# Response of aggregate associated organic carbon, nitrogen and phosphorous to re-vegetation in agro-pastoral ecotone of northern China

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## ABSTRACT

The agro-pastoral ecotone is an ecologically fragile transition zone and suffers from extensive human-induced land-use changes. To understand how soil organic carbon (OC) and nutrients will respond to re-vegetation practices in such an ecotone zone, we present the response of OC, nitrogen (N) and phosphorus (P) in both bulk soils and water-stable aggregates to typical re-vegetation patterns in an agro-pastoral ecotone of northern China. Three re-vegetation patterns, i.e., cropland converted to natural grassland, woodland and artificial grassland at different times (6 to 40 years), were selected. The paired croplands were also selected as the control for each re-vegetation pattern. The measured soil metrics include the proportions of each type of water-stable aggregate, the mean weight diameter and geometric mean diameter of the aggregates, and the concentrations of OC, N and P in bulk soils and each aggregate fraction. The results showed that the three re-vegetation patterns significantly increased the mass proportion of macro-aggregates, the values of mean weight diameter and geometric mean diameter, and the concentrations of OC and N in topsoils (0–10 cm). The accumulation of OC and N in bulk soils was mainly due to the accumulation in macro-aggregates. Furthermore, increases in OC and N were greater after conversion to legume vegetation than to non-legume vegetation, and were highest at approximately 20 years after the conversion. However, concentrations of P in bulk soils and aggregates were similar among the three re-vegetation patterns and the three aggregate fractions, and were minimally affected by the conversion. These results highlighted the potential of legume vegetation to increase OC and N in surface soils and aggregates, and indicated no response of soil P to re-vegetation in an agro-pastoral ecotone of northern China.

## 1. Introduction

The agro-pastoral ecotone of northern China was characterized by the cropland and grassland, and has long been deemed as an ecologically fragile transition zone (Li et al., 2015; Li et al., 2018). This ecotone suffers from severe soil erosion, grazing, over-cultivation and human-induced land-use changes that lead to land degradation and greenhouse gas emissions (Liu et al., 2017). In addition, this ecotone is very sensitive to climate change, and the climatic warming in this ecotone has exceeded global averages (Li et al., 2015). The increased annual temperature, longer dry and hot days, and decreased precipitation have led to a reduction in the ecosystem productivity and sustainability.

Cultivation causes the depletion of soil organic matter and emissions of carbon (C) and nitrogen (N) into the atmosphere, which in turn exacerbates soil degradation through soil compaction, crusting, erosion, nutrient depletion, acidification and a reduction in the activity and diversity of soil microorganisms (Lal, 2004). Re-vegetation has been regarded as the most widely accepted and effective approach to protect soils from degradation (Bienes et al., 2016; Kroepfl et al., 2013). Re-vegetation improves soil physical, chemical and biological properties (e.g., increased nutrient pools, aggregate stability, enzyme activity, and decreased soil bulk density), which all contribute to the restoration of soils (An et al., 2009). For instance, IIzquierdo et al. (2005) reported that planting either native or exotic trees in a degraded mining site increased soil organic C (OC) and N and improved the aggregation of soil particles through enmeshing soil particles with hyphae and roots. Zhao et al. (2017) found that the shrub plantation markedly improved the microstructure of soil aggregates with increased total porosity and

elongated pores on the Loess Plateau of China. Moreover, the effects of re-vegetation in soils varied with vegetation types and the times after the conversion. For instance, Gei and Powers (2013) found that soils under leguminous species (*Acosmium panamense*) had the highest level of soil total C and N among six species in plantation habitats in northwestern Costa Rica. Cheng et al. (2015) reported that total C in bulk soils and aggregates increased with recovering time during the revegetation succession in the Loess Plateau of China. A better understanding of the response of soil OC and nutrient cycling in an agro-pastoral ecotone would help to understand the mechanism underlying the restoration of this degraded ecosystem and would give proper suggestions on land use management to policy makers.

Soil aggregates significantly influence the turnover and availability of OC and nutrients, as well as their responses to land management (Rabot et al., 2018; Six et al., 2000). For example, soil aggregates are important agents of soil OC and N retention, and provide physical protection for OC and N from microbial decomposition and loss (Udom et al., 2016; Wei et al., 2013). Furthermore, the concentrations of OC and N in aggregates were hypothesized to be increased with the increasing size of aggregates according to the hierarchy theory (Elliott, 1986; Qiu et al., 2015), and the responses of soil OC and N to land management were supposed to be related to changes in OC and N associated with macro-aggregates (Cambardella and Elliott, 1993; Wei et al., 2013). However, the distribution patterns of OC and N among aggregate fractions and their responses to land-use change were not examined in the agro-pastoral ecotone. This knowledge is urgently needed given the important roles of OC and N in mediating the sustainability of the ecosystem and their responses to global change and land management.

Current studies regarding the effects of land-use change on soil nutrients have mainly focused on OC and N (e.g. Adesodun et al., 2007; Wei et al., 2013), while the response of phosphorous (P) in bulk soils and aggregates was less investigated. Phosphorous plays an essential role in regulating plant growth and the biogeochemical cycles of OC and N due to its role in mediating the molecular structure and energy transfer during the activities of plant and soil microbes (Marschner, 2012; Richardson et al., 2011), and therefore, it has important effects on ecosystem functionality (Ellsworth et al., 2017). For example, a recent study by Ellsworth et al. (2017) showed that P availability may potentially constrain $CO_2$-enhanced productivity in P-limited forests in western Austria. However, the knowledge gap of P distribution among aggregates and its response to land-use change hinders our ability to understand the interactions among nutrients and their regulations on ecosystem processes, and to model biogeochemical cycles.

Based on the above considerations, we generate the following hypotheses: (H1) re-vegetation would result in the accumulation of OC, N and P in bulk soils and aggregates, and the accumulation in bulk soils would result from the accumulation in macro-aggregates; (H2) the effects of re-vegetation would be greater after conversion to legume vegetation than to non-legume vegetation; and (H3) the effects of re-vegetation would be enhanced over time. To test these hypotheses, we present results of the OC, N and P concentrations in bulk soils and water-stable aggregates in three typical re-vegetation patterns, i.e., cropland converted to natural grassland, woodland and artificial grassland, in the agro-pastoral ecotone of northern China. We also examined the relationships of the changes in OC, N and P in bulks soils with those in each aggregate fraction to understand the contribution of aggregates to the responses of OC and nutrients in bulk soils. Furthermore, the effect of the re-vegetation age on the response was assessed.

