Distribution of soil carbon and nitrogen along a revegetational succession on the Loess Plateau of China

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ABSTRACT

Changes in the natural abundance of δ^{13}C and δ^{15}N in soil are proposed as indicators in the evolution of ecosystems caused by land management and/or vegetational succession. Such changes are seldom known due to the lack of comparative data on the levels of δ^{13}C and δ^{15}N in soils. To understand the distribution of C and N during revegetational succession, we investigated the changes in the levels of δ^{13}C, δ^{15}N, organic carbon (OC), and total nitrogen (TN) for soils sampled from three incremental depths (0–10, 10–20, and 20–40 cm) during a revegetational succession from alfalfa (*Medicago sativa*, C$_3$ plant) to the natural vegetation of bunge needlegrass (*Stipa bungeana*, C$_3$ plant) in the northern Loess Plateau. The levels of δ^{13}C and δ^{15}N increased gradually with the succession of the alfalfa community and decreased with the succession to the bunge needlegrass community, indicating that the dynamics of the δ^{13}C/δ^{15}N isotopic signatures differed within the different revegetational stages of succession. A pronounced inverse relationship developed between the soil water storage (0–40 or 0–100 cm depths) and the natural abundance of δ^{13}C/δ^{15}N. The use of this relationship may be of value to scientists modelling nutrient cycling within different ages of vegetational succession across the semiarid regions of the Loess Plateau. Concentrations and stocks of OC and TN increased with revegetational age during the two successional stages, suggesting that revegetation enhanced the sequestration of OC and N in soils by reducing soil erosion and increasing inputs of organic materials. Our results highlight the utility of stable isotopes in studying C and N dynamics along revegetational succession on degraded land and emphasise the improvement in soil OC and N by well-managed strategies of vegetational restoration in the northern Loess Plateau.

1. Introduction

The revegetation of degraded land is one of the principal strategies for the control of soil erosion and recovery of ecosystems in the frail regions where ecosystems can be easily disturbed by anthropogenic activities or severe environmental conditions. Revegetation can also greatly influence soil quality, cycling of carbon (C) and nitrogen (N), land management, and regional socioeconomic development (Eaton et al., 2008; Fu et al., 2010). The development of managed grasslands and forestlands accelerates the restoration of ecosystems, promotes the area's livestock industry (She et al., 2009), and affects the C and N cycles and pools stored in soils (Eaton et al., 2008; Fu et al., 2010; Wei et al., 2010). However, excessive planting of nonnative species may exacerbate deficits of soil water, adversely affecting the succession of vegetation (Li, 1983; Wang and Zhang, 2003) and thus C and N dynamics. Altered C and N cycles and pools in turn influence the production of biomass and ecosystemic functioning (Foster et al.,

2003; Wei et al., 2010). Elucidating the dynamics of organic carbon (OC) and N in soils along such successions may therefore have important implications for the sustainable management of land resources and the improvement of model simulations and predictions of future global C cycling.

The northern Loess Plateau of China is mainly characterised by a semiarid climate and has been affected by various forms of human activities, including over-grazing and large-scaled monoculture (Fu et al., 2000). During the past century, degradation and fragmentation of ecological environments have been accelerated due to an expanding human population and a changing lifestyle. To improve the frail natural ecosystems on the Loess Plateau and alleviate the degradation of land, the Chinese government has launched a series of nationwide conservation projects, including eco-environmental revegetation, focused on the recovery of damaged ecosystems (Wang, 2002). Recommendations included carefully monitoring managed artificial grassland successions to control soil erosion, increasing stocks of soil OC and N and preventing the occurrence of dried soil layers on the plateau (Fu et al., 2010; Wang et al., 2010). Alfalfa (*Medicago sativa*) has been widely used in the restoration of degraded ecosystems due to its rapid growth, protection of the soil surface, and fixation of N in the soil (She et al., 2009). Many

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studies, however, have reported that alfalfa grassland degrades quickly without careful management because of intense competition from native grasses and excessive consumption of water (Li and Huang, 2008; Li et al., 2006). This vegetational succession may affect the processing of soil OC and N decomposition. Little is known, though, about long-term changes in soil OC and N during vegetational succession on the Loess Plateau. This information can be useful for estimating the temporal distribution of stocks of soil OC and N and for evaluating OC and N dynamics during a succession from managed to natural communities in semiarid regions.

In terrestrial ecosystems, the natural stable isotopic ratio of C and N (δ^{13}C and δ^{15}N) in bulk soil has been increasingly applied to help meet the challenge of closing the gaps in our understanding of the dynamics of OC and N (Arrouays et al., 1995; Awiti et al., 2008; Boutton et al., 1998; Ehleringer et al., 2000; Evans, 2001; Högberg, 1997; Nadelhoffer et al., 1996; Wynn et al., 2006). For example, Arrouays et al. (1995) modelled the turnover of OC in soils of cleared temperate forests converted to maize cropping by measuring the natural abundance of ^{13}C. Awiti et al. (2008) studied the dynamics of forest-derived and maize-derived C by noting the changes in levels of δ^{13}C between soils of forest and cropland. Revegetation of degraded lands provides an opportunity to use the natural abundance of ^{13}C and ^{15}N to evaluate the dynamics of soil OC and N. Revegetation can thus help us to understand the long-term impact of the reconstruction of degraded systems on C and N sequestration, because the signatures of stable isotopes and the processes of C and N cycling in ecosystems naturally evolve over periods of time (Lambers et al., 2008; Selmants and Hart, 2008; Vitousek, 2004). Patterns of change in the isotopic ratio of C and N are rather inconsistent along vegetational succession; some types show increasing trends (Osono et al., 2006; Wedin et al., 1995), whereas others show decreasing trends (Benner et al., 1987). This inconsistency may be attributed to changes in environmental conditions, isotopic differences between surface litter and root-derived soil organic matter, selective preservation of recalcitrant components depleted in ^{13}C such as lignin, kinetic fractionation during the maturation of soil organic matter, etc. (Balesdent et al., 1993; Ehleringer et al., 2000; Swap et al., 2004; Wynn et al., 2006). Similarly, revegetation of degraded lands can affect the dynamics of soil nitrogen, in particular by leguminous plants that can fix atmospheric N_2. Patterns of N transformation (N-mineralisation, nitrification, denitrification, N-immobilisation, and N-assimilation) have significant effects on the natural abundance of ^{15}N in soils (Högberg, 1997; Hopkins et al., 1999; Nadelhoffer and Fry, 1994). However, global δ^{13}C and δ^{15}N data sets spanning different ages of vegetational restoration are currently lacking (Ehleringer et al., 2000), although information is available for understanding the dynamics of C and N along revegetational chronosequences.

