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# Changes in above- and below-ground nitrogen stocks and allocations following the conversion of farmland to forest in rocky desertification regions

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### ABSTRACT

Afforestation of degraded land is one of the principal strategies for preventing soil erosion and promoting ecosystem recovery in fragile regions, especially in rocky desertification areas. In China, millions of hectares of farmland have been converted into forest in order to arrest and reverse rocky desertification under the Grain for Green Program (GGP). This study evaluated implications of land-use change from annual maize cultivation to perennial Zanthoxylum bungeanum plantations (1-, 4-, 7-, and 10-year-old) in the karst region of Guizhou province, southwest China. The study analyses the variations of biomass and nitrogen (N) storages as well as N distributions in biomass components and soil depths. Results showed that the N content in components of Z. bungeanum ranged from 0.31% to 3.24% with a mean value of 1.75%, which was lower than that of maize (2.13%) in the same region. The biomass N storage measured for the maize cropland was 210.59 kg ha<sup>-1</sup>, while this value increased linearly with stand ages for the four Z. bungeanum plantations (0.94, 108.31, 212.20, and 262.12 kg ha<sup>-1</sup>, respectively). The average amount of soil N storage in the Z. bungeanum plantations (9.33 t ha<sup>-1</sup>) was significantly lower than in the adjacent intensively managed maize cropland (10.04 t ha<sup>-1</sup>). This is mainly due to long-term organic and inorganic fertilizer inputs in the farmland stage. Total ecosystem N storage averaged 10.25 t ha<sup>-1</sup> in the maize cropland, and 9.38, 9.82, 9.05, and 9.67 t ha<sup>-1</sup> in the 1-, 4-, 7- and 10-year-old Z. bungeanum plantations, respectively. Soils accounted for 97% of total ecosystem N storage in both land-use systems. This study suggests that the reduction of surface soil disturbance during plantation management practices plays a crucial role in improving the N storage. Data of annual plantation area and biomass N accumulation rates under the GGP indicate that Guizhou province was a net N sink with  $2.35 \times 10^8$  kgN in 2010, corresponding to 41.45% of N (NO<sub>X</sub>-N and NH<sub>3</sub>-N) emissions in that year. Besides increasing N sequestration over time (as these forest mature), the large-scale plantations of Z. bungeanum have the potential to restore severely degraded soils in the karst region of SW China.

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# 1. Introduction

Rocky desertification, a process characterized by serious soil erosion and widespread bedrock exposure, is one of the most serious land degradation issues in karst areas (Xiong et al., 2009). Rocky desertification in southern China and sandy desertification in northern China are two major environmental problems that hamper local sustainable development (Wang and Li, 2007). In

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http://dx.doi.org/10.1016/j.agee.2016.07.019 0167-8809/© 2016 Elsevier B.V. All rights reserved. response to these environmental crises, a range of policies have been developed to support forest management strategies aimed at the prevention of land degradation and the creation of new forests, mainly on barren, degraded or former agricultural land (Madsen, 2002; Smith et al., 2000). Therefore, a nationwide Grain for Green Program (GGP), also known as the Conversion of Cropland to Forest and Grassland Program (CCFGP) (Ostwald et al., 2007), was initiated by the Chinese government in 1999. The GGP is the largest ecological restoration project in China with the intention of preventing further soil erosion, controlling desertification, and improving land quality. In practice, the GGP helped to restore the ecological functionality in degraded areas by returning cultivated slopes and barren lands to pastures and forests. Over the past ~10 years, the replacement of annual crops by perennial plants has led to a marked improvement of the natural environment. According to the Annual Reports on the Development of Chinese Forestry,  $2.34 \times 10^7$  ha of lands, including  $8.27 \times 10^6$  ha of croplands and  $1.51 \times 10^7$  ha of barren lands, have been planted under the GGP during the period of 1999–2010 (State Forestry Administration, 1999–2011). Therefore, the broad areal extent of GGP in the degraded region of China will lead to an increase in new forests and ultimately contribute to improvements in regional biogeochemical cycles.