## 2. Materials and methods

### 2.1. Study site

This study was conducted in the Liudaogou watershed (38°46′–38°51′N, 110°21′–110°23′E) on the northern Loess Plateau of China. The site is located in the center of the agro-pastoral ecotone of northern China. The watershed has an area of 6.89 km$^2$ and an elevation ranging from 1081 to 1274 m. The region is characterized by a semiarid continental climate with a mean annual temperature of 8.4 °C (ranging from −9.7 °C in January to 23.7 °C in July), a mean annual precipitation of 437 mm (77% occurs from June to September) and a mean frost-free period of 169 days. The soil at the study site is a Calcaric Regosol according to the FAO/UNESCO system. The contents of sand, silt and clay in the study soils are 72% (± 4%), 15% (± 3%) and 13% (± 2%), respectively, with a texture of sandy loam.

Before the 1970s, the study site was characterized by grassland and cropland. The dominant native grasses are bunge needlegrass (*Stipa bungeana* Trin.) and Dahurian bushclover (*Lespedeza daurica* (Laxm.) Schindl.). The crop in the cropland was millet (*Panicum miliaceum* L.) for > 50 years. Manure fertilizer was applied in the cropland before the 1970s, while chemical fertilizers were used after that time. From the late 1970s, large amounts of the legume shrub, Korshinsk pea shrub (*Caragana korshinskii* Kom.), and legume grass, purple alfalfa (*Medicago sativa* L.), have been planted in degraded cropland to prevent soil erosion and degradation.

### 2.2. Field investigation and soil sampling

This study consisted of three re-vegetation patterns, i.e., cropland converted to natural grassland, shrub woodland and artificial grassland. Each re-vegetation pattern has various conversion ages and each conversion age was composed of a paired cropland and re-vegetated land. The natural grasslands selected in this study were converted from cropland 10 and 20 years ago and are dominated by a native perennial grass, bunge needlegrass. The woodlands selected were converted from cropland 10, 20 and 40 years ago and are dominated by Korshinsk pea shrub. The artificial grasslands were converted from cropland 6, 10, 15 and 20 years ago and are dominated by purple alfalfa. The grassland is not under pasture, and the biomass is not harvested. The adjacent cropland was selected as the control to assess the effects of re-vegetation at each conversion age. Cropland is still under tillage. The crops in the cropland are maize (*Zea mays* L.) and millet (*Panicum miliaceum* L.). Crops are not rotated in this site due to the relatively short growing season. In the study site, chemical N and P fertilizers are not applied to re-vegetated lands, but they are applied into cropland, which has the potential to increase soil N and P in cropland. The chemical N and P fertilizer used were diammonium hydrogen phosphate and dipotassium hydrogen phosphate. The amounts applied varied among years, with a mean rate of 360 (± 26) kg N ha$^{-1}$ yr$^{-1}$ and 135 (± 17) kg P ha$^{-1}$ yr$^{-1}$, respectively (Ge et al., 2019). A previous study in the same site showed that 3–11% of fertilizer N will be lost through volatilization, and 8–61% of fertilizer N will leach out of the 0–100 cm soil depth (Fu et al., 2010). Furthermore, the N use efficiency of cropland in this site was 82.3% (Wang, 2005). Hence, the application of chemical N fertilizer will not result in the accumulation of N in cropland soils. Previous measurements in a cropland that received 60 kg P$_2$O$_5$ ha$^{-1}$ year$^{-1}$ in the study site showed that the concentration of P in soils did not change over time from 2007 (0.47 g kg$^{-1}$, Fu et al., 2009) to 2014 (0.45 g kg$^{-1}$, unpublished data). Therefore, selecting adjacent cropland as a control to assess the effect of re-vegetation on soil OC, N and P was deemed applicable in this study.

China's agro-pastoral ecotone has suffered severe soil erosion and responds sensitively to global warming, both could significantly influence C and nutrient dynamics in soils (Bokhorst et al., 2007; Koerner and Basler, 2010; Quinton et al., 2010; Wei et al., 2017). Disentangling the effects of land-use change, soil erosion and global change would provide appropriate assessment on soil C and nutrient dynamics in this ecotone. The experimental design of current study allows us to test the effects of re-vegetation patterns and conversion ages, but not the interaction among soil erosion, global change and land-use change. Herein we investigated the effects of land-use change on soil OC, N and P without considering the effects of soil erosion and global change, but do not intent that re-vegetation is more important than the other two factors in influencing soil C and nutrients in this region. We recommend that the effects of soil erosion and global change should be separated in further research.
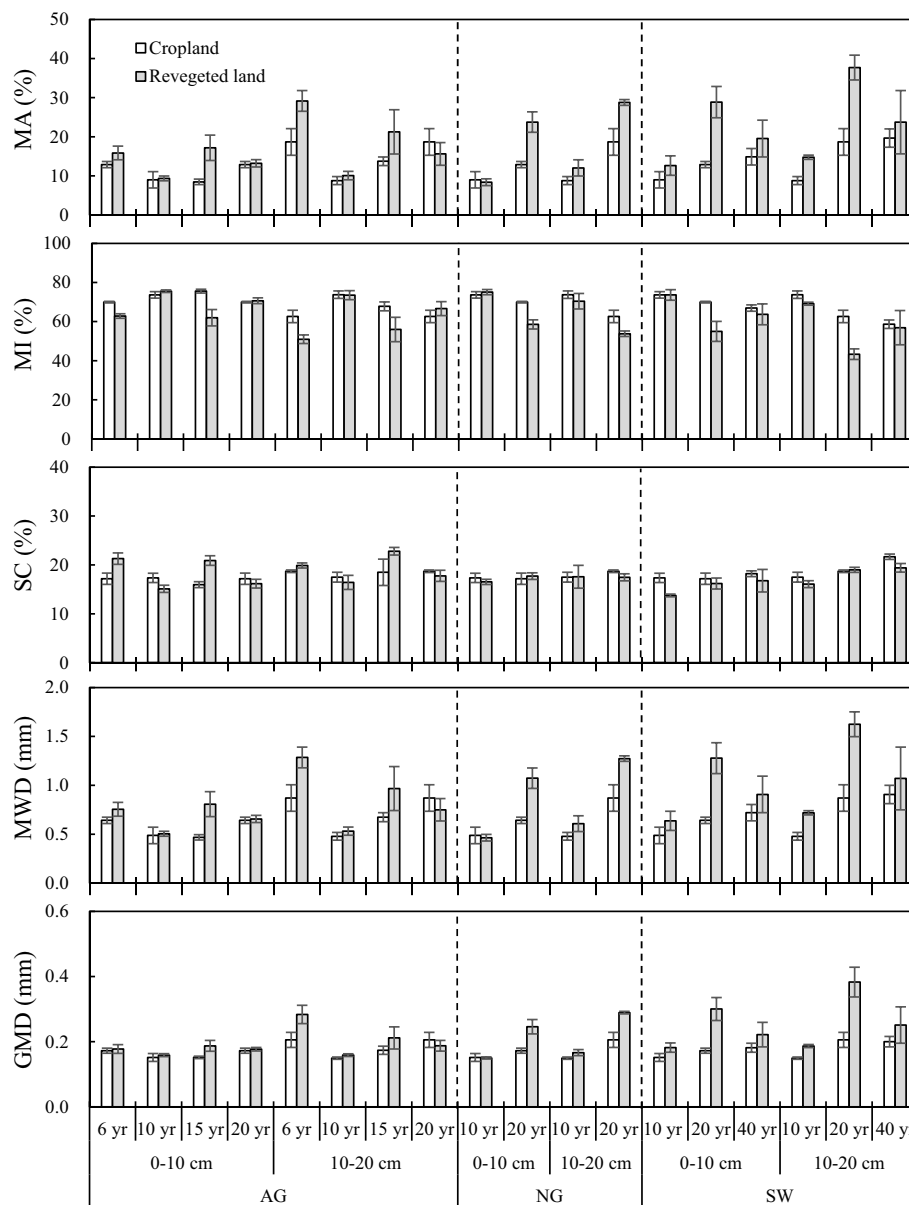
**Fig. 1.** Proportions of each aggregate fraction, the mean weight diameter (MWD) and geometric mean diameter (GMD) of soil water-stable aggregates for each age and pattern of re-vegetation at the 0–10 and 10–20 cm depths. AG: converting cropland to artificial grassland; NG: converting cropland to natural grassland; SW: converting cropland to pea shrub woodland. MA: macro-aggregates; MI: micro-aggregates; SC: silt + clay fraction. Error bars were two standard errors of the mean.

