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Conversion of cropland to forage land and grassland increases soil labile carbon and enzyme activities in northeastern China

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ABSTRACT

Soil labile carbon (C) and enzyme activities are valuable indicators of changes in soil quality and health. Understanding the changes in soil labile C and enzyme activities under different land uses is important to maintain soil quality and health and for sustainable land use. The primary objective of this study was to investigate the short-term influences of different land uses on SOC, soil labile C and enzyme activities in semiarid alkaline grassland of northeastern China. The experiment was organized as a block design with four replications of each land use treatment. Land use treatments were corn cropland (Corn), alfalfa forage land (Alfalfa), *Lyemus chinensis* grassland (AG), *Lyemus chinensis* grassland for mowing (AG + Mow) and restored grassland (RG), which were applied for five years. Total soil organic carbon (SOC), three labile C pools (oxidizable labile C; water-extractable organic C; microbial biomass C) and the activities of four soil enzymes (catalase; urease; alkaline phosphatase; invertase) were determined at the 0–20 cm depth in the five land use treatments. Results showed that soil labile C and enzyme activities were sensitive indicators of land use change. Conversion of cropland to forage land and grassland increased SOC (40.42%), soil labile C measures (25.50%) and enzyme activities (55.60%). However, the responses of different forms of soil labile C and enzyme activities to different land uses were not similar. Under Corn, AG + Mow, AG and RG land uses, the geometric means of labile C (27.01%, 10.95%, 17.52% and 5.11%, respectively) and enzyme activities (40.92%, 13.54%, 11.38% and 7.38%, respectively) were lower than those under Alfalfa, demonstrating that soil labile C and enzyme activities improved more under Alfalfa than under other land uses in northeastern China. Significant correlations were also obtained between SOC, soil labile C measures and enzyme activities. To conclude, soil labile C and enzyme activities can be expected to gradually increase with the conversion of cropland to grasslands and forage land, and planting to alfalfa offers a profitable and sustainable solution to our requirement for pairing forage production with rapid restoration of soil quality in the areas in which soils are not suitable for growing crops in the Songnen Grassland.

1. Introduction

The Songnen Grassland is located in the eastern part of the northern agro-pastoral zone in China. The coexistence of agriculture and animal husbandry in this area leads to the diversification of land uses. The areas with high quality soils are used as cropland to produce food, whereas the areas in which the soils are not suitable for growing crops remain as grasslands. In recent decades, to meet the increasing demands for food from a rapidly growing population, grassland has been converted to cropland in the areas with soils with less salt and alkali content. However, the removal of natural vegetation, with the conversion from grassland to cropland, accelerates substantially land salinization and alkalization. A survey showed that 26.4% of the grassland has been converted to cropland during the past 30 years because of the increasing population (Liu et al., 2009). Land degradation leads to the decline of grain production, and some of the degraded croplands have been abandoned. Additionally, with the yearly increase in grain production in China in recent years, particularly for corn production, yield oversupply resulted, causing in a substantial increase of inventory and a remarkable decline in the benefit of planting corn (Chen et al., 2016). The corn price in northeastern China decreased more than 50% from approximately 2.20 CNY kg^{-1} (0.32 USD kg^{-1} or

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0.30 EUR kg^{-1}) in 2015 to approximately 1.06 CNY kg^{-1} (0.15 USD kg^{-1} or 0.14 EUR kg^{-1}) in early 2017. Therefore, adjusting the agricultural planting structure is very important to improve the efficiency and sustainable development of agriculture in northeastern China. Considering the poor soil conditions and semiarid climate, the conversion of cropland to forage or planted grassland to meet the requirements for high quality forage grass in this region can help not only to restore soil quality and health, but also can promote the income of farmers and the development of animal husbandry (Zhou et al., 2012). *Leymus chinensis* is the dominant native specie in the Songnen Grassland and is usually harvested as hay in mowed grasslands or used as forage grass in grasslands for grazing animals (Yu et al., 2014). Alfalfa is high quality forage due to the high N and protein content and can also maintain or even improve soil fertility through biological N fixation (Li et al., 2016a). Alfalfa has been introduced into the agricultural ecosystem in the Songnen Grassland because of the additional relatively high saline and alkaline tolerance. However, the effect of changes in land uses on soil quality has yet to be elucidated in the Songnen Grassland.

Change in land use is the primary factor that influences the quantity and quality of soil organic carbon (SOC). However, changes in total SOC are difficult to detect because of the heterogeneous nature of the mixture of forms with different levels of stability (Zhao et al., 2016). Soil labile C pools, such as microbial biomass C, water extractable organic C and oxidizable labile C, can respond more rapidly to land use changes than total SOC and therefore have been suggested for use as early indicators of changes in SOC and soil quality (Guidi et al., 2014; Gabarron-Galeote et al., 2015a). Soil enzyme activities are very sensitive and provide immediate and precise information on small changes in soil properties, including SOC, nutrients, pH and salinity (Torres et al., 2015), and they can forecast changes in these soil properties earlier than other soil analyses (Liu et al., 2017). Thus, soil enzyme activities, such as catalase, invertase, urease and phosphatase, are usually measured as sensitive and important indicators to reflect the effect of land use change on soil quality and health (Raiesi and Beheshti, 2015; Wang et al., 2016). Although increasing interest has stimulated great efforts to monitor the changes in soil labile C and enzyme activities under different land uses, large and different responses of soil labile C pool and enzyme activities to land use change are observed due to the diverse regional climates, soil types, initial soil conditions and land management practices (Poeplau and Don, 2013; Raiesi and Beheshti, 2014; Machmuller et al., 2015; Veres et al., 2015). Furthermore, the short-term effects of land use changes on soil quality and health have received relatively less attention, particularly in the easily affected and alkaline soils with a high content of exchangeable Na or free soda (Guo et al., 2015).