The impact of afforestation and reforestation on carbon (C) and nitrogen (N) cycles, has garnered significant attention in recent years (Fortier et al., 2015; Li et al., 2012; Ritter, 2007). A shift in land-use from agriculture to forestry induces major changes in the N cycle, including inputs, internal cycling, and losses. The N cycle in agricultural soils is characterized by an open cycle. N fertilizer is supplied regularly in large amounts and approximately the same amount of N leaves the ecosystem via leaching or harvested crops. However, the soil properties of former cropland after afforestation are slowly modified towards conditions found in closed forests because of a lack of the intense cultivation (high fertilization, annual tillage, weed control, etc.) as well as due to the effect of trees themselves (Zhang et al., 2012). In the initial phase of afforestation, plantations on former cultivated land have higher N status as a legacy of former fertilization, which supports continued high mineralization and nitrification in the mineral soil (Jug et al., 1999). Even a century after afforestation, soils in plantations on former cropland have N-cycling characteristics that differ significantly from those of nearby soils with unbroken forest cover (Compton et al., 1998; Jussy et al., 2002). Mobilized N can be fixed in the biomass of above- and below-ground vegetation for a longer period of time. Namely, plant biomass after afforestation might act as an important sink for N and lead to an overall increase in N retention within the system (Heilman et al., 1995). Moreover, afforestation of agricultural land is likely to have a large effect on the N equilibrium, resulting in changing flows of N from plant biomass fractions and soil organic matter pools. Therefore, the magnitude and duration of the net gains or losses of stored N in different stand-aged forest and the effects of the change in landuse on N cycling is of particular interest (Hansen, 2002). Despite the importance of these changes in the N stocks and cycles following the conversion of agriculture to forestry, the effects of widely applied afforestation remains unclear. Furthermore, little is known about changes in N stocks and distributions in former framlands and recently converted forests under the GGP, particularly in the karst region.

Located in the central karst regions of southwest China, Guizhou province is subject to severe rocky desertification. In 2000, approximately  $3.25 \times 10^4$  km<sup>2</sup> of the province had outcrops of carbonate rocks, making up 17.10% of its total land area (Jiang et al., 2014). To prevent soil erosion and restore ecosystem functionality, Guizhou is high on the list of provinces for implementation of the GGP and provides a particularly good context for evaluating the influence of land-use change on the dynamics of biomass and N stocks in this area. By the end of 2011,  $1.20 \times 10^6$  ha of land in Guizhou province, including  $4.39 \times 10^5$  ha of steep cropland and  $7.60 \times 10^5$  ha of barren land has been planted with trees under the GGP, accounting for 4.97% of the national GGP-lands (including 5.31% of total steep croplands and 4.80% of total barren lands, respectively) (State Forestry Administration, 1999–2011). It is expected that  $7.03 \times 10^5$  ha of steep croplands (>25°) will be converted to forests during the period from 2014 to 2020 (Guizhou Provincial Forestry Department, 2014). Therefore, the large-scale of actual and potential implementation of the GGP in the karst region of Guizhou province involves a shift from this high level of human interference in the annual cycle of cultivating and harvesting crops to a lower level interference in a much longer forest cycle (Hansen, 2002). This process will lead to an expansion of new forests and will ultimately change regional N cycle as well as reduce of N leaching over the long term. An improved quantification of the change in biomass and N pool allocation is fundamental to the understanding of the effect of land-use and land cover change on ecosystem function at a regional and national scale. This understanding is required for scientists, managers, and policy makers to play their roles in the implementation of the GGP.

In this study, we examined the changes of vegetation biomass, ecosystem (plant and soil) N storages, and allocations following the conversion of farmland (*Zea mays* L.) to forestland (1-, 4-, 7-, and 10-years old *Z. bungeanum*) under the GGP in the Hua Jiang Karst Canyon of Guizhou province. The focus of this study was to: 1) compare the changes of above- and below-ground biomass between cropland and forest; 2) quantify the N stocks in plant and soil in both before and after land-use changes; and 3) implicate the N pools by restoring the degraded karst regions under the GGP in Guizhou province.