In September 2014, soil sampling was conducted for each re-vegetation pattern at each conversion age (three to five replications, 1 × 1 m plots), which included a paired cropland and re-vegetated land. All the sampling plots have similar physical-geographical conditions. Seven samples were randomly collected from the 0–10 and 10–20 cm depths in each plot to form a composite sample. Visible pieces of organic material were removed, and then the moist soil samples were taken to the laboratory and air dried.

### 2.3. Laboratory analysis

Soil water-stable aggregates were obtained by wet sieving through 0.25 and 0.053 mm sieves following the procedures described by Cambardella and Elliott (1993). A 200 g air-dried and un-ground soil sample was spread on top of a 0.25-mm sieve submerged in deionized water. Soil samples were immersed in the water for 10 min and then sieved by moving the sieve 3 cm vertically 50 times over 2 min. The material remaining on the 0.25-mm sieve was transferred to a glass pan

(macro-aggregates > 0.25 mm, MA). The soil plus water that passed through the sieve was poured onto the 0.053-mm sieve, and the process was repeated. Three fractions were obtained for each soil sample: macro-aggregates (> 0.25 mm, MA), micro-aggregates (0.25–0.053 mm, MI) and silt + clay (< 0.053 mm, SC). All of these fractions were oven-dried at 40 °C and then weighed. The recovery rate of soil mass, OC, N and P were 98.3%, 100.3%, 102.6% and 102.8%, respectively.

The remaining air-dried, undisturbed soils and dried aggregate samples from each plot were ground to pass through 0.25-mm sieves for the measurement of OC, N and P concentrations. The concentrations of OC, N and P in both bulk soils and aggregate fractions were determined using the Walkley-Black method, the Kjeldahl method, and the sulfuric acid and perchloric acid digestion method (Page et al., 1982), respectively.

### 2.4. Data analysis

The mean weight diameter (MWD) and geometric mean diameter (GMD) were calculated to assess the effects of re-vegetation on soil

structure as (Kemper and Rosenau, 1986):

$$MWD = \sum_{i=1}^{n} x_i \times w_i \qquad (1)$$

$$GMD = \exp\left[\frac{\sum_{i=1}^{n} w_i \times \ln x_i}{\sum_{i=1}^{n} w_i}\right] \qquad (2)$$

where $w_i$ is the weight fraction (%) of the $i$th aggregate fraction and $x_i$ is the mean diameter of each class (mm).

A three-way analysis of variance (ANOVA) was conducted to test the effects of re-vegetation, conversion age and soil depth on (1) the proportions of each aggregate fraction, MWD and GMD and (2) the concentrations of OC, N and P in bulk soils and in each aggregate fraction. An ANOVA was conducted for each re-vegetation pattern. Regression analysis was used to establish the relationships between (1) the concentrations of OC, N and P in bulk soils and those in each aggregate fraction and (2) the changes in the concentrations of OC, N and P (ΔOC, ΔN and ΔP) after re-vegetation and the changes of those in each aggregate fraction. The variance analyses and regression analysis were conducted using JMP 10 (SAS Institute, Cary, USA).

## 3. Results

### 3.1. Effects of re-vegetation on the distribution of soil aggregate fractions

Averaged across all sources of variation, MI dominated the soil mass in the study region, with a proportion of 65% (38–78%), while MA and SC accounted for 16% (5–44%) and 18% (13–24%) of the soil mass, respectively. Re-vegetation significantly increased the proportion of MA and the values of MWD and GMD, but decreased the proportion of MI (Fig. 1, Table 1). These effects varied with the patterns of re-vegetation, with greater percent changes after converting cropland to shrub woodland than to the natural or artificial grassland (Fig. 1). For example, averaged across various re-vegetation ages and soil depths, converting cropland to natural grassland resulted in 43.3%, 33.0% and 19.6% increases in MA, MWD and GMD but an 8.6% decrease in MI, converting to artificial grassland resulted in 36.0%, 26.6% and 13.3% increases in MA, MWD and GMD, but a 6.7% decrease in MI, while converting to shrub woodland resulted in 75.1%, 57.3% and 43.6% increases in MA, MWD and GMD, respectively, but a 10.9% decrease in MI.

The response of aggregate distribution to each re-vegetation pattern varied with age (Fig. 1, Table 1). After converting cropland to natural grassland, the increases in MA, MWD and GMD and decrease in MI were greater after 20 years of re-vegetation than those after 10 years of re-vegetation at the 0–10 and 10–20 cm depths ($P < 0.05$ for R × A). After converting to shrub woodland, the changes were enhanced in the first 20 years of re-vegetation but decelerated in the following 20 years at both depths ($P < 0.05$ for R × A). After converting to artificial grassland, the changes were greater after 15 years of re-vegetation than those after 6, 10 and 20 years of re-vegetation at the 0–10 cm depth but decelerated with the age of re-vegetation at the 10–20 cm depth ($P < 0.05$ for R × A).

### 3.2. Effects of re-vegetation on organic carbon, nitrogen and phosphorous in bulk soils

The responses of OC and N in bulk soils to re-vegetation varied with soil depth (Table 2, Fig. 2). Generally, re-vegetation resulted in an increase of OC and N in bulk soils at the 0–10 cm depth, and the highest increase occurred after 20 years of conversion (Fig. 2). For the 10–20 cm depth, OC and N decreased initially but increased at the later stage after converting to natural grassland and shrub woodland, always decreasing over the 20 years after converting to artificial grassland. However, the responses of OC and N to re-vegetation among conversion ages were not statistically significant at the 10–20 cm depth.

