Soil, Leaf and Root Ecological Stoichiometry of *Caragana korshinskii* on the Loess Plateau of China in Relation to Plantation Age

Quanchao Zeng¹, Rattan Lal², Yanan Chen¹, Shaoshan An¹,³*

¹ College of Natural Resources and Environment, Northwest A&F University, Yangling, P.R. China, ² Carbon Management and Sequestration Center, The Ohio State University, Columbus, Ohio, United States of America, ³ State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Northwest A&F University, Yangling, Shaanxi, China

* shan@ms.iswc.ac.cn

**Abstract**

*Caragana korshinskii*, a leguminous shrub, a common specie, is widely planted to prevent soil erosion on the Loess Plateau. The objective of this study was to determine how the plantation ages affected soil, leaf and root nutrients and ecological stoichiometry. The chronosequence ages of *C. korshinskii* plantations selected for this study were 10, 20 and 30 years. Soil organic carbon (SOC) and soil total nitrogen (STN) of *C. korshinskii* plantations significantly increased with increase in the chronosequence age. However, soil total phosphorous (STP) was not affected by the chronosequence age. The soil C: N ratio decreased and the soil C: P and N: P ratios increased with increasing plantation age. The leaf and root concentrations of C, N, and P increased and the ratios C: N, C: P, and N: P decreased with age increase. Leaf N: P ratios were >20, indicating that P was the main factor limiting the growth of *C. korshinskii*. This study also demonstrated that the regeneration of natural grassland (NG) effectively preserved and enhanced soil nutrient contents. Compared with NG, shrub lands (*C. korshinskii*) had much lower soil nutrient concentrations, especially for long (>20 years) chronosequence age. Thus, the regeneration of natural grassland is an ecologically beneficial practice for the recovery of degraded soils in this area.

**Introduction**

Ecological stoichiometry is the study of the balance of energy and multiple chemical elements in ecological interactions [1]. Ecological stoichiometry connects evolutionary patterns affecting organismal elemental compositions to large-scale consequences and constraints associated with energy and material flows in ecosystems [2]. Ecological stoichiometry, especially for carbon (C), nitrogen (N) and phosphorus (P), plays an important role in analyzing composition, structure and function of a concerned community and ecological system [3]. It has been used in different different systems to explore the limited nutrition for varied plant groups [2, 4–9]. Generally, green leaf N:P ratio can indicate plant nutrition limitation, providing suggestive guide to the practice of vegetation management [3, 9–11]. Leaf N:P ratio (mass ratio) has been
suggested to be useful for indicating the shift between N— and P -limitation [12]. Researchers based on fertilization experiments proposed that low leaf N:P ratios (<14) reflect N limitation, that high N:P ratios (>16) likely reflect P limitation[3, 13, 14]. This threshold value has been widely used in different ecosystems.

Vegetation restoration is an effective and beneficial soil conservation practice that can enhance the contents of soil nutrients [15, 16]. The Loess Plateau was the largest eroded land area of China [17]. In 1999, the Chinese government initiated the “Grain for Green” program to address the environmental and economic developmental problems[18]. The program is aimed at converting degraded farmland into forest, shrub, and grassland [19, 20]. Natural grasslands such as the Yunwu Mountain have been protected by establishing shrub land and forest land over large areas [21, 22]. Vegetation restoration has improved soil quality over the last 30 years. Vegetation succession increases small macro-aggregates, bulk soil carbon(C) and aggregate C with an increase in duration of the plantation [23, 24]. Wang et al. (2015)[25] reported that after 15 years of conversion, the natural restoration process must be utilized properly to enhance the accumulation of soil organic carbon (SOC) upon conversion from cropland to grassland. Research studies thus far indicate that vegetation restoration strongly impacts soil quality and the attendant ecosystem services.