# 2. Materials and methods

# 2.1. Site description

The study site is situated on the Hua Jiang Karst Canyon (HJKC) of Guizhou province, southwest China, located in the range of 25°39.2′–41′N latitude and 105°36.5′–46.5′E latitude. The total area of HJKC is 51.62 km<sup>2</sup>, of which 87.92% is karst. This area has been considered as a typical representative of karst region in SW China. The terrain, soil type, and climatic condition in this region are described in more detail by Cheng et al. (2015).

The landscape in the HJKC is characterized by cultivated land and recently converted forest land. The cultivated land is mainly upland maize (No. 778 Andan) field, and the area of cultivated land is about 1051.0 ha (Gao and Xiong, 2015). In the process of maize cultivation, the field will receives significant amounts of farmyard manure (15,000 kg ha<sup>-1</sup>) and multi-element compound fertilizer  $(750 \text{ kg ha}^{-1})$ , which were used as base fertilizers. Additionally, urea is applied to the field as topdressing fertilizers during seedling and huge bellbottom periods at 150 kg  $ha^{-1}$  and 225 kg  $ha^{-1}$  levels, respectively. However, the use of nitrogen fertilizer is markedly reduced and sometimes ceased altogether following the conversion of cropland to forest under the GGP. The recently converted forest is mainly consist of Z. bungeanum with an area of 1786.9 ha (Li et al., 2010). Z. bungeanum is a suitable species for plantation/ afforestation to restore the rocky desertification, due to its fast growth and adaptability to poor habitat, drought tolerance, and calcareous soil. Furthermore, the fruit of the Z. bungeanum tree can be sold to supplement the income of farmers in poor karst areas of Guizhou. Indeed, the HJKC has become a demonstration of how rocky desertification may be arrested under the GGP in Guizhou province and all over China, because ecological restoration under the GGP in this region increased vegetation coverage from 21.4% in 2001 to 53.2% in 2005 (Chen et al., 2007).

#### 2.2. Biomass estimation

Because most of the land was discontinuous in rocky desertification regions, the area of the sampled fields was relatively small. It was about 0.10 ha for the maize field and approximately 0.25 ha for the *Z*. *bungeanum* plantation. Biomass of the maize was measured by placing three random one-meter-square  $(1 \text{ m} \times 1 \text{ m})$  quadrats (blocks) in the fields, and the height (H) and circumference at the base of the stem (CBS) of each plant were measured in the quadrats (Table 1). Total biomass of maize was measured by cutting,

#### Table 1

Stand characteristics following the conversion of cropland (Zea mays L.) to forest (Z. bungeanum) (mean  $\pm$  SD).

Characteristics	Years afforested					
	0 (cropland)	1	4	7	10	
Н	$2.25\pm0.22$	$\textbf{0.67} \pm \textbf{0.08}$	$2.95\pm0.22$	$3.12\pm0.47$	$3.12\pm0.47$	
CBS	$10.25\pm2.33$	$\textbf{2.77} \pm \textbf{0.46}$	$17.53\pm3.16$	$24.73 \pm 0.87$	$26.58\pm3.76$	
CD	none	$0.42\pm0.10$	$\textbf{3.58} \pm \textbf{0.43}$	$4.68\pm0.50$	$4.81 \pm 1.05$	
D	40,000	1665	1250	950	850	

H: height of plant (m), CBS: circumference at basic stem (cm), CD: crown diameter (m), D: density of plantation (plants ha<sup>-1</sup>).

excavating, drying and weighing all the plant materials within the squares. Above-ground biomass of maize was divided into three components (leaf, stem, and fruit). After cutting off their aboveground parts, the root of maize was sampled in a cube of 100 cm (length, distance between rows of plants)  $\times$  25 cm (width, distance between plants within a row)  $\times$  80 cm (depth). Fresh weights of each component were weighed separately in the field for every quadrat, and subsamples for each component were collected for moisture content and N concentration analysis.

