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Impacts of afforestation on plant diversity, soil properties, and soil organic carbon storage in a semi-arid grassland of northwestern China

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ABSTRACT

Grassland afforestation, occurring in many parts of the world, can modify the nature and transformations of soil organic carbon and associated soil properties, which in turn can affect plant diversity and ecosystem function. Afforestation area has grown rapidly over the last few decades in the semi-arid grasslands of the Qilian Mountains in northwestern China in an effort to restore mountain vegetation. However, ecological consequences of this land use change are poorly known. We investigated the effects of grassland afforestation on plant diversity, soil properties, and soil organic carbon and nitrogen storage at the soil depth of 0–70 cm. Our results showed that afforestation decreased percent cover and aboveground biomass, and increased plant diversity of herbaceous community. Afforestation also decreased soil bulk density and pH, and increased soil water content. Generally, afforestation favored an increase in soil organic carbon, total nitrogen, and organic carbon storage, and resulted in a significant increase in total phosphorus in the surface soil (0–5 cm), although a slight decrease ($P > 0.05$) was observed in the subsoil. In addition, afforestation significantly increased soil C:N ratio in the upper soil. Results of this study demonstrate the potential for afforestation to increase soil organic carbon and nitrogen storage in semi-arid grasslands of Qilian Mountains. This has important implications for C sequestration in this area.

1. Introduction

Land-use and land-cover changes have attracted increasing scientific interest in the past decades in relation to their contribution to global change and potential impacts on carbon (C) dioxide sequestration, soil quality, ecosystem function, and long-term sustainability (DeFries et al., 2004; Laganiere et al., 2010; Sauer et al., 2012; Deng et al., 2014a, 2014b; Bárcena et al., 2014; Deng and Shangguan, 2016). Grassland afforestation, mainly with coniferous trees, has expanded rapidly in the last decades (Otto and Simpson, 2005; Rudel et al., 2005; Chen et al., 2008; Wei et al., 2009; Hewitt et al., 2012; Vassallo et al., 2013); the reasons for this land-use change included an increasing demand for timber production, and a growing need to control soil erosion, restore vegetation, and mitigate CO_2 emissions (Chen et al., 2007; Chen et al., 2008; Hewitt et al., 2012; Vassallo et al., 2013). Rapid expansion of plantation areas highlights the need to understand ecological consequences of this land-use change for the maintenance of long-term nutrient availability, sustainable productivity, and C sequestration (Berthrong et al., 2012).

Conversion of native grasslands to forest plantations modifies primary production, ecosystem structure (Vassallo et al., 2013), the quantity and quality of litter inputs, root turnover (including exudates), and microclimatic conditions such as moisture and temperature (Nosetto et al., 2005; Laganiere et al., 2010). Shifts in plant species can lead to changes in soil properties and C stocks, which in turn have the potential to affect biomass production and ecosystem function (Jackson et al., 2002; Foster et al., 2003; Jobbágy and Jackson, 2003; Chen et al., 2008; Wei et al., 2009; Wang et al., 2016). Moreover, the "nutrient pumping" effect, observed following the conversion of grasslands to forests, results in the redistribution of nutrients, with decreasing concentrations at intermediate depths and increasing at the soil surface (Jobbágy and Jackson, 2004; Farley and Kelly, 2004).

Current understanding of ecological consequences of grassland afforestation includes plot-scale changes in plant diversity and community structure, soil organic carbon (SOC) and total nitrogen (TN) stocks, soil moisture, acidity, soil nutrient status, and microbial community structure (Jobbágy and Jackson, 2003; Farley and Kelly, 2004; Alrababah et al., 2007; Chen et al., 2008; Berthrong et al., 2009a; Berthrong et al., 2009b; Wei et al., 2009; Berthrong et al., 2012; Hewitt et al., 2012; Deng et al., 2016). However, extensive uncertainty remains at region scales, especially for arid and semi-arid regions (Hu et al., 2008; Zhang et al., 2013), and the results vary greatly with climate, forest age and type, and soil type and management practices (Jobbágy and Jackson, 2003; Farley and Kelly, 2004; Chen et al., 2008; Hu et al., 2008; Berthrong et al., 2009b; Wei et al., 2009; Wei et al., 2010; Deng et al., 2014a, 2014b Bárcena et al., 2014). For example, afforestation in a temperate grassland of Inner Mongolia resulted in an initial loss of

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total soil nitrogen (TN) during the first few years, but a gradual recovery with increasing stand age (Hu et al., 2008). Similar trends for soil N stocks were observed in Ecuadorian grasslands (Farley and Kelly, 2004). However, a global meta-analysis showed that SOC were reduced with afforestation of grasslands but not significantly ($P > 0.05$) (Shi et al., 2013). Jackson et al. (2002) found a significant and negative relationship between precipitation and changes in SOC and TN when grasslands were invaded by woody vegetation, with drier sites gaining, and wetter sites losing both SOC and TN. Soils frequently become more acidic with afforestation (Jobbágy and Jackson, 2003; Farley and Kelly, 2004; Berthrong et al., 2009a); however, detailed studies were conducted mainly in New Zealand, America and Australia (Berthrong et al., 2009a), and these effects remain uncertain in other regions. Additionally, many studies have focused on the effects of grassland afforestation in the topsoil (e.g., 0–20 cm) due to historical practices and the ease of sampling, and the responses in deep soil layers are still poorly understood (Chang et al., 2012). However, increasing evidence suggests that the SOC contents in the subsoil are also sensitive to changes in land use and management (Chang et al., 2012; Shi et al., 2013; Deng et al., 2014a, 2014b). Thus, data are needed from different regions and from the subsoil to increase the understanding of the ecological consequences of grassland afforestation.

