

Spatio-temporal distribution dynamics of diverse soil nitrogen pools and patterns of nitrogen and carbon sequestration after slope farmland conversion to *Zanthoxylum bungeanum* plantations

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Research Article

Keywords: land-use conversion, *Zanthoxylum bungeanum*, mineral-associated nitrogen, organic nitrogen pools, farmland abandonment, soil nitrogen sequestration

Posted Date: September 6th, 2022

DOI: <https://doi.org/10.21203/rs.3.rs-2006198/v1>

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Abstract

Aims

Plant-soil interactions, and regulatory roles of soil nitrogen (N) fractions in availability and the magnitudes of N sequestration, therein the interplay of soil C-N in cold arid regions is poorly characterized.

Methods

Post-afforestation and land-abandonment dynamics of C and N sequestration, and total inorganic N (TIN) availability were identified by quantifying changes in diverse N fraction, and their distributions patterns in 0–100 cm soil profile across a chronosequence of *Zanthoxylum bungeanum* (28-year (H28), 20-year (H20), 15-year (H15), and 8-year (H8) old) plantations, and abandoned-land (GL), originally converted from former farmland (FL) in cold-arid valley in Southwest China.

Results

Afforestation and GL favored gains in labile and non-labile (LON and NLON) N fractions and total N stocks. Concentrations of LON fractions and TIN was comparatively higher at 0–40 cm. Gains in NLON fractions and total organic N (TON) was significantly higher in the deep soil, as confirmed by correlation and redundancy analysis. N and C sequestration was synchronous ($r=0.948$), with cumulative (0–100 cm) increase of 1.149–1.277 folds in H28 compared to H8, at an average sequestration rate of $1.336 - 0.121 \text{ Mg ha}^{-1} \text{ yr}^{-1}$, respectively. N pool management index (NPMI) correlated positively with soil TON, TIN, available phosphorus, potassium, and organic N fractions. NPMI improved significantly ($P < 0.05$) with the plantations age.

Conclusion

Plantations age and soil depths significantly influence ecosystems N dynamics. Furthermore, TON, NPMI, N fractions, and TIN can be useful indicators to gain comprehensive insights on ecosystems N restoration patterns.

Introduction

Land degradation defined as persistent loss of ecosystem services, has become an increasingly severe environmental and socio-economic problem throughout the world (Hussain et al., 2021). The arid/semi-arid biome accounting for 41% of the earth's land area, is in particular vulnerable to land degradation (Li et al., 2017). Agriculture is ranked at top among anthropogenic activities that cause degradation of soil

properties and function, resulting in 562 M ha of global land degradation (Wu et al., 2021). As an important terrestrial sink, forest ecosystems cover about 4 billion ha worldwide and store approximately 70% of all soil organic carbon (C), and $3.5\text{--}5.5 \times 10^5$ Tg of nitrogen (N) (Liu et al., 2018; Wang et al., 2020).

Afforestation on formerly arable land restores degraded soils, contributes significantly to the local economic development, and helps mitigate global warming (Gregory et al., 2016; Zhang et al., 2020). Soil N is one of the determinative factors of restoring degraded ecosystems, and substantial evidence suggests that soil N is responsible for the limitation or co-limitation of primary productivity and C sequestration (Guan et al., 2015; Tang et al., 2018; Li et al., 2019). N limitation to plant productivity is relatively common in arid and semi-arid ecosystems (Gelfand and Yakir, 2008; Tang et al., 2018). Afforestation in arid regions is an effective solution to increase soil N storage and thus resultant C sequestration (Liu et al., 2018). The majority of existing research has only focused on the post-afforestation C sequestration and hydrothermal activity. However, soil N dynamics and C–N interactions, which may be critical in deciding whether the C sink can be sustained over the long run in arid ecosystems are largely ignored (Schaeffer et al., 2003; Kicklighter et al., 2019; Nyameasem et al., 2020). The arid regions of Southwest China, such as the transition zone between Sichuan Basin and the eastern edge of the Qinghai-Tibet Plateau, the hotspots of land degradation, have experienced a massive land-use conversion of marginal farmlands (FL) to forest plantations under grain for green program of China (GGP) (Cheng et al., 2015; Liao et al., 2020). However, previous research on FL conversion in this region has mostly emphasized soil C accumulation; apart from total N (TN), other N cycling variables are rarely reported.

Soil N fluxes are primarily controlled by the small but highly bio-reactive labile organic N pools (Belay-Tedla et al., 2009). Fractionating soil organic matter (SOM) into distinct N pools based on stability and functional significance is presented to be a useful method for investigating post-afforestation implications on N dynamics (Galdo et al., 2003; Gregorich et al., 2006; Darby et al. 2020). These pools include the light fraction organic matter N (LFON) separated based on specific density ($1.6\text{--}2.0 \text{ g cm}^{-3}$), particulate organic matter ($> 53 \mu\text{m}$) associated N (PON: represents an easily decomposable pool); microbial biomass (MBN: a highly labile, dynamic and rapidly recycling N pool), soluble organic N (SON), and alkali hydrolysable N (AHN), all represent highly mineralizable N pools; the substances that are non-labile (NLON) and more stable to decomposition; heavy fraction organic N (HFON), and mineral-associated N (MAON) are generally regarded as highly stable components of N pools with a relatively long turnover time (Stevenson, 1994; Qiu et al., 2015; Li et al., 2019; Cai et al., 2021).

These fractions are highly susceptible to management practices and land-use systems. For example, LFON is sensitive to management practices, correlates with the rate of N mineralization, functions as a short-term sink for mineral N, and could be a good indicator of a soil's capacity to supply N (Compton and Boone, 2002; Malhi et al., 2003; Tan et al., 2007). Soil organic N accounts for as much as 95% of total N in the topsoil and enters the soil as POM-N through above-ground plant litter and root litter (Martínez et al., 2017). Similarly, Drescher et al. (2020) and Roberts et al. (2009) attributed soil AHN as potentially

mineralizable N and reported AHN was variable across sites and soil depths. Furthermore, earlier research has also demonstrated that ecosystems stabilize SOM by increasing biochemically resistant compounds to sequester N and C long-term (Filley et al., 2008; Zhang et al., 2020). The soil MAON, for example, represents approximately 82% of the total N in most soils, serves as an important and stable pool of SOM, and is susceptible to management practices (Cai et al., 2021). Thus, analyzing distinct N pools simultaneously regarding post-afforestation years can shed light on patterns of soil N cycling that no single pool can adequately describe.

Post-afforestation research on labile organic N fractions and their temporal and depth variability is still quite limited, particularly in arid regions, and the dynamic balance between labile and recalcitrant pools is also rarely addressed (Assefa et al., 2017). Additionally, existing research is mainly limited to shallow soil layers; for example, Yang et al. (2004) and Gelaw et al. (2014) asserted that there is no significant difference in soil organic C (SOC) and N concentrations below 40 cm soil depth, because soil disturbances are limited to the topsoil. Substantial amounts of N and C are stored in subsoil layers, only focusing on the topsoil would not accurately assess TON and SOC changes following land-use conversion (Gregory et al., 2016; Zhang et al., 2020). Moreover, as forest stands mature, the relative occurrence of organic N fractions and mineral N availability may change because of the N requirements for tree growth, litter inputs, and soil microbial activities (Assefa et al., 2017; Yang et al., 2019; Darby et al., 2020; Xia et al., 2021). Increases in soil N storage generally boost SOC stocks and sink strength, which becomes visible as plantations age (Chang et al., 2014). Thus, examining the temporal and vertical distribution patterns of labile and physically protected organic N pools may result in a paradigm shift in our current understanding of the mechanisms behind the long-term persistence of soil sink strength and N supply capacity.

Zanthoxylum bungeanum colloquially referred to as "Chinese prickly ash," is a calcium-loving, drought-tolerant, photophilous tree thrives in arid climates and regions that have suffered serious land degradation (Moberg and Persson, 2011). Besides, due to its high adaptability and rapid growth, it is one of the key species that has been effectively used in GGP for restoring previously depleted farmlands of southwest China. Currently, *Z. bungeanum* is planted on 1087,667 ha in China, yielding 380,000 tons of dry pericarp and a value of 14.8 billion yuan, benefiting 45 million people (Liu et al., 2021). However, no prior work has thoroughly examined the vertical and temporal patterns of N storage in soil and how the rate of soil N sequestration alters following afforestation with *Z. bungeanum* plantations? What regulatory roles do highly labile and physically protected organic N fractions play in the N storage under predominantly arid conditions? Furthermore, whether the relative distribution of labile and non-labile N pools within the vertical soil profile and the N pool management index (NPMI) can be used as sensitive indicators for assessing the success of afforestation has not been addressed. To bridge these gaps, the current study was aimed to 1) investigate the mineral and subsurface soil profile (0–100 cm) in order to determine spatio-temporal patterns of total N, and SOC storage, sequestration rates, and vertical distributions; 2) fractionate labile and recalcitrant N pools, and explore their regulatory roles in soil TON dynamics; 3) and identify reliable indicators to evaluate soil quality.