The former crop was maize for the sampled Z. bungeanum fields. Based on field investigation, three standard sampling plots (blocks)  $(20 \text{ m} \times 20 \text{ m})$  were established, and the H and CBS of every Z. bungeanum tree in each plot were measured (Table 1). The above-ground biomass of the standard tree in sampling plots was divided into six components: stem, branch, leaf, bark, thorn and fruit, using the segmenting method. The below-ground (root) biomass was obtained using total excavation method. Most roots of the Z. bungeanum plantations were distributed within the canopy zone. Therefore, after cutting the tree from the stump, we excavated all roots within the radius of one-half of their crown diameter from the tree center. The maximum depth excavated varied between 80 and 150 cm until no more roots were visible. Roots were separated with soil by washing with water and dried by exposing to the air. The fresh weights of all biomass components (stem, branch, leaf, bark, thorn, fruit, and root) were measured insitu, and then samples (500-1000g) of every component for each standard tree were taken to the laboratory for moisture content and N concentration analysis.

# 2.3. Soil sampling

Soil samples were taken up to a depth of 30 cm with three replicates for maize and *Z. bungeanum* plantations. At each sampling point, soil samples were collected at three depths of 0-10, 10-20, and 20-30 cm using a soil corer. Bulk density for each depth was determined by weighing the whole sample and then drying subsamples at 105 °C to a constant weight (the cylinder method).

# 2.4. N concentration analysis

Each subsample of the different plant components in maize and in a chronosequence of *Z. bungeanum* plantations was oven-dried at 60 °C to a constant weight. Dry weights of every component were determined. The components were then mechanically ground to pass through a 0.5 mm mesh screen. Soil samples were dried at 70 °C for 48 h, milled, and sieved with a 0.25 mm mesh screen. The powder samples of plant components and mineral soils were analyzed for N concentrations using a Vario Macro Elemental Analyzer (Elementar Analysensysteme GmbH, Germany).

#### 2.5. Calculation of N storage

The N storage in the plant components and soil depths is estimated as follows:

Nitrogen storage (NS) in vegetation  $(kg ha^{-1})=\sum$  (nitrogen content in different components  $(kg kg^{-1}) \times$  biomass in different components  $(kg ha^{-1})$ )

NS in soil depth layers  $(\text{kg ha}^{-1}) = \sum (\text{N content at different depths}) \times (\text{kg kg}^{-1}) \times \text{soil bulk density at different depths}) \times 0.10 (m) \times 10,000 (m^2 ha^{-1}))$ 

# 2.6. Statistical analysis

Differences between biomass, N content, and N storage following the conversion of cropland to forest were identified and compared using a mixed model analysis of variance (ANOVA) (PROC. MIXED in SAS), in which stand age was considered a fixed effect while block was used as a random effect. When a significant difference was observed using the mixed model ANOVA, The means were compared using the Tukey's HSD (honest significant difference) test for multiple comparisons between stand ages. The relationship between biomass, biomass N storage and base of the stem was obtained using a power function fitting, and R-squared values of the model parameters were measured to determine goodness of fit. For all the statistics, significance level was set at p < 0.05.

# 3. Results

#### 3.1. N concentrations in plant components following the GGP

There were obvious differences in N contents among different plant components and among the same components from stands of different ages. The mixed model ANOVA revealed that significant differences were observed by stand age for the N concentrations in stem and thorn. However, there were no significant differences in root, branch, bark, leaf and fruit among stand ages (Table 2). Following the conversion of cropland to forest, leaves had the highest N concentration with a mean value of 3.65% and 3.13% while stems had the lowest with a mean of 0.89% and 0.39% in maize and *Z. bungeanum* plantations, respectively. The average N concentration decreased following order: leaf > fruit > root  $\approx$  stem (maize) and leaf > fruit > bark > root > branch > thorn > stem (*Z. bungeanum*) (Table 2).