The Qilian Mountains, located in the northern margin of the Tibetan Plateau, are the source of several key inland rivers in northwestern China, including the Heihe, Shiyang, and Shule. The mountains were designated as a National Nature Reserve in 1988 for their key role in maintaining regional ecological security. Forests, dominated by Qinghai spruce (*Picea crassifolia*), and grasslands are the main landscape types in this area (Wang et al., 2001). However, forest cover decreased from 22.4% in 1949 to 12.4% during the 1990s due to deforestation associated with increasing demand for timber production, and with global warming (Wang and Cheng, 1999). Loss of forest cover affected hydrological processes, and had important consequences for sustainable development in the region (Wang and Cheng, 1999; He et al., 2012). Since the 1970s, the area of afforestation has been increasing in an effort to restore mountain vegetation, and many semi-arid grasslands were converted to *P. crassifolia* plantation forests (He et al., 2012). Although this land use has grown rapidly over the past four decades, little is known about the effects of change in vegetation cover on plant diversity, soil properties, and soil C stocks in these ecosystems. Thus, the objectives of the present study were to investigate the effects of grassland afforestation on: (1) plant composition and diversity of herbaceous community; (2) soil properties; and (3) soil C and N storage.

2. Methods

2.1. Study area

The study site was located in the Dahuang Mountain Forest Reserve (100°22′E, 38°43′N, 2919 m a.s.l.) in the Qilian Mountains. The area is situated approximately 45 km southeast of Shandan County, Gansu Province, in northwestern China. The site has a semiarid and cold temperate climate, with a mean annual temperature (MAT) of about 1 °C and mean annual precipitation (MAP) of about 400 mm, falling mainly between July and September. The main parent material is calcareous rock, which is overlaid by a relatively thin soil layer (<1 m deep) (Jiang et al., 2013). Native vegetation patterns are closely related to topographic aspects, and represent a mosaic of grassland, forest, and small areas of scrubland. Forests, dominated by the *P. crassifolia*, are distributed on shaded, north-facing slopes; grasslands are mainly found on sunny, south-facing, and semi-shaded, east- or west-facing slopes. Since the 1970s, most of the grasslands on east-facing and west-facing slopes have been converted into *P. crassifolia* plantation forests, and the forest cover has increased from 24.6% to 52.8%. Differences in topographical aspects and vegetation patterns induced divergent soils properties. Soils are classified according to the FAO classification system as Haplic Kastanozems on sunny and semi-shaded slopes, and Haplic Phaeozems on shaded slopes (IUSS Working Group. WRB, 2014).

2.2. Experimental design, soil sampling, and vegetation survey

In early August 2014, two study sites were selected, one on west-facing and one on east-facing slope (Table 1). At each site, native grasslands and *P. crassifolia* plantation forests occurred directly adjacent to each other. Site conditions (e.g. topography and vegetation patterns) and management practices of the selected grasslands and plantations were typical in the region. Three replicate sample plots of 30×30 m^2 were randomly located in each grassland and forest for a total of 15 sample plots (Table 1). Geographic coordinates and elevations of each plot were obtained using a global positioning system (GPS) with differential correction.

Within each plot, five randomly-located soil profiles were excavated (after removing the surface litter layer), and soil samples were collected at depths of 0–5, 5–15, 15–30, 30–50, and 50–70 cm. In addition, undisturbed soil cores were obtained from each layer for the measurements of bulk density using a standard container with the volume of 100 cm^3. In forest plots, tree height, diameter at breast height (DBH), tree crown area, and the number of trees per plot were measured. We measured canopy height at 25 points in each plot, using a telescopic measuring rod (5 cm precision). We also measured leaf area index (LAI) using CI-110 with a fisheye lens (Juarez et al., 2009) at 1.5 m above ground every 3 m along the diagonal of each plot to evaluate the light conditions. Ten quadrats of 1×1 m^2 were randomly located to investigate species composition and percent cover of the herbaceous community (main understory vegetation); subsequently, the herbaceous layer was harvested at 2–3 cm above ground and oven-dried at 65 °C to a constant weight to determine aboveground biomass of understory vegetation. Vegetation cover for the herbaceous layer was visually estimated by two experienced observers. For grassland plots, species composition, percent cover, and aboveground biomass of the plant community were also investigated separately in ten quadrats of 1×1 m^2.

Importance values (IV, Eq. (1)) for herbs were calculated using the following equations (Zhao et al., 2009):

$$IVi = \frac{(RAi + RCi + RFi + RBi + RHi)}{5} \quad (1)$$

where *IVi*, *RAi*, *RCi*, *RFi*, *RBi* and *RHi* were the importance values (%), relative density (%), relative coverage (%), relative frequency (%), relative biomass (%), and relative height (%) of species *i*, respectively.

Plant diversity analysis was conducted for the herbaceous plant community. Species richness index (R), Shannon index (H), and Pielou evenness index (E) were calculated as biodiversity indicators at the quadrat level. Species richness (R) was calculated as the number of species identified in each quadrat, while Shannon index (H, Eq. (2)) and Pielou evenness index (E, Eq. (3)) were calculated using the following equations (Spellerberg and Fedor, 2003; Deng et al., 2014a, 2014b):

$$H = -\sum_{i=1}^{n} (Pi \ln Pi) \quad (2)$$

$$E = \frac{H}{\ln S} \quad (3)$$

Where *S* is the total species numbers of the herbaceous community, and *Pi* is the proportional density of species *i* (number of individuals of species *i* divided by the total number of individuals of all species).