Our previous study showed that legume introduction in the Songnen Grassland can significantly increased SOC and N storage through changing plant community structure and SOC storage was greater under treatment with higher legume content (Li et al., 2016a). Moreover, we also found that land uses had a significant effect on the total SOC and SOC fractions with increasing degrees of oxidizability (very labile C, labile C and stable C), and grassland colonization of croplands increased total SOC content and the percentages of labile C and stable C to total SOC in the Songnen Grassland (Yu et al., 2017). However, the changes in the forms and quantities of soil labile carbon pools under different land uses in this grassland remain unknown. The responses of soil enzyme activities to different land uses are also unknown. Therefore, the changes in soil labile C and enzyme activities under different land uses in the Songnen Grassland require further investigation. In this research, we hypothesized that the short-term of planting of grass including legume forage grass and reduction in the disturbances of croplands could lead to increased soil C sequestration and enzyme activities and therefore improve soil quality in northeastern China. To address this hypothesis, the objectives of this study were to (1) compare the changes in soil labile C pools and enzyme activities of alkaline soils between grasslands and cropland; and (2) examine whether land use for legume forage was better than other grasslands for the restoration of soil C and enzymes in alkaline soils of the Songnen Grassland.

2. Materials and methods

2.1. Study site

The study was conducted at the Changling Ecological Research Station for Grassland Farming (44°33′ N, 123°31′ E, 145 m a.s.l). The station is located in the south of the Songnen Grassland, a typical semiarid agro-pastoral transitional zone in northeastern China. The area is characterized by a temperate, semiarid continental monsoon climate. The annual average air temperature is 5.9 °C, and annual average precipitation is 427 mm from 1980 to 2013, and approximately 70%–80% of total precipitation occurs between June and September. The pan evaporation approximates 1600 mm. The frost free period is approximately 140 days. The soil type is alkali-saline with a soil texture of 23% sand, 35% silt and 42% clay, which is classified as Solonetz in the World Reference Base for Soil Resources (IUSS Working Group, 2015). The soils contain high content of free sodium bicarbonate ($NaHCO_3$) and sodium carbonate (Na_2CO_3), and the pH of the soils is between 8.0 and 11.0. Mites and collembola are the two dominant soil meso- and micro-fauna communities. *Actinedida, Gamasida,* and *Oribatida* are the dominant mites suborder, which accounted for 50.22% of total soil animal, while *Istotomidae, Paronellidae* and *Pseudachorutidae* are the dominant collembola family, which account for 25.25% of total soil animal (Yin et al., 2003; Yan et al., 2015). The dominant native species include *Leymus chinensis, Chloris virgata, Puccinellia spp. and Polygonum gracilius*. The vegetation coverage measures 50%-90%, with 100–360 g m^{-2} of aboveground biomass in the peak season (Yu et al., 2014).

Farmers in the study area traditionally grow rain-fed maize and soybean [*Glycine max* (L.) Merr.]. These crops are usually sown in early May and harvested in late September to early October. The common practice is to plow the soil at least twice before the crop growing season down to 20 cm using a rotary tiller mounted with the tractor. Fertilizers (50–96 kg N ha^{-1}, 20–45 kg P ha^{-1}, and 30–45 kg K ha^{-1}) are applied twice a year at sowing and mid July.

2.2. Experimental design and soil sampling

The experiment was established in early May 2011 at a cropland and run for five years until soil sampling. Because of the continuous plowing, soil conditions in the cropland before this experiment were homogeneous. The initial SOC content is 7.82 ± 0.29 g C kg^{-1} at the 0–10 cm depth and 4.86 ± 0.27 g C kg^{-1} at the 10–20 cm depth. The initial soil bulk density is 1.47 ± 0.07 g cm^{-3} at the 0–10 cm depth and 1.51 ± 0.04 g cm^{-3} at the 10–20 cm depth. Five land use treatments were designed in a block design with four replications. The five treatments were corn cropland (Corn), alfalfa forage land (Alfalfa), artificial grassland of *Lyemus chinensis* (AG), artificial grassland of *Lyemus chinensis* and mowing for hay every year (AG + Mow) and restored grassland (RG). The block size was approximately 60 m × 50 m whereas the plot size was 12 m × 50 m for Corn and Alfalfa treatments and 6 m × 50 m for the AG, AG + Mow and RG treatments. There was a 2 m buffer between the blocks and a 1 m buffer between the plots. The cropland was under continuous corn monoculture since 2011. The Corn plots followed the tradition cropland practice in the Songnen Grassland, which consist of plowing the soil at least twice before the crop growing season down to 20 cm and fertilizers (74 kg N ha^{-1}, 22 kg P ha^{-1}, and 41 kg K ha^{-1}) were applied twice a year at sowing and mid July. The corn straw was removed from the plots after harvest, while the corn residues (including root, stem base and aerial root, which is 137 g m^{-2}) were incorporated into soil during plowing. The Alfalfa plots were set up in May 2014. Before

2014, these plots were croplands without tillage (no tillage) and other practices were the same as for the described conventional cropland. However, the growth of corn was very bad in no-tillage cropland in 2011–2013 due to the poor soil condition and short-term land use. Considering the poor natural conditions and the development of local animal husbandry, we changed the no-tillage cropland to alfalfa forage land in May 2014 with the sowing density of approximately 1200 seeds m^{-2}. The above-ground (307 g m^{-2}) and below-ground (321 g m^{-2}, 0–20 cm depth) biomass were kept in Alfalfa plots to restore soil fertility in 2014 and 2015. Seeds of *Lyemus chinensis* were sowed in May 2011 with a density of approximately 2000 seeds m^{-2} in the two treatment plots of artificial grasslands. Reseeding had a positive effect to recover the vegetation and the aboveground biomass reached approximately 100–120 g m^{-2} in the early September 2011. The aboveground biomass was removed from the AG + Mow plots once a year at the peak season and the vegetation in AG plots was kept as litter to return into the soils. The above-ground biomass and below-ground biomass at 0–20 cm depth was 380.7 and 455.9 g m^{-2} in AG plots, respectively, and was 488 and 638 g m^{-2} in AG + Mow plots, respectively. The cropland was abandoned in 2011 in the RG plots to restore grassland without any disturbance. The dominant species in the RG plots include *Chloris virgata, Sonchus brachyotus, Chenopodium glaucum,* and *Phragmites communis*, which account for more than 85.00% of the aboveground biomass. After several years' restoration, the above- and below-ground biomass in RG plots reached about 348 and 397 g m^{-2}, respectively. No fertilizer was used in Alfalfa, AG + Mow, AG and RG plots.