*C. korshinskii* is the most common shrub which had been established over large areas on the Loess Plateau. Because most studies concerning soil nutrients and aggregate stability in this area, little is known about C:N:P stoichiometry in *Caragana korshinskii* leaf, roots, and soil under different duration age. The duration of the establishment of this shrub varies between 10 and 30 years. Therefore, it is pertinent to assess variations in soil, leaf and root ecological stoichiometry with changes in duration of the plantation. These results will provide well understanding of the practice of planting *Caragana korshinskii* from ecological stoichiometry aspect. In this study, we focused on *C. korshinskii* along a successional gradient and a natural grassland with 30 duration years in northwest China. Previous studies had showed that duration age had positive on soil nutrients, especially for the storage of carbon and nitrogen in the grassland [23–25]. Based on the previous studies, we addressed following hypotheses: (1) plantation ages had the positive effects on soil nutrients and ecological stoichiometry; (2) natural restoration might be better method to improve soil nutrients compared with establishing *C. korshinskii* for the limited precipitation in drylands.

**Materials and Methods**

**Sample sites**

*C. korshinskii* areas were located in the Shanghuang watershed (106°26’–106°30’ E, 35°59’–36°02’N) in the Loess Plateau, Ningxia province of China, which covered an area of 8.19 hm². This region had a semi-arid climate. The average annual temperature (MAT) was 6.9˚C and the average annual precipitation (MAP) was about 419 mm (1982–2002 data) [26]. A severe decline in soil quality and accelerated erosion, serious threats to human wellbeing, were attributed to decline in the vegetative cover because of over-gazing, intensive cultivation and other anthropogenic perturbations. Towards an attempt to address the environmental and economic developmental issues, Shanghuang watershed had been used to conduct restoration projects. Thus, vegetation restoration had been attempted by establishing *C. korshinskii*. Thus, the most popular shrub had been planted in the watershed over a wide range of duration. The watershed was highly diverse physiographically and in terrain characteristics spatially (90% of the area is covered by hills, 51% of the land is located at altitude of 1534.3–1822.0 m, and the terrain was closely dissected and sharp-edged with steep slopes) [27]. Restoration of vegetation since circa 1984 had strongly changed the watershed into a demonstration zone for successful land
reclamation and comprehensive management. Therefore, the sampling sites for this study ran-
ger in age duration for 10, 20 and 30 years since the establishment of *C. korshinskii* plantations after imposing the grazing exclusion.

Another sample site was a natural grassland in the same area, which was a control experiment to compare the effects of vegetation restoration between shrub land and natural grassland. The two sample areas had similar climate (MAP and MAT) and soil types (Entisols, U.S. A. taxonomy). Natural grassland was selected “YunWu Mountain Nature Reserve” where was near Shanghuang watershed. It had a well-protected *Stipa bungeana* population, with an area of 6700 ha, an altitude of 1800–2100 m and an MAT of 6–7˚C. The MAP is 455 mm, most of that occurred during the summer (from June to September). In this area, most plants were herb with natural succession, with the constructive species of *Stipa bungeana* and *Stipa grandis*. Other companion species were *Thymus mongolicus*, *Potentilla bifurca*, *Artemisia vestita*, *Agropyron dasystachys*, *Heteropappus altaicus*, etc [22].

We state that no specific permissions were required for these locations/activities. We conform that the field studies did not involve endangered or protected species.

**Sampling and analyses**

We chose three different ages of plots of *C. korshinskii* and a natural grassland site without human disturbance as a control which could compare the differences of *C. korshinskii* land and grass land duration on soil nutrients and ecological stoichiometry. There were three field plots for every site as repeats. Three subplots (20 × 20 m) were established for each plantation age and NG site, with a total of 36 subplots. In each subplot, we collected three soil and plant samples, respectively. The dominant species and geographic information in each sampling site were showed in Table 1.