Materials And Methods

Site description

The study area is located in Shengou village, Feihong town, Mao County, Aba Tibetan and Qiang Autonomous Prefecture, Sichuan Province (103°43, 31°50'N), at an altitude range of 2647–2663 m. The mean annual temperature is 11.0 °C, and average annual precipitation is 486.3 mm. The soil is yellowish brown to brown classified as Haplustepts (USDA). The vegetation is dominated by drought-tolerant mesophytes. The local conventional crops mainly include *Solanum tuberosum* and *Zea maize*. Slope farmlands (FL) were converted to Chinese prickly ash (*Zanthoxylum bungeanum*) plantations (aged 8–28 years). Abandoned-land (GL) is a naturally formed grassland after the abandonment of FL, and its formation time is also approximately 28 years. The *Zanthoxylum bungeanum* plantations are monocultures. The dominant plant species in GL are primarily annual herbs. The selected slope FL and GL are continuous and have never been converted to other land uses. The slope FL and Chinese prickly ash plantations have similar fertilization management, while the GL plots have never been fertilized, irrigated, or used for grazing. Weeds are manually eradicated in FL as well as *Z. bungeanum* plantations.

Experimental design and sampling

Soil samples were collected from six distinct sites, including four different age monocultures of Chinese prickly ash (including 8-year (H8), 15-year (H15), 20-year (H20) and 28-year (H28) old plantations), and adjacent cultivated FL and abandoned-GL, farmlands were selected as control. All the selected plots have similar soil types (Haplustepts), physiographic conditions, slope gradient (21–24°) and aspect (SW 123°–140°). The mean height, the average ground diameter, and the canopy densities were 2.46 m, 7.9 cm and 74.1 % for H8, respectively; 2.93 m, 14.1 cm and 79.3% for H15, respectively; 3.24 m, 19.4 cm and 81.7% for H20, respectively; 3.26 m, 24.5 cm and 85.5% for H28, respectively. Whereas, the average grass height and vegetation cover in GL plots was 0.81 m and 65.3%, respectively. Leaf litter biomass was recorded as 4.36 Mg/ha, 3.45 Mg/ha, 3.01 Mg/ha, 1.94 Mg/ha, and 4.55 Mg/ha in H28, H20, H15, H8, and GL respectively. The average fine root biomass was recorded as 20.3 Mg/ha, 11.1 Mg/ha, 4.7 Mg/ha, 1.5 Mg/ha, 1.2 Mg/ha and 38.8 Mg/ha, for H28, H20, H15, H8, FL, and GL respectively.

Three sampling sites (10 × 10 m) were set at random in each plot and before collecting soil samples, litter layer was removed. Stratified soil samples were collected in three replications using the quadrat method (Deng et al., 2013) with a hand auger (5 cm in diameter), from the soil surface to a depth of 100 cm at 20 cm interval as follows: 0–20, 20–40, 40–60, 60–80 and 80–100 cm. Soil samples were returned to the laboratory, and manually sieved through a 2-mm mesh to remove roots and other debris. Soil from each specific layer was thoroughly mixed to generate composite samples. Each plot's fresh soil samples were evaluated for soil MBN, ammonium ($\text{NH}_4^+\text{-N}$), and nitrate ($\text{NO}_3^-\text{-N}$) contents. The total soil inorganic N (TIN) concentration was calculated as the sum of $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$ (Li et al., 2019). The remaining soil was air-dried, ground, and sieved before subjecting to other chemical analyses.

Analyses of soil physico-chemical properties

Soil total organic N (TON) concentration was determined by micro-Kjeldahl method described by Gong et al. (2011) and total organic C (TOC) was determined by potassium dichromate ($K_2Cr_2O_7$) oxidation at 170–180 °C followed by titration with 0.1 mol L⁻¹ ferrous sulfate (Yang et al., 2012). Soil MBN content was measured using chloroform fumigation-extraction method (Hassan et al., 2018). Following the extraction, MBN content was measured by determining N in the filtrate using a CN elemental Analyzer (Vario TOC select, Elementar, Germany). The soluble organic N (SON) was calculated as the difference between total N and TIN in the extracted solution (Jiang et al., 2010). The HFON and LFON contents were determined by the procedures described by Huang et al. (2011). Soil PON content was assessed by the methods of Cambardella and Elliott (1994), described by Chen et al. (2018). Soil MAON content was determined according to the methods of Hassink (1997) as described by Cai et al. (2021). AHN in the soil was determined by reduction-diffusion method as described by Hassan et al. (2017). Soil N was divided according to oxidation and extraction, with 333 mmol L⁻¹ $KMnO_4$ solution into labile (LON) and non-labile (NLON) organic N, using the procedure of Blair et al. (1995) and Westerhof et al. (1998) as described by Gong et al. (2011). Soil bulk density (BD) was determined by following Darby et al. (2020). Available phosphorus (AP) was measured according to Hassan et al. (2016), and available potassium (AK) concentration was determined by following procedure of Gong et al. (2011).

Calculations

The stocks of soil N fractions and SOC for each land-use type and soil layer was calculated according to Eq. (1) described by Li et al. (2019):

$$N_{\text{stock}} = BD \times N_{\text{concentration}} \times D / 10 \quad \text{Eq..... (1)}$$

where N_{stock} represents stock (kg ha⁻¹) of N fractions including MBN, TON, TIN and so on; BD is the soil bulk density (g cm⁻³); $N_{\text{concentration}}$ is concentration (g kg⁻¹) of respective N fraction; and D is the soil sampling thickness (i.e. 20 cm). The cumulative stock at a 0–100 cm soil depth is the sum of the stocks in all of the soil layers.

Total organic N storage (Mg N ha⁻¹) of soil at 0–20 cm, 20–40 cm, 40–60 cm, 60–80 cm or 80–100 cm depth and cumulative TON storage (0–100 cm) was calculated on area basis. The amount of sequestered organic N (Mg N ha⁻¹) in 0–20 cm, 20–40 cm, 40–60 cm, 60–80 cm or 80–100 cm depth and cumulative TON storage (0–100 cm) was estimated after subtracting the TON storage of the control (cultivated FL) from that of other land-use types (Gong et al., 2011). While the N accumulation rate or N sequestration rate for each land-use type was calculated by dividing the total N stock by the number of years since the start of accumulation (post-afforestation years i.e. 8, 15, 20 and 28 years, and land abandonment years i.e. 28 years), assuming a constant N accumulation rate (Leblans et al., 2014).

The mean concentrations (g kg⁻¹) in 0–100 cm soil depth for each organic N fractions and TIN were calculated by the following equation 2:

The mean average concentration = Total stocks/ Total soil weight Eq.....(2)

where the total stocks and total soil weight are the sum of the stocks and soil weight of 0–100 cm soil depth.

N liability (L), liability index (LI), N pool index (NPI) and N pool management index (NPMI) was calculated for each land-use type according to Gong et al. (2011) by using the following formulas:

$$\text{NPI} = (\text{sample TON}) / (\text{reference soil TON}) \quad \text{Eq..... (3)}$$

The TON of cultivated FL was used as reference soil TON

Sample lability index (LI) was determined according to following equation:

$$LI = (\text{sample lability}) / (\text{reference soil lability}) \quad \text{Eq..... (4)}$$

The lability of cultivated FL was used as reference soil lability

where lability (L) is L_N / N_{LN} Eq..... (5)

LN is labile N ($333 \text{ mmol L}^{-1} \text{ KMnO}_4$ extracted N), and NLN is non-labile nitrogen.

These two indices were used to calculate NPMI as

$$\text{NPMI} = \text{NPI} \times \text{LI} \times 100 \quad \text{Eq..... (6)}$$

Statistical analysis

Excel 2016 was used to sort the data, and SPSS 17.0 was used to conduct all statistical analyses (SPSS Science, Chicago, USA). Redundancy analysis (RDA) was applied to investigate the relationship among TON, TIN and TOC contents, soil organic N fractions, AK, AP contents and soil bulk density. CANOCO 5.0 was used to conduct RDA (Microcomputer Power, Ithaca, NY, USA). Graphs, and Pearson's correlation analysis was produced by OriginPro 2019b 64Bit.

Results

Basic soil properties

Land-use change had no significant ($P < 0.05$) effects on soil BD. Soil BD, in general, was low for 0–20 cm and increased as the soil depth increased across all land-use types (Fig. 1). Converting FL to *Z. bungeanum* plantations and GL significantly ($P < 0.05$) affected soil TOC contents (Fig. 1). The highest TOC content was observed for GL, and significantly ($P < 0.05$) positive impacts on soil TOC content were also observed as a result of FL conversion to *Z. bungeanum* plantations (Fig. 1). TOC content increased as the plantations age and ranked as H28 > H20 > H15 > H8. Generally, higher TOC contents were observed at 0–20 cm, however; the gains in TOC content at deeper soil horizons were considerably higher

than the topsoil (Fig. 1). Similarly, FL conversion to *Z. bungeanum* plantations and GL also significantly ($P < 0.01$) improved soil AK and AP concentrations, and their spatial distributions followed a general trend of decline as the soil depth increased (Fig. 1).