Soil samples were collected in early September 2015 to a depth of 20 cm at two intervals of 0–10 and 10–20 cm with a 4 cm diameter soil core sampler after removing the aboveground biomass and litter. A sample for each depth was composted by mixing sub-samples from five randomly selected locations (0.5 m × 0.5 m) at least 6 m apart from one another and at a 1 m distance from the plot boundary within each plot. Soil samples were gently mixed, and the root materials and other visible debris were removed. One sub-sample was stored field-moist in a cooler at 4 °C for water-extractable organic carbon (WEOC) and microbial biomass carbon (SMBC) analyses within 10 days. Another sub-sample was air-dried, passed through a 2-mm sieve, and then ground to pass through a 0.25-mm sieve for oxidizable labile carbon (OLC), SOC and soil enzyme activity analyses.

2.3. Soil analyses

Total SOC content under different land uses was determined by wet oxidation with K_2CrO_7 in the presence of sulfuric acid at 170–180 °C (Yeomans and Bremner, 1988). The OLC was estimated using a modified method defined by Chan et al. (2001). For the determination, 10 mL of concentrated sulfuric acid (36 N H_2SO_4) was added to an Erlenmeyer flask, which contained 0.50 g of soil sample and 10 mL of 0.167 M $K_2Cr_2O_7$, resulting in a sulfuric acid-aqueous solution ratio of 1:1. After the reaction, the excess dichromate was determined by titrating against 0.50 M $FeSO_4$. The amount of dichromate consumed by the soil sample was used to calculate the content of OLC. The WEOC was determined using a modified method described by Li et al. (2016b), which was an extraction performed by shaking 10.00 g of field-moist soil with 50 mL of 0.50 M K_2SO_4 solution for 30 min at a speed of 250 rpm. The extracted solutions were subsequently centrifuged for 10 min at 5000 rpm, and organic carbon in the supernatant was measured using an automated TOC-VCPH (Total Organic Carbon Analyzer, SHIMADZU Company). The SMBC was determined using the chloroform fumigation-extraction method (Jenkinson and Powlson, 1976). Paired 10.00 g field-moist soil samples that were either un-fumigated or fumigated with alcohol-free $CHCl_3$ for 24 h were extracted with 50 mL of 0.50 M K_2SO_4 solutions. Organic carbon in the extracted supernatant was analyzed using an automated TOC-VCPH (Total Organic Carbon Analyzer, SHIMADZU Company). The efficiency factor of 0.38 (Kec) was used to calculate the content of SMBC.

The activities of three hydrolytic enzymes and one oxidative enzyme were measured using modified determination methods as previously described (Guan, 1986; Wang et al., 2008). Soil catalase (EC 1.11.1.6, CAT) activity was determined by back-titrating residual H_2O_2 with a standard solution of $KMnO_4$. Briefly, three grams of soil was added into a 100 mL triangular flask with 40 mL of DI water and 5 mL of 0.30% H_2O_2. After shaking for 30 min, 5 mL of 1.50 M H_2SO_4 was added into the flask to stabilize the residual H_2O_2. After filtration, 25 mL of filtrate was titrated with standard solution of 0.05 M $KMnO_4$ until the mixture changed to pink. The CAT activity was expressed as ml $KMnO_4$ per g dry soil. Soil urease (EC 3.5.1.5, URE) activity was measured using 10.00% urea solution as the substrate. Two grams of soil and 0.4 mL of toluene were added into a 50 mL flask. After 15 min, 10 mL of 10.00% urea solution and 10 mL of citrate buffer (pH 6.7) were mixed with the soil sample. The mixture was incubated at 37 °C for 24 h. After filtration, 5 mL of filtrate was transferred to a 50 mL flask and then mixed with 10 mL of DI water, 4 mL of sodium phenolate and 3 mL of sodium hypochlorite. After the mixture was diluted to 50 mL, the concentration of NH_3-N released was assayed using a spectrophotometer at 578 nm. The URE activity was expressed in mg NH_3-N per g of dry soil per 24 h. Soil alkaline phosphatase (EC 3.1.3.1, ALP) activity was measured by disodium phenyl phosphate colorimetry. Briefly, two grams of soil was added into a 50 mL flask and incubated for 12 h at 37 °C after the addition of 0.4 mL of toluene and 10 mL of disodium phenyl phosphate and borate buffer (pH 9.0). After filtration, 5 mL of filtrate was transferred to a 50 mL flask, and 5.0 mL of the buffer, 3 mL of 2.50% potassium ferricyanide and 3 mL of 0.50% 4-aminoantipyrine were added to facilitate the reaction. The mixture was shaken and diluted to 50 mL. After 30 min, the concentration of phenol released over 12 h was assayed colorimetrically at 510 nm. The ALP activity was expressed in mg phenol per g of dry soil per 12 h. Soil invertase (EC 3.2.1.26, INV) activity was determined by 3, 5-dinitro salicylic acid colorimetry and using 8.00% sucrose as the substrate. Briefly, three grams of soil was incubated for 24 h at 37 °C with 15 mL of 8.00% sucrose, 5 mL of phosphate buffer (pH 5.5) and six drops of toluene. After filtration, 2 mL of the filtrate and 3 mL of 3, 5-dinitrosalicylic acid were added to a 50 mL flask and heated at 100 °C for 5 min. The heated solution was diluted to 50 mL. The concentration of glucose released was assayed using a spectrophotometer at 508 nm. The INV activity was expressed in mg glucose per g of dry soil per 24 h. For the incubation of each enzyme, two controls (one without substrate and the other without soil sample) were also included.