We therefore investigated the natural abundances of ^{13}C, ^{15}N, OC, and total nitrogen (TN) in soils at depths of 0–10, 10–20, and 20–40 cm from restored grasslands of different ages in the northern Loess Plateau. The objectives were to evaluate the application of stable-isotopic techniques in studying C and N dynamics in the restored ecosystems along revegetational succession and to assess the improvement of soil OC and N by well-managed strategies of vegetational restoration in the northern Loess Plateau.

2. Materials and methods

2.1. Location of study site

The study was conducted in a Liudaogou catchment on the northern Loess Plateau of China (latitude 38°46′ to 38°51′ N and longitude 110°21′ to 110°23′ E) at the Shenmu Erosion and Environmental Research Station of the Institute of Soil and Water Conservation, Chinese Academy of Sciences. The altitude and area of the catchment are 1081–1274 m and 6.89 km², respectively. This area is characterised

by a semiarid climate with a mean annual temperature of 8.6 °C and an annual precipitation of 414 mm (from 1961 to 2009; see Fig. 1). The soil is loessial and is formed over a deep (up to 100 m) loessial layer. The soil is characterised as low cohesion, low water retention, and is prone to erosion. The study site is representative of the transitional region subjected to severe erosion by both wind and water. To remedy the severe erosion, widespread vegetational restoration has been implemented in the region during the past several decades by planting arid-land perennial vegetation, including purple alfalfa (*M. sativa*) and korshinsk peashrub (*Caragana korshinskii Kom*).

2.2. Sampling-site selection and botanical surveys

To study the changes in soil C and N for long-term recovery of vegetation, the method of space-for-time substitution, or chronosequence (Foster and Tilman, 2000; Li et al., 2005), was used to select different sampling plots with various ages of revegetation. In this study, an existing successional sere, i.e. a series of stages of a particular plant succession, was selected in a relatively homogeneous field (i.e. with similar bedrock, slope gradient, and parent material), where alfalfa had been grown in a randomised plot design for periods ranging from three to 48 years. Slope gradient generally varied from 5° to 15° (Table 1). Eight successional stages (3, 8, 10, 18, 22, 28, 41, and 48 years) had been grown in plots no smaller than 500 m². Alfalfa (a C_3 plant) was planted in the degraded lands without fertilization and irrigation. The farmers cut the alfalfa one or two times each year for livestock. Li and Shao (2005) and She et al. (2009) divided the revegetational succession into three stages: artificial alfalfa community (1–7 years), transitional community from alfalfa to bunge needlegrass
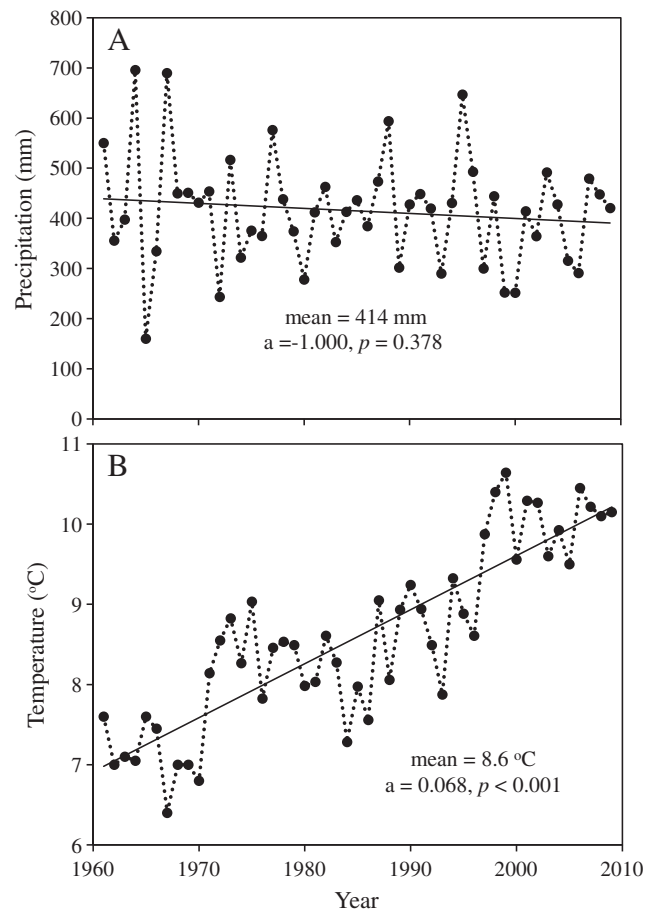

Fig. 1. Changes in annual precipitation and annual temperature between 1961 and 2009 for the study area of the Loess Plateau (the slope of the linear regression of the data is given by "a", "m" indicates the mean precipitation/temperature of this period).

Table 1
Geographical and vegetational characteristics of the sampling sites representing a revegetational gradient. Biomass values followed by different lower-case letters within columns are significantly different at $P<0.05$.

Vegetational stage	Revegetational age	Elevation (m)	Geographical coordinates Ψ (N), λ (E)	Slope	Accompanying species (C_3)	Aboveground biomass (g m^{-2})	Belowground biomass (g m^{-2})
Purple alfalfa (Medicago sativa) dominated community							
ALF-1	3	1205	38°47′33.1″ 110°21′52.6″	10°	*Stipa bungeana* Trin.; *Lespedeza davurica*	245.7 ± 27.6b	387.6 ± 43.5ab
ALF-2	8	1205	38°47′45.8″ 110°21′53.9″	8°	*Stipa bungeana* Trin.; *Artemisia scoparia*	298.7 ± 14.2a	391.2 ± 18.8ab
ALF-3	10	1180	38°47′39.3″ 110°21′51.0″	8°	*Stipa bungeana* Trin.; *Artemisia scoparia*; *Polygala tenuifolia*	295.3 ± 25.1a	450.8 ± 38.3a
ALF-4	18	1222	38°47′31.6″ 110°21′50.0″	5°	*Stipa bungeana* Trin.; *Artemisia eriopoda*	237.3 ± 24.4b	367.5 ± 37.8b
Bunge needlegrass (Stipa bungeana Trin.) dominated community							
BNE-1	22	1179	38°47′39.0″ 110°21′50.8″	8°	*Poa sphondylodes* Trin.; *Melilotus albus* Desr.; *Lespedeza davurica*	154.7 ± 26.6c	265.5 ± 45.6c
BNE-2	28	1223	38°47′29.5″ 110°21′50.9″	5°	*Melilotus albus* Desr.; *Lespedeza davurica*; *Polygala tenuifolia*; *Heteropappus altaicus* (Willd.)	162.3 ± 15.5c	284.4 ± 27.2c
BNE-3	41	1253	38°47′26.3″ 110°22′10.4″	15°	*Lespedeza davurica*; *Cleistogenes chinensis*; *Oxytropis bicolor*; *Polygala tenuifolia*	232.7 ± 24.2b	372.9 ± 3 8.8b
BNE-4	48	1207	38°47′39.0″ 110°22′02.1″	12°	*Lespedeza davurica*; *Astragalus scaberrimus* Bunge	302.7 ± 15.5a	392.4 ± 20.1ab

(*Stipa bungeana*) (7–15 years), and secondary natural grassland community dominated by *S. bungeana* (15 years or more). In our study, the transitional community defined by Li and Shao (2005) and She et al. (2009) was included in the artificial alfalfa community because it is still dominated by alfalfa due to careful management. After 18 years, alfalfa was replaced by a stable natural community dominated by *S. bungeana*. The bunge needlegrass community was protected from being disturbed by humans with natural revegetation to improve the potential for the sequestration of soil OC and to alleviate the degradation of land. In particular, by the early 1980s, the Chinese government imposed polices for some fragile ecosystems, including the northern Loess Plateau, and livestock grazing was banned from the agro-pastoral ecotone.