Converting cropland to natural grassland and shrub woodland

**Table 1**

Results of the variance analysis (*P*-values) for the effects of re-vegetation (R), re-vegetation age (A) and soil depth (D) on soil water-stable aggregates in each re-vegetation pattern.

|  | R | A | D | R × A | R × D | A × D | R × A × D |
|---|---|---|---|---|---|---|---|
| **Conversion to artificial grassland** | | | | | | | |
| MA | 0.016 | 0.402 | 0.003 | 0.189 | 0.529 | 0.366 | 0.181 |
| MI | 0.010 | 0.333 | 0.003 | 0.178 | 0.640 | 0.646 | 0.339 |
| SC | 0.079 | 0.426 | 0.081 | 0.439 | 0.909 | 0.458 | 0.719 |
| MWD | 0.017 | 0.406 | 0.003 | 0.190 | 0.526 | 0.358 | 0.177 |
| GMD | 0.028 | 0.324 | 0.004 | 0.158 | 0.330 | 0.176 | 0.094 |
| **Conversion to natural grassland** | | | | | | | |
| MA | 0.023 | 0.087 | 0.179 | 0.140 | 0.833 | 0.458 | 0.784 |
| MI | 0.046 | 0.546 | 0.165 | 0.396 | 0.654 | 0.635 | 0.773 |
| SC | 0.502 | 0.012 | 0.257 | 0.262 | 0.338 | 0.702 | 0.806 |
| MWD | 0.023 | 0.080 | 0.180 | 0.134 | 0.841 | 0.452 | 0.784 |
| GMD | 0.009 | 0.006 | 0.168 | 0.025 | 0.948 | 0.252 | 0.667 |
| **Conversion to pea shrub woodland** | | | | | | | |
| MA | 0.005 | 0.101 | 0.153 | 0.650 | 0.784 | 0.731 | 0.814 |
| MI | 0.027 | 0.030 | 0.050 | 0.697 | 0.695 | 0.583 | 0.677 |
| SC | 0.011 | < 0.001 | < 0.001 | 0.867 | 0.455 | 0.214 | 0.282 |
| MWD | 0.004 | 0.105 | 0.158 | 0.648 | 0.788 | 0.737 | 0.819 |
| GMD | 0.003 | 0.322 | 0.248 | 0.822 | 0.637 | 0.827 | 0.967 |

MA: macro-aggregate; MI: micro-aggregate; SC: silt + clay fraction; MWD: mean weight diameter of water stable aggregates; GMD: geometric mean diameter of water stable aggregates.

resulted in an initial decrease and later increase of P in bulk soils at both the 0–10 and 10–20 cm depths (Fig. 2). Converting to artificial grassland resulted in an increase in P at the 0–10 cm depth in most ages, with the highest increase after 15 years of conversion. However, this re-vegetation did not affect soil P at the 10–20 cm depth, except for 15 years of re-vegetation where soil P was significantly increased (Fig. 2).

The effects of re-vegetation on soil OC and N in the 0–10 cm soils varied with conversion patterns (Fig. 2). The higher increase in soil OC was observed after converting cropland to natural grassland and shrub woodland, while the higher increase in N was observed after converting cropland to shrub woodland and artificial grassland. The effect of re-vegetation on soil P was similar among the three conversion patterns.

### 3.3. Effects of re-vegetation on organic carbon, nitrogen and phosphorous in aggregate fractions

The re-vegetation resulted in significant increases in the concentrations of OC and N associated with aggregates at the 0–10 cm depth but did not affect them at the 10–20 cm depth (Figs. 3 and 4, Table 2), and these effects varied with the aggregate fraction, re-vegetation pattern and conversion age. For example, the increases in OC and N were higher in MA ($+2.82$ and $+0.28\,\mathrm{g\,kg^{-1}}$) than those in MI ($+0.72$ and $+0.07\,\mathrm{g\,kg^{-1}}$) and SC ($+0.17$ and $+0.06\,\mathrm{g\,kg^{-1}}$) when averaged across re-vegetation patterns and ages. The increases in OC and N in MA and SC were greater after converting cropland to shrub woodland and artificial grassland than converting to natural grassland.

The responses of OC and N in aggregates to re-vegetation age varied with the re-vegetation pattern and soil depth. After converting cropland to natural grassland, the increases in OC and N were only observed in MA at the 0–10 cm depth after the first 10 years of conversion and were observed in all aggregate fractions and soil depths after 20 years of conversion. After converting to shrub woodland and artificial grassland, OC and N in each aggregate fraction increased at the 0–10 cm depth but were not affected at the 10–20 cm depth after conversion. Furthermore, these effects increased over time after converting to natural grassland, declined over time after converting to shrub woodland, but did not vary with age after converting to artificial grassland (Figs. 3 and 4). The concentrations of P associated with aggregates were not affected by re-vegetation, except for the P in MA, which increased after converting to artificial grassland ($+11.66\%$, $P = 0.04$), and in MI, which decreased

**Table 2**
Results of the variance analysis (*P*-values) for the effects of re-vegetation (R), re-vegetation age (A) and soil depth (D) on soil properties in each re-vegetation pattern. The three-way interaction was not provided.