Table 1
Geographical and vegetation characteristics for the *Picea crassifolia* plantation forest - adjacent grassland plot pairs. Values (±SE) followed by different lower-case letters within columns are significantly different at $P < 0.05$.

Aspect (°)	Altitude (m)	Slope (°)	Land use	Plot number	Plantation age (year)	Stand density (trees ha^{-1})	Height (m)	DBH (cm)	Crown area (m^2)	LAI	Herbaceous Total cover (%)	Herbaceous Aboveground biomass (g m^{-2})
SW70	2600	38–43	Grassland	3	–	–	–	–	–	–	90.5 ± 2.84a	126.54 ± 11.71a
			Forest	3	33	2833 ± 189	4.2 ± 0.59	6.3 ± 0.51	3.61 ± 0.24	1.15 ± 0.28	72.5 ± 4.86b	80.10 ± 7.94b
			Forest	3	45	2967 ± 38	6.3 ± 0.23	8.4 ± 0.25	5.51 ± 0.11	1.76 ± 0.31	46.5 ± 5.80c	44.19 ± 5.31c
NE87	2635	40–42	Grassland	3	–	–	–	–	–	–	93.5 ± 3.37a	108.75 ± 16.06a
			Forest	3	35	2917 ± 101	4.7 ± 0.53	6.9 ± 0.34	4.52 ± 0.17	1.43 ± 0.32	67.5 ± 6.35b	73.80 ± 13.43b

2.3. Soil analysis

Soil samples were air-dried, and then passed through a 2 mm soil sieve; the weight and volume of gravels (>2 mm) were measured. Soil water content before air drying was measured gravimetrically and expressed as a percentage of soil water to dry soil weight (oven-dried at 105 °C to a constant weight). Soil bulk density was calculated as the ratio of weight of undisturbed cores, oven-dried at 105 °C to a constant weight, to the container volume. Soil texture was determined by the wet sieve method (Chaudhari et al., 2008). Soil pH was determined using the method of acidity agent (soil-water ratio of 1:5) (PHS-3C pH acidometer, China) (Deng et al., 2014a, 2014b). Subsamples of soils were analyzed for SOC, TN and TP. Soils were finely ground to pass through a 0.10 mm sieve. SOC was determined by the $K_2Cr_2O_7$-H_2SO_4 oxidation method of Walkley-Black (Nelson et al., 1982). TN was measured with the Kjeldahl method (Bremner and Mulvaney, 1982). TP was determined colorimetrically after wet digestion with $H_2SO_4 + HClO_4$ (Parkinson and Allen, 1975).

2.4. Calculation of soil C and N storage

We calculated SOC stocks (Eq. (4)) and TN stocks (Eq. (5)) for each soil depth using the following equations (Rytter, 2012; Zhang and Zhao, 2015):

$$SOCD = \sum_{i=1}^{n} Ti \times BDi \times SOCi \times \frac{(1-Pi)}{100} \tag{4}$$

$$TND = \sum_{i=1}^{n} Ti \times BDi \times TNi \times \frac{(1-Pi)}{100} \tag{5}$$

where *SOCD* and *TND* were the SOC (kg m^{-2}) and TN stocks (kg m^{-2}), respectively, of a soil profile; *n* was the number of soil layers considered; *SOCi* and *TNi* were the SOC (g kg^{-1}) and TN concentrations (g kg^{-1}), respectively, at layer *i*; *Ti*, *BDi* and *Pi* were the soil thickness (cm), bulk density (g cm^{-3}), and volumetric percentage (%) of coarse fragments (>2 mm), respectively, at layer *i*.

2.5. Statistical analysis

All data were expressed as mean ± standard error. We used one-way analysis-of-variance (ANOVA) to examine differences in vegetation cover, aboveground biomass, plant diversity of herbaceous community, soil properties, and SOC and TN stocks between *P. crassifolia* plantation forests and adjacent grasslands. As the values of vegetation cover followed an abnormal distribution, the data was normalized by logit transformation before adopting ANOVA. The least-significant-difference test (LSD) was performed when significant differences were detected by ANOVA for the west-facing slope. Significant differences were evaluated at the 0.05 level. All statistical analyses were performed using the software program SPSS, ver. 17.0 (SPSS Inc., Chicago, IL, USA).

3. Results

3.1. Comparison of herbaceous communities in forest and adjacent grassland

Afforestation resulted in a significant decrease ($p < 0.05$) in percent cover and aboveground biomass, and a significant increase ($p < 0.05$) in species number of the herbaceous community in comparison to the adjacent grasslands on both east- and west-facing slopes (Table 1; Fig. 1). On the west-facing slope, cover and aboveground biomass decreased, while species number increased with plantation development. Meanwhile, the Shannon and Pielou evenness indices for the herbaceous community were also higher after afforestation, especially on the east-facing slope ($p < 0.05$) (Fig. 1).

3.2. Changes in soil physical and chemical properties

Generally, afforested grasslands were associated with a reduction in soil pH and bulk density across the sampled soil depth (0–70 cm) on both east- and west-facing slopes (Table 2). On the west-facing slope, soil pH and bulk density decreased with forest age, and a significant ($p < 0.05$) decrease was observed in plots 45 years after afforestation; on the east-facing slope, a significant ($p < 0.05$) decrease in soil bulk density was also observed after 35 years of afforestation. Afforestation resulted in higher soil water content. On the west-facing slope, 45-year-old afforestation resulted in significantly ($p < 0.05$) increased soil water content in the 0–30 cm soil layer; a significant ($p < 0.05$) increase was also observed in the 0–15 cm soil layer in the 35-year-old afforestation on the east-facing slope. However, no significant ($p > 0.05$) difference was observed in soil texture following grassland afforestation (data not shown).