Vegetational surveys were conducted from September to October 2009. At each restored grassland site, three randomised circular sample plots of 3.14 m² (1 m radius) were surveyed, and the plants were separated and counted. Each sample plot was at least 20 m from its nearest neighbour. Edge effects were avoided by not sampling within a 10-m buffer zone around the field boundaries. The aboveground biomass was taken, after identification, by clipping within the sample plot and then dried at 65 °C in an oven for 72 h to estimate dry weight. After aboveground plant residues were collected, five soil cores (10 cm diameter) were collected from 0 to 40 cm soil depths to estimate belowground biomass in each sample plot. All cores were transported to the laboratory and carefully washed on a 60-mesh sieve to separate the roots from the soil as soon as possible. The washed roots were oven-dried at 65 °C for 72 h to estimate dry weight. The geographic and vegetational characteristics of the sites are described in Table 1. All the species in the sample sites are C_3 species.

2.3. Soil sampling and laboratory analyses

In early October 2009, undisturbed soil samples were taken at the eight studied sites at three depth increments: 0 to 10 cm, 10 to 20 cm, and 20 to 40 cm. Three core samples (5 cm) were collected from each site and mixed to form a composite sample of about 1 kg and taken to the laboratory. The soil samples were air-dried, crushed, and passed through 1.0 mm and 0.25 mm sieves before chemical analysis. Analytical methods described in ISSCAS (1981) were chosen for determining the chemical properties of the soil. Samples passed through a 0.25 mm mesh were used to determine soil OC, TN, and CaCO₃ contents. Soil OC was measured by wet digestion with a mixture of potassium dichromate and concentrated sulphuric acid. TN was measured by the semi-macro Kjeldahl method. CaCO₃ was

determined by dissolving soil samples in HCl and measuring the amount of CO₂ released. Soil pH was measured at a soil (passed through a 1 mm mesh) to water mass ratio of 1:2.5 by using a pH meter equipped with a calibrated combined glass electrode. Samples passed through a 1 mm mesh were used to determine soil mechanical composition (%) by laser diffraction using a Mastersizer 2000 (Malvern Instruments, Malvern, England). Soil bulk density (g cm^{-3}) of each studied site was measured using a soil bulk sampler with a 5.0 cm diameter by 5.0 cm height stainless steel cutting ring within 0–10, 10–20, 20–40, 40–60, 60–80, and 80–100 cm soil depths (3 replicates) at points adjacent to the soil sampling quadrants by measuring the original volume of each soil core and the dry mass after oven-drying at 105 °C. Soil water content (θ) was simultaneously determined gravimetrically. Soil water storage (mm) for different soil layers was calculated from the θ and bulk-density data using the trapezoidal rule (Fig. 2). Selected physico-chemical properties in different profiles under different revegetational years are shown in Table 2.

Stocks of OC (SOC) and TN (STN) (which mean soil OC and TN quantity per unit area, Mg ha^{-1}) were calculated as follows: Stocks of soil OC, SOC$_i$, for a given soil layer i was calculated using the following equations:

$$SOC_i = OC_i * BD_i * E_i * (1 - F_i/100)/10$$

$$STN_i = TN_i * BD_i * E_i * (1 - F_i/100)/10$$

where i is the ith layer, and OC$_i$, TN$_i$, BD$_i$, E$_i$ and F$_i$ are the soil OC concentration (g kg^{-1}), TN concentration (g kg^{-1}), bulk density (g cm^{-3}), thickness (cm), and proportion (%) of coarse (>2 mm) fragments in the ith layer, respectively. The occurrence of coarse particles in the loessial soils of the study region was rare, so F$_i$ was considered to be negligible (Liu et al., 2011). We could determine SOC$_i$ and STN$_i$ to a depth of 40 cm more reliably by using the data obtained for the three layers, 0–10 cm, 10–20 cm, and 20–40 cm, which included the measured soil bulk densities. In this research, SOC$_{(0-40\ cm)}$ and STN$_{(0-40\ cm)}$ were calculated as the sum of the SOC$_{(0-10\ cm)}$, SOC$_{(10-20\ cm)}$, and SOC$_{(20-40\ cm)}$; and STN$_{(0-10\ cm)}$, STN$_{(10-20\ cm)}$, and STN$_{(20-40\ cm)}$, respectively.