The average N concentration of fruit was 2.57% in the maize crops, which was in line with that of fruit (2.32%-2.67%) in the *Z. bungeanum* forests. The N concentration of stem was significantly greater in maize cropland (0.89%) compared to 0.49%, 0.31%, 0.34% and 0.42% in 1-, 4-, 7-, and 10-year-old *Z. bungeanum* plantations, respectively (Tukey's HSD). Although mean N concentration of thorn was similar among stand ages, there were significant differences (p < 0.01) between 1-year-old stand and 7-, 10-year-old stands (Table 2).

#### 3.2. Biomass allocation

In the maize fields, the amount of total plant biomass (TPB) was 9.90 t ha<sup>-1</sup>. Most of the TPB was stored in the stem and leaf,

1	2
т	2

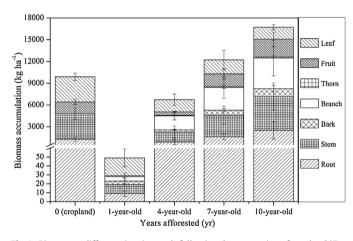
Table 2

Years afforested	Root	Stem	Branch	Thorn	Bark	Leaf	Fruit	Whole plant
0	$0.87\pm0.09^a$	$0.89\pm0.12^a$	none	none	none	$\textbf{3.65}\pm\textbf{0.70}^{a}$	$2.57\pm0.14^a$	2.13
(cropland)								
1	$1.65\pm0.47^{a}$	$0.49\pm0.05^{\rm b}$	$1.81\pm0.36^{a}$	$1.06\pm0.08^a$	$2.08\pm0.25^a$	$3.23\pm0.17^{a}$	none	2.10
4	$1.78\pm0.56^{\text{a}}$	$0.31\pm0.04^{b}$	$1.10\pm0.21^{a}$	$0.96\pm0.03^{ab}$	$2.43\pm0.04^{a}$	$2.88\pm0.11^{a}$	$2.32\pm0.13^a$	1.62
7	$2.19\pm0.04^a$	$0.34\pm0.06^{\rm b}$	$1.40\pm0.42^{a}$	$0.86\pm0.08^{bc}$	$2.30\pm0.14^{a}$	$3.20\pm0.27^a$	$2.62\pm0.18^{a}$	1.76
10	$1.95\pm0.63^a$	$0.42\pm0.16^b$	$1.00\pm0.06^a$	$0.82\pm0.09^c$	$2.22\pm0.49^a$	$3.24 \pm 0.22^{a}$	$2.67\pm0.23^a$	1.53
Variations								
Stand age	3.22 <sup>ns</sup>	18.91***	4.15 <sup>ns</sup>	7.84 <sup>°</sup>	0.84 <sup>ns</sup>	1.21 <sup>ns</sup>	1.71 <sup>ns</sup>	
Block	0.01 <sup>ns</sup>	0.25 <sup>ns</sup>	0.71 <sup>ns</sup>	1.73 <sup>ns</sup>	1.16 <sup>ns</sup>	1.54 <sup>ns</sup>	0.08 <sup>ns</sup>	

N concentration (%) of biomass components following the conversion of cropland (Zea mays L.) to forest (Z. bungeanum).

The effects of stand age on N concentration of biomass components were examined using a mixed model that included stand age as a fixed effect, with block as a random effect. Means in a column followed by the same letter are not significantly different at p < 0.05 (Tukey's HSD test). Significance of effects are shown as F value followed by P category: ns, p > 0.05; \*\*, p < 0.05; \*\*\*, p < 0.001.

representing 35.35 and 35.15% of TPB, respectively (Fig. 1). TPB following afforestation of maize croplands with *Z. bungeanum* increased markedly with stand age, from  $0.05 \text{ t ha}^{-1}$  in the 1-yearold plantation to 6.76, 12.22, and 16.71 t ha<sup>-1</sup> in 4-, 7-, and 10-yearold plantations, respectively. The distribution of biomass in *Z. bungeanum* forests varied with the stand age. Most of the biomass was stored in the leaves of the 1-year-old plot (40% of TPB), while most of the biomass was stored in the branches of the 4- and 7- year-old plots (28.25 and 25.53%, respectively). However, in the 10-year-old plot, biomass was mainly stored in the stems, which represented 27.95% of TPB (Fig. 1). Following the conversion of farmland to forest, stand age had significant effects on the biomass accumulation in root, stem, bark, leaf and TPB (Table 3).