Afforestation also had significant effects on SOC and TN (Table 2). SOC concentrations in plantation forests were significantly ($p < 0.05$) greater than in adjacent grasslands across the sampled soil depth (0–70 cm) on both east- and west-facing slopes, and SOC concentration increased with plantation development on the west-facing slope. TN exhibited trends similar to those of SOC. On the west-facing slope, TN concentration increased with forest age, and 45-years after afforestation, a significant ($p < 0.05$) increase in TN was observed in all, except in the 50–70 cm soil layer. On the east-facing slope, a significant ($p < 0.05$) increase was observed across the sampled soil depth (0–70 cm) after 35 years of afforestation. Plantation forests had greater TP concentration in comparison to the adjacent grasslands in the topsoil (0–15 cm), and a significant ($p < 0.05$) increase was observed in the 0–5 cm surface soil layer in the 45-year-old afforestation on the west-facing and east-facing slopes. A non-significant ($p > 0.05$) decrease in TP concentration was observed in the subsoil (at 30–70 cm soil depth on the west-facing slope and at 15–70 cm soil depth on the east-facing slope). In addition, afforestation also induced changes in soil C:N ratio, especially in the upper soil. On the west-facing slope, afforestation significantly ($p < 0.05$) increased soil C:N ratio except for the 30–50 cm soil layer, and C:N ratios were not significantly ($p > 0.05$) different between afforestation of 33 and 45 years of age. On the east-facing

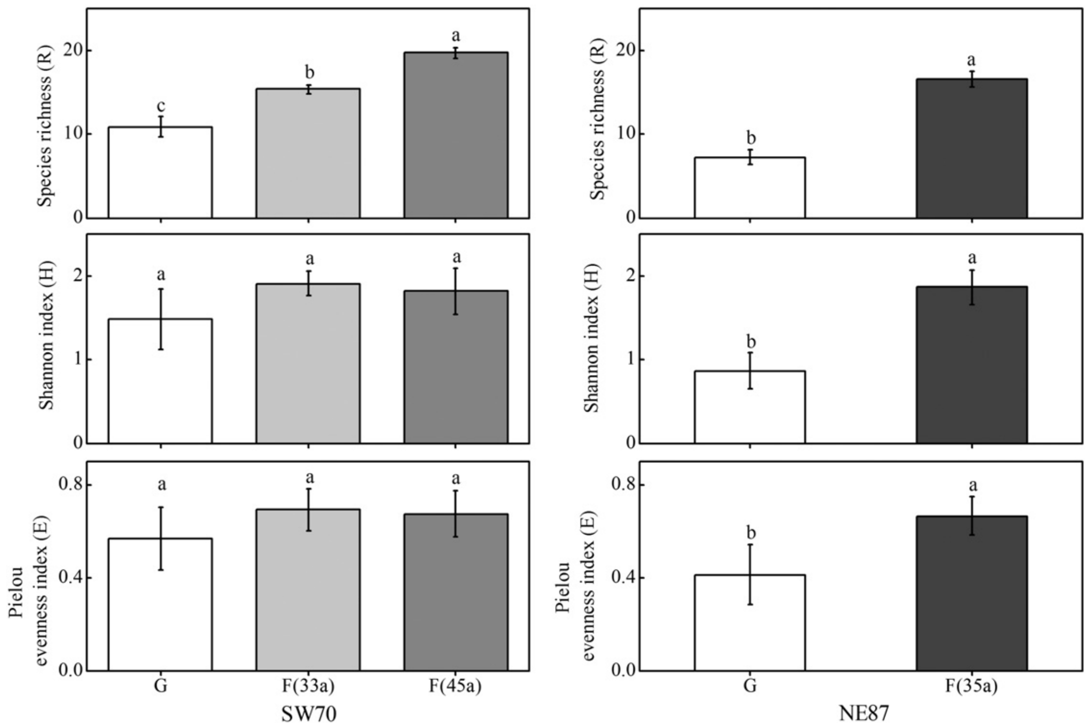

Fig. 1. Comparison of species richness index (R), Shannon index (H) and Pielou evenness index (E) for the herbaceous community between the paired *Picea crassifolia* plantation forests and adjacent grasslands. Different lower-case letters above the bars indicate significant differences at *P* < *0.05*. F(33a), F(45a) and G represent afforestation for 33 and 45 years, and the adjacent grasslands on the west-facing slope. F(35a) and G represent afforestation for 35 years and adjacent grasslands on the east-facing slope.

Table 2
Comparison of soil organic carbon, soil total nitrogen, soil total phosphorus, soil carbon/nitrogen (C:N) ratios, pH value, soil water content and soil bulk density between the paired *Picea crassifolia* plantation forests and adjacent grasslands at different depths. Values (±SE) followed by different lower-case letters within rows are significantly different at *P* < *0.05*.