2.4. Statistical analyses

A single measure of soil labile carbon and one enzyme are not sufficient to reveal the changes within the soil environment. The geometric mean is a general index to integrate information from variables that possess different units and ranges of variation, which can reflect the responses of these variables to factors of change (Wang et al., 2012b; Raiesi and Beheshti, 2014). Here, the geometric means of labile C (GMC) and enzyme activities (GME) were calculated for each land use and soil depth as follow:

$$GMC = (OLC \times WEOC \times SMBC)^{1/3} \quad (1)$$

where OLC, WEOC and SMBC are oxidizable labile C, water-extractable organic C and microbial biomass C, respectively.

$$GME = (CAT \times URE \times INV \times ALP)^{1/4} \quad (2)$$

where CAT, URE, INV and ALP are the activities of catalase, urease, invertase, and alkaline phosphatase, respectively.

All statistical analyses were conducted with the SPSS 16.0 statistical software package for Windows (SPSS, Inc., Chicago, IL, USA). One-way analysis of variance (ANOVA) was used to test the effects of land-use

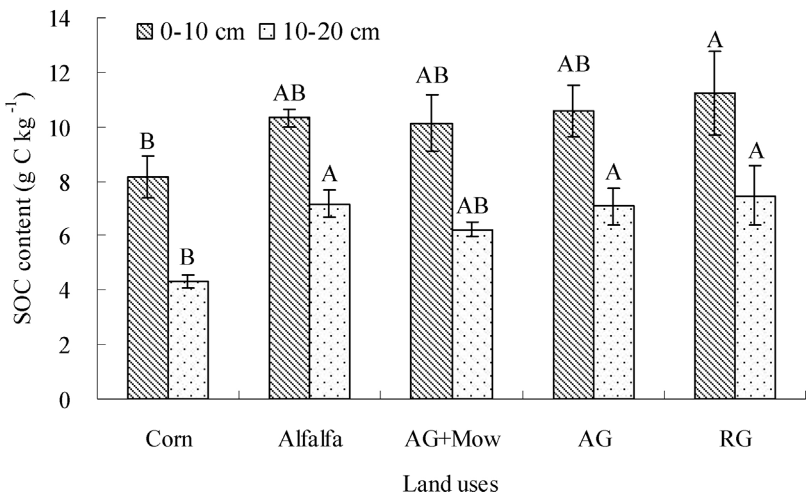

Fig. 1. Contents of SOC under different land uses. Values with the same uppercase letters within a land use are not significantly different at $P < 0.05$. The bars represent standard errors. Corn, corn cropland; Alfalfa, alfalfa grassland; AG + Mow, *Lyemus chinensis* grassland for mowing; AG, *Lyemus chinensis* grassland; RG, restored grassland.

treatment on soil labile C pools and soil enzyme activities, and Fisher's least significant difference test (LSD) was used to test the differences of soil labile C pools and soil enzyme activities among land use treatments. The analyses were performed at the $P < 0.05$ significance level. Correlation matrices between soil labile C pools and soil enzyme activities were reported based on Pearson correlation coefficients with levels of significance at $P < 0.05$, $P < 0.01$and $P < 0.001$.

3. Results and discussion

3.1. Changes in SOC and soil labile C measures under different land uses

Our results confirmed that conversion of cropland to grasslands or forage land increased SOC content. Compared with Corn, SOC content in Alfalfa, AG + Mow, AG and RG increased by 2.17, 1.96, 2.41 and 3.07 g C kg^{-1}, respectively, at the 0–10 cm depth and by 2.84, 1.89, 2.74 and 3.14 g C kg^{-1}, respectively, at the 10–20 cm depth (Fig. 1). However, the SOC content increased significantly only at 0–10 cm and 10–20 cm depths in RG and at the 10–20 cm depth in Alfalfa and AG. The increase in SOC content in the grasslands and forage land was also demonstrated in the comparison of the current SOC content after 5 years of conversion with the initial SOC content before conversion. Compared with the initial SOC content of 6.34 g C kg^{-1} at the 0–20 cm depth, SOC content in Alfalfa, AG + Mow, AG and RG increased significantly by 2.40, 1.82, 2.47 and 3.00 g C kg^{-1}, respectively. The increases of SOC content might be explained by the recovery of above- and below-ground biomass and the decrease of tillage disturbance, which directly increase the inputs of organic C into the soil and reduce the loss of SOC caused by soil erosion. This result is generally consistent with that of Machmuller et al. (2015) in the southeastern United States, who found similar increases in SOC content after six years when cropland was converted to grassland. Among the land use treatments of Alfalfa and grasslands, no significant differences in SOC contents were found. This result could be partially explained by the management practices of these land use treatments. Because the conversion from cropland to alfalfa planting has been only two years, the effect of this land use on C sequestration was very limited. Although the above- and below-ground biomass in AG + Mow was higher than those in other land uses, the removal of the above-ground biomass reduced the C input from plants to the soil and resulted in no significant difference in C inputs between AG + Mow and other land uses.

Soil labile C has higher turnover rates than SOC, therefore the dynamics of soil labile C content can be more easily detected under short- and medium-term land management practices than total SOC (Li et al., 2016b). The changes in OLC contents among various land uses can be used to evaluate their responses to land management practices (Purakayastha et al., 2008). Soil tillage results in lower contents of oxidizable labile C than soils under NT cropland and grassland due to the breakup of the protection of soil aggregates and accelerated OLC decomposition by soil heterotrophic activity (Chan et al., 2001; Barreto et al., 2011). In the present study, the contents of OLC in Alfalfa, AG + Mow, AG and RG increased significantly by 35.62%, 24.30%, 25.97% and 52.69% at the 0–10 cm depth, respectively, and by 47.75%, 33.56%, 43.60% and 69.55% at the 10–20 cm depth, respectively, compared with Corn. The increases in OLC content under these land uses indicated that revegetation of the grasslands and forage land contributed to improving the SOC contents in soils. These findings are generally consistent with the results of Benbi et al. (2015) in semiarid subtropical regions and they found a similar increase of OLC content in undisturbed soils (undisturbed for more than 40 years) compared with cultivated soils due to the higher inputs of above- and below-ground biomass in undisturbed soils. The highest OLC contents at 0–10 cm and 10–20 cm depths were all found in RG, indicating that this land use treatment was more effective than other land uses in restoring OLC. The higher species diversity in RG and the input of more easily oxidized organic C components into the soil than other land uses might explain the increase in OLC content. The percentage of OLC to total SOC ranged from 61.3% to 69.2% for the 0–20 cm depth in all land uses (Table 1); thus, the SOC content in study area was characterized by a predominant OLC fraction. The percentages of OLC to SOC observed in this study are similar to those observed by Chan et al. (2001) in semiarid western New South Wales in which all values were in the range from 63.0% to 67.0% under different land uses. By contrast, our results for the proportions of OLC were higher than those reported by Bhattacharyya et al. (2015) in the western Indo-Gangetic plains and by Benbi et al. (2015) in northern India, with values that only ranged from 38.8% to 55.6%. These differences in the percentages of OLC to SOC can be primarily attributed to different land use practices, environmental conditions, climatic and initial soil properties in these semiarid regions (Bhattacharyya et al., 2015). Unlike the increase of OLC content in the grasslands and forage land, the percentages of OLC to SOC among the treatments at 0–10 cm and 10–20 cm depths were not significantly different, except under AG at the 0–10 cm depth, which suggested that conversion of cropland to forage land and grasslands increased both the quantity of the OLC and the stable organic C (unoxidizable organic C) in the Songnen Grassland. In previous studies that reported gains in stable C related to grasslands in temperate climate regions, the gains in SOC were assumed to be primarily influenced by root material rather than plant litter in the short-term following land use change (Poeplau and Don, 2013; Gabarron-Galeote et al., 2015a). Moreover, SOC is easily