Spatio-temporal patterns of soil N sequestration and availability

Land-use change from FL to GL and *Z. bungeanum* plantations significantly improved ($P < 0.05$) soil TON content and stocks across all soil depths (Fig. 2). Soil TON contents also increased with the plantations age. Generally higher TON contents were recorded for the topsoil. However, post-afforestation gains in the TON content in the deeper soil profile were higher than in the topsoil (Fig. 2). The TON content increased by 1.257-fold at 80–100 cm than 1.124-fold at 0–20 cm soil depth in H28 compared to H8. The land-use change also significantly ($P < 0.05$) affected soil TON stocks, and differences were more pronounced with the number of post-afforestation years. TON stocks (0–100 cm) in soil followed a similar trend of increase as the plantations aged, and the highest soil TON stock, 1.920 Mg ha^{-1} , was recorded for H28, which is 1.248-folds higher than the FL (Fig. 2). Initially, TON losses were observed in H8 plantations. However, as the age of plantations increased, TON stocks recovered, and plantations sequestered TON at an average rate of $0.046\text{--}0.121 \text{ Mg ha}^{-1} \text{ yr}^{-1}$. There were no significant differences in TON stocks between GL and H28.

The availability of the TIN also improved significantly upon land-use change from FL to GL and *Z. bungeanum* plantations ($P < 0.05$). The distribution of TIN followed a similar pattern to the TON contents i.e., increased with the post-afforestation years and decreased with the soil depth (Fig. 2). The highest TIN content (0–100 cm) was observed for GL, which is 1.632-folds higher than FL. The TIN content of *Z. bungeanum* plantations also increased steadily as the plantations age increased, and an overall (0–100 cm) increase of 1.302-folds and 1.499-folds was observed in H28 compared to H8 and FL, respectively. Similarly, higher TIN concentrations were observed at 0–20 cm than deeper soil layers (Fig. 2). FL abandonment to GL resulted in the highest TIN stocks, 0.377 Mg ha^{-1} , with a cumulative increase (0–100 cm) of 1.563-fold compared to the FL. Plantations age also had a strong impact on TIN stocks, and an overall increase of 1.258-fold of TIN stocks in H28 was observed compared to H8.

Spatio-temporal dynamics of soil organic N pools

Land-use change from FL to GL and *Z. bungeanum* plantations significantly affected concentrations of most labile organic N fractions i.e. MBN, SON, and AHN, and their contents varied significantly with the soil depth ($P < 0.05$). Farmland abandonment to GL resulted in the highest increase in the MBN, SON, and AHN concentrations (Fig. 3). Plantations' age also significantly affected their contents, and an overall trend of increase was observed as the plantations aged. The maximum post-afforestation increase in the concentrations of MBN, SON, and AHN contents in H28, is 1.508-folds, 1.423-folds, and 1.458-folds higher than the FL, respectively (Fig.3). Soil depth also had significant impacts on the dynamics of these organic N fractions ($P < 0.05$), and the highest concentrations were observed at 0–20 cm, and their concentrations declined as the soil depth increased (Fig. 3).

Land-use change as well as soil depths significantly ($P < 0.05$) improved concentrations of physically protected organic N (PON, MAON, LFON, HFON) pools (Fig. 4). Conversion of FL to GL resulted in the highest gains of PON, HFON, MAON, and LFON contents. FL conversion to *Z. bungeanum* plantations also significantly improved PON, HFON, MAON, and LFON contents and the highest concentrations were observed for H28, which is 1.301-folds, 1.094-folds, 1.115-folds, and 1.223-folds respectively, higher than H8, at 0–20 cm. However, post FL conversion gains in their concentrations at deeper soil horizons were significantly higher than the topsoil.

Soil labile and non-labile organic nitrogen and nitrogen pool management index

Land-use conversion from FL to *Z. bungeanum* plantations and GL significantly ($P < 0.05$) influenced LON and NLON contents (Fig. 5). Soil depths also significantly affected LON and NLON contents; higher contents were observed in the topsoil (0–20 cm), however the gains in subsoil layers were comparatively higher. For instance, 28-year post-afforestation (H28), increases in LON and NLON contents were 1.228–1.022 folds at 0–20 cm while 1.409–1.191 folds at 80–100 cm respectively, compared to H8. The gains in NLON contents were relatively lower compared to the increases in LON content, and an overall increase (0–100 cm) of 1.388-folds and 1.219-folds was observed in LON and NLON content of H28, respectively, compared to the FL (Fig. 5). However, compared to the LON, the NLON content was higher at all soil depths and across all land-use types.

Land-use conversion from FL to *Z. bungeanum* plantations and GL significantly influenced ($P < 0.05$) labiality of N (LN), labiality index (LI), N pool index (NPI), and N pool management index (NPMI) at 0–100 cm soil depth (Fig. 6). Soil LN and LI significantly varied among all land-use types generally, LN was recorded highest for GL and continuously improved with the plantation age, and highest was recorded for H28, which is 1.143-folds higher than H8. Similarly, plantations' age also significantly improved soil NPI and NPMI. The highest NPI and NPMI were also recorded for GL, followed by H28 > H20 > H15 > H8 (Fig. 6). Land-use conversion from FL to *Z. bungeanum* plantations increased soil NPMI by 1.424-folds in H28. NPMI of managed plantations and FL, also correlated positively with all organic N fractions, and soil AP, and AK contents.

Relationship between soil properties and soil N pools

The relationship among the soil physicochemical properties, labile and non-labile organic N fractions, TON, TIN, and TOC average concentrations (0–100 cm), was examined by Redundancy analysis (RDA) and Pearson correlation analysis (Fig. 7 and Fig. 8). RDA biplot was used to assess factors that influenced soil TON, TOC, and TIN contents across land-use types at a soil depth of 0–100 cm. Soil TON, TOC, and TIN contents were used as response variables, while BD, AP, AK, PON, AHN, MBN, SON, LFON, MAON, and HFON were used as the explanatory variables. The eleven selected soil variables explained the variation in soil TON, TOC and TIN contents, and these variables accounted for 91.4% of the variance. The first two RDA axes explained 85.2% and 90.4% of data variations, respectively. RDA showed that gains in soil TON content were related to HFON, LFON, MAON, and PON, while gains in soil TOC content were most closely associated with AHN, MBN, LFON, and PON, which was further confirmed by Pearson

correlation analysis (Fig. 8). The vector angles of soil organic nitrogen pools with TON and TOC contents were smaller than 90°. Moreover, correlation coefficients indicated positive relations of these fractions with N and C concentrations. The angles of soil BD with TON and TOC were greater than 90°, indicating negative relations. The results showed that soil, MAON, HFON, LFON, AHN, PON, MBN, and SON explained most of the variations in soil N gains, thus are the most important contributors to the variations in ecosystems TON pool stability and TOC sequestration.

Discussion

Synchronous soil N and C sequestration

Previous studies have attributed land-use change as one of the most important human effects on the rate of changes in TON and TOC concentrations and stocks (Tan et al., 2007; Li et al., 2012; Guan et al., 2015). The current study showed significant gains in soil TOC and TON after afforestation on previously cultivated FL (Fig. 1 and Fig. 2). Lemenih et al. (2005) and Zhang et al. (2020) showed 19% and 15%, respectively, increases in soil TON stocks after afforestation on agricultural land. Similarly, Laganière et al. (2010) and Assefa et al. (2017) advocated higher soil TON stocks in tree plantations is because trees have plenty of litterfall and extensive root system, which increase organic matter inputs and reduce losses from soil erosion. Similarly, 2.243-fold increase in leaf litter stock and 16.917-fold increase in fine root biomass was observed in H28, as compared to H8. Similar to other studies (Chang et al., 2014; Assefa et al., 2017; Liu et al., 2018), the current study also showed that the gains in TOC and TON stocks contents strongly depend on the number of post afforestation years.