Soil properties	Depth (cm)	SW70			NE87	
		Grassland	Afforestation (33a)	Afforestation (45a)	Grassland	Afforestation (35a)
Soil organic carbon ($g\ kg^{-1}$)	0–5	33.32 ± 2.98c	42.70 ± 1.83b	50.29 ± 1.89a	36.58 ± 2.91b	58.14 ± 3.70a
	5–15	31.00 ± 2.24c	40.08 ± 1.48b	45.97 ± 1.19a	33.04 ± 3.57b	48.71 ± 2.47a
	15–30	29.09 ± 1.01c	37.69 ± 1.56b	41.37 ± 1.23a	31.09 ± 2.42b	45.70 ± 1.50a
	30–50	27.80 ± 1.17b	34.74 ± 2.49a	35.96 ± 2.50a	26.85 ± 1.65b	37.19 ± 1.52a
	50–70	25.42 ± 0.70b	29.63 ± 1.41a	30.39 ± 2.09a	23.71 ± 2.12b	29.16 ± 1.40a
Soil total nitrogen ($g\ kg^{-1}$)	0–5	2.63 ± 0.14c	3.07 ± 0.11b	3.43 ± 0.07a	3.02 ± 0.11b	4.23 ± 0.21a
	5–15	2.48 ± 0.08c	2.94 ± 0.10b	3.21 ± 0.05a	2.88 ± 0.10b	3.90 ± 0.14a
	15–30	2.44 ± 0.19b	2.74 ± 0.13ab	3.02 ± 0.24a	2.68 ± 0.13b	3.75 ± 0.28a
	30–50	2.31 ± 0.10b	2.63 ± 0.11a	2.70 ± 0.15a	2.36 ± 0.10b	3.15 ± 0.14a
	50–70	2.13 ± 0.13a	2.28 ± 0.20a	2.26 ± 0.16a	2.08 ± 0.07b	2.44 ± 0.10a
Soil total phosphorus ($g\ kg^{-1}$)	0–5	0.77 ± 0.02b	0.80 ± 0.03ab	0.85 ± 0.03a	0.78 ± 0.02b	0.84 ± 0.02a
	5–15	0.75 ± 0.04a	0.76 ± 0.05a	0.80 ± 0.04a	0.74 ± 0.04a	0.77 ± 0.03a
	15–30	0.73 ± 0.05a	0.75 ± 0.04a	0.72 ± 0.03a	0.72 ± 0.02a	0.70 ± 0.04a
	30–50	0.69 ± 0.02a	0.67 ± 0.03a	0.64 ± 0.02a	0.65 ± 0.04a	0.64 ± 0.02a
	50–70	0.62 ± 0.03a	0.59 ± 0.05a	0.56 ± 0.03a	0.56 ± 0.03a	0.53 ± 0.05a
C/N	0–5	12.67 ± 0.62b	13.93 ± 0.56ab	14.65 ± 0.47a	12.11 ± 0.15b	13.74 ± 0.26a
	5–15	12.48 ± 0.07b	13.66 ± 0.70a	14.32 ± 0.15a	11.47 ± 0.14b	12.51 ± 0.37a
	15–30	11.94 ± 0.28b	13.77 ± 0.69a	13.70 ± 0.80a	11.63 ± 0.42a	12.18 ± 0.26a
	30–50	12.04 ± 0.34a	12.86 ± 0.59a	13.33 ± 0.59a	11.37 ± 0.57a	11.81 ± 0.18a
	50–70	11.96 ± 0.63b	12.98 ± 0.21a	13.44 ± 0.15a	11.42 ± 0.78a	11.97 ± 0.19a
pH value	0–5	8.62 ± 0.08a	8.42 ± 0.14ab	8.19 ± 0.12b	8.49 ± 0.11a	8.40 ± 0.16a
	5–15	8.60 ± 0.12a	8.49 ± 0.21ab	8.35 ± 0.07b	8.51 ± 0.06a	8.31 ± 0.10b
	15–30	8.60 ± 0.05a	8.53 ± 0.06ab	8.42 ± 0.06b	8.53 ± 0.14a	8.42 ± 0.06a
	30–50	8.73 ± 0.11a	8.61 ± 0.08ab	8.47 ± 0.05b	8.59 ± 0.10a	8.49 ± 0.23a
	50–70	8.77 ± 0.05a	8.68 ± 0.14ab	8.61 ± 0.09b	8.70 ± 0.13a	8.59 ± 0.07a
Soil water content (%)	0–5	13.99 ± 0.71c	18.61 ± 0.86b	24.08 ± 1.97a	17.87 ± 0.65b	25.81 ± 1.49a
	5–15	15.79 ± 1.30b	19.79 ± 0.94ab	22.59 ± 3.87a	19.71 ± 0.64b	23.42 ± 1.85a
	15–30	16.75 ± 3.36b	21.31 ± 0.71ab	23.16 ± 2.56a	22.00 ± 1.27a	24.84 ± 1.35a
	30–50	18.11 ± 1.87a	20.98 ± 1.78a	21.77 ± 2.46a	20.90 ± 0.95a	22.47 ± 1.41a
	50–70	15.94 ± 2.25a	16.97 ± 0.67a	16.63 ± 0.72a	17.69 ± 1.00a	19.17 ± 1.71a
Soil bulk density ($g\ cm^{-3}$)	0–5	1.08 ± 0.03a	0.95 ± 0.06b	0.83 ± 0.02c	1.05 ± 0.05	0.78 ± 0.06b
	5–15	1.09 ± 0.04a	0.98 ± 0.02b	0.90 ± 0.08b	1.04 ± 0.06a	0.90 ± 0.03b
	15–30	1.11 ± 0.07a	1.01 ± 0.05ab	0.94 ± 0.05b	1.09 ± 0.08a	0.89 ± 0.05b
	30–50	1.06 ± 0.04a	0.98 ± 0.07ab	0.93 ± 0.04b	1.07 ± 0.04a	0.92 ± 0.05b
	50–70	1.07 ± 0.06a	1.02 ± 0.05ab	0.95 ± 0.03b	1.17 ± 0.07a	0.95 ± 0.08b

slope, a significant ($p < 0.05$) increase in soil C:N was observed in the 0–15 cm soil layer after afforestation.