Table 1
Variations of soil labile C content and its percentage to SOC under different land use. Results are shown as mean (± SE). Values with the same uppercase letters within a land use are not significantly different at $P < 0.05$. Corn, corn cropland; Alfalfa, alfalfa grassland; AG + Mow, *Lyemus chinensis* grassland for mowing; AG, *Lyemus chinensis* grassland; RG, restored grassland.

Land uses	Content of labile C (g kg^{-1} soil)		Percentages of labile C to SOC (%)	
	0–10	10–20	0–10	10–20
	Oxidizable labile carbon (OLC)			
Corn	5.39(± 0.55)B	2.89(± 0.07)B	66.34(± 3.35)AB	67.38(± 2.77)A
Alfalfa	7.31(± 0.19)AB	4.27(± 0.19)A	70.81(± 0.51)AB	60.29(± 3.49)A
AG + Mow	6.70(± 0.72)AB	3.86(± 0.17)AB	66.30(± 2.58)AB	62.09(± 0.70)A
AG	6.79(± 0.87)AB	4.15(± 0.47)A	63.64(± 2.44)B	59.00(± 4.03)A
RG	8.23(± 1.13)A	4.90(± 0.69)A	72.53(± 3.26)A	65.94(± 3.46)A
	Water-extractable organic carbon (WEOC)			
Corn	137.13(± 5.13)A	131.34(± 7.89)B	1.63(± 0.14)A	2.92(± 0.23)A
Alfalfa	137.15(± 5.90)A	153.82(± 3.00)B	1.29(± 0.04)A	2.05(± 0.14)A
AG + Mow	148.37(± 1.21)A	190.73(± 13.69)A	1.60(± 0.19)A	3.06(± 0.36)A
AG	144.67(± 6.54)A	174.88(± 6.39)AB	1.43(± 0.15)A	2.68(± 0.39)A
RG	143.26(± 7.07)A	178.70(± 16.10)AB	1.37(± 0.22)A	2.69(± 0.65)A
	Soil microbial biomass carbon (SMBC)			
Corn	295.83(± 10.13)B	162.09(± 33.15)AB	3.55(± 0.41)B	3.61(± 0.76)A
Alfalfa	533.29(± 16.50)A	256.14(± 19.26)A	5.01(± 0.08)A	3.38(± 0.16)A
AG + Mow	427.59(± 58.54)AB	148.39(± 31.32)B	4.46(± 0.14)AB	2.32(± 0.37)A
AG	391.07(± 73.23)B	117.25(± 13.57)B	3.72(± 0.23)B	1.74(± 0.15)A
RG	395.19(± 30.52)AB	213.30(± 51.20)AB	3.79(± 0.68)B	3.19(± 1.17)A

incorporated and stabilized into mineral soils and is sequestered with the increase in soil aggregates (Guidi et al., 2014).

Compared with the OLC, WEOC was only a small proportion of SOC, ranging from 1.29% to 1.63% for the 0–10 cm depth and from 2.05% to 3.06% for the 10– 20 cm depth in all land uses (Table 1). Different land uses did not influence the content of WEOC, except at the 10–20 cm depth in AG + Mow, or the percentages of WEOC to SOC at the two soil depths. WEOC is the most mobile fraction and can be leached to the subsoil in the soil solution (Walmsley et al., 2011). Additionally, slow decomposition by microorganisms and high sorption by soil minerals may lead to the enrichment of WEOC in deeper soil layers (Chantigny, 2003; Wang et al., 2008). Support for this scenario is provided by the higher contents of WEOC at the 10– 20 cm depth in all land uses in this study (Table 1). The accumulation of WEOC from topsoil to subsoil likely led to the small differences in contents of WEOC in the topsoil among the different land uses. Marinari et al. (2010) also observed that WEOC was not significantly affected under different land uses, and they concluded that WEOC as an indicator was unable to discern the effect of land use changes on soil quality in temperate regions.