In natural grassland, we just collected soil samples. Thus, a total of 72 soil samples (under *C. korshinskii* and NG), and 54 samples of each of root and leaf for only of *C. korshinskii* were collected in this study area in mid-August 2014. Soil and plant samples were collected as previous studies described [9, 28]. Briefly, five soil cores (0–20 cm and 20–40 cm depths) were collected from each quadrat using a 3-cm diameter soil auger and mixed to obtain a composite sample. After removing stones, roots and small animals, soil samples were air-dried and sieved through a 0.15 mm sieve for the analysis of soil organic carbon, total nitrogen and total phosphorus. Plant samples included green leaf and root samples. In each subplot, we collected green leaf and root from 5 well-grown *C. korshinskii*. All the roots were fine roots with the diameter of < 0.5 mm. Leaf and root were watering with distilled water and dried at 65˚C for about 72 hours until reached a consistent weight. After over-dried, leaf and root samples grounded using a ball mill and sieved through a 0.15 mm sieve.

Concentrations of total nitrogen (TN) of the plant tissues and of soil total nitrogen (STN) were determined colorimetrically using the Kjeldahl acid-digestion method (KDY-9830) after

| Revegetation ages/(year) | Height/m   | Stand density/ (tree/ hm²) | Crown area/m² | Main types of herb species                                           |
|--------------------------|------------|-----------------------------|---------------|---------------------------------------------------------------------|
| 10                       | 1.31±0.06  | 3500±1.53                   | 1.24±0.02     | *Stipa bungeana*, *Heteropappus altaicus*, *Thymus mongolicus*, *Artemisia frigida*, *Lespedeza davurica* |
| 20                       | 1.54±0.08  | 4500±6.08                   | 1.55±0.14     | *Stipa bungeana*, *Heteropappus altaicus*, *Artemisia frigida*, *Lespedeza davurica*, *Artemisia scoparia* |
| 30                       | 1.59±0.12  | 3300±5.29                   | 1.69±0.08     | *Stipa bungeana*, *Heteropappus altaicus*, *Lespedeza davurica*, *Artemisia scoparia* |
| Natural grassland (NG)   | -          | -                           | -             | *Thymus mongolicus*, *Stipa bungeana*, *Lespedeza davurica*          |

doi:10.1371/journal.pone.0168890.t001
extraction with 0.02 mol/L sulfuric acid [29]. Total plant P (TP) was measured using colorimetric analysis (UV 2800) after digestion with H$_2$SO$_4$ and H$_2$O$_2$ [30]. Soil total P (STP) was measured using a spectrophotometer after wet digestion with H$_2$SO$_4$ and HClO$_4$ followed by colorimetric analysis (UV 2800) [31]. The organic C concentration in soils and plants was measured using a modified Mebius method [32]. More specifically, 0.5 g of soil samples (or 0.02 g of plant samples) were digested with 5 ml of 1 mol/L K$_2$Cr$_2$O$_7$ and 5 ml of concentrated H$_2$SO$_4$ at 180˚C for 5 min, followed by titration with standardized FeSO$_4$ (0.2 mol/l).

**Data analysis**

We used one-way analysis of variance (ANOVA) to analyze the effects of duration age on nutrients and stoichiometric characteristics of the soil, root, leaf. Linear regression analysis was used to test the relationships between the soil and plant stoichiometric characteristics. All the significant differences were showed at the level of 0.05. SPSS 20.0 software (SPSS, Inc., Chicago, IL, USA) was used for the statistical analysis. Soil and plants nutrients concentrations expressed as g/kg on a dried weight basis and all the C:N:P ratios were mass ratios. Figures were conducted by Origin 9.0.

**Results**

**Soil nutrients and soil C:N:P characteristics**

The plantation age had significant effects on the soil organic carbon (SOC) and soil total N (STN) concentrations (Fig 1). Concentrations of SOC (g/kg) for 10, 20 and 30 yr duration were 10.79, 12.66 and 13.85 in the 0-20-cm soil layer compared with 8.13, 8.49, and 8.97 in 20-40-cm layer, respectively. Concentrations of STN increased with increase in the plantation age, and the highest value was measured at 30 yr. Concentration of STN (g/kg) for 30 yr was, 1.57 and 1.05 in 0–20 cm and 20–40 cm-soil layer, respectively. In comparison, concentration of STP had a narrow range (0.46–0.50 g/kg) in 0-20-cm and 20-40-cm soil layers. While concentrations of SOC and STN changed significantly ($P<0.05$) with increase in the plantation age (from 10 yr to 30yr), that of STP had no significant effect. However, concentrations of SOC and STN under NG were approximately 2-fold higher than those measured under *C. korshinskii* plantations. Similarly, concentration of STP under NG was much higher than that measured under three *C. korshinskii* sites of 10 yr, 20 yr, and 30yr.