The analysis of $\delta^{13}C$ and $\delta^{15}N$ was implemented using a C/N/O/H/S analyser coupled to an isotope-ratio mass spectrometer (MAT253, Thermo Fisher Scientific Company, US) at the State Key Laboratory of Hydrology-Water Resources and Hydraulic Engineering at Hohai University in Nanjing. Stable ^{13}C isotope abundance is typically expressed as $\delta^{13}C$ in per mil (‰), as the relative ratio of the heavy isotope ^{13}C to the light isotope ^{12}C in a sample, relative to the Vienna
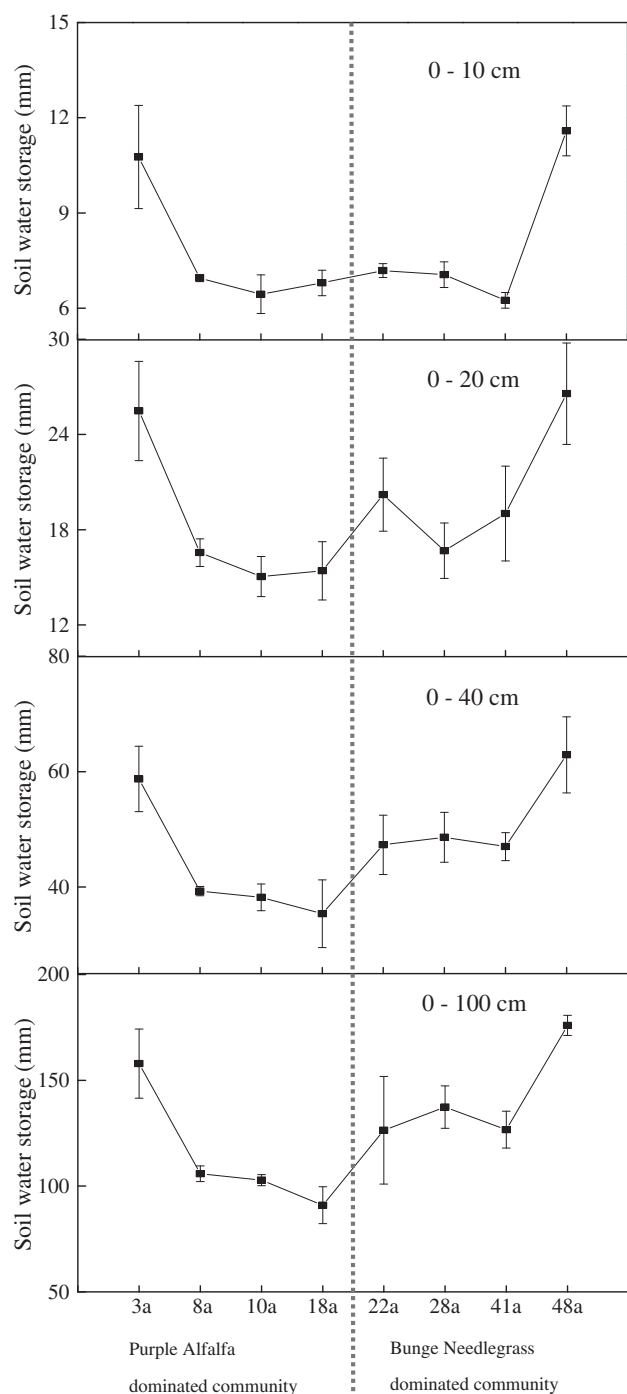
Fig. 2. Changes in soil water storage (SWS) at 0–10, 0–20, 0–40 and 0–100 cm depths along revegetational succession.

Pee Dee Belemnite (VPDB) standard. δ^{15}N was expressed relative to the air standard (N_{air}). The δ^{13}C and δ^{15}N values were calculated as δ (‰) = 1000 * [($R_{sample}/R_{standard}$) − 1] with R defined as the ^{13}C/^{12}C or ^{15}N/^{14}N ratio (Awiti et al., 2008).

2.4. Statistical analysis

Data were analysed to provide mean and standard deviation for each variable measured at every depth for sites representing the different stages of revegetation. One-way analysis of variance (ANOVA) followed by the LSD test ($p < 0.05$) was used to examine

the effect of revegetational ages on soil δ^{13}C, δ^{15}N, OC, and TN. If significant effects were observed by ANOVA, a least significant difference [LSD (0.05)] test was used. All statistical analyses were carried out with SAS software (SAS Institute Inc., Cary, NC, USA).

3. Results

3.1. Characteristics of vegetational succession and soil properties

Vegetational succession in the sites began after the transition of land from degraded to restored land, represented as the 1-year site. However, in this study, the 1-year or 2-year alfalfa communities were not investigated, thus the vegetational succession in the sites hypothetically began after three years' alfalfa planting. Species in the successional were changed over time; however, non-C$_4$ species have been observed in the sampling field (Table 1). With the increase of revegetational age, the occurrence of *S. bungeana* increased. After approximately the 18th year, it became the predominant species, and the community was recognised as a bunge needlegrass community. We divided the plant community succession into two stages: alfalfa (ALF) community and bunge needlegrass (BNE) community.

The aboveground and belowground biomasses changed along the revegetational succession. Generally, biomass increased at the early revegetational stage and declined after about eight years. These changes can be attributed to the reduction of alfalfa, because the unstable alfalfa monoculture was invaded by highly competitive native species that increased the competition for limited resources, while native species may have benefited from increased levels of N fixed by the alfalfa plants (She et al., 2009); however, biomass began to increase after the vegetation developed to a BNE community with increasing restorational ages (Table 1).

Selected soil physico-chemical properties (soil texture, soil pH, soil bulk density, and soil CaCO$_3$ concentration) for different soil layers do not vary significantly with revegetational age (Table 2). Inconsistent with other soil properties, soil water storage (SWS) changed significantly along the vegetational chronosequence of the restoration. The SWS generally decreased at the early revegetational stage, but increased after alfalfa was replaced by *S. bungeana* (Fig. 2). During the first eight years, alfalfa was the predominant species, which consumed more water due to its high rate of evapotranspiration and deep root system (Dunin et al., 2001; Li and Huang, 2008). Therefore, SWS was lowest for the first successional stage compared with other stages. Compared to alfalfa, however, the BNE community had relatively low evapotranspiration, and the relatively low biomass led to the increase of the SWS in the 0–100 cm layer due to supplementation from precipitation.

3.2. Soil δ^{13}C and δ^{15}N at different stages of revegetation

The values of δ^{13}C and δ^{15}N varied significantly with soil depth and revegetational stage (Fig. 3). At the 0–40 cm depth, δ^{13}C increased gradually with revegetational age in the ALF community, with a range of −23.7‰ to −22.5‰ ($p < 0.05$). However, ^{13}C decreased during the process of natural rehabilitation in the BNE community, with a range of −22.5‰ to −24.4‰. Changes in δ^{13}C were mostly significant ($p < 0.05$) for the 0–10 and 10–20 cm soil layers among different stages during the process of vegetational rehabilitation, and 20–40 cm layers were slightly changed, but the changes were mostly not statistically significant (Fig. 3A).

Soil δ^{15}N showed a similar trend to δ^{13}C (Fig. 3B). The δ^{15}N in soil depths of 0–10 cm and 10–20 cm increased significantly with revegetational age in the ALF community (ranging from 0.75‰ to 2.58‰) but decreased significantly in the BNE community (ranging from 1.90‰ to 0.44‰) (Fig. 3B). However, δ^{15}N changed slightly with revegetational ages in the 20–40 cm soil layer, indicating that the effects of revegetational age on soil δ^{13}C and δ^{15}N were mainly

Table 2
Selected physico-chemical properties in different profiles under different revegetational ages.