| | Conversion to artificial grassland | | | | | | Conversion to natural grassland | | | | | | Conversion to pea shrub woodland | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | R | A | D | R×A | R×D | A×D | R | A | D | R×A | R×D | A×D | R | A | D | R×A | R×D | A×D |
| OC | 0.001 | 0.418 | < 0.001 | 0.929 | 0.015 | 0.984 | 0.023 | 0.088 | < 0.001 | 0.449 | 0.354 | 0.644 | 0.002 | 0.541 | < 0.001 | 0.196 | 0.001 | 0.637 |
| N | 0.005 | 0.179 | < 0.001 | 0.820 | 0.027 | 0.710 | 0.135 | 0.215 | < 0.001 | 0.664 | 0.238 | 0.855 | 0.005 | 0.020 | < 0.001 | 0.278 | 0.008 | 0.475 |
| P | 0.049 | 0.010 | 0.007 | 0.765 | 0.587 | 0.795 | 0.010 | 0.474 | 0.012 | 0.817 | 0.971 | 0.994 | 0.707 | 0.069 | 0.053 | 0.298 | 0.839 | 0.813 |
| OC-MA | < 0.001 | 0.703 | < 0.001 | 0.977 | < 0.001 | 0.171 | 0.021 | 0.395 | < 0.001 | 0.567 | 0.043 | 0.182 | < 0.001 | 0.026 | < 0.001 | 0.112 | < 0.001 | 0.020 |
| OC-MI | 0.001 | 0.368 | < 0.001 | 0.876 | < 0.001 | 0.713 | 0.098 | 0.184 | 0.001 | 0.340 | 0.226 | 0.925 | 0.003 | 0.350 | < 0.001 | 0.193 | < 0.001 | 0.938 |
| OC-SC | 0.363 | 0.513 | < 0.001 | 0.716 | 0.016 | 0.937 | 0.823 | 0.011 | < 0.001 | 0.540 | 0.686 | 0.949 | 0.100 | 0.876 | < 0.001 | 0.069 | 0.009 | 0.413 |
| N-MA | < 0.001 | 0.155 | < 0.001 | 0.835 | < 0.001 | 0.515 | 0.023 | 0.454 | < 0.001 | 0.878 | 0.055 | 0.414 | < 0.001 | 0.288 | < 0.001 | 0.034 | < 0.001 | 0.250 |
| N-MI | 0.005 | 0.225 | < 0.001 | 0.802 | 0.002 | 0.612 | 0.124 | 0.195 | 0.009 | 0.780 | 0.346 | 0.721 | 0.044 | 0.078 | < 0.001 | 0.187 | 0.022 | 0.505 |
| N-SC | 0.009 | 0.149 | < 0.001 | 0.829 | 0.188 | 0.615 | 0.417 | 0.094 | < 0.001 | 0.332 | 0.983 | 0.823 | 0.084 | 0.249 | < 0.001 | 0.043 | 0.359 | 0.941 |
| P-MA | 0.090 | 0.292 | 0.037 | 0.850 | 0.571 | 0.966 | 0.122 | 0.789 | 0.049 | 0.487 | 0.629 | 0.638 | 0.915 | 0.139 | 0.055 | 0.207 | 0.522 | 0.900 |
| P-MI | 0.948 | 0.259 | 0.641 | 0.522 | 0.300 | 0.668 | 0.735 | 0.189 | 0.797 | 0.092 | 0.915 | 0.387 | 0.363 | 0.024 | 0.887 | 0.022 | 0.583 | 0.940 |
| P-SC | 0.342 | 0.089 | 0.189 | 0.766 | 0.226 | 0.521 | 0.154 | 0.518 | 0.049 | 0.155 | 0.715 | 0.791 | 0.694 | 0.022 | 0.598 | 0.410 | 0.388 | 0.839 |

OC, N and P: concentrations of organic carbon, nitrogen and phosphorous in bulk soils; OC-MA, OC-MI and OC-SC: concentrations of organic carbon in macro-aggregate (MA), micro-aggregate (MI) and silt + clay (SC) fractions, respectively; N-MA, N-MI and N-SC: concentrations of nitrogen in MA, MI and SC fractions, respectively; P-MA, P-MI and P-SC: concentrations of phosphorous in MA, MI and SC fractions, respectively.

after converting to natural grassland ($-13.65\%$, $P = 0.02$) (Fig. 5).

In this study, the OC, N and P in each aggregate fraction and their responses to re-vegetation were significantly correlated with OC, N and P in bulk soils (Tables 3 and 4). The MA-associated OC and N had a greater contribution (slopes of the relationships in Tables 3 and 4) to OC and N in bulk soils after re-vegetation than MI- and SC-associated OC and N, while each aggregate fraction-associated P had a similar contribution to P in bulk soils after re-vegetation (Tables 3 and 4).

## 4. Discussion

Our results demonstrated that all three re-vegetation patterns increased OC and N at the 0–10 cm depth, and the accumulation in bulk soils was mainly due to the accumulation in MA, supporting H1. The conversion of cropland to artificial grassland or woodland (both are legume plants) resulted in significant increases in OC and N in each aggregate fraction in surface soils, supporting H2. However, most effects of re-vegetation differed with re-vegetation ages and were highest at approximately 20 years after conversion, rejecting H3.

### 4.1. The effects on soil aggregates

In this study, MI dominated the soil mass, which was determined by the soil texture. The high content of sand ($72 \pm 4\%$) in the studied soils prevents the aggregation of soil particles and thus resulted in the lower proportion of MA. This could be supported by the negative relationship between sand and MA ($r = -0.601$, $P < 0.0001$) and the positive relationship between sand and MI ($r = 0.688$, $P < 0.0001$) in our study. This result was consistent with observations from van Der Heijden et al. (2006), in which soil fractions of < 1 mm dominated soil mass ($> 70\%$) in a loamy sand soil, and from Gao et al. (2013), in which MI dominated the soil mass in aspen (*Populus simonii* Carr.)
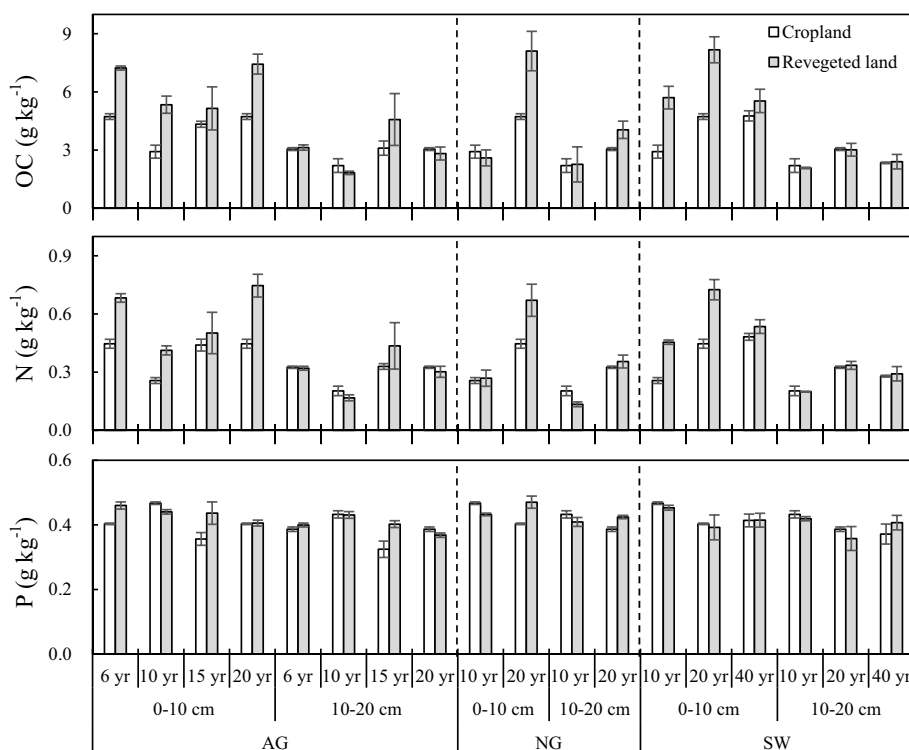