Initially, losses of soil TON stocks were observed in H8 (20–80 cm) than in the original FL (Fig. 2). Wang et al. (2019) and Chang et al. (2014) also reported a decline in soil TON and TOC contents during the early stages of FL restoration because of the disturbance during site preparation, the lower productivity of new vegetation, the scarcity of leaf litter and low root biomass. Li et al. (2012) reported significant soil C and N stock increases were found 30–50 years after afforestation, and before these time points, C and N stocks were either depleted or unchanged. Vesterdal et al. (2002) attributed it to young stands' low organic matter inputs and high rates of C decomposition inherited from farmland to meet the needs of rapidly growing stands. According to Compton and Boone (2000), there is typically a time lag for soil C accumulation because, during afforestation in North Carolina, nearly all of the increase in ecosystem C went into standing biomass, but not soil C. Furthermore, according to Xia et al. (2021) and Darby et al. (2020), as plant biomass accumulation slows in mature stands, soils may re-accumulate N that is no longer required for new plant biomass, supporting larger soil N storage in older stands. Similarly, in the current study, 28-years later, afforestation resulted in cumulative (0–100 cm) 23.128 Mg ha⁻¹ gains in TON stocks at an accumulation rate of 0.121 Mg ha⁻¹ yr⁻¹, which is 1.167-fold higher than H8 (Fig. 2), implying *Z. bungeanum* plantations are capable of restoring soil N and C in this arid region.

Soil TON follows similar temporal patterns of SOC sequestration, and slight improvements in TON stocks can significantly increase soil SOC gains (Leblans et al., 2014; Liu et al., 2018). Bingham and Cotrufo

(2016) further established that soil N plays an indispensable role in C sequestration by improving the stability of SOM, particularly in the mineral soil layer. Similarly, the current study showed that TOC sequestration was 6.796-fold higher than TON sequestration in H28 (Fig. 2). Zhang et al. (2020) showed SOC sequestration is synchronous to TON. In the present study, this is also evident from the significantly ($P < 0.05$) positive correlation observed between TOC and TON ($r = 0.948$) contents and further strengthened by the RDA (Fig. 7 and Fig. 8).

Similar to several other studies (e.g., Chen et al., 2010; Justine et al., 2017; Xia et al., 2021), results of the current study also revealed that higher TON and TOC concentrations were found in the upper 0–40 cm soil layers compared to deep soil horizons (Fig. 1 and Fig. 2). However, the gains in soil C and N concentrations below 0–40 cm were potentially higher compared to the topsoil. Gregory et al. (2016) also reported higher C and N sequestration potential of deep soil. Liao et al. (2020) reported that C and N pools in the subsoil (below 0–30 cm) are assumed to be more stable than the topsoil due to differences in the composition of SOM and environmental conditions. In addition, Li et al. (2019) reported that soil N is one of the determinative factors of restoring degraded ecosystems. Leblans et al. (2014) also advocated that N accumulation accelerates soil development. The current study revealed strong interaction in soil C and N under *Z. bungeanum* plantations which improved significantly as the age of the plantations increased. Thus, planting *Z. bungeanum* is an effective approach for restoring ecosystems N gains and C sequestration under the current land-use change scenario.

Post land-use changes in concentrations of soil organic N fractions and TIN availability

Soil organic N fractions constitute biologically active N pools, act as sources of easily mineralized N, and contribute to the stable N pools (Batlle-Aguilar et al., 2011). These organic N pools are susceptible to land-use change (Magid et al., 2010; Zhou et al., 2020). In the current study, land-use change from FL to *Z. bungeanum* plantations and GL evidently favored gains in MBN, AHN, SON, PON, MAON, LFON, and HFON contents (Fig. 3 and Fig. 4). Mao and Zeng (2010) investigated the impact of cropland conversion to tree-based plantations and reported improved soil quality and fertility was due to increased POM-N and MBN contents. Similarly, in the present study, afforestation with *Z. bungeanum* enhanced resulted in overall (0–100 cm) gains of 1.619-folds and 1.591-folds in PON and MBN contents respectively, in H28 compared to the original FL (Fig. 3 and Fig. 4). Soil OM is both a substrate *for* and a product *of* soil microorganisms resultantly, microbial necromass is estimated to be the most abundant form of SOM (Kallenbach et al., 2016). Wild et al. (2015) and Li et al. (2019) further argued MBN affects N dynamics because the microbial demand for C and N drives SOM decomposition and ecosystem C and N sequestration. Similarly, in the current study, MBN was the most abundant labile organic N fraction following afforestation, suggesting that FL conversion to *Z. bungeanum* plantations is conducive to soil N restoration.

Several studies have also regarded MBN, AHN, SON, and PON as potential sources of mineralized N i.e., plant-available TIN in soil solutions, which in the current study is also supported by the RDA (Fig. 7) and correlation analysis (Fig. 8). Similar to other studies (e.g., Mendham et al., 2004; Belay-Tedla et al., 2009;

Mao and Zeng, 2010; Maharjan et al., 2017; Li et al., 2019), soil TIN contents were most significantly correlated to AHN, PON, MBN, and LFON (Fig. 8). Increased soil labile organic N fractions accumulation upon afforestation and FL abandonment is linked with the reduced disturbance caused by tillage and increased biomass return to the ecosystem in the form of above and belowground plant litter (Magid et al., 2010; Asseefa et al., 2017; Zhou et al., 2020). Increments in biomass and litter return thus ultimately labile organic N fractions accumulation is directly controlled by the post-afforestation duration (Mendham et al., 2004). In the present study, concentrations of labile N fractions increased significantly ($P < 0.05$) as the plantations aged (Fig. 3), which coincides with the other similar studies (Mao and Zeng, 2010; Darby et al., 2020; Xia et al., 2021), and indicated positive impacts of *Z. bungeanum* plantations soil N pool stability and N supply capacity.

The scientific community is divided on the role of LFON and PON in the course of N cycling. Several reports support these two fractions play an important role in stabilizing soil N pools as well as serve as short term N sink (Compton and Boone, 2002; Mendham et al., 2004; Tan et al., 2007) and upon demand serve as highly labile N source (Gregorich et al., 2006; Yan et al., 2007). In the current study, LFON ($r = 0.981$) and PON ($r = 0.983$) correlated positively with N contents suggesting their key role in the N sequestration (Fig. 8). The increased PON and LFON post-afforestation can positively affect soil physical structure and aggregation and make soil organic C and N pools more stable (Mendham et al., 2004; Tan et al., 2007; Gartzia-Bengoetxea et al., 2009). Soil LFON and PON cumulative contents (0–100 cm) increased by 1.329-fold and 1.394-fold in H28 compared to the youngest H8 plantations, respectively (Fig. 4). Liao et al. (2020), and Gregory et al. (2016) attributed increased stability of soil N to the increased PON and LFON fractions in the subsurface soils. Similarly, results of the current study showed that soil depth significantly affected vertical dynamics of labile organic N fractions, and with the plantations age significant ($P < 0.05$) gains in HFON, LFON, PON, SON, AHN and MBN contents were observed across all soil depths (Fig. 3 and Fig. 4).

Land-use change improved labile soil organic N concentrations across all soil depths; however, increases in HFON and MAON concentrations were much greater and improved continuously with the age of the *Z. bungeanum* plantations (Fig. 4). The fractions of N consisting of SOM intimately associated with minerals, decrease microbial and enzymatic capacity to decompose organic substrates (Plaza et al., 2019), thus serve as sinks of organic N in the soil (Whalen et al., 2000; Tan et al., 2007;), and in principal are the long-term N sources for plants and microbes (Qiu et al., 2015). Hassink (1997) and Cai et al. (2021) showed increased soil MAON contents significantly improved N pool stability and capacity of soils to preserve N and C long-term. In the present study this is supported by RDA (Fig. 7), and confirmed by the significant ($P < 0.05$) positive correlation observed for HFON ($r = 0.997$, $r = 0.933$) and MAON ($r = 0.999$ and $r = 0.943$) with soil TON and TOC, respectively (Fig. 8). Thus, it can be asserted that post-afforestation increases in HFON and MAON played a significant role in increasing the stability and storage capacity of TON sinks in soils. The land-use-related variations in these pools are generally linked to the changes in OM inputs with post-afforestation years, as increased aboveground biomass output and litter inputs increase with the plantation age (Laganière et al., 2010; Qiu et al., 2015; Feyissa et al., 2020; Xia et al., 2021). Cai et al. (2021) reported increased MAON contents improved soil aggregation and

structural stability; previous studies have also shown *Z. bungeanum* plantations as an effective measure to cease soil erosion and reclaim degraded soils (Cheng et al., 2015).

Additionally, while the topsoil contained higher concentrations of HFON and MAON, the increases in the subsurface layers were much more significant than those in the topsoil (Fig. 4), indicating that *Z. bungeanum* plantings are well-suited for ecosystem restoration projects. However, to date, there has not been a comprehensive examination of the temporal and vertical dynamics of MAON and HFON following afforestation, particularly under arid conditions. Therefore, variations in MAON and HFON must be included in future studies to gain insights into the N sink stabilization paradigm for long-term N sequestration in intensively managed plantations.