The soil ecological C:N:P stoichiometry also varied with the plantation age (Fig 1). The soil C:N ratios for plantation age of 10, 20 and 30 yr under *C. korshinskii* were 9.12, 9.07, and 8.80 in the 0-20-cm soil layer, respectively. However, the C:N ratio was rather narrow (8.51–8.59) in 20-40-cm layer. Overall, along the plantation age significantly impact the soil C:N ratio. In fact, the trend of the soil C:P ratio was also similar to that for soil N:P ratio (Fig 1). The soil N:P ratio significantly increased with increase in the plantation age for both soil depths (Fig 1). The C:N:P ratio also increased with increase in the plantation age (Table 2).

**Leaf and root C, N and P contents and C:N:P characteristics**

The plantation age had a significant effect on leaf total C, total N and total P concentrations. The highest concentrations (g/kg) of total C (475.2), total N (42.3) and total P (1.9) were observed under the 30-year-old and the lowest under 10-year-old plantation of *C. korshinskii* (Fig 2). Concentration (g/kg) of nutrients in roots ranged from 452.3 to 466.3 for total C, 21.2 to 27.0 for total N, and 1.6 to 1.9 for total P.

Increase in the plantation age decreased the ratio of nutrients in the leaves from 12.0 to 11.2 for C:N, 291.3 to 247.1 for C:P, and 24.3 to 22.0 for N:P. Trends in the ratio of nutrients in the
Fig 1. Stoichiometric characteristics of soil C, N and P as affected by soil depth and plantation age. Values are the means ± SE of three plots. Values designated by different lowercase letters were significantly different among different duration ages, and different capital letters indicated significant difference soil layers, respectively (P < 0.05).

doi:10.1371/journal.pone.0168890.g001
roots with regards to the planation age for C:N, C:P and N:P exhibited trends similar to those of the leaves. The plantation age had a significant effect on the leaf and root ecological stoichiometry \((P<0.05)\).

The highest leaf C:N:P ratios were observed in the 30-year-old and the lowest in the 10-year-old plantation (Table 2). Trends of the root C:N:P ratios were similar to those of the leaves (Table 2). In general, the plant C:N:P ratios increased with increase in the plantation age.

### Relationships between soil versus leaf and root stoichiometry

The leaf C:P ratio decreased linearly with the soil C:P ratio \((r = -0.925, P<0.01\) for the 0-20-cm; \(r = -0.736, P = 0.024\) for 20-40-cm layer) (Fig 3b). The N:P ratio in soil under \textit{C. korshinskii} varied linearly with the leaf N:P ratio \((r = -0.905, P<0.01\) for the 0-20-cm; \(r = -0.768, P = 0.016\) for 20-40-cm layer) (Fig 3c). In the 0-20-cm layer, the soil C:N ratio increased linearly with the root C:N ratios \((r = 0.768, P = 0.016\) (Fig 3d). The soil N:P ratios decreased linearly with the root N:P ratios \((r = -0.945, P<0.01\) for 0-20-cm; \(r = -0.826, P = 0.006\) for 20-40-cm layer) (Fig 3e). The soil C:P ratios decreased linearly with the root C:P ratios \((r = -0.974, P<0.01\) for 0-20-cm; \(r = -0.868, P = 0.002\) for 20-40-cm layer) (Fig 3f).