3.3. Changes in soil organic carbon and nitrogen storage

Afforestation resulted in a significant ($p < 0.05$) increase in SOC stocks on both east- and west-facing slopes, compared to the adjacent grasslands (Fig. 2). On the west-facing slope, SOC stocks tended to increase with stand development, however, no significant ($p > 0.05$) difference was observed between the 33 and 45-year-old afforestation plots.

In contrast to the cumulative SOC storage, TN stocks did not differ significantly ($p > 0.05$) between plantation forests and adjacent grasslands on the west-facing slope (Fig. 2). On the east-facing slope, afforestation significantly ($p < 0.05$) improved TN stocks at 0–30, 0–50, and 0–70 cm soil depths, while no significant ($p > 0.05$) increase was observed at the 0–5 and 0–15 cm soil depths (Fig. 2).

4. Discussion

4.1. Effect of afforestation on the herbaceous community

Alrababah et al. (2007) observed that afforestation in semi-arid Mediterranean grasslands significantly decreased vegetative cover and diversity, and that vegetative cover was very low to completely absent under a dense tree cover. A global meta-analysis of the effects of afforestation on grassland biodiversity also showed that conversion of natural and semi-natural grasslands to forests resulted in a decrease in species richness and diversity (Bremer and Farley, 2010). The decline in plant diversity and richness with grassland afforestation has been attributed to several factors including the exclusion of shade-intolerant native species by plantation canopy cover, allelopathy, and the physical barrier of litter (particularly pine litter) to germination (Alrababah et al., 2007; Buscardo et al., 2008; Bremer and Farley, 2010). We found in this study that afforestation had significant and negative impacts on the cover and aboveground biomass of the herbaceous community (Table 1), confirming the results of previous research. However, we also observed a marked increase in species richness in afforested grasslands, and the Shannon and Pielou evenness indices were also higher after afforestation. Chiarucci and De Dominicis (1995) observed similar trends following afforestation of natural grasslands in Italy, and they attributed the increase in total species richness to the expansion of generalist or exotic species, such as those that prefer shady environments. In the present study, we also found that the importance values of shade-intolerant native species, such as *Echinocereus cylindricus* and *Achnatherum splendens*, decreased with plantation development, but they did not disappear; at the same time, the shade-tolerant species, such as *Carex vulpina* and *Urtica triangularis* expanded with plantation development (Appendix A). The result of these successional processes would have resulted in the increase in overall species richness and diversity.

4.2. Effects of afforestation on soil organic carbon and nitrogen storage

A net SOC and TN gain has been generally observed following afforestation of SOC-depleted systems, such as croplands, at global, and at regional scales in temperate zones (Chen et al., 2007; Laganiere et al., 2010; Deng et al., 2014a, 2014b; Bárcena et al., 2014). However, extensive uncertainty remained regarding the effects of afforestation of more SOC-rich systems such as grasslands (Guo and Gifford, 2002; Jackson et al., 2002; Farley and Kelly, 2004; Berthrong et al., 2009b; Hu et al., 2008; Shi et al., 2013). We found that afforestation significantly improved SOC stocks in semi-arid grasslands of the Qilian Mountains. Our results confirmed those of Jackson et al. (2002), who found that

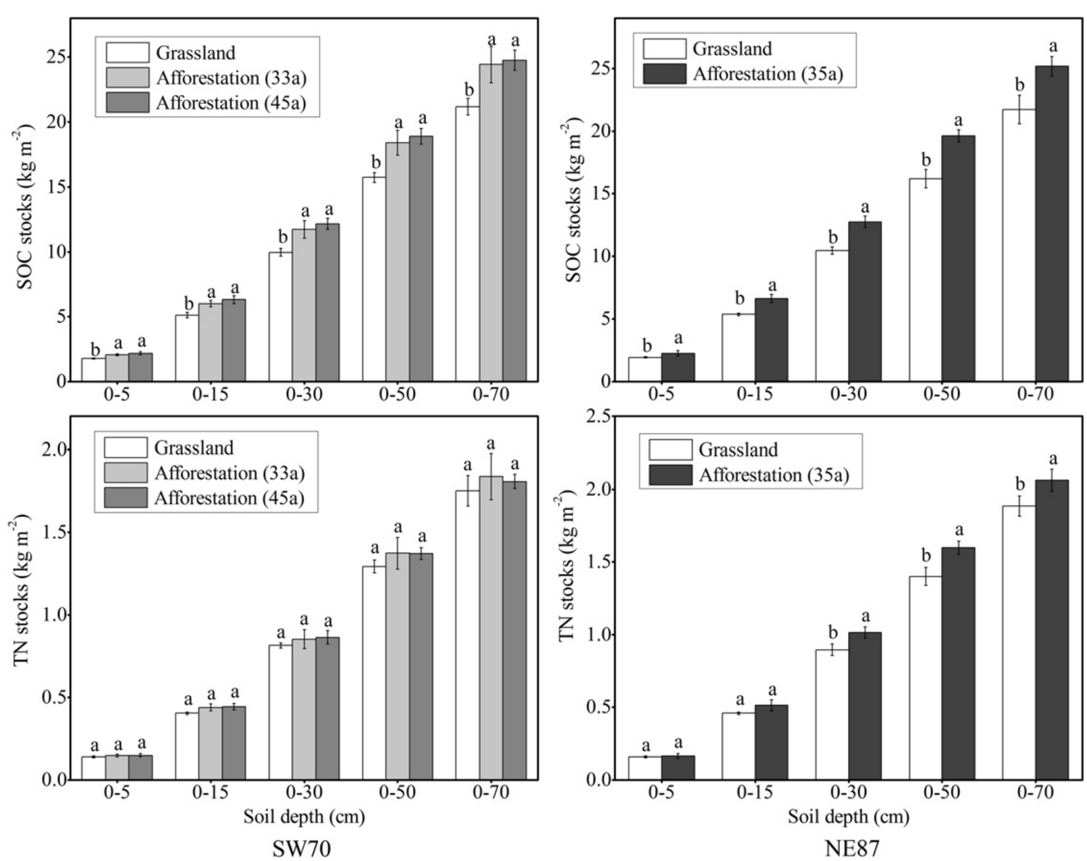

Fig. 2. Comparison of soil organic carbon (SOC) and soil total nitrogen (TN) stocks between the paired *Picea crassifolia* plantation forests and adjacent grasslands. Different lower-case letters above the bars indicate significant differences at $P < 0.05$.