Heterogeneous vertical distribution dynamics of LON, NLON and NPMI

The soil N pool management index (NPMI) is derived from Blair et al. (1995), who first combined the soil TOC pool and C lability index to evaluate the capacity of management systems in promoting soil quality. Later on, Westerhof et al. (1998) and Gong et al. (2011) adapted this for soil N and suggested NPMI is a useful index to evaluate and compare the changes that occur in total and labile N as a result of management practices, with an emphasis on the changes in labile and non-labile N in SOM. Thus NPMI can provide a useful parameter to assess the capacity of management systems to improve soil quality (Westerhof et al., 1998; Gong et al., 2011). However, these indexes have largely been ignored in ecological restoration studies. The current study showed that converting FL to *Z. bungeanum* plantations and GL considerably improved ecosystems N lability, NPI, LI, and NPMI (Fig. 6). Based on the previous reports (Gami et al., 2009; Lal, 2016; Jiang et al., 2021), the higher value of the C pool management index implies that soil is being restored, improved and maintained. The strong stoichiometric association between soil SOC and TON, also suggests the values of NPMI may also serve as valuable indicator to reflect the status of soil quality.

The trend of NPMI among soil horizons can be further established by the relative soil LON and NLON stocks distribution along the soil depth. Soil LON and NLON exhibited heterogeneous vertical distribution patterns across the soil profile in all land-use systems (Fig. 5). Higher distribution of soil labile N in the topsoil layers significantly increases soil N supply capacity (Chen et al., 2018). In their experiment, Chen et al. (2018) found that labile N in 0–30 cm soil profile correlated positively with N mineralizing enzymes and soil N supply capacity. In the current study, surface soils had higher soil LON compared to the subsurface soil (Fig. 5), similarly, in the present study, the higher value of NPMI for H28 might be due to lack of disturbance and reduced mineralization of labile N pools (Compton and Boone, 2002; Mao and Zeng, 2010; Zhou et al., 2020). Feyissa et al. (2020) suggested that the higher aboveground biomass production and litter inputs could increase substrate inputs and labile C and N concentrations in the topsoil. Previous studies have established that ecosystems, to sequester N and C on a long-term basis, need to stabilize SOM, and one of the mechanisms is by increasing compounds that are biochemically resistant towards microbial decay (Filley et al., 2008; Belay-Tedla et al., 2009; Feyissa et al., 2020). The present study revealed that the NLON was the dominant N form in the soil, and it correlated strongly with

the NPMI ($r = 0.886$, $P < 0.05$), indicating increased N pool stability and long-term N storage. Previously post-afforestation improvements in soil available nutrients have been regarded as indicators of soil quality and fertility (Gao and Huang, 2020; Tellen and Yerima, 2018). In the current study, NPMI exhibited significant positive correlation with soil available nutrients AP ($r = 0.900$, $P < 0.05$), and AK ($r = 0.865$, $P < 0.05$). Therefore, these results imply that long-term afforestation with *Z. bungeanum* plantations on previously cultivated FL could promote soil N sequestration, and NPMI could be used to estimate ecosystem N restoration and the status of soil quality.

Conclusion

Farmland conversion to GL and *Z. bungeanum* plantations favored TON and TOC storage as well as TIN availability, implying that soil restoration can play significant roles in improving C and N sink. Post-afforestation years significantly increased soil MBN, SON, AHN, LFON, MAON, PON, and HFON contents at all soil depths, indicating improved N pools stability and labile sources for plant-available inorganic N. The NLON was the dominant form of N in *Z. bungeanum* plantations, suggesting that N storage in these soils can be sustained long-term. Comparatively higher gains in HFON, MAON, and NLON in the subsurface soil also showed the higher N sequestration potential of the subsoil and further suggested that deep soil has a greater role in N and C sequestration. In addition, soil NPMI demonstrated soil N supply and storage capacity improved with continuous planting of *Z. bungeanum* plantations on former depleted farmlands. Thus obtaining credible estimates of spatio-temporal variability on soil TON storage, organic N fractions, and TIN availability along with NPMI may serve as useful indicators for impact assessment of land-use change and sustainable establishment of intensively managed plantations. Furthermore, subsoil along with mineral soil and temporal patterns of different organic N fractions must be included in future studies across diverse land-use systems and climate zones for a thorough understanding of the mechanisms explaining long-term impacts of ecosystems N changes under land-use conversion.

Declarations

Acknowledgements

This work was supported financially by The National Key Research and Development Program of China (Program No. 2018YFD1000605, and 2020YFD1000700) and The Projects of Science and Technology Department of Sichuan Province (Grant No. 2016NYZ0035, 2019ZHFP0002, and 2021YFYZ0032).

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Figures

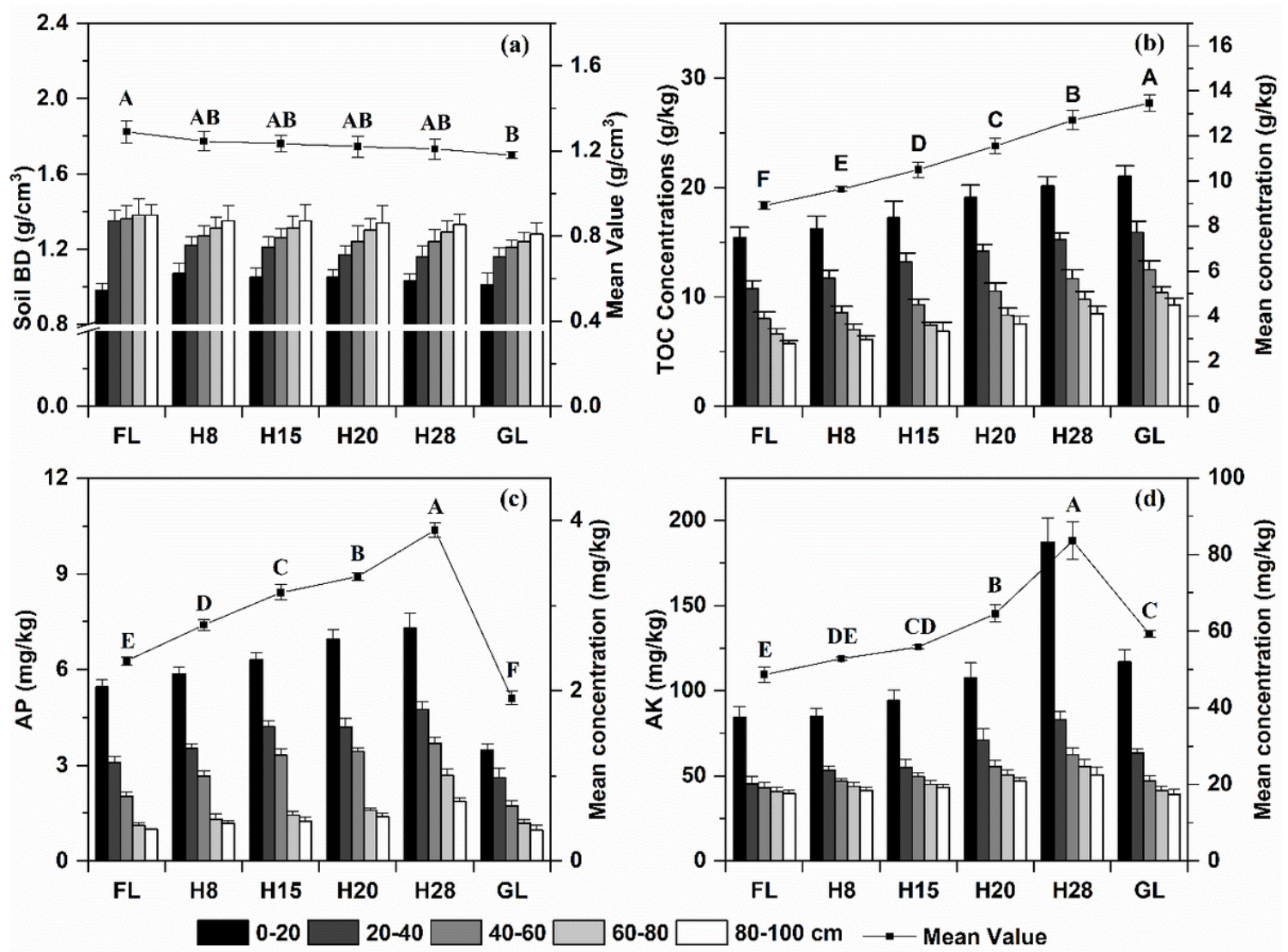


Figure 1

Dynamics of soil bulk density (a), total organic carbon (b), available phosphorus (c) and available potassium (d) contents and mean concentrations at the soil depth of 0–100 cm in the *Zanthoxylum bungeanum* chronosequences (including H8, H15, H20 and H28), farmland (FL) and grassland (GL). Different uppercase letters indicate that the cumulative concentrations values (0–100 cm soil depth) at different sites are significantly different ($P < 0.05$ and $n = 3$).