### Discussion

The effects of plantation ages of \textit{C. korshinskii}

Vegetation restoration was one of the most effective method to solve the soil erosion problem, especially on the Loess Plateau of China [33, 34]. Shrubs (i.e. \textit{C. korshinskii}) were one of the most common vegetation types in the Shanghuang watershed and had been used to improve the soil quality, including soil aggregates and nutrient availability. The data from the present study showed that vegetation age had positive effects on soil nutrients, and these results were in accord with those reported for the Shapotou area [35], Yaoledianzi village in northeast China [36], Wulanaodu area in northeast China [37], and Yanchi country in Ningxia province, northwest China [38]. Revegetation from abandoned farmland significantly influenced soil total C and total N, resulting in a C:N ratio of about 10 after 30 years of abandonment [39]. The observations in this study showed that the C:N ratio of soil under \textit{C. korshinskii} ranged from 8.80 to 9.11. In the first 20 years, soil C:N ratio was nearly constant, and significant higher than the soil in the duration of 30 years. Soil total C concentration was significantly correlated with the soil total N, indicating similar trends for soil C and N storage. This coupling relation led to a narrow range for soil C:N ratio.

The N:P ratio in leaf was an indicator of N or P limitation in terrestrial ecosystems [13]. The relationships between leaf N:P ratios and plant properties have also been used to indicate their responses to environmental change or human disturbance [40]. In this study, the N:P ratios decreased with increase in plant age, and all the ratios were higher than 20. Review of previous

### Table 2. Stoichiometric characteristics of plant and soil C:N:P as affected by plantation age and soil depth.

| Variable | 10-year-old | 20-year-old | 30-year-old | NG |
|----------|-------------|-------------|-------------|----|
| Leaf     | 291:24:1    | 258:23:1    | 247:22:1    | -  |
| Root     | 659:31:1    | 530:27:1    | 434:25:1    | -  |
| Soil depth |            |             |             |    |
| 0–20 cm  | 22:2:1      | 25:3:1      | 28:3:1      | 30:4:1 |
| 20–40 cm | 18:2:1      | 18:2:1      | 19:2:1      | 24:3:1 |

doi:10.1371/journal.pone.0168890.t002
Fig 2. Stoichiometric characteristics of leaf and root C, N and P as affected by plantation age. Values are the means ± SE of three plots. For each plant issue, means with different letters are significantly different based on ANOVA and Scheffe’s test (P < 0.05). Note: Values designated by different lowercase letters were significantly different among different duration ages, and different capital letters indicated significant difference between leaf and root, respectively (P < 0.05).

doi:10.1371/journal.pone.0168890.g002
Fig 3. Relationships between soil, leaf and root stoichiometric characteristics of *C. korshinskii*.

doi:10.1371/journal.pone.0168890.g003
studies [3, 14], showed that C. korshinskii was limited by P nutrition, and the severity of limita-
tion decreases with the increase in age of the shrub, implying that older C. korshinskii plants
need a much lower available P requirement for growth. Therefore, soil available P content
would increase strongly with increasing C. korshinskii age, and which had positive conse-
quences for soil quality. However, it was interesting that soil total P concentration was not
decreased with age increase. Because soil parent materials were the main resources of total P
content[41]. Thus C. korshinskii soil total P had no significant changes with age increase.

The difference between C. korshinskii and natural grassland

Land use was another factor affecting soil nutrients. Grasslands played an important role in
the global C and N cycles. The data showed that soil total C and total N were higher in the
older grasslands (30 years) and the values were much higher than in a soil with similar aged C.
korshinskii plots. Thus, vegetation type had a strong effect on the soil C and N storage. Li et al.
(2013) [42] reported that C input was greater than C output in an enclosed grasslands. Soil N
fixation by C. korshinskii and subsequent N released by litter decomposition could increase
soil N content [36]. Carbon fixation via photosynthesis and the subsequent transfer of C to the
soil via leaf, litter and root turnover contributes to soil C accumulation [43]. A larger above-
ground biomass indicated that more litters returned to the soil and the roots exudates would
contain more nutrients.