**Fig. 2.** Concentrations of organic carbon (OC), nitrogen (N) and phosphorous (P) in bulk soils for each age and pattern of re-vegetation at the 0–10 and 10–20 cm depths. AG: converting cropland to artificial grassland; NG: converting cropland to natural grassland; SW: converting cropland to pea shrub woodland. Error bars were two standard errors of the mean.
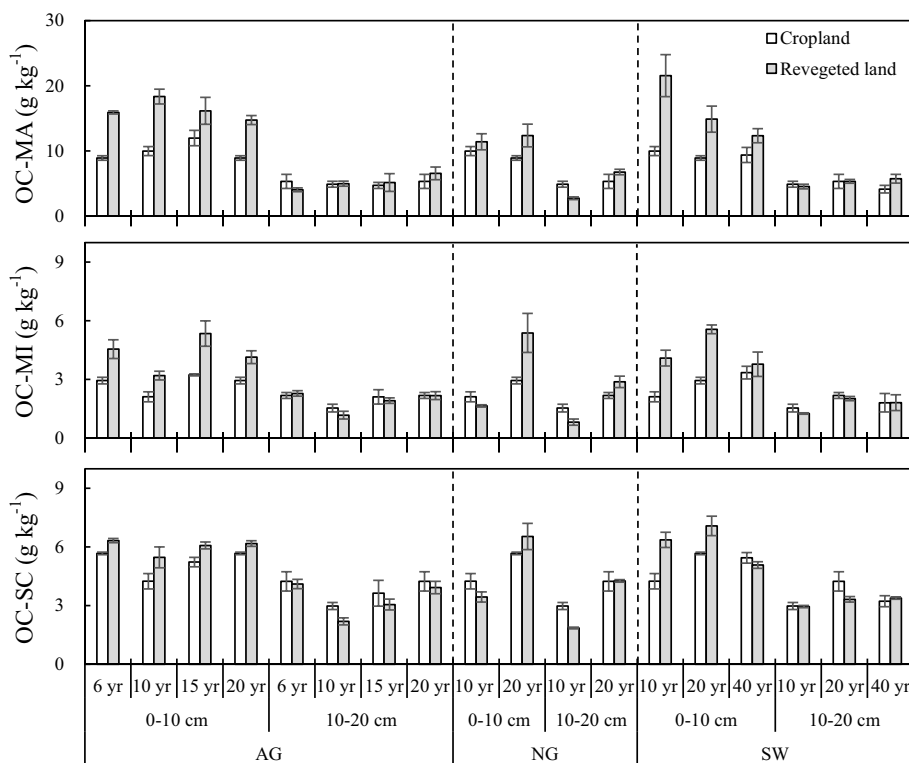
**Fig. 3.** Concentrations of organic carbon (OC) in each aggregate fraction for each age and pattern of re-vegetation at the 0–10 and 10–20 cm depths. AG: converting cropland to artificial grassland; NG: converting cropland to natural grassland; SW: converting cropland to pea shrub woodland. MA: macro-aggregates, MI: micro-aggregates, SC: silt + clay fraction. Error bars were two standard errors of the mean.

woodlands in semi-arid loamy and sandy soils. Furthermore, the significant positive correlation of sand to MI suggests that the MI measured in this study might be the sand per se, rather than a micro-aggregate. In this case, the physical protection of MI by soil organic matter might be smaller than that of MA.

The increases in the MA, MWD, and GMD and the decrease in MI after re-vegetation were the results of the enhanced aggregation of MI and SC into MA and thus the improvement of soil structure. The

enhanced aggregation and the improvement in soil structure are caused by the direct effect of fine roots and by the indirect effect of the association of the roots with external hyphae (Jastrow et al., 1998; Cheng et al., 2015). Previous studies in the same region as this study indicated the large amount of root biomass for the three species used here (Cheng et al., 2007; Su et al., 2017), supporting this explanation. On the other hand, increased OC concentration after re-vegetation (Fig. 2) could also contribute to aggregation of soil particles as evidenced by the positive
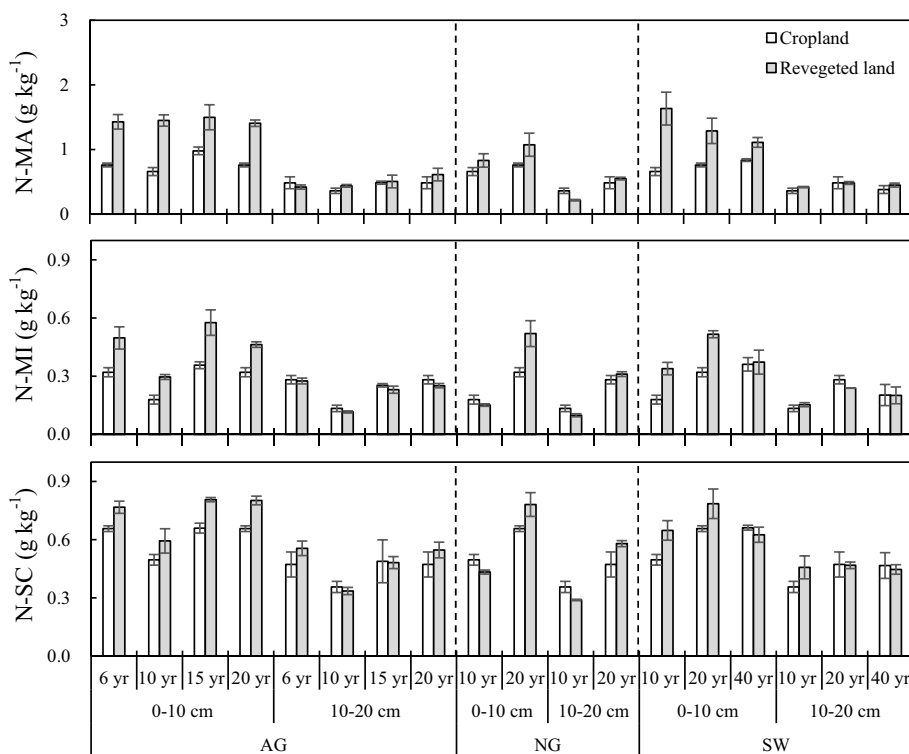