Revegetational ages	Layer (cm)	Texture (%)			Bulk density ($g\, cm^{-3}$)	pH	$CaCO_3$ ($g\, kg^{-1}$)
		Clay <0.002 mm	Silt 0.2-0.002 mm	Sand >0.2 mm			
3	0–10	12	78	10	1.39 ± 0.03	8.2	85
	10–20	12	76	12	1.43 ± 0.08	8.1	86
	20–40	16	81	3	1.52 ± 0.02	8.2	99
	Mean	14	78	8	1.45 ± 0.02	8.2	90
8	0–10	16	80	4	1.42 ± 0.04	8.2	79
	10–20	26	72	2	1.52 ± 0.01	8.2	71
	20–40	24	74	2	1.54 ± 0.01	8.1	82
	Mean	21	76	3	1.50 ± 0.01	8.2	81
10	0–10	12	82	6	1.46 ± 0.04	7.9	71
	10–20	27	70	3	1.52 ± 0.02	7.9	67
	20–40	26	72	2	1.53 ± 0.01	7.9	96
	Mean	22	74	4	1.50 ± 0.01	7.9	78
18	0–10	15	80	5	1.48 ± 0.04	8.2	79
	10–20	24	72	4	1.51 ± 0.06	8.2	96
	20–40	24	71	5	1.57 ± 0.05	8.2	126
	Mean	21	74	5	1.52 ± 0.03	8.2	100
22	0–10	19	72	9	1.39 ± 0.10	8.2	88
	10–20	24	74	2	1.50 ± 0.03	8.3	89
	20–40	27	67	6	1.52 ± 0.04	8.3	86
	Mean	23	71	6	1.47 ± 0.03	8.3	88
28	0–10	13	83	4	1.44 ± 0.03	8.4	88
	10–20	15	82	3	1.50 ± 0.09	8.3	106
	20–40	21	74	5	1.48 ± 0.04	8.3	122
	Mean	16	80	4	1.47 ± 0.03	8.3	105
41	0–10	15	80	5	1.44 ± 0.05	8.3	82
	10–20	17	81	2	1.49 ± 0.02	8.3	99
	20–40	26	72	2	1.50 ± 0.03	8.2	93
	Mean	19	78	3	1.48 ± 0.03	8.3	91
48	0–10	13	80	7	1.40 ± 0.04	8.2	89
	10–20	24	73	3	1.52 ± 0.05	8.1	92
	20–40	23	75	2	1.55 ± 0.05	8.1	109
	Mean	20	76	4	1.49 ± 0.05	8.1	97

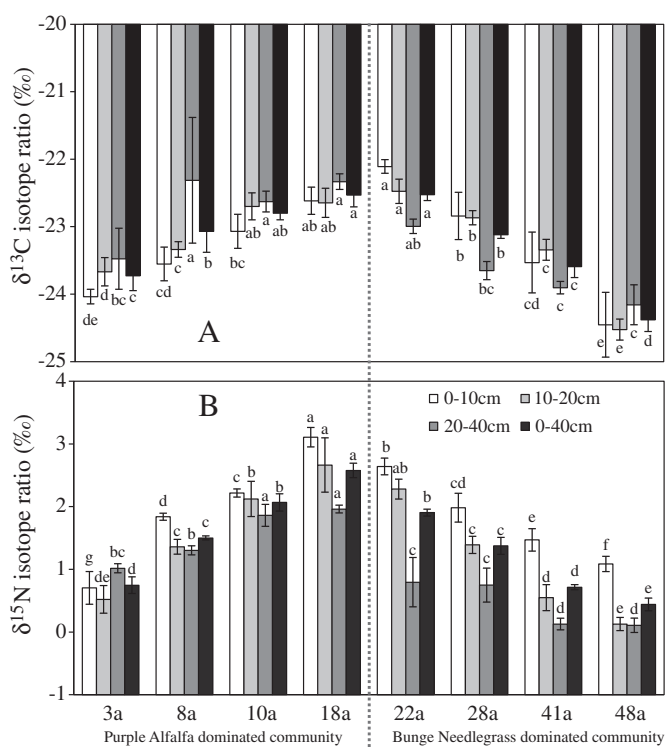**Fig. 3.** Changes in soil $\delta^{13}C$ and $\delta^{15}N$ in different soil layers along revegetational succession. Values are means ± 2SD with the sample size $n = 3$. Values followed by a different lower-case letters are significantly different within each soil depth at $P<0.05$.

exerted in surface soil. A sharp decline of $\delta^{13}C$ and $\delta^{15}N$ in the 20–40 cm soil layer, however, was observed after the ALF community was replaced by BNE, suggesting that species replacement had an influence on $\delta^{13}C$ and $\delta^{15}N$ in the sub-surface soils (Fig. 3). Relationships between soil $\delta^{13}C$, $\delta^{15}N$ at 0–40 cm depth, and SWS for 0–10, 0–20, 0–40, and 0–100 cm depths were described as linear functions (Table 3). Results showed that the SWS and $\delta^{13}C$ of soil are negatively correlated for the four soil layers (Table 3; $p<0.05$). A negative correlation between $\delta^{15}N$ and SWS for the 0–40 and 0–100 cm depths, is consistently significant (Table 3; $p<0.01$).

3.3. Concentrations of soil OC and TN at different stages of revegetation

Generally, concentrations of soil OC increased with revegetational age. At 0–40 cm, the concentration of OC increased with revegetational age from 2.01 $g\, kg^{-1}$ to 2.79 $g\, kg^{-1}$ for the ALF community and from 2.57 $g\, kg^{-1}$ to 5.53 $g\, kg^{-1}$ for the BNE community. The concentrations of OC were higher in upper soils than in deeper soils, indicating that soil OC had a tendency to accumulate in surface soils for the ALF community (Fig. 4A). However, soil OC gradually began to increase along the revegetational succession, with a short-term reduction of 0.53 $g\, kg^{-1}$ after the ALF community developed to the BNE community. Inconsistent with the ALF community, the concentration of soil OC in the BNE community significantly increased by 3.14 $g\, kg^{-1}$, 3.88 $g\, kg^{-1}$, and 1.87 $g\, kg^{-1}$ at 0–10, 10–20, and 20–40 cm depths, respectively (Fig. 4A). This indicates that the BNE community is more beneficial for relatively deep accumulation of carbon compared to the ALF community.

Similar to the soil OC, concentrations of soil N increased with revegetational time, with a sharp decline after ALF developed to a BNE community (Fig. 4B). Soil N increased from 0.20 $g\, kg^{-1}$ to 0.36 $g\, kg^{-1}$ in the ALF community and from 0.18 $g\, kg^{-1}$ to

Table 3
Linear and quadratic regression analyses of the $\delta^{13}C_{0-40\ cm}$, $\delta^{15}N_{0-40\ cm}$, $SOC_{0-40\ cm}$ (stocks of soil OC in the 0–40 cm depth) and $STN_{0-40\ cm}$ (stocks of soil TN in the 0–40 cm depth) using soil water storage at different soil depths and revegetational ages, respectively.