Microbial biomass C, as one form of soil labile C, has been proposed as an important indicator of changes in soil quality under different land uses (Sheng et al., 2015). SMBC content at the 0–10 cm depth in Alfalfa, AG + Mow, AG and RG increased by 80.27%, 44.54%, 32.19% and 33.59%, respectively, compared with Corn (Table 1). The higher SMBC contents observed in Alflafa and grasslands could be due to the continuous inputs of leaf litter and root materials, which provide sufficient energies and substrates for soil microorganisms to stimulate the growth and activities of microbial populations (Sheng et al., 2015). Additionally, the inputs of above- and below-ground biomass into the surface soil in all land uses might have resulted in the much higher SMBC contents in the 0–10 cm depth than those in the 10–20 cm depth (Table 1). In the 10–20 cm depth, the SMBC content in Corn was not significantly different from those in other land uses, which suggested that habitat improvement is also important for the growth of microorganisms. Furthermore, the percentages of SMBC to SOC at the 0–10 cm depth were not significantly different among the land use treatments, whereas the value at the 10–20 cm depth in Corn was higher than those at that depth in other land uses (Table 1). Soil tillage in Corn increases soil porosity and creates better environmental conditions for the growth and activity of the microbial biomass. Additionally, the increase in soil porosity due to soil continuous tillage can prevent the upward movement of CO_3^{2-} and HCO_3^-, thereby reducing the soil sodicity and making the habitat more suitable for microorganisms (Yu et al., 2014). Among the land use treatments, SMBC contents at both soil depths in Alfalfa were the highest as expected, indicating that planting alfalfa in the study area provided a better option to accumulate soil SMBC, which was likely because biological N fixation by alfalfa provided more nitrogen and substrate for microbial growth.

The geometric mean of labile C (GMC) synthesized the responses of OLC, WEOC and SMBC to land uses in this study and can be used as a sensitive indicator to monitor the changes in soil C quantity or quality. In general, land use treatments exerted a clear influence on the GMC as presented in Fig. 2. Among the different land uses, the highest values of GMC at 0–10 cm and 10–20 cm depths were found in Alfalfa, which showed that soil labile C was more sensitive to alfalfa planting than other land uses. Compared with Corn, the GMC values in Alfalfa, AG + Mow, AG and RG increased by 34.42%, 21.31%, 16.39% and 22.95% at the 0–10 cm depth, respectively, and by 41.03%, 23.08%, 7.69% and 41.03% at the 10–20 cm depth, respectively. As the overall

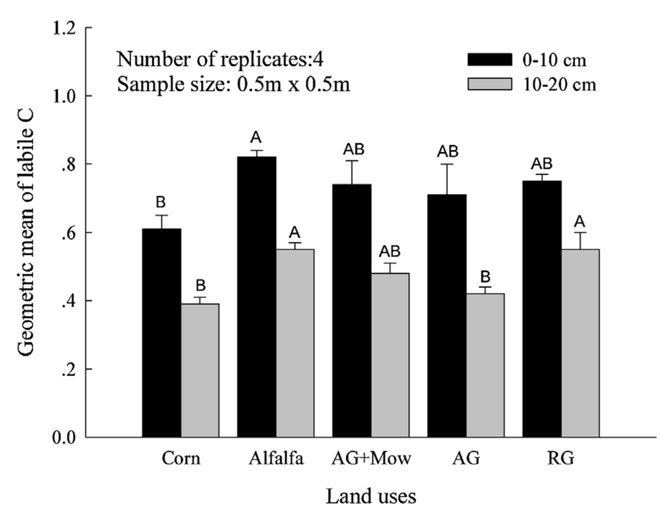

Fig. 2. The geometric mean of labile C under different land uses. Values with the same uppercase letters within a land use are not significantly different at $P < 0.05$. The bars represent standard errors.

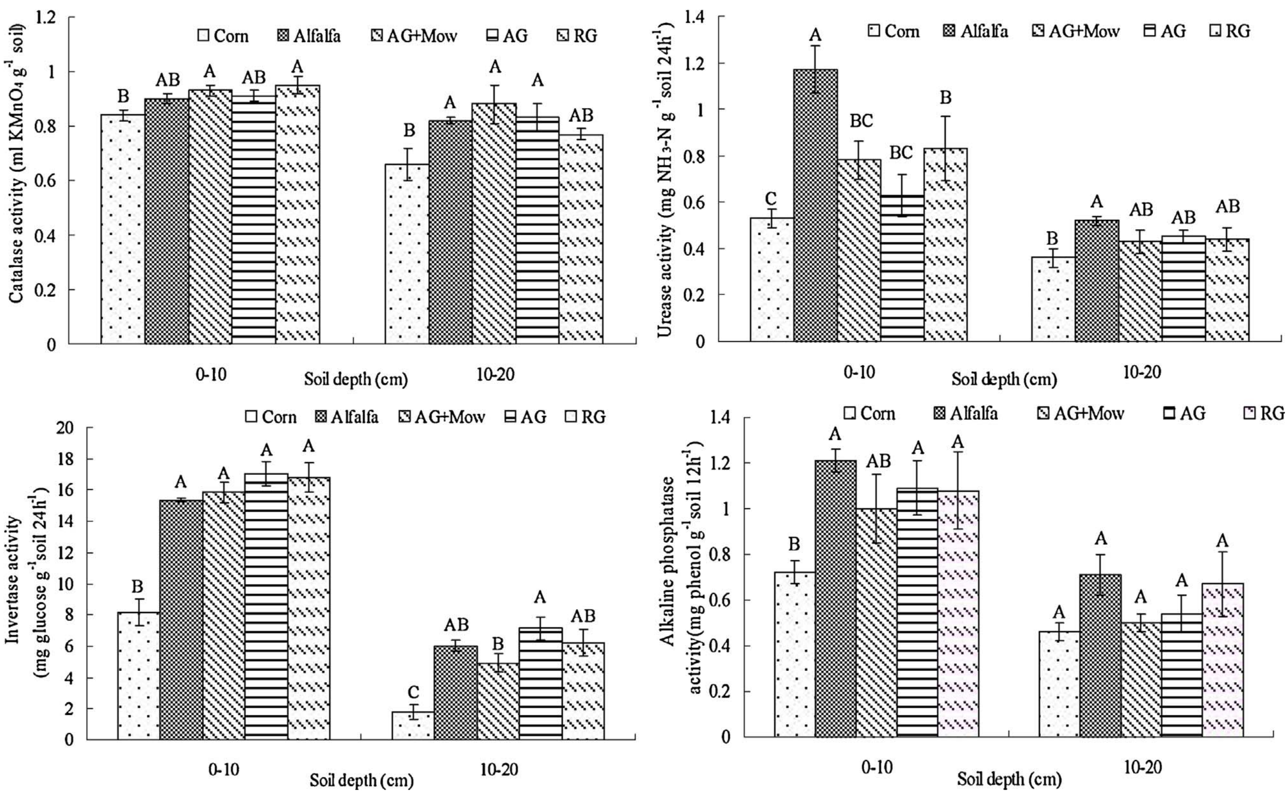

Fig. 3. Soil enzyme activities at 0–10 cm and 10–20 cm depth under different land use. Results are shown as mean (± SE). Values with the same uppercase letters within a land use are not significantly different at $P < 0.05$. The bars represent standard errors.

response index of labile C measures, the increases of GMC in the forage land and grasslands also indicated that their establishment contributed to the sequestration of soil labile C and that alfalfa planting offered a better choice than the other land use treatments to rapidly increase soil organic C in northeastern China.