**Fig. 4.** Concentrations of nitrogen (N) in each aggregate fraction for each age and pattern of re-vegetation at the 0–10 and 10–20 cm depths. AG: converting cropland to artificial grassland; NG: converting cropland to natural grassland; SW: converting cropland to pea shrub woodland. MA: macro-aggregates, MI: micro-aggregates, SC: silt + clay fraction. Error bars were two standard errors of the mean.
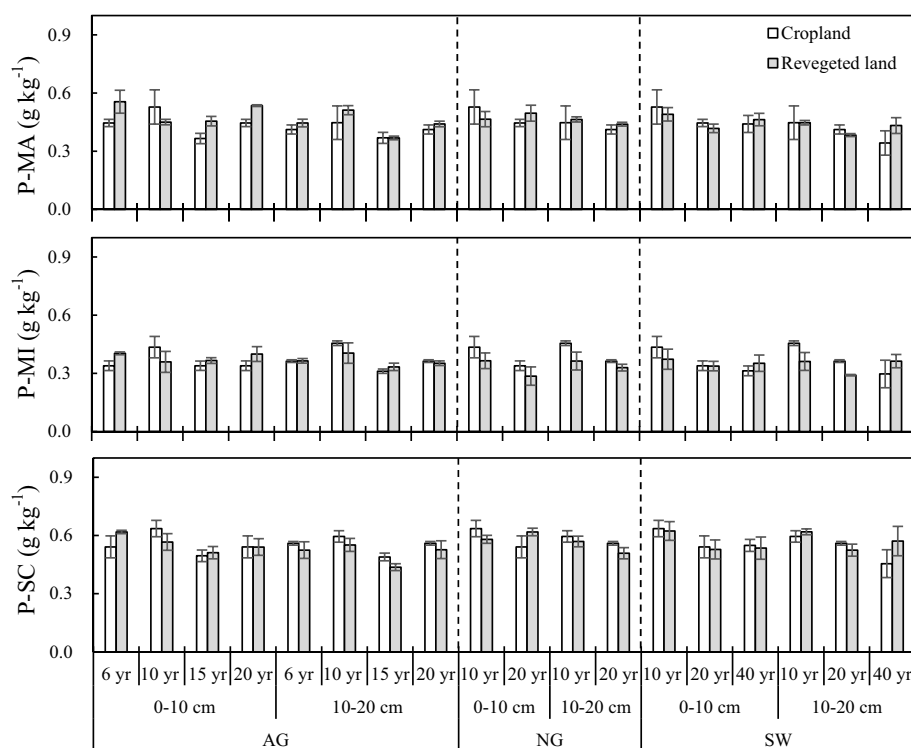
**Fig. 5.** Concentrations of phosphorous (P) in each aggregate fraction for each age and pattern of revegetation at the 0–10 and 10–20 cm depths. AG: converting cropland to artificial grassland; NG: converting cropland to natural grassland; SW: converting cropland to pea shrub woodland. MA: macro-aggregates, MI: micro-aggregates, SC: silt + clay fraction. Error bars were two standard errors of the mean.

**Table 3**

Relationships ($y = a \times x + b$) between organic carbon, nitrogen and phosphorous in aggregates and those in bulk soils across the re-vegetation patterns, ages and soil depths.

| X | Y | a | SE(a) | b | SE(b) | $R^2$ | RMSE | n | P |
|---|---|---|---|---|---|---|---|---|---|
| OC | OC-MA | 1.68 | 0.17 | 1.93 | 0.77 | 0.44 | 3.55 | 120 | < 0.001 |
| | OC-MI | 0.60 | 0.03 | 0.32 | 0.14 | 0.74 | 0.66 | 120 | < 0.001 |
| | OC-SC | 0.59 | 0.04 | 2.08 | 0.18 | 0.65 | 0.81 | 120 | < 0.001 |
| N | N-MA | 1.75 | 0.15 | 0.07 | 0.06 | 0.52 | 0.27 | 120 | < 0.001 |
| | N-MI | 0.69 | 0.04 | 0.02 | 0.02 | 0.74 | 0.07 | 120 | < 0.001 |
| | N-SC | 0.75 | 0.05 | 0.26 | 0.02 | 0.68 | 0.08 | 120 | < 0.001 |
| P | P-MA | 0.14 | 1.22 | − 0.05 | 0.06 | 0.39 | 0.06 | 120 | < 0.001 |
| | P-MI | 0.13 | 0.75 | 0.05 | 0.05 | 0.22 | 0.06 | 120 | < 0.001 |
| | P-SC | 0.13 | 1.14 | 0.08 | 0.05 | 0.40 | 0.06 | 120 | < 0.001 |

OC, N and P: concentrations of organic carbon, nitrogen and phosphorous in bulk soils; MA, MI and SC: macro-aggregates, micro-aggregates and silt + clay fraction, respectively. **a** and **b**: slope and intercept of the linear relationship; SE: standard error of slope or intercept of the relationship; RMSE: root mean square error.

**Table 4**

Relationships ($Y = a \times x + b$) between changes in organic carbon, nitrogen and phosphorous in aggregates after re-vegetation and changes in OC, N and P in bulk soils across the re-vegetation patterns, ages and soil depths.

| X | Y | a | SE(a) | b | SE(b) | $R^2$ | RMSE | n | P |
|---|---|---|---|---|---|---|---|---|---|
| ΔOC | ΔOC-MA | 1.28 | 0.29 | 1.29 | 0.58 | 0.25 | 3.58 | 60 | < 0.001 |
| | ΔOC-MI | 0.50 | 0.07 | 0.11 | 0.14 | 0.47 | 0.88 | 60 | < 0.001 |
| | ΔOC-SC | 0.40 | 0.07 | − 0.31 | 0.13 | 0.39 | 0.81 | 60 | < 0.001 |
| ΔN | ΔN-MA | 1.47 | 0.29 | 0.15 | 0.05 | 0.31 | 0.30 | 60 | < 0.001 |
| | ΔN-MI | 0.51 | 0.08 | 0.02 | 0.01 | 0.42 | 0.08 | 60 | < 0.001 |
| | ΔN-SC | 0.43 | 0.09 | 0.02 | 0.01 | 0.28 | 0.09 | 60 | < 0.001 |
| ΔP | ΔP-MA | 1.01 | 0.24 | 0.01 | 0.01 | 0.24 | 0.10 | 60 | < 0.001 |
| | ΔP-MI | 0.82 | 0.19 | − 0.02 | 0.01 | 0.24 | 0.08 | 60 | < 0.001 |
| | ΔP-SC | 1.06 | 0.21 | − 0.01 | 0.01 | 0.31 | 0.09 | 60 | < 0.001 |

ΔOC, ΔN and ΔP: changes in concentrations of organic carbon, nitrogen and phosphorous after re-vegetation; MA, MI and SC: macro-aggregates, micro-aggregates and silt + clay fraction, respectively. **a** and **b**: slope and intercept of the linear relationship; SE: standard error of slope or intercept of the relationship; RMSE: root mean square error.

correlation between soil OC and MWD (MWD = 0.632 + 0.034 × OC, $P = 0.038$). These results were consistent with observations in other regions (Wei et al., 2013; Qiu et al., 2015), but the extent of the improvement was relatively lower in this study than others due to the lower input of organic materials and the sandy texture of soils.

We observed that the response of aggregate distributions and soil structure to re-vegetation varied with conversion patterns and ages (Fig. 1), probably due to the differences in the changes of soil moisture and plant productivity and thus the suitability of the newly established ecosystem. The plantation of pea shrub and alfalfa in the study region resulted in a great depletion of the soil water reservoir (Li and Huang, 2008), causing the soil moisture to always be below the wilting point (thus the dried soil layer) at the later stage of pea shrub or alfalfa (Li and Huang, 2008; Liu and Shao, 2014; Jia et al., 2017). The over-depletion of the soil water reservoir decelerated the accumulation of above and belowground biomass (Jia et al., 2017) because the ecosystem in the study region is mostly limited by the availability of soil moisture (Li and Huang, 2008). However, the dried soil layer was not

found in the natural grassland (Liu and Shao, 2014; Jia et al., 2017), and the above and belowground biomass always increased with the age of the natural grassland (Jia et al., 2017). Therefore, natural or native vegetation might be an option for re-vegetation to improve the soil structure in this transition region.