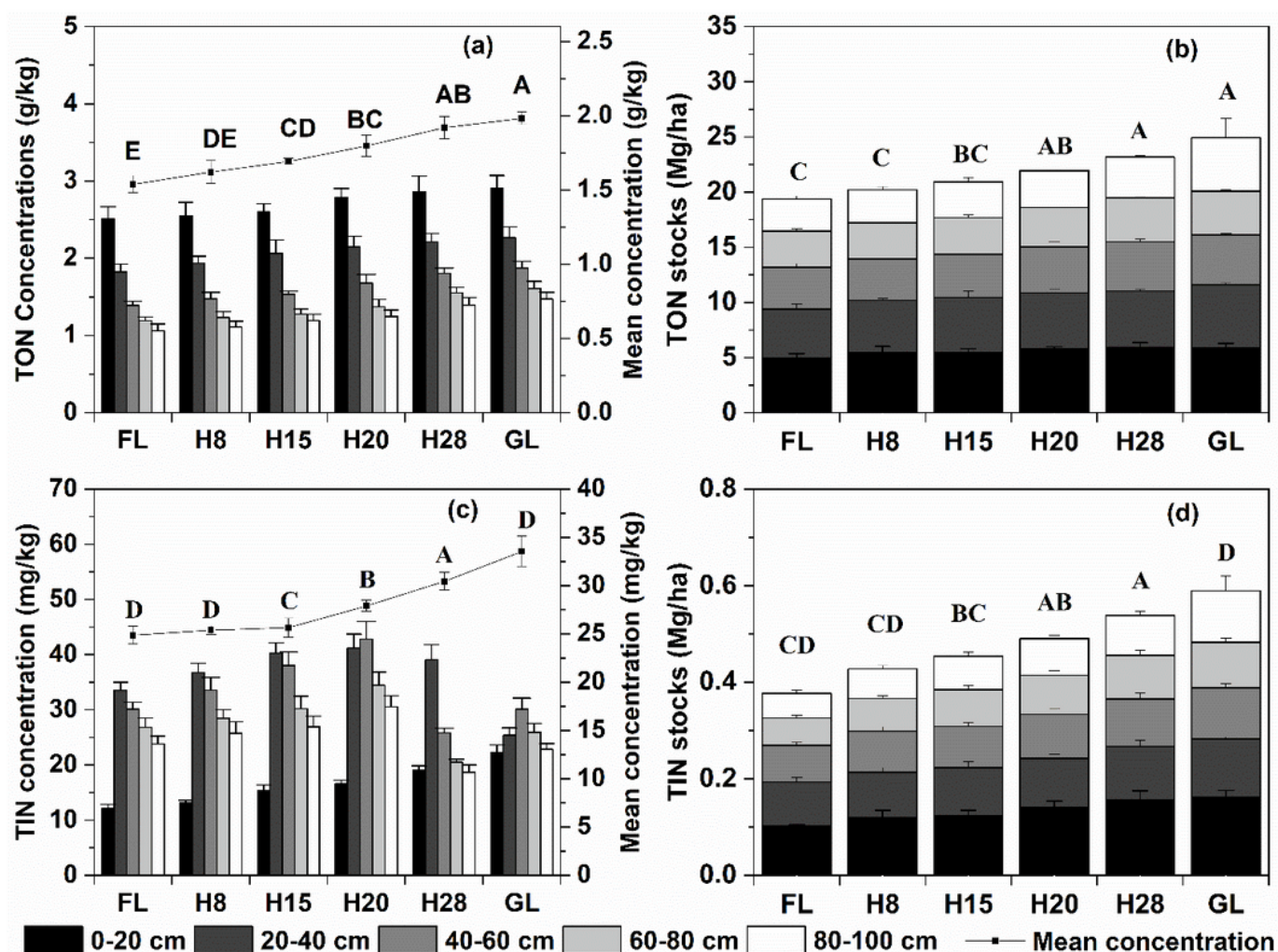


Figure 2

Spatio-temporal distribution dynamics of soil total organic nitrogen content (a), total organic nitrogen stocks (b), total inorganic nitrogen content (c), and total inorganic nitrogen stocks (d) at the soil depth of 0–100 cm in the *Zanthoxylum bungeanum* chronosequences (including H8, H15, H20 and H28), farmland (FL) and grassland (GL). Data are means, error bars represent standard deviations ($P < 0.05$, $n = 3$).

Different uppercase letters indicate that the cumulative contents (0–100 cm soil depth) of total organic and inorganic nitrogen at different sites are significantly different ($P < 0.05$, $n = 3$).

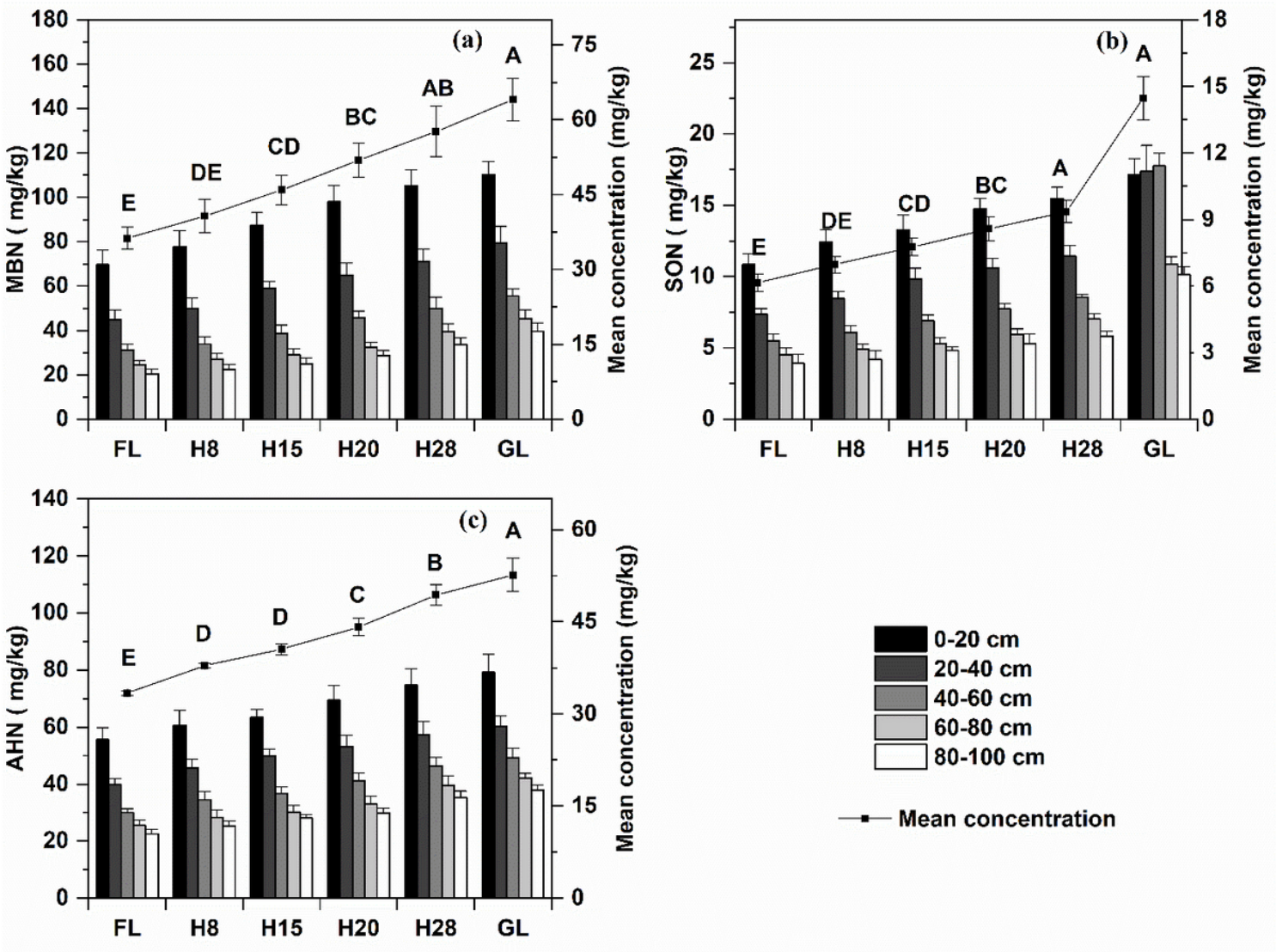


Figure 3

Spatio-temporal distribution dynamics of soil microbial biomass nitrogen (a) soluble organic nitrogen (b), and alkali-hydrolysable nitrogen content (c) at the soil depth of 0–100 cm in the *Zanthoxylum bungeanum* chronosequences (including H8, H15, H20 and H28), farmland (FL) and grassland (GL). Data are means, error bars represent standard deviations ($P < 0.05$, $n = 3$). Different uppercase letters indicate that the cumulative (0–100 cm soil depth) concentration values at different sites are significantly different ($P < 0.05$, $n = 3$).