3.2. Changes in soil enzyme activities under different land uses

Many studies reported that soil enzyme activities respond rapidly to changes in land use (Wang et al., 2008; Raiesi and Beheshti, 2014; Rui et al., 2014). Our results showed that soil enzyme activities varied with different land uses at 0–10 cm and 10–20 cm depths (Fig. 3). At the 0–10 cm depth, land use treatments had a significant effect on the activities of URE ($F = 6.45$, $P < 0.010$) and INV ($F = 25.86$, $P < 0.001$), whereas no significant differences were found for CAT ($F = 2.96$, $P = 0.055$) and ALP ($F = 2.50$, $P = 0.087$), indicating that URE and INV were more sensitive than CAT and ALP to land use treatments in the 0–10 cm depth. However, the activities in Alfalfa, AG + Mow, AG and RG increased by 7.14%, 10.71%, 8.33% and 13.10% for CAT and by 68.06%, 38.89%, 51.39% and 50.0% for ALP, respectively, compared with Corn, suggesting that CAT and ALP activities were also sensitive to land use. The increased activities of the four measured enzymes in Alfalfa and the grasslands in the present study indicated that biological activity was greatly promoted by plant colonization. Planting grass is well known to improve soil quality by increasing above- and belowground biomass inputs (Gabarron-Galeote et al., 2015b). The C input from vegetation or root material to soils provides a well-developed habitat and more stable food web for soil microbes, and therefore, the activities of microorganisms increase and more soil enzymes are produced (An et al., 2009; Veres et al., 2015). Additionally, soil enzymes can also adhere to SOC and its fractions to form humus-protein complexes that can protect enzymes from decomposition (Allison and Jastrow, 2006). Compared with the other soil enzymes, the effect of different land uses was greater on URE activities. The highest URE activity was found in Alfalfa (1.17 mg NH_3-N g^{-1} soil 24 h^{-1}), followed by RG, AG + M and AG, and Corn with the lowest values (0.53 mg NH_3-N g^{-1} soil 24 h^{-1}). As an N cycle enzyme, URE plays an important role in soil nitrogen utilization because of the hydrolysis of urea to ammonia-nitrogen. Therefore, the highest URE activity in Alfalfa was not surprising, because this forage is an important N fixing legume, which likely increased the nitrogen content and improved the URE activity. These results are consistent with the findings of Wang et al. (2012a) who found similar patterns for higher URE activity under two legume sites of *Caragana korshinskii* and *Robinia pseudoacacia* than under other land uses.

In the 10–20 cm depth, land uses had a significant effect on the activities of CAT ($F = 3.44$, $P < 0.050$) and INV ($F = 10.52$, $P < 0.001$), whereas no significant differences were found for URE ($F = 1.84$, $P = 0.174$) and ALP ($F = 1.55$, $P = 0.238$) among the five land use treatments, which indicated that INV and CAT were more sensitive than URE and ALP to land uses in the 10–20 cm depth. Land use treatments had no significant effect on ALP activities at both soil depths of 0–10 cm and 10–20 cm, suggesting that ALP activity was less sensitive to different land uses than other enzyme activities. However, three types of phosphatase are found in soils, which are primarily determined by the soil pH (Guan, 1986). The soil pH in our study sites ranged from 9.0 to 10.5, which was very high (Yu et al., 2014) and likely determined the minimal change in ALP activities. By contrast, land use treatments had significant effects on INV activities at both soil depths. Li et al. (2015) also observed the sensitivity of INV activities to different land uses in soils of the NW Loess Plateau of China. Soil INV activities are closely associated with the C cycle and reflect the intensity of C metabolism (Liu et al., 2014). The increases of C input from vegetation biomass and in labile C pools might be the primary reason for the sensitivity of INV to different land uses.

In the present study, four enzymes involved in soil C, N and P transformations were evaluated, but the sensitivity of soil enzyme activities to land use treatments was different. The geometric mean of

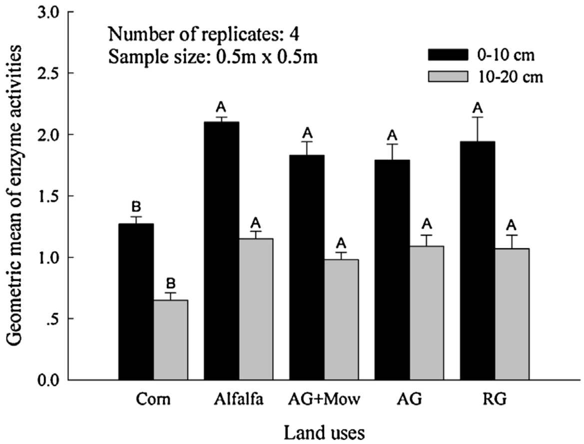

Fig. 4. The geometric mean of soil enzyme activities under different land uses. Values with the same uppercase letters within a land use are not significantly different at $P < 0.05$. The bars represent standard errors.

enzyme activities (GME) integrated information of the various soil enzymes, and can indicate the overall levels of enzyme and general microbiological activities (Hinojose et al., 2004). Compared with Corn, the GME value in Alfalfa, AG + Mow, AG and RG increased by 65.35%, 44.09%, 40.94% and 52.76%, respectively, in the 0–10 cm depth and by 76.92%, 50.77%, 67.69% and 64.62% in the 10–20 cm depth, respectively (Fig. 4). The significant increases in GME were primarily due to the higher sustained inputs of above- and belowground biomass in the forage land and grasslands than in the cropland, which stimulated the growth and activity of microbial populations to produce more soil enzymes (Sheng et al., 2015). Similarly, Wang et al. (2012b) and Raiesi and Beheshti (2015) observed that soil GME values were reduced by short-term (3 years) and long-term (more than 40 years) cultivation; therefore, this index could explain the changes in soil quality following land use changes. Among the forage land and grasslands, the highest value of GME at the 0–20 cm depth was found in Alfalfa, followed by RG, and with lower GME values under the land uses AG and AG + Mow. The reasons for the differences were primarily due to the different planting regimes and human management practices, which provided different forms of energy, substrates and habitats for soil microbes, leading to the production of different types and amounts of soil enzymes. Additionally, the differences in microclimates under the different land use treatments might affect soil enzyme activities.