SOC and TN increased at drier sites and decreased at wetter sites in afforested grasslands, and of Berthrong et al. (2009b), who reported similar results along a precipitation gradient (650 to 1450 mm $year^{-1}$). Results of a modeling study by Kirschbaum et al. (2008) suggested that the different response of SOC to afforestation for different precipitation levels was tied to the alterations in the nitrogen cycle; the model indicated that soils in drier regions stored C through increases in soil C:N, while in more humid areas, increased decomposition and N losses through leaching lead to C losses. In our study, the increase in SOC was accompanied by an increase in TN content and C:N, supporting model results to some extent.

The dynamics of C stored in soils depend on the balance between inputs, primarily from plant leaf and root detritus, and outputs through decomposition (Davidson and Janssens, 2006). Litter of coniferous trees, with higher amounts of secondary compounds such as lignin and polyphenols, decomposes more slowly than that of grass, which typically has lower phenolic and lignin concentrations (Corbeels et al., 2003; Berthrong et al., 2009b). Thus, increased SOC stocks in afforested grasslands may be partially explained by the reduced decomposition rates of conifer litter. Furthermore, an initial loss in soil C stocks was observed during the initial 4–5 years after grassland afforestation (Zhang et al., 2010; Deng et al., 2014a, 2014b), due to low litterfall production of young forests and accelerated decomposition rates resulting from mechanized site preparation (Aguilar et al., 1988; Wei et al., 2009). However, accumulation of soil C was observed within 20–30 years after afforestation and was attributed to increasing annual inputs through net primary production with stand development (Davis and Condron, 2002; Thuille and Schulze, 2006; Hu et al., 2008; Berthrong et al., 2012). Similar patterns were observed for soil N stocks in Ecuadorian grasslands (Farley and Kelly, 2004). In our study, accumulation of an organic layer was observed in afforested grasslands, thus the larger annual C inputs through litter production may have contributed to the increase in SOC stocks.

4.3. Effect of afforestation on soil properties

Results of this study indicated that grassland afforestation altered soil physical and chemical properties in both top- and subsoil. In addition to higher SOC and TN contents, and higher soil moisture and TP, lower soil pH and bulk density were observed in afforested grasslands. Our results provided evidence that soil properties in the subsoil were also sensitive to afforestation in semi-arid grasslands, and underscored the necessity to include subsoil when assessing ecological consequences of grassland afforestation.

A decrease in soil pH has been observed following the conversion of grasslands to forests (Jobbágy and Jackson, 2003; Farley and Kelly, 2004; Berthrong et al., 2009a), and we observed a similar pattern in our study. Several mechanisms have been proposed to explain acidification following the afforestation of grasslands, including: (1) increases in organic acid inputs from coniferous trees, such as acidic litter, canopy leachates, and decomposition products, and (2) accumulation of cations in tree biomass contributing to acidification in the rooting zone (Ugolini et al., 1988; Jobbágy and Jackson, 2003; Farley and Kelly, 2004). Vertical patterns of acidification in the soil profile can provide an indication of the dominant mechanism (Jobbágy and Jackson, 2003; Farley and Kelly, 2004). Thus, if acidification is present in the surface soil, then organic acid inputs are the likely drivers; however, if a pattern of maximum acidification is present in the subsoil, then acidification is driven by the accumulation of cations in tree biomass (Farley and Kelly, 2004). In our study, soil pH tended to be lower in afforested grasslands in both topsoil and subsoil, and a significant decrease was observed in plots 45 years after afforestation. These results suggest that the decline in pH may be linked to both mechanisms - organic acid inputs and cation accumulation in the biomass.

Afforestation in our study resulted in an increase in soil water content, especially in the topsoil (Table 2). Generally, pine roots tend to be distributed in deeper soil horizons than grass roots (Farley and Kelly, 2004). We found that grass roots were distributed in the 0–30 cm soil layer, while those of *P. crassifolia* were present mainly below 20–30 cm. We concluded that the significant increase in soil water content that we observed in the topsoil may be explained by the decrease in cover and biomass of grasses following afforestation. In our study, surface temperatures were cooler in the afforested than in the grass areas (data not shown); lower temperatures may decrease evaporation from surface soil and increase soil water content. Generally, lower bulk density, observed in the topsoil following afforestation, was attributed to a higher content of organic matter (Ritter, 2007). Our results confirmed those of previous research, and a decrease in bulk density was observed across the sampled soil depth. In addition, the decline in bulk density and increase in SOC would increase the water holding capacity, which would have resulted in the increase in soil water content to some extent.