At the same age, soil under natural grassland had the highest total C, total N and total P,
suggesting that, compared with natural grassland, C. korshinskii was not the most suitable spe-
cie for restoration of eroded lands in this region. Firstly, C. korshinskii required more amount
of water and nutrients to maintain its growth than grasses for the higher living biomass. Sec-
ondly, given that plant growth was main limited by water in the dryland [44], the annual pre-
cipitation was limited in this area. Finally, soils of the studied areas were deficit in nutrients
before planting C. korshinskii because of over grazing, which also limited the growth of C. kor-
shinskii. Therefore, natural restoration may be the best approach to restore degraded soils to
attain the desired outcome albeit taking a long time. While establishing C. korshinskii
improved soil quality over a short time period, it had negative effects on the soil on the long-
term basis. Consequently, planting C. korshinskii was not the best choice to improve soil qual-
ity for Shanghuang watershed in the long run.

Conclusion

C. korshinskii, a leguminous shrub, is a dominant native plant species that is widely planted to
prevent soil erosion on the Loess Plateau. The results showed that C. korshinskii growth altered
concentration of nutrients in soil, leave and root. The leaf N:P ratios were higher than 20
regardless of the age of the shrub. Therefore, the growth of C. korshinskii was limited by P
availability. Compared with natural grassland, planting Caragana korshinskii scrubland may
not be a good choice for vegetation restoration as its soil fertility was lower than the natural
grasslands. Accumulation of soil nutrients under C. korshinskii is a slow process and takes a
long time.

Supporting Information

S1 File. Supporting information about data.
(XLSX)
Acknowledgments

This study was supported by the National Natural Science Foundation of China (41671280,41171226) and the Non-profit Industry Research Project of Chinese Ministry of Water Resources (201501045). We also thank the anonymous reviewers and editor for their great help in improving the quality of the manuscript.

Author Contributions

Conceptualization: SA RL.

Data curation: QZ.

Formal analysis: QZ YC.

Funding acquisition: SA.

Investigation: QZ YC.

Methodology: QZ YC SA.

Project administration: QZ SA.

Resources: SA RL.

Software: YC QZ.

Supervision: SA.

Validation: QZ RL.

Visualization: YC SA.

Writing – original draft: QZ YC.

Writing – review & editing: RL SA.

References

1. Elser J, Sterner R, Gorokhova E, Fagan W, Markow T, Cotner J, et al. Biological stoichiometry from genes to ecosystems. Ecology Letters. 2000; 3(6):540–50.
2. Sterner RW, Elser JJ. Ecological stoichiometry: the biology of elements from molecules to the biosphere: Princeton University Press; 2002.
3. Koerselman W, Meuleman AF. The vegetation N: P ratio: a new tool to detect the nature of nutrient limitation. Journal of applied Ecology. 1996:1441–50.
4. Yuan Z, Chen HY, Reich PB. Global-scale latitudinal patterns of plant fine-root nitrogen and phosphorus. Nature communications. 2011; 2:344. doi: 10.1038/ncomms1346 PMID: 21673665
5. Wardle DA, Bardgett RD, Klimomos JN, Setalâ H, Van Der Putten WH, Wall DH. Ecological linkages between aboveground and belowground biota. Science. 2004; 304(5677):1629–33. doi: 10.1126/science.1094875 PMID: 15192218
6. Wang SQ, Yu GR. Ecological stoichiometry characteristics of ecosystem carbon, nitrogen and phosphorus elements. Acta Ecologica Sinica. 2008; 8:3937–47.
7. Elser J, Fagan W, Kerkhoff A, Swenson N, Enquist B. Biological stoichiometry of plant production: metabolism, scaling and ecological response to global change. New Phytologist. 2010; 186(3):593–608. doi: 10.1111/j.1469-8137.2010.03214.x PMID: 20298486
8. Li Y, Li Q, Guo D, Liang S, Wang Y. Ecological stoichiometry homeostasis of Leymus chinensis in degraded grassland in western Jilin Province, NE China. Ecological Engineering. 2016; 90:387–91.
9. Zeng Q, Li X, Dong Y, An S, Darboux F. Soil and plant components ecological stoichiometry in four steppe communities in the Loess Plateau of China. CATENA. 2016; 147:481–8.
10. Rong Q, Liu J, Cai Y, Lu Z, Zhao Z, Yue W, et al. Leaf carbon, nitrogen and phosphorus stoichiometry of Tamarix chinensis Lour. in the Laizhou Bay coastal wetland, China. Ecological Engineering. 2015; 76:57–65.