**Fig. 1.** Biomass at different time intervals following the conversion of cropland (*Zea mays* L.) to forest (*Z. bungeanum*) (kg ha<sup>-1</sup>, mean  $\pm$  SD) (error bars indicate the standard deviation).

Table :	3
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Mixed model ANOVA for the effects of stand age on biomass allocations.

Components	Stand age F statistics P values		Block		
			F statistics	P values	
Root	6.90	0.0105	0.52	0.6142	
Stem	6.59	0.0119	0.55	0.5961	
Branch	3.82	0.0763	0.37	0.7057	
Bark	9.94	0.0096	0.26	0.7781	
Thorn	1.62	0.2813	0.89	0.4597	
Leaf	9.94	0.0034	1.78	0.2297	
Fruit	4.75	0.0502	0.89	0.4597	
ТРВ	7.63	0.0078	0.58	0.5808	

The effects of stand age on biomass allocations were examined using a mixed model that included stand age as a fixed effect, with block as a random effect. Summary table indicates variance ratios (F-statistics) and P-values; TPB: total plant biomass.

The Tukey's HSD test indicated that a significant difference (p < 0.05) was found in TPB between maize plantation and 1-year-old *Z. bungeanum* plantation. However, there were no significant differences in TPB between maize plot and 4-, 7- or 10-year-old plots. In addition, the below-ground biomass (BGB)/ above-ground biomass (AGB) ratio was 0.15 in the annual maize cultivation, which is lower but not significantly different than that (0.18) of the perennial *Z. bungeanum* plantation.

# 3.3. Nitrogen accumulation in the ecosystem

The plant N pool in a given component of maize and Z. bungeanum may be derived from the biomass production and N concentration of the component (Tables 4 and 5). Total biomass N storage (TBNS) refers to N stored in TPB (both AGB and BGB). The average amount of TBNS was  $210.59 \text{ kg} \text{ ha}^{-1}$  for the annual maize, and it decreased in the following order: leaf (60.42%)>fruit (19.69%)>stem (14.59%)>root (5.30%) (Fig. 2). The average TBNS was 0.94, 108.31, 212.20, and 262.12 kg N ha<sup>-1</sup> for the 1-, 4-, 7- and 10- year-old Z. bungeanum plots, respectively, corresponding to an annual TBNS rate of 0.94, 35.79, 34.63, and 16.64 kg N ha<sup>-1</sup> year<sup>-1</sup>. The mixed model ANOVA indicated that stand age had significant influences (p < 0.05) on N storage in the stem, branch, bark, leaf, AGB and TPB. However, there were no significant differences in thorn, fruit and root (BGB) among stand ages (Table 4). A highly significant difference (p < 0.01) in TBNS was observed between maize and 1-year-old Z. bungeanum, but the values measured for maize and other stand ages were not significantly different.

For TBNS in the *Z. bungeanum* plantations, significant differences (p < 0.05) were discovered between 10-year-old, 7-year-old and 1-year-old stands as well as between 10-year-old and 4-year-old stands (Tukey's HSD). However, there were no significant differences in TBNS among other stand ages (Table 5). The N stored

Table 4				
Mixed model ANOVA	for the effects	of stand	age on N	storage.

Components	Stand age		Block	Block		
	F statistics	P values	F statistics	P values		
Stem	11.61	0.0021	0.73	0.5118		
Branch	6.48	0.0260	0.32	0.7383		
Bark	4.87	0.0477	0.42	0.6774		
Thorn	1.60	0.2849	0.89	0.4571		
Leaf	8.46	0.0057	0.43	0.6649		
Fruit	4.59	0.0538	1.02	0.4152		
AGB	8.84	0.0049	0.68	0.5314		
Root (BGB)	3.55	0.0601	0.64	0.5506		
ТРВ	7.49	0.0082	0.44	0.6607		

The effects of stand age on N storage in biomass components were examined using a mixed model that included stand age as a fixed effect, with block as a random effect. Summary table indicates variance ratios (F-statistics) and P-values; AGB: above-ground biomass; BGB: below-ground biomass; TPB: total plant biomass.