C and N	Soil depth (cm)	Equations	R^2
$\delta^{13}C_{0-40\ cm}$			
	0–10	$y = -0.24 \times x - 21.33$	0.596*
	0–20	$y = -0.11 \times x - 21.16$	0.545*
	0–40	$y = -0.06 \times x - 20.60$	0.704**
	0–100	$y = -0.02 \times x - 20.79$	0.719**
$\delta^{15}N_{0-40\ cm}$			
	0–10	$y = -0.23 \times x + 3.25$	0.415‡
	0–20	$y = -0.12 \times x + 3.70$	0.496‡
	0–40	$y = -0.06 \times x + 4.48$	0.716**
	0–100	$y = -0.02 \times x + 4.26$	0.728**
$SOC_{0-40\ cm}$			
	0–10	$y = 2.30 \times x^2 - 39.58 \times x + 175.76$	0.676‡
	0–20	$y = 0.22 \times x^2 - 8.47 \times x + 93.64$	0.401‡
	0–40	$y = 0.04 \times x^2 - 3.69 \times x + 95.14$	0.508‡
	0–100	$y = 0.01 \times x^2 - 1.24 \times x + 86.85$	0.631‡
		$y = 0.01 \times z^2 - 0.32 \times z + 13.75$	0.748*
$STN_{0-40\ cm}$			
	0–10	$y = 0.10 \times x^2 - 1.72 \times x + 8.53$	0.226‡
	0–20	$y = 0.02 \times x^2 - 0.93 \times x + 10.68$	0.598‡
	0–40	$y = 0.004 \times x^2 - 0.41 \times x + 11.22$	0.833*
	0–100	$y = 0.001 \times x^2 - 0.13 \times x + 9.49$	0.914**
		$y = 0.0002 \times z^2 - 0.01 \times z + 1.34$	0.063‡

(a) y, values of $\delta^{13}C_{0-40\ cm}$ (‰), $\delta^{15}N_{0-40\ cm}$ (‰), $SOC_{0-40\ cm}$ (Mg ha^{-1}) and $STN_{0-40\ cm}$ (Mg ha^{-1}); (b) x, soil water storage (mm); (c) z, revegetational age; (d) R^2, coefficients of determination; (e) *, $P < 0.05$; **, $P < 0.01$; ‡, $P > 0.05$.

0.33 g kg^{-1} for BNE community at 0–40 cm, respectively (Fig. 4B). For the ALF community, data showed that increases in soil N were mostly significant ($p < 0.05$) for the three layers among different years. Changes in soil N at 20–40 cm were not statistically significant for the BNE community. This suggests that ALF can more effectively

sequester N in relatively deep soil compared to the BNE community. Furthermore, N showed a similar trend to OC in accumulating in surface soils (0–20 cm).

3.4. Stocks of soil OC and TN at different stages of revegetation

For the ALF community, stocks of OC (SOC) in the 0–40 cm depth increased with revegetational age, with an annual rate of increase of 0.30 Mg ha^{-1} yr^{-1} (Fig. 5A). The increases in SOC with revegetational age, however, were not significant after eight years. SOC decreased after ALF was replaced by BNE. A similar increasing trend was observed in the BNE community, with an annual rate of increase of 0.66 Mg ha^{-1} yr^{-1} in 0–40 cm soils (Fig. 5A). Increases of SOC mainly occurred in surface soil (0–10 cm) during vegetational succession, with average rates of 0.14 Mg ha^{-1} a^{-1} over 15 years in ALF land and 0.20 Mg ha^{-1} a^{-1} over 26 years in BNE land (Fig. 5A). Consistent with SOC, the stocks of total N (STN) in 0–40 cm soils in ALF and BNE communities increased from 1.01 Mg ha^{-1} to 1.95 Mg ha^{-1} and 1.02 Mg ha^{-1} to 1.82 Mg ha^{-1}, with rates of increase of 0.06 Mg ha^{-1} a^{-1} and 0.03 Mg ha^{-1} a^{-1}, respectively. The STN decreased by 0.93 Mg ha^{-1} after ALF was replaced by a BNE community (Fig. 5B). For the ALF community, increases of STN were statistically significant in the three soil layers. However, the changes of STN in the BNE community were statistically significant at 0–10 and 10–20 cm depths. Combined with the relatively higher rates of increase in STN for the ALF community compared with BNE, ALF had a greater potential than BNE for stocking soil N due to the ability to fix atmospheric N_2. SOC and STN for the 0–40 cm depth, as a quadratic function of revegetational age, and SWS for 0–10, 0–20, 0–40, and 0–100 cm depths were determined (Table 3). The regression results of SOC by SWS for the four soil layers were not significant; however, the quadratic function with revegetational age was statistically significant ($R^2 = 0.748$; $p < 0.05$). In contrast, regression results of STN by revegetational age were not significant;
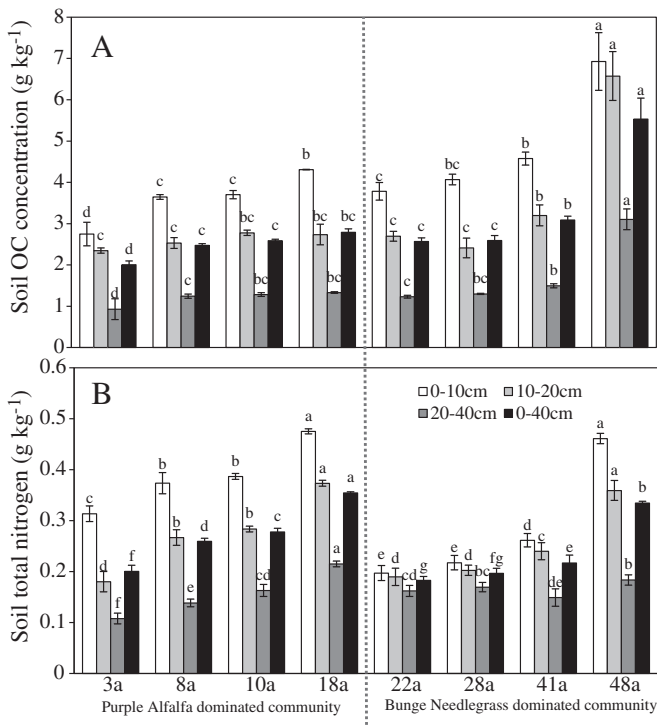