11. Pan FJ, Zhang W, Liu SJ, Li DJ, Wang KL. Leaf N:P stoichiometry across plant functional groups in the karst region of southwestern China. Trees-Struct Funct. 2015; 29(3):883–92.

12. Schreeg LA, Santiago LS, Wright SJ, Turner BL. Stem, root, and older leaf N: P ratios are more responsive indicators of soil nutrient availability than new foliage. Ecology. 2014; 95(8):2062–8. PMID: 25230458

13. Güsewell S. N: P ratios in terrestrial plants: variation and functional significance. New phytologist. 2004; 162(2):243–66.

14. Zhang L-X, Bai Y-F, Han X-G. Differential responses of N: P stoichiometry of Leymus chinensis and Carex korshinskyi to N additions in a steppe ecosystem in Nei Mongol. Acta Botanica Sinica. 2004; 46(3):259–70.

15. Zheng F-L. Effect of vegetation changes on soil erosion on the Loess Plateau. Pedosphere. 2006; 16(4):420–7.

16. Zhou H, Peng X, Peth S, Xiao TQ. Effects of vegetation restoration on soil aggregate microstructure quantified with synchrotron-based micro-computed tomography. Soil and Tillage Research. 2012; 124:17–23.

17. Feng X, Fu B, Lu N, Zeng Y, Wu B. How ecological restoration alters ecosystem services: an analysis of carbon sequestration in China’s Loess Plateau. Scientific reports. 2013; 3.

18. Chen X, Zhang X, Zhang Y, Wan C. Carbon sequestration potential of the stands under the Grain for Green Program in Yunnan Province, China. Forest Ecology and Management. 2009; 258(3):199–206.

19. Wolf B, Kiese R, Chen W, Grote R, Zheng X, Butterbach-Bahl K. Modeling N2O emissions from steppe in Inner Mongolia, China, with consideration of spring thaw and grazing intensity. Plant and soil. 2012; 350(1–2):297–310.

20. Deng L, Liu GB, Shangguan ZP. Land-use conversion and changing soil carbon stocks in China’s ‘Grain-for-Green’ Program: a synthesis. Global Change Biology. 2014; 20(11):3544–56. doi: 10.1111/gcb.12508 PMID: 24357470

21. Teffesse Z, De Boer W, Baars R, Prins H. Changes in soil nutrients, vegetation structure and herbaceous biomass in response to grazing in a semi-arid savanna of Ethiopia. J Arid Environ. 2011; 75(7):662–70.

22. Cheng J, Cheng J, Hu T, Shao H, Zhang J. Dynamic changes of Stipa bungeana steppe species diversity as better indicators for soil quality and sustainable utilization mode in Yunwu Mountain Nature Reserve, Ningxia, China. CLEAN–Soil, Air, Water. 2012; 40(2):127–33.

23. Cheng M, An S. Responses of soil nitrogen, phosphorous and organic matter to vegetation succession on the Loess Plateau of China. Journal of Arid Land. 2015; 7(2):216–23.

24. Cheng M, Xiang Y, Xue Z, An S, Darboux F. Soil aggregation and intra-aggregate carbon fractions in relation to vegetation succession on the Loess Plateau, China. Catena. 2015; 124:77–84.

25. Wang D, Liu Y, Shang ZH, Tian FP, Wu GL, Chang XF, et al. Effects of Grassland Conversion From Cropland on Soil Respiration on the Semi-Arid Loess Plateau, China. CLEAN–Soil, Air, Water. 2015; 43(7):1052–7.