#### Table 5

N pools in biomass, soil, and ecosystem following the conversion of cropland (Zea mays L.) to forest (Z. bungeanum) (kg N ha<sup>-1</sup>, mean  $\pm$  SD).

Components	Years afforested	Years afforested						
	0 (cropland)	1	4	7	10			
Stem	$30.72\pm5.31^{\text{a}}$	$0.06\pm0.04^{c}$	$4.22\pm2.30^{c}$	$9.91\pm4.18^{bc}$	$20.96\pm12.60^{ab}$			
Branch	none	$0.09\pm0.08^{\rm b}$	$20.83\pm6.88^{ab}$	$39.40\pm8.38^a$	$41.08\pm21.15^a$			
Bark	none	$0.08\pm0.07^{\rm b}$	$\textbf{7.59} \pm \textbf{1.52}^{b}$	$16.22\pm5.68^{ab}$	$25.04\pm14.50^a$			
Thorn	none	$0.01\pm0.01^a$	$0.74 \pm 0.55^{a}$	$0.94\pm0.4^{a}$	$1.09 \pm 1.09^{a}$			
Leaf	$127.24 \pm 33.39^{a}$	$0.57\pm0.41^{c}$	$49.12 \pm 23.51^{bc}$	$63.13 \pm 46.57^{b}$	$53.00\pm9.14^{b}$			
Fruit	$41.47 \pm 6.70^{ab}$	none	$10.88 \pm 1.87^{b}$	$46.96 \pm 19.66^{ab}$	$67.21\pm30.64^a$			
AGB	$199.43 \pm 32.38^{a}$	$0.81\pm0.61^{c}$	$93.39 \pm 31.25^{\rm b}$	$176.56 \pm 76.15^{ab}$	$208.38 \pm 70.20^{a}$			
Root (BGB)	$11.15 \pm 2.10^{b}$	$0.13\pm0.03^{\rm b}$	$14.92\pm2.38^b$	$35.65\pm8.02^{ab}$	$53.74 \pm 41.51^{a}$			
TPB	$210.59 \pm 33.64^{ab}$	$0.94\pm0.63^{c}$	$108.31 \pm 33.46^{bc}$	$212.20 \pm 73.61^{ab}$	$262.12 \pm 110.11^{a}$			
Soil	$10043.65\pm$	$9378.63\pm$	9715.87±	$8833.90\pm$	$9410.03\pm$			
(0-30 cm)	294.79	395.35	351.37	224.54	596.32			
Ecosystem	10254.24	9379.57	9824.18	9046.10	9672.15			

Means in a row followed by different letters are significantly different at p < 0.05 (Tukey's HSD test). AGB: above-ground biomass; BGB: below-ground biomass; TPB: total plant biomass.

in the AGB steadily increased from 0.81 kg N ha<sup>-1</sup> in the 1-year-old plot to 208.38 kg N ha<sup>-1</sup> in the 10-year-old plot. The corresponding values for the BGB were 0.13, 14.92, 35.65, and 53.74 kg N ha<sup>-1</sup> in the 1-, 4-, 7-, and 10-year-old plots, respectively, representing 13.8%, 13.8%, 16.8% and 20.5% of TBNS in the four stand ages of *Z. bungeanum* plantations (Table 5; Fig. 2). Most of the N in the AGB was stored in the leaves and fruits, while only a small amount was associated with stems and thorns. There was also a marked increase in fruit N storage with stand age. The contribution of fruit N storage to TBNS changed from zero in the 1-year-old stand to 25.64% in the 10-year-old stand (Table 5; Fig. 2).