Overall, the higher values of GME and GMC in the forage land and grasslands than those in the cropland and the highest values in Alfalfa at both soil depths indicated that plant colonization had a positive effect on the recovery of soil labile C and enzyme activities and that alfalfa planting might be the optimum land use to rapidly increase soil quality in the alkaline soils of northeastern China with soil pH more than 8.0. Additionally, production of alfalfa, as high quality forage, can also provide an economic return for the landowner and therefore, is a win-win strategy to solve land degradation and increase the income of farmers in the Songnen Grassland. Profitable forage production and soil restoration potential depends on high yields of alfalfa and soil degradation level. Field trials performed in southwestern Wisconsin showed that yields of alfalfa yields drop sharply when soil pH falls below 6.7 (Undersander et al., 2011). Therefore, carefully selecting field with suitable soil and environment conditions, and using appropriate planting practices before conversion of cropland to alfalfa land is important to ensure longer economic and environmental profitable in other semiarid agroecosystems. Changes in soil labile C and enzyme activities due to changes in land uses can influence physical, chemical and biological properties of soil and affect the ecology processes in soil ecosystems (Syswerda et al., 2011). According to many studies on grassland restoration, soil quality and food web function require 6–18 or even 23 years to stabilize (An et al., 2009; Yin et al., 2014; Machmuller et al., 2015). Five years after conversion in our study, SOC content at the 0–20 cm depth increased by 2.52 g C kg^{-1} (40.42%) in the grasslands, which remains 2.24 g C kg^{-1} less than the peak SOC content in a healthy grassland (Yu et al., 2014); therefore, four to five more years might be required to recover SOC contents to the peak content based on the current accumulation rate. Thus, soils in the Songnen Grassland may require at least 10 years to recover after conversion from cropland to grassland.

3.3. Correlations of soil labile C with soil enzyme activities

Pearson correlations between soil labile C pools and enzyme activities are listed in Table 2. The significant correlations among soil labile C pools and SOC indicate that contents of soil labile C were largely dependent on the quantity of SOC in soils (McLauchlan and Hobbie, 2004). This finding is consistent with the reports of Benbi et al. (2015) for cultivated and undisturbed soils in northern India and also that of Demisie et al. (2014) for red soils in China.

Decomposition of SOC and its fractions is an important microbial-mediated process by which soil microbes produce soil enzymes to catalyze the mineralization of SOC and convert the nutrients from organic forms into inorganic forms (Demisie et al., 2014). Thus, contents of SOC and its fractions can be adequate predictors of changes in soil enzyme activities and microbial biomass (Xu et al., 2015). Significant positive correlations between SOC, OLC, SMBC and four enzyme activities in the present study confirmed the importance of SOC and its fractions in maintaining enzyme activities (Table 2). These positive correlations are consistent with those of Liu et al. (2014) who found similar positive relationships between soil C content and soil enzyme activities. However, the WEOC showed no obvious correlation with soil CAT activity (r = 0.136, P > 0.05), and a slight negative correlation with other soil enzyme activities (r < 0.506, P < 0.05) (Table 2), most likely attributed to the easily oxidizable nature of WEOC and because WEOC was both the primary substrate and product of soil enzymatic reactions (Li et al., 2016b).

Table 2
Correlation between soil labile C and different soil enzyme activities.

	SOC	OLC	SMBC	WEOC	CAT	URE	INV
OLC	0.962***	1					
SMBC	0.755***	0.789***	1				
WEOC	−0.383*	−0.4*	−0.484***	1			
CAT	0.637***	0.627***	0.419*	0.136NS	1		
URE	0.763***	0.804***	0.81***	−0.467***	0.446***	1	
INV	0.903***	0.883***	0.837***	−0.366*	0.666***	0.738***	1
ALP	0.896***	0.912***	0.808***	−0.506***	0.548***	0.803***	0.853***

*, **, *** indicate significant difference at $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively. NS indicate no significant difference at $P < 0.05$. SOC, soil organic carbon; OLC, oxidizable labile carbon; SMBC, soil microbial biomass carbon; WEOC, water-extractable organic carbon; CAT, soil catalase activity; URE, soil urease activity; INV, soil invertase activity; ALP, soil alkaline phosphatase activity.

4. Conclusions

This study provided insight into the responses of soil labile C and enzyme activities to the different land use treatments in an agro-pastoral transitional zone in northeastern China. Results showed that soil labile C and enzyme activities were sensitive to different land uses. Although the range of responses of different soil labile C pools and enzyme activities to different land uses was different, the higher values of GMC and GME for the land use treatments of Alfalfa and grasslands than those for Corn indicated that the forage land and grasslands contributed to the increases in soil C content and enzyme activities. Based on the highest values of GMC and GME in Alfalfa treatments, alfalfa planting may be a rare win-win strategy by combining profitable forage production and rapid improvement of soil quality in north-eastern China. The following conclusions were reached in this study: (1) Land use treatments had different effects on SOC content, soil labile C pools and soil enzyme activities, primarily through the effects of planting regimes and management practices; (2) Soil labile C and enzyme activities were useful indicators of soil quality and health following land use changes; (3) With the colonization of plants and reduction in disturbance, particularly the planting of leguminous species, soil C content and enzyme activities can be expected to gradually recover in semiarid agroecosystems.

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