Fig. 4. Changes in concentrations of soil OC and TN in different soil layers along revegetational succession. Values are means ± 2SD with the sample size $n = 3$. Values followed by a different lower-case letters are significantly different within each soil depth at $P < 0.05$.
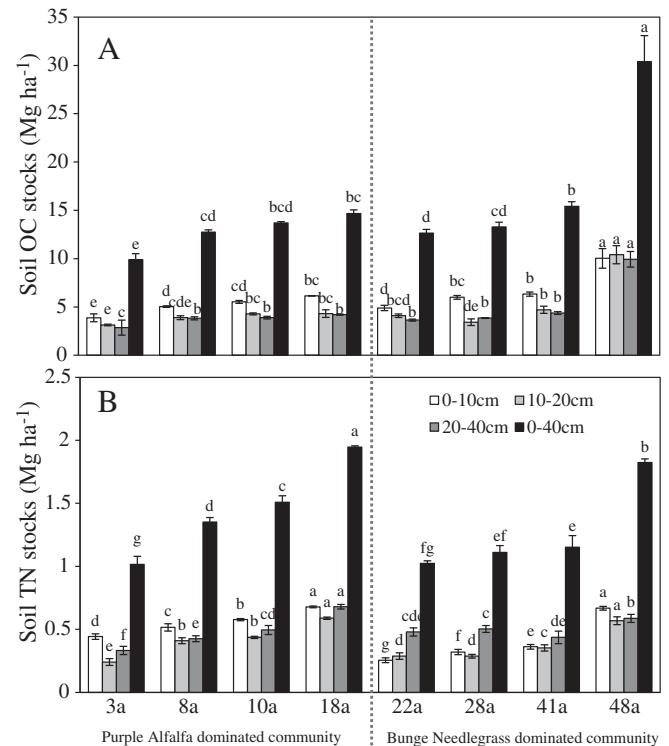

Fig. 5. Changes in stocks of soil OC and TN in different soil layers along revegetational succession. Values are means ± 2SD with the sample size $n = 3$. Values followed by a different lower-case letters are significantly different within each soil depth at $P < 0.05$.

however, STN could be significantly fitted by SWS for the 0–40 ($R^2 = 0.833$; $p < 0.05$) and 0–100 cm ($R^2 = 0.914$; $p < 0.05$) depths.

4. Discussion

4.1. Effects of revegetational succession on soil $\delta^{13}C$ and $\delta^{15}N$

The differences in the response of soil $\delta^{13}C$ and $\delta^{15}N$ to revegetational succession can be ascribed to biotic and abiotic factors (Awiti et al., 2008). The natural abundances of $\delta^{13}C$ and $\delta^{15}N$ in 0–40 cm soil layers varied with vegetational succession in the ALF and BNE communities. The level of $\delta^{13}C$ increased in ALF soils, but decreased in BNE soils with the increase in revegetational age. In general, the enrichment of $\delta^{13}C$ in soils has been attributed to: (i) change in land use and land cover, for instance, conversion of forest to cropland (Awiti et al., 2008) or a C_3 plant community is replaced by a C_4 plant community (Boutton et al., 1998); (ii) decomposition and humification processes associated with isotopic fractionation, since decomposition of organic structure can promote enrichment of ^{13}C in surface soil (Agren et al., 1996); (iii) increases of $\delta^{13}C$ in plant tissues, such as leaves and roots (Balesdent et al., 1993; Ehleringer et al., 2000). In this study, alfalfa and bunge-needlegrass communities are pure C_3 communities during the entire revegetational succession. Furthermore, the accompanying species occurring in the investigated plots are less than 5% of the total number of species, and their combined biomass is less than 10% of the total biomass (data not shown). The influence of the accompanying species on changes of soil $\delta^{13}C$ could therefore be negligible. Reports suggest that stress from drought may increase the $\delta^{13}C$ of plant tissues, especially in arid or semiarid regions (Austin and Vitousek, 1998; Dawson et al., 2002; Swap et al., 2004; Wang et al., 2003). Changes in annual precipitation during the complete succession in the study area were not significant (Fig. 1); however, SWS decreased significantly in the ALF community during succession due to greater evapotranspiration, which considerably exceeded the available precipitation. However, SWS increased after alfalfa was replaced by *S. bungeana* (Fig. 2). In the water-limited systems, water-use efficiency (WUE) also influences the natural abundance of isotopic C in soils (Swap et al., 2004). The exhaustion of SWS may change the isotopic fractionation in plants that relates to the assimilation of C and diffusion of CO_2 (Dawson et al., 2002; Swap et al., 2004). Soil texture, pH, and carbonate content may influence the levels of $\delta^{13}C$ in soil (Stevenson et al., 2005; Xu et al., 2006). Nevertheless, we observed no significant relationships between these variables and $\delta^{13}C$; the influence of soil pH, texture, and carbonate content were therefore be neglected in this study. The changes in abundance of $\delta^{13}C$ with revegetational age are partly similar or greater in the 10–40 cm compared to the 0–10 cm layer, suggesting the largest changes may occur in the deeper soils. This may point to the influence of the carbonates and accumulation of dust at the soil surface where more biochemical reactions occur compared to deeper soils. These reactions may affect the evolution of carbonates and stable-isotopic C along the revegetational succession (Ali and Atekwana, 2011; Nordt et al., 1998). The relationships between $\delta^{13}C$ and SWS along the revegetational succession showed that SWS in the 0–40 and 0–100 cm layers accounted for approximately 70% of the variation in the levels of $\delta^{13}C$ (Table 3). The result is consistent with those observed by Farquhar (1991) and Admundson et al. (1994) who reported that ecosystemic communities adapted to arid environments are expected to have higher abundance of ^{13}C. The different dynamics of soil $\delta^{13}C$ in ALF and BNE communities might be primarily attributed to the replacement of the dominant plant species and the availability of water. When ALF was replaced by BNE, the availability of soil water generally increased (Fig. 2). Alfalfa with a high WUE should have been enriched relative to *S. bungeana*, which has a lower WUE along the revegetational succession.

The abundance of $\delta^{15}N$ in soils of the ALF community increased with vegetational age, which could be due to the symbiotic fixation of N_2 in alfalfa. An increase in soil $\delta^{15}N$ may occur in many ways, such as by increased rates of mineralization, nitrification (Högberg, 1997; Nadelhoffer and Fry, 1994), humification, and decomposition (Liao et al., 2006). Additionally, the enrichment of $\delta^{15}N$ in soils in arid regions has been demonstrated for sites within the Atacama Desert, in South America and the deserts of southern Africa and southwestern USA (Austin and Vitousek, 1998; Evans and Ehleringer, 1993; Swap et al., 2004). Such enrichments suggest difference in biogeochemical processing and cycling of N caused by stress from drought, which produces a more open cycling of N at direr sites, with smaller losses relative to turnover as the humidity increases. Further, the intense periods of microbial activity during initial wet conditions are not sufficiently long to process the entire available N with revegetational succession due to depletion of soil water, so that the isotopic abundance of the residual N gradually increases over time (Swap et al., 2004). The symbiotic fixation of N_2 and the depletion of soil water along revegetational succession could therefore be the main reasons for the enrichment of $\delta^{15}N$ in lands dominated by alfalfa. The $\delta^{15}N$ in BNE soils decreased with revegetational age due to the decreased uptake of ^{15}N by plants (Nadelhoffer and Fry, 1994), since the input of ^{15}N in soils requires the growth of N-fixing plants.