26. Xue Z, Cheng M, An S. Soil nitrogen distributions for different land uses and landscape positions in a small watershed on Loess Plateau, China. Ecological Engineering. 2013; 60:204–13.

27. An S-S, Huang Y-M, Zheng F-L, Yang J-G. Aggregate characteristics during natural revegetation on the Loess Plateau. Pedosphere. 2008; 18(6):809–16.

28. Bao SD. Soil and agricultural chemistry analysis. China Agriculture Press, Beijing; 2000.

29. Sparks DL, Page A, Helmke P, Looepert R, Soltanpou R, Tabatabai M, et al. Methods of soil analysis. Part 3-Chemical methods: Soil Science Society of America Inc.; 1996.

30. Thomas R, Sheard R, Moyer J. Comparison of conventional and automated procedures for nitrogen, phosphorus, and potassium analysis of plant material using a single digestion. Agron J. 1967; 59(3):240–3.

31. Parkinson J, Allen S. A wet oxidation procedure suitable for the determination of nitrogen and mineral nutrients in biological material. Communications in Soil Science & Plant Analysis. 1975; 6(1):1–11.

32. Nelson D, Sommers LE. Total carbon, organic carbon, and organic matter. Methods of soil analysis Part 2 Chemical and microbiological properties. 1982; (methodssofsoiland2):539–79.

33. An S, Mentler A, Mayer H, Blum WEH. Soil aggregation, aggregate stability, organic carbon and nitrogen in different soil aggregate fractions under forest and shrub vegetation on the Loess Plateau, China. Catena. 2010; 81(3):226–33.
34. Zhao G, Mu X, Wen Z, Wang F, Gao P. Soil erosion, conservation, and eco-environment changes in the loess plateau of China. Land Degradation & Development. 2013; 24(5):499–510.

35. Yu Y, Lin Q, Shi Q, Liu J. Changes of habitat and vegetation in man-made vegetation area of Shapotou section along Baotou-Lanzhou railway. Acta Ecologica Sinica. 2001; 22(3):433–9.

36. Su YZ, Lin Zhao H. Soil properties and plant species in an age sequence of Caragana microphylla plantations in the Horqin Sandy Land, north China. Ecological Engineering. 2003; 20(3):223–35.

37. Cao C, Jiang D, Teng X, Jiang Y, Liang W, Cui Z. Soil chemical and microbiological properties along a chronosequence of Caragana microphylla Lam. plantations in the Horqin sandy land of Northeast China. Applied Soil Ecology. 2008; 40(1):78–85.

38. Liu J-B, Zhang Y-Q, Wu B, Qin S-G, Jia X, Feng W. Changes in Soil Carbon, Nitrogen, and Phosphorus along a Chronosequence of Caragana microphylla Plantation, Northwestern China. Pol J Environ Stud. 2014; 23(2):385–91.

39. Deng L, Shangguan Z-P, Sweeney S. Changes in soil carbon and nitrogen following land abandonment of farmland on the Loess Plateau, China. PloS one. 2013; 8(8):e71923. doi: 10.1371/journal.pone.0071923 PMID: 23940793

40. Olde Venterink H, Wassen M, Verkroost A, De Ruiter P. Species richness–productivity patterns differ between N-, P-, and K-limited wetlands. Ecology. 2003; 84(8):2191–9.

41. Mage SM, Porder S. Parent material and topography determine soil phosphorus status in the Luquillo Mountains of Puerto Rico. Ecosystems. 2013; 16(2):284–94.

42. Li M, Zhang X, Pang G, Han F. The estimation of soil organic carbon distribution and storage in a small catchment area of the Loess Plateau. Catena. 2013; 101:11–6.

43. Leifeld J, Kögel-Knabner I. Soil organic matter fractions as early indicators for carbon stock changes under different land-use? Geoderma. 2005; 124(1):143–55.

44. Ren H, Xu Z, Huang J, Lü X, Zeng D-H, Yuan Z, et al. Increased precipitation induces a positive plant-soil feedback in a semi-arid grassland. Plant and Soil. 2015; 389(1–2):211–23.