Due to a greater P demand and uptake by trees than by grasses, grassland afforestation with coniferous trees have often resulted in a decrease in soil TP (Chen et al., 2008). In the present study, a small decrease in TP was observed in the subsoil following afforestation; however, afforestation increased TP contents in the topsoil (0–15 cm). The “nutrient pumping” effect (Jobbágy and Jackson, 2004; Farley and Kelly, 2004) has been observed following conversion of grasslands to forests in several studies; it had been suggested that pine roots, which tend to reach greater depths than those of grasses, can absorb nutrients from lower soil horizons; these nutrients return to the surface soil through litterfall and throughfall. Thus, we concluded that the increase in TP content in the surface soil in our study may be explained by “nutrient pumping” effect following grassland afforestation.

4.4. Implications for land management

Generally, grassland afforestation altered plant composition and diversity of the herbaceous community, and soil properties in both top and subsoil in the study area. Results from this study also demonstrated the potential for afforestation to increase soil organic carbon and nitrogen storage in semi-arid grasslands, which has important implications for C sequestration in the area. In the arid and semi-arid region of northwestern China, water supply is the main factor limiting sustainable development of planted forests, and water deficit had often been observed following afforestation (Wang et al., 2001; Wang et al., 2011; Zhu et al., 2015). A previous study on this site showed that severe water deficit developed in the subsoil 29 years after afforestation at tree density of 4458 trees ha^{-1}, while no significant differences in soil moisture were observed after 25 years at density of 2725 trees ha^{-1} (Zhu et al., 2015). In our study, we did not observe a water deficit in the subsoil, and a significant increase was observed in the topsoil at a density ranging from 2833 to 2967 trees ha^{-1}, which was close to the average stand densities (about 3000 trees ha^{-1}) of afforested areas in the region. Thus, we conclude that it is feasible to pursue C sequestration and maintain sustainable development of planted forests in this semiarid area if appropriate stand density is chosen for afforestation. However, to determine key theoretical thresholds for the establishment of planted forests in the area, long-term observations at a number of sites need to be conducted to reveal the dynamics of soil organic C, soil moisture, and nutrients following grassland afforestation. Furthermore, nutrient availability is an important factor to consider.

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Appendix A. Importance values (% plots) of the herbaceous community surveyed among the different-aged *Picea crassifolia* plantation forests and adjacent grasslands on two slopes.

		SW70			NE87	
Species	Family	Grassland	Afforestation (33a)	Afforestation (45a)	Grassland	Afforestation (35a)
Elymus cylindricus	*Gramineae*	34.5	13	3.1	30.2	12.8
Achnatherum splendens	*Gramineae*	21.2	8.2	7.3	15.2	7.9
Carex vulpina	*Cyperaceae*	4.9	18	31.8		21
Medicago archiducis-nicolai	*Leguminosae*	19.7	8.8	3.4	2.2	7.4
Oxytropis kansuensis	*Leguminosae*		3	3.7		4.1
Oxytropis glabra	*Leguminosae*		3.9			2.3
Oxytropis latibracteata	*Leguminosae*		1.5	1.3		
Oxytropis ochrocephala	*Leguminosae*					1.2
Artemisia mongolica	*Compositae*	11.5	9.4	7.4	12.7	3.2
Taraxacum mongolicun	*Compositae*	1.4	3.3	1.5	1.7	1.4
Leontopodium leontopodioides	*Compositae*	2.1	1.3	2.6		3.5
Leontopodium haplophylloides	*Compositae*	2.8	6.3	4.5		5.7
Taraxacum asiaticum	*Compositae*		2.3	1.1		1.1
Anaphalis flavescens	*Compositae*			2.5		
Cirsium arvense	*Compositae*			1.3		
Saussurea epilobioides	*Compositae*			1.5		
Anaphalis nepalensis	*Compositae*			1.6		
Anaphalis aureo-punctata	*Compositae*			2.2		
Urtica triangularis	*Urticaceae*	1.8	8.8	13.7		16.9
Potentilla acaulis	*Rosaceae*	5.6	7.3	8.4		16.6
Sibbaldia tetrandra	*Rosaceae*		2.8	2.3	2.8	1.5
Potentilla bifurca	*Rosaceae*	2.1	1.5	1.1	2.2	1.6
Potentilla multicaulis	*Rosaceae*		1	2.2	2	1.3
Potentilla anserina	*Rosaceae*		0.9			1.5
Potentilla conferta	*Rosaceae*					1.3
Potentilla supina	*Rosaceae*		1.2			1.4
Lilium pumilum	*Liliaceae*	2.2	2.4	1.4		1.6
Polygonum viviparum	*Polygonaceae*			1.6		2.1
Polygala sibirica	*Polygalaceae*	1.5	1.5			
Geranium sibircum	*Geraniaceae*			2.4		1.9
Iris lactea	*Iridaceae*		3.7	1.8		
Dracocephalum heterophyllum	*Labiatae*	1.8				1.1
Silene pterosperma	*Caryophyllaceae*	2.6			2.6	
Stellaria graminea	*Caryophyllaceae*	1.3			3.5	
Chenopodium glaucum	*Chenopodiaceae*	1.7			2.7	
Ceratoides latens	*Chenopodiaceae*			1.5		
Circaeaster agrestis	*Ranunculaceae*			1.4		
Aconitum tanguticum	*Ranunculaceae*			1.3		
Anemone rivularis	*Ranunculaceae*			1.5		
Pulsatilla ambigua	*Ranunculaceae*					1.1
Cynoglossum gansuense	*Boraginaceae*			2.1		
Lycopsis orientalis	*Boraginaceae*	1.9				
Stellra chamaejasme	*Thymelaeaceae*	2.8				
Morina chinensis	*Dipsacaceae*	1.8				
Rubia cordifolia	*Rubiaceae*			2		
Meconopsis quintuplinervia	*Papaveraceae*		1.5			

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