Swap et al. (2004) showed an inverse relationship between humidity and the enrichment of $\delta^{15}N$ in soils under C_3 vegetation. In our study, another explanation is the increased recovery of soil water in the BNE community (Fig. 2): the BNE community is nutrient-limited because wetter ecosystems have a lower availability of nutrients compared to drier ecosystems (Swap et al., 2004). The ALF community along the revegetational succession, with lower SWS, therefore showed an enrichment of $\delta^{15}N$, while the BNE community showed a depletion of $\delta^{15}N$. Like $\delta^{13}C$, $\delta^{15}N$ was linearly related with SWS for the 0–10, 0–20, 0–40, and 0–100 cm depths, and SWS at the 0–40 and 0–100 cm depths accounted for more than 70% of the variance in the relationship (Table 3). These results demonstrate that the dynamics of $\delta^{13}C$ and $\delta^{15}N$ during revegetational succession are related to the storage of soil water in semiarid ecosystems. Therefore, soil isotopic fractionation varied with the revegetational communities, and such differences might be attributed to the changes of plant species, soil water, and nutrient availability.

4.2. Effects of revegetational succession on soil OC and TN

The levels of soil OC and TN increased with revegetational age in the two communities, indicating the accumulation of soil OC and TN by revegetation in the transitional region of erosion by wind and water. The accumulation of nutrients and organic matter in surface soils results from complex interactions between biotic processes moderated by plants and soil biota and abiotic processes driven by environmental processes (Hooper et al., 2000). Altered sizes, composition and dynamics of microbial communities and altered vegetational biomass along the revegetational chronosequence may impact the biogeochemical cycles of soil C and other nutrients, resulting in increases in the storage of soil C (Cleveland et al., 2004; Li et al., 2005).

The conservation of soil and water on sloping lands is an important factor determining the enrichment of OC and N in soils. Erosion by wind and water can cause significant losses of soil OC and nutrients (Jacinthe et al., 2001). Martinea-mena et al. (2002) reported that the magnitude of loss of soil OC was dominated by erosion rather than mineralisation after the initial years of change in land use. Revegetation with grass in the study region, which suffered severe erosion by water during the rainy seasons and by wind on windy days (Tang, 2004), provided an important protection against soil erosion, indicating a potential to sequestrate soil OC and N and to reduce the losses of soil OC and N through runoff and transport of sediments. These results are in agreement with a recent study conducted in the same area (Fu et al., 2010). Additionally, the leaves of grasses are enriched with nutrients from the dust (Wezel et al.,

2000), and the roots are distributed in the surface soil layer, increasing soil organic C and nutrients.

All these processes can contribute to the accumulation of C in the soils. During the transitional stage, however, levels of soil OC and TN decreased significantly due to the reduction in the return of aboveground and belowground biomass to the soil (Table 1). Our results suggest that SOC and STN varied with time along the revegetational succession, which may help to determine whether grassland is a sink or a source of CO_2 during the successional process.

The annual increasing rate of SOC for the 0–40 cm layer during the succession of the ALF community (0.30 Mg ha^{-1} a^{-1}) was lower than that for the BNE community (0.66 Mg ha^{-1} a^{-1}), because alfalfa in the study area is mainly used for feeding stock. The farmers cut the alfalfa one or two times per year. Therefore, the input of new input C into soils is lower in ALF community compared with BNE community, which could be evidenced by the changes of natural abundance of ^{13}C in soils. Generally, the natural abundance of ^{13}C in soil organic matter would increase due to the mineralisation and isotopic fractionation in C_3 plant community. However, the new input of C would decrease the ^{13}C abundance, which makes the ^{13}C in a relatively stable level. We observed an increased ^{13}C with stand age in ALF soils, indicating the decreasing of new input C. However, after replaced by BNE community, ^{13}C decreased to the initial level in ALF soils, indicating increase in new input C in ALF soils.

Furthermore, the availability soil water increased after the ALF community was replaced by the BNE community, which may accelerate the decomposition of organic material (Austin, 2002; Fang et al., 2005), resulting in more input of soil organic C and nutrients. Despite the depletion of nutrients in the ALF community by harvesting, harvesting can also increase the storage of soil OC by the accumulation of root-derived organic matter and the reduction of soil and water erosion. The rates of increase of OC in surface soils (0–10 cm) for ALF communities (0.14 Mg ha^{-1} a^{-1}) and BNE communities (0.20 Mg ha^{-1} a^{-1}) were relatively smaller compared to the earlier results (0.25 Mg ha^{-1} a^{-1}) over 14 years in the southern part of China (Zhang and Peng, 2006). The lower rates could be ascribed to the differences in climatic conditions and plant species between the regions (Wei et al., 2009). At our study site, annual temperature and precipitation are lower than in southern China, and the study site had less input of soil organic C and nutrients, fixation of N by alfalfa, and release of N by the decomposition of litter.

The relationships between STN and SWS along the revegetational succession showed that SWS at the 0–40 and 0–100 cm depths accounted for approximately 83.3% and 91.4% of the variation of STN, respectively, but no significant relationships were observed between STN and revegetational age. In contrast, revegetational age accounted for 74.8% of the variation in SOC, but no significant relationships between SOC and SWS for the four soil layers were found. The prediction of SOC is therefore more closely related to revegetational age, while STN is also limited by more-complicated factors, e.g. plant species and local conditions. In general, revegetation effectively sequestered N in the transitional area over a long timescale by reducing soil erosion as well as soil OC. Vegetational restoration resulted in an effective control of soil OC and loss of nutrients and subsequent amelioration of the eco-environment. Knowledge of changes in soil OC and TN along revegetational successions may thus contribute to a mechanistic understanding and projection of C and N cycles in response to the evolution of ecosystemic communities in semiarid regions.

5. Conclusions

This study indicates different dynamics of δ^{13}C/δ^{15}N within different revegetational stages during a restored grassland succession. A pronounced inverse relationship was observed between soil water storage (0–40 or 0–100 cm depths) and the natural abundance of δ^{13}C/δ^{15}N under C_3 vegetation in the northern Loess Plateau. This relationship may be of value to scientists modelling nutrient cycling within different ages of vegetational succession. The study also showed an increase in stocks of OC and TN with revegetational age, increasing for the two stages of community succession by controlling losses of soil and water and increasing inputs of organic matter. The rates of increase, however, vary with the different stages of succession due to changes in plant species, soil conditions, and microclimate. This information may thus be helpful in improving model simulations of C and N and projections of future dynamics of grassland ecosystems as a sink or source for CO_2 during successional processes. Our research reinforces the use of stable-isotopic techniques to study the dynamics of C and N in terrestrial systems and demonstrates that vegetational restoration may have the potential to sequester C and N in the northern Loess Plateau of China.

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