### 4.2. The effects on organic carbon and nitrogen in bulk soils

The increases in soil OC and N after re-vegetation could be ascribed to the return of organic materials to the soil as plant litter, dead roots, and root exudates, which results in an increase in soil OC and N pools (Cleveland et al., 2004). However, our results showed that the accumulation of OC and N mainly occurred in surface soils (0–10 cm), indicating that subsurface soils (10–20 cm) respond slowly to land-use change. The lack of the response of OC and N concentrations in the 10–20 cm depth could also be related to the relatively low precipitation in the study region, which led to little transportation of OC and N from

topsoil to deep soil. This explanation was supported by our previous observation in a wetter site, in which the conversion of cropland to forest resulted in a significant increase in the soil OC and N associated with each aggregate at both the 0–10 and 10–20 cm depths (Qiu et al., 2015). An alternative explanation is that the low precipitation and temperature in this study lead to less decomposition and thus less incorporation of OC and N with soil particles; hence, there was less of an increase in OC and N at both the 0–10 and 10–20 cm depths (although the increase at the 0–10 cm depth was statistically significant in this study) compared with our previous observations in a wetter and warmer site (Qiu et al., 2015).

We showed that the increases in OC and N in topsoils were highest at approximately 20 years after re-vegetation, probably because the release of OC and N from litter returned to the soils at a maximum after ca. 20 years after conversion. In the study region, the biomass accumulations of grasses and semi-shrubs were highest at 8 to 10 years after conversion, while it will take 7 to 15 years for these litters to decompose (Wu et al., 2013; Cheng, 2014). However, our results was inconsistent with observation by Parfitt et al. (2013) that OC and N continuously accumulated within 70 years in the grass-legume pastures of New Zealand. This difference might be due to the extra input of OC and N from grazers and higher mean annual precipitation (600–1500 mm) in their study than this study (437 mm). Furthermore, the higher increases of soil OC were observed in natural grassland and pea shrub woodland than in the artificial grassland, primarily due to the higher biomass of the natural grassland and the greater increase of MA and the improvement in soil structure (higher increase in MWD and GMD) in pea shrub woodland, which accelerated the input of organic material into soils and the incorporation of OC with soil particles (Wei et al., 2013). The higher increase in soil N in pea shrub woodland and artificial grassland was due to the higher N concentrations in organic materials from pea shrubs and alfalfa, which have the capacity to fix N from the atmosphere and thus result in higher N input into soils.

### 4.3. The effects on organic carbon and nitrogen in soil aggregates

The conversion of cropland to artificial grassland or woodland significantly increased the concentrations of OC and N in each aggregate fraction at the 0–10 cm depth. This increase could be ascribed to the accumulation of organic materials from litter, root exudates and dead roots, which release OC and N during decomposition (Six et al., 2000; Ayoubi et al., 2012), and thus, they are incorporated with soil particles. The increases in OC and N concentrations in MA could also be attributed to the aggregation of soil particles. This process not only uses organic materials as binding agents and thus results in the accumulation of organic matter in MA (Wei et al., 2013; Qiu et al., 2015) but also physically protects the organic matter occluded into the MA from the contact with microbes and oxygen and thus decreases the loss of OC and N from decomposition (Zimmermann et al., 2012; O'Brien and Jastrow, 2013). The accumulation of organic matter within MA and the physical protection posed by MA for organic matter also provides an explanation for that the increases in OC and N concentrations were higher in MA than MI and SC as observed in this study (Figs. 3 and 4). The increases in OC and N in the SC fraction could be attributed to the chemical combination or adsorption of newly input organic material with clay or silt particles (Caravaca et al., 2004). However, the re-vegetation did not affect OC and N associated with aggregates at the 10–20 cm depth, primarily due to the relatively low precipitation in the study region, as discussed in the previous section.

The increases in concentrations of OC and N associated with aggregates were greater after converting cropland to shrub woodland and artificial grassland than to natural grassland, probably due to the inclusion of legumes in the woodland and artificial grassland. The organic materials derived from legumes have high N contents (Parr et al., 2011), which increased the N concentration in each aggregate fraction. Furthermore, legume-derived organic materials have a low C/N ratio, which favors the decomposition of organic materials (Migliorati et al., 2015); hence, there were greater accumulations of OC in bulk soils and aggregates.

### 4.4. The effects on phosphorous in bulk soils and aggregates

In this study, the decrease in soil P in bulk soils at the initial stage of re-vegetation (Fig. 2) was mainly due to the depletion of P by plant uptake, while the increase in the later stage was due to the return of P from plant litter and roots (Stevenson and Cole, 1999). These results suggest that soil P is more inert than soil OC and N in responding to land-use change in this agro-pastoral ecotone because P release during decomposition of organic materials is less than that of OC and N (Manzoni et al., 2010). Our observation was in line with early result from Yang et al. (2012) that the conversion of the natural forestland to cultivated land moderately increased the total P (from 770 mg kg$^{-1}$ to 1014 mg kg$^{-1}$) in the 0–20 cm soil layer in the Sanjiang Plain of China. This reason could also provide an explanation for our observation that the effect of re-vegetation on soil P was not responsive to conversion patterns.

We found that P in most aggregates was not affected by re-vegetation, primarily due to the relatively slow turnover rate of P in the ecosystem (Cardoso and Kuyper, 2006). The distribution of P among soil aggregates and its response to land-use change were less reported compared with those of OC and N; therefore, further research regarding this issue should be conducted to obtain a general pattern. Although soil P in cropland did not change over time in this study (see the Materials and Methods section), the application of chemical P fertilizers usually results in an increase of P in soils (Lehmann et al., 2001; Lekberg and Koide, 2005), and using cropland as a control would underestimate the effects of re-vegetation on soil P. Therefore, the effects of P fertilizer should be explicitly disentangled when using this approach to assess the effects of cropland-based land-use change on soil P.

## 5. Conclusions

In this study, we assessed the changes in OC, N and P in bulk soils and water-stable aggregates following conversion of cropland to artificial and natural grasslands and pea shrub woodland in an agro-pastoral ecotone of northern China. We found that re-vegetation significantly improved the soil structure and increased OC and N in both bulk soils and aggregates but had minimal effects on soil P. The increases in OC and N were greater after conversion to legume vegetation than non-legume vegetation and were highest approximately 20 years after conversion. Furthermore, the changes in soil OC and N after re-vegetation were mainly due to the changes in macro-aggregates. Therefore, any re-vegetation practice would have the potential to increase OC and N sequestration in this agro-pastoral ecotone, and an appropriate management approach should be recommended at the right conversion ages (ca. 20 years) to maintain the maximum effect on soil OC and N.

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