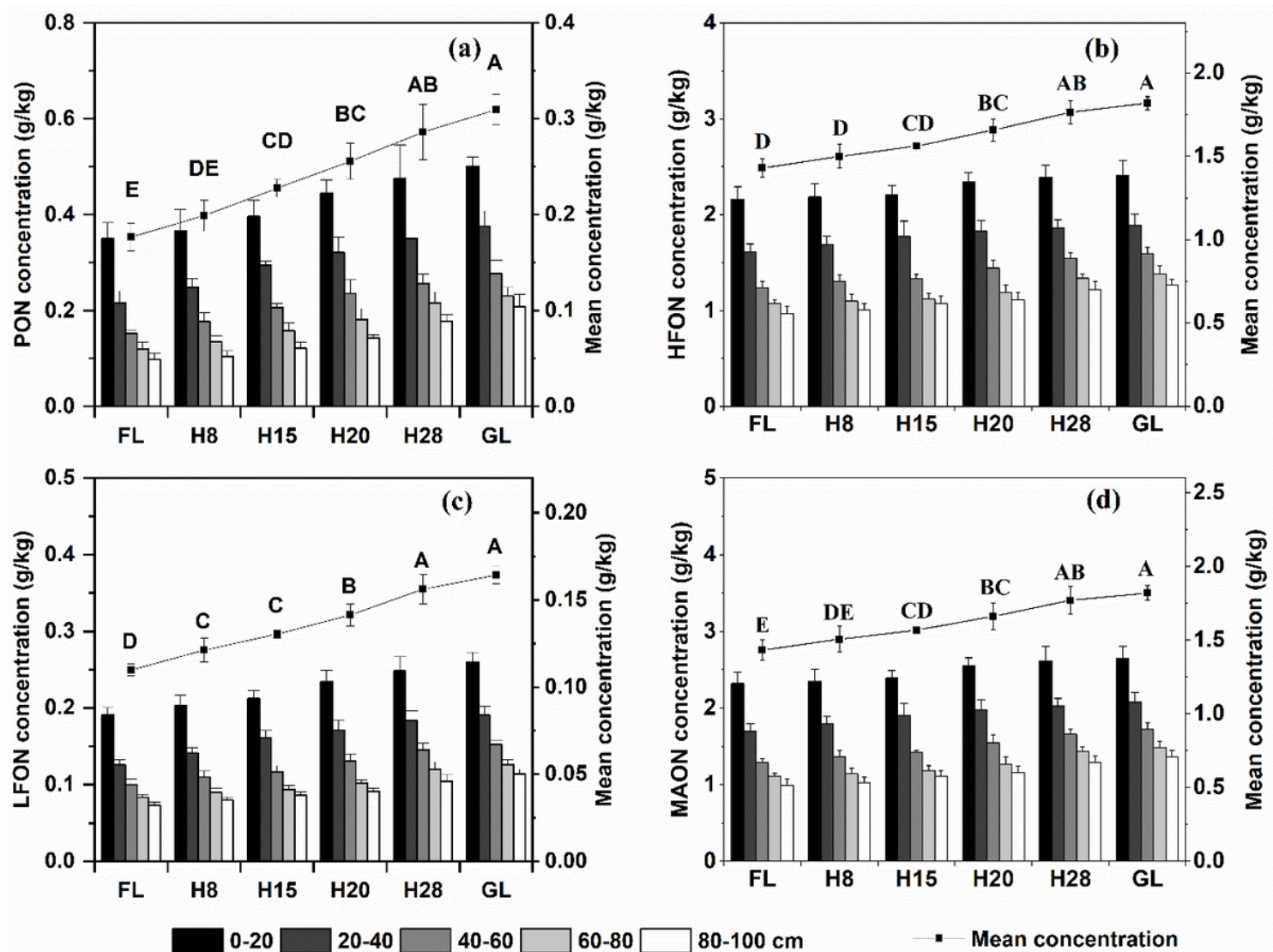


Figure 4

Spatio-temporal distribution dynamics of soil particulate organic nitrogen (a), heavy fraction organic nitrogen (b), light fractions organic nitrogen (c), and mineral-associated organic nitrogen content (d), at the different soil depths from 0–100 cm in the *Zanthoxylum bungeanum* chronosequences (including H8, H15, H20 and H28), farmland (FL) and grassland (GL). Vertical bars for each column represent standard deviations of the mean ($P < 0.05$, $n = 3$). Different uppercase letters indicate that the cumulative concentrations values (0–100 cm soil depth) at different sites are significantly different ($P < 0.05$, $n = 3$).

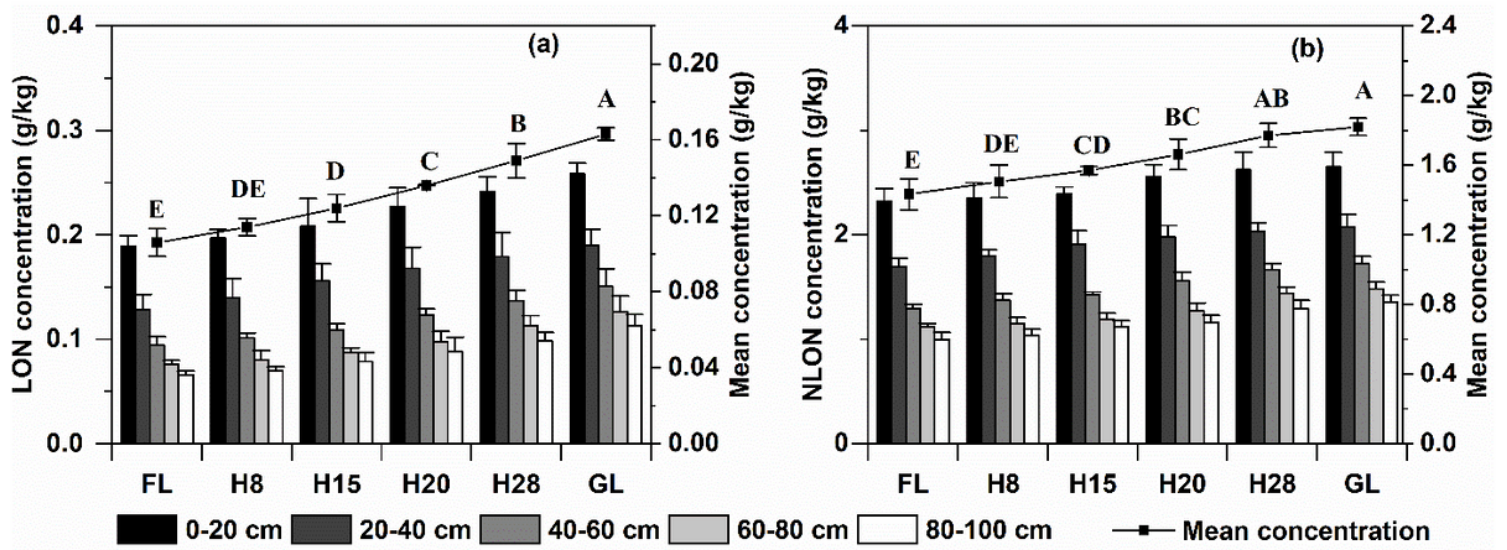


Figure 5

Spatio-temporal distribution dynamics of soil labile organic nitrogen (a), non-labile organic nitrogen (b) at the different soil depths from 0–100 cm in the *Zanthoxylum bungeanum* chronosequences (including H8, H15, H20 and H28), farmland (FL) and grassland (GL). Vertical bars for each column represent standard deviations of the mean ($P < 0.05$, $n = 3$). Different uppercase letters indicate that the cumulative concentrations values (0–100 cm soil depth) at different sites are significantly different ($P < 0.05$, $n = 3$).

