, T_4 = Z. \text{ mays root}; T_5 = Trifolium repens \text{ shoot}; T_6 = T. repens \text{ root}; T_7 = T$

5 Poplus euramericana leaves; $T_8 = Rubinia pseudoacacia$ leaves; $T_9 = Elagnus$ umbellate leaves incubated under controlled laboratory conditions.

6 LSD represents the least significant difference ($p \le 0.05$) among incubation periods (within rows) and among the treatments (within column).

Table 4. Pearson linear correlation coefficients between initial quality characteristics of the plant

	\mathbf{N}_{\min}	TN	LG	PP	C:N	LG:N	PP:N
TN	0.89**						
LG	-0.84**	-0.66*					
PP	-0.42ns	-0.10ns	0.62*				
C:N	-0.69*	-0.80**	0.73*	0.07ns			
LG:N	-0.68*	-0.76**	0.77**	0.14ns	0.99**		
PP:N	-0.73*	-0.77**	0.82**	0.29ns	0.99**	0.98**	
LG+PP:N	-0.70*	-0.76**	0.79**	0.19ns	0.99**	1.00**	0.99**

residues and net N mineralization

** and * represent significant level at $p \le 0.01$ and $p \le 0.05$, respectively; ns means non-significant at a p<0.05 N_{min}, N mineralization; TN, total nitrogen; LG, lignin; PP, Polyphenols



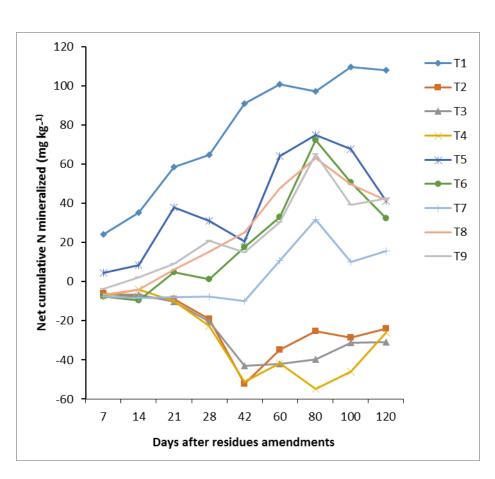




Figure 1 Net cumulative N mineralized (NCNM) from the added plant residues at different incubation periods. The legends at the top represent $T_1 = Glycine max$ shoot, $T_2 = G$. max root; $T_3 = Zea mays$ shoot, $T_4 = Z$. mays root; $T_5 = Trifolium repens$ shoot; $T_6 = T$. repens root; $T_7 = Poplus euramericana$ leaves; $T_8 = Rubinia$ *pseudoacacia* leaves; $T_9 = Elagnus$ umbellate leaves

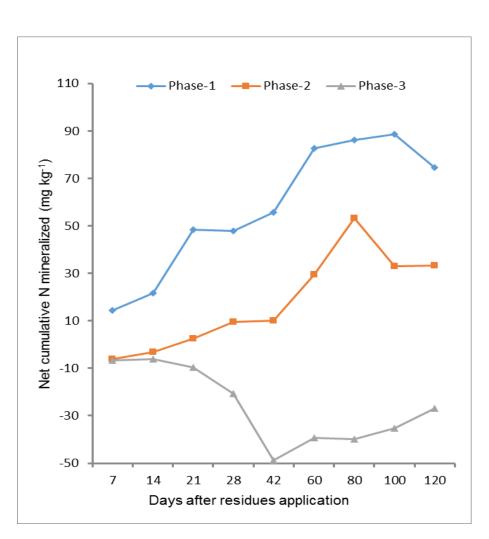


Figure 2 The mineralization – immobilization turnover (MIT) of added plant residues representing three
 phases during 120 days incubation.

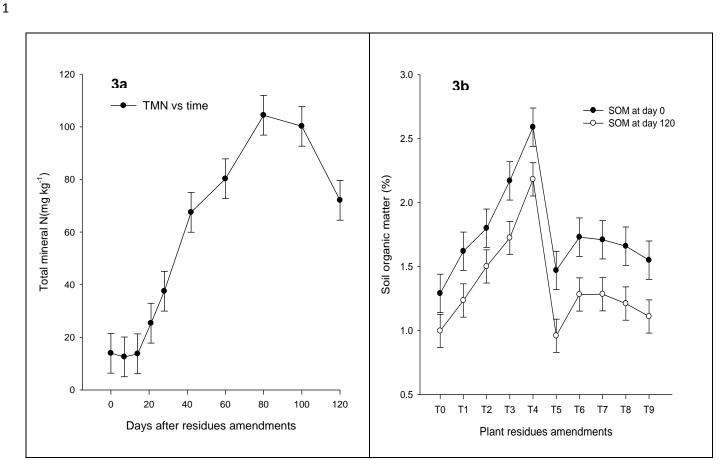




Figure 3. Mineralization trend of added plant residues across timings (3a) and soil organic matter (SOM) turnover of different plant residues (3b) recorded at the start of the experiment at day 0 and at the end of incubation at day 120. Vertical line on each major line represents the LSD ($P \le 0.05$) between incubation periods

5 and between each treatment, respectively.