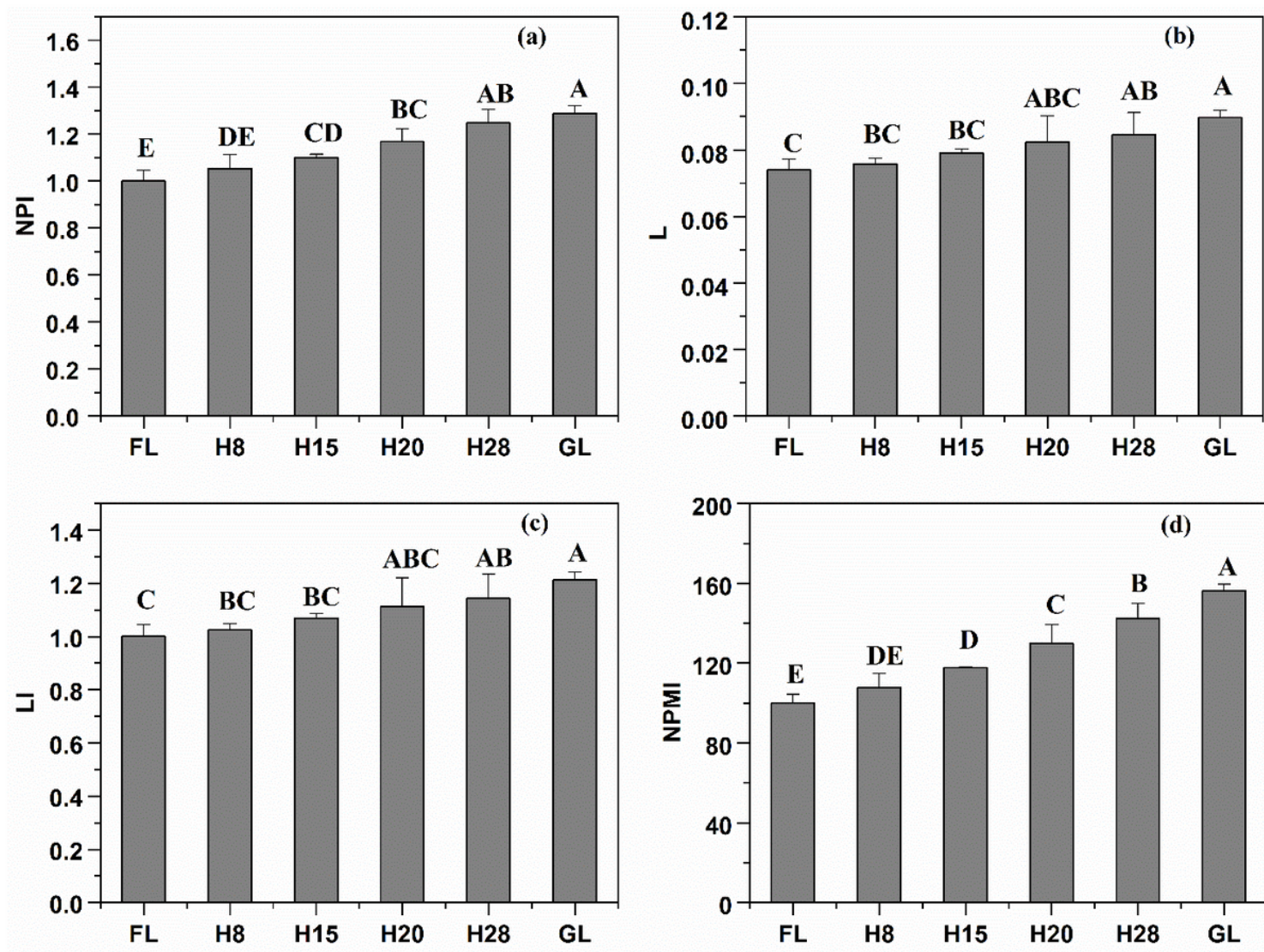


Figure 6

Soil nitrogen pool index (a), nitrogen lability (b) nitrogen lability index (c), and nitrogen pool management index (d), for 0–100 cm in the *Zanthoxylum bungeanum* chronosequences (including H8, H15, H20 and H28), farmland (FL) and grassland (GL). Vertical bars for each column represent standard deviations of the mean ($P < 0.05$, $n = 3$). Different uppercase letters indicate that the cumulative concentrations values (0–100 cm soil depth) at different sites are significantly different ($P < 0.05$, $n = 3$).

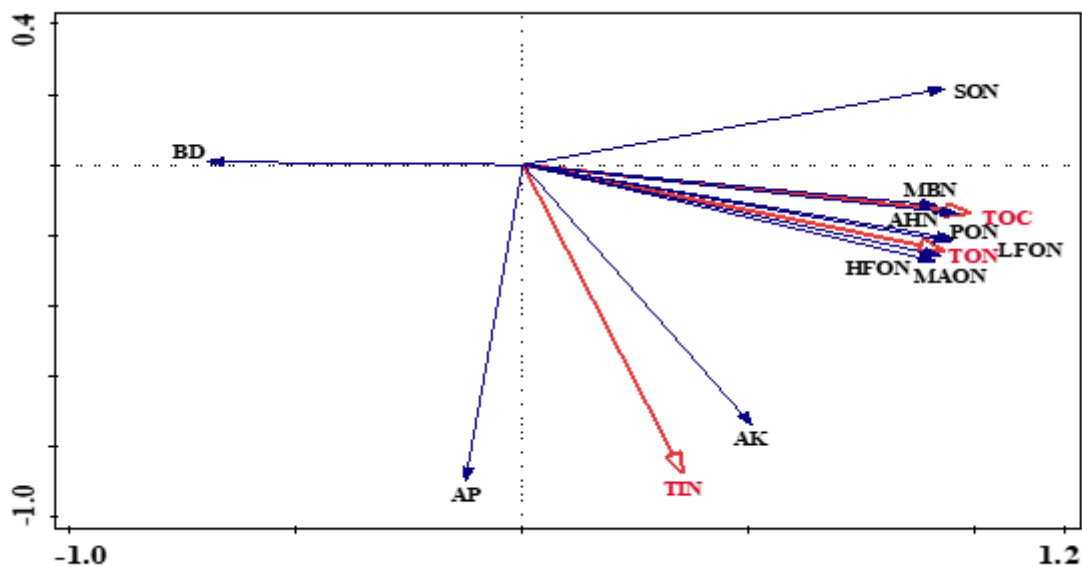


Figure 7

Ordination plots of the results from the redundancy analysis (RDA) to identify the relationships among total organic carbon (TOC), total organic nitrogen (TON), total inorganic nitrogen (TIN) and soil physicochemical properties. SON: soluble organic nitrogen, AHN: alkali-hydrolysable nitrogen, MBN: microbial biomass nitrogen, PON: particulate organic nitrogen, MAON: mineral-associated organic nitrogen, HFON: heavy fraction organic nitrogen, LFON: light fraction organic nitrogen, BD: bulk density, AK: available potassium, and AP: available phosphorus.

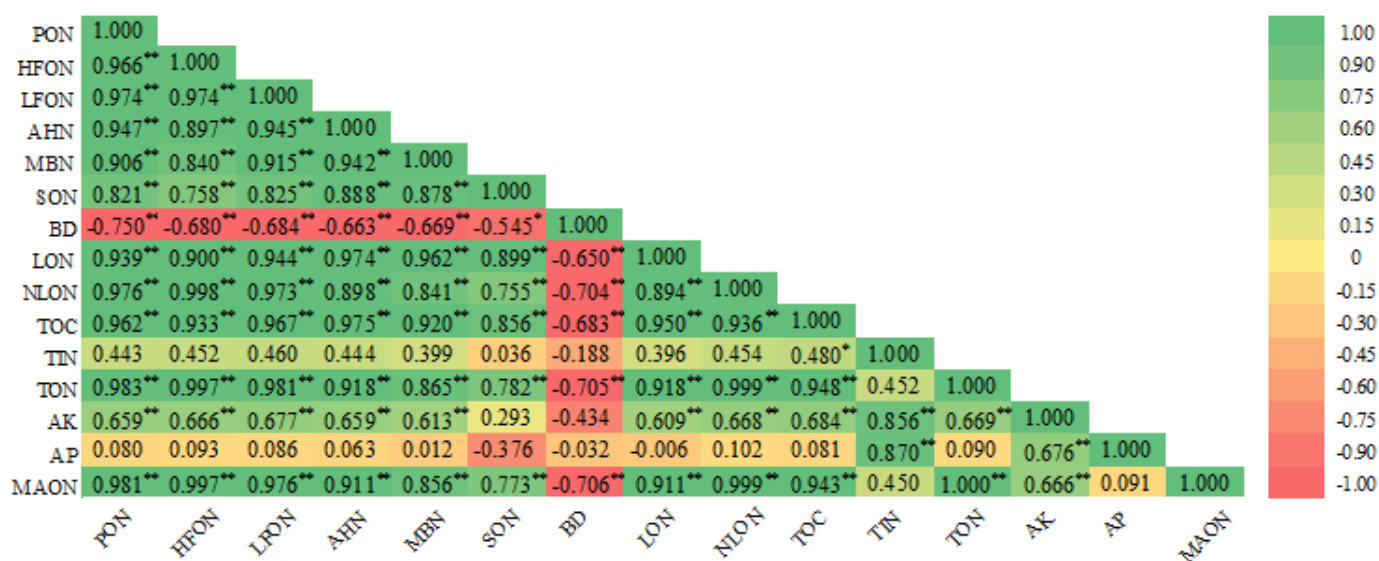


Figure 8

Heatmap of Pearson correlation coefficients (r) among soil particulate organic nitrogen (PON), heavy fraction organic nitrogen (HFON), light fraction organic nitrogen (LFON), alkali-hydrolysable nitrogen

(AHN), microbial biomass nitrogen (MBN), soluble organic nitrogen (SON), bulk density (BD), labile organic nitrogen (LON), non-labile organic nitrogen (NLON), total organic carbon (TOC), total organic nitrogen (TON), total inorganic nitrogen (TIN), available potassium (AK), available phosphorus (AP), and mineral-associated organic nitrogen (MAON) . * $P < 0.05$, and ** $P < 0.01$.