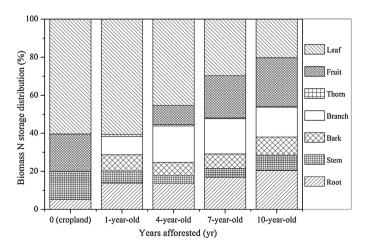
Table 5 shows soil N storage (SNS) of a chronosequence of *Z. bungeanum* forest following the conversion of maize cropland, based on calculations in which the soil bulk density, total nitrogen concentration and depth were taken into account. The average amount of SNS in the maize fields was  $10.04 \text{ t N ha}^{-1}$ , which was significantly higher (p < 0.05) than that ( $9.33 \text{ t N ha}^{-1}$ ) in the *Z. bungeanum* forests. In the *Z. bungeanum* plantations, average N storage in the soil (0-30 cm) ranged from  $8.83 \text{ t N ha}^{-1}$  to  $9.72 \text{ t N ha}^{-1}$ , which did not increase with stand age. The highest SNS was measured for the 4-year-old stand and the lowest was recorded in the 7-year-old stand. Irrespective of vegetation type and stand age, the SNS decreased with increasing soil depth, being greater in the top layer (0-10 cm) than in deep layers (10-20 and 20-30 cm) (Fig. 3).

The ecosystem nitrogen storage (ENS) (TBNS plus SNS) amounted to  $10.25 t N ha^{-1}$  in the maize plantation. After the

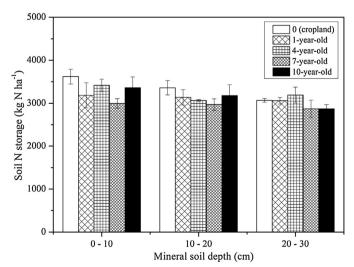
afforestation, the average of ENS was  $9.48 \text{ t N ha}^{-1}$  across all stand age groups of *Z. bungeanum* plantations. In both the cropland and recently converted forests, most of the N accumulated in the soil rather than in the biomass (Table 5). Indeed, 97.95% of the ENS in the annual maize cultivation was stored in the soil. This proportion in the *Z. bungeanum* plantation decreased with stand age with 99.99%, 98.90%, 97.65%, and 97.29% in the 1-, 4-, 7-, and 10-year-old plots, respectively.

# 3.4. Regression models for biomass N storage after afforestation

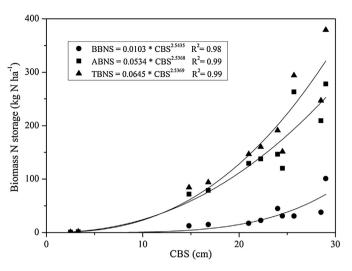
In the *Z. bungeanum* plantation, allometric equations using power functions were selected to estimate N storage for the various components using the circumference at the base of the stem (CBS). These were highly predictive (Fig. 4). Statistically, all the correlations are highly significant (p < 0.01). Among the tree components the correlation was highest ( $p \le 0.001$ ) for the total biomass N storage (TBNS), and above-ground biomass N storage (ABNS), which can explain approximately 99% variations. For the below-ground biomass N storage (BBNS), the model can explain more than 98% of the variation. However, the models that used height (H), crown diameter (CD) and CBS<sup>2</sup> × H as predictive variables of ABNS, BBNS, and TBNS, failed to show a good fit.



**Fig. 2.** Percentage distribution of biomass N storage in various components following the conversion of cropland (*Zea mays* L.) to forest (*Z. bungeanum*).



**Fig. 3.** N storage at different depths of soil profiles in a chronosequence of forest succession (*Z. bungeanum*) following the conversion of cropland (*Zea mays* L.) (error bars indicate the standard deviation).



**Fig. 4.** Models selected for the estimation of above-ground biomass nitrogen storage (ABNS), below-ground biomass nitrogen storage (BBNS), and total biomass nitrogen storage (TBNS) in the *Z. bungeanum* stands (CBS: circumference at the base of the stem).

#### 4. Discussion

# 4.1. Plant nitrogen content

The mean N concentration in the *Z. bungeanum* plantations ranges from 1.53% to 2.10%, which is higher than the 1.40% and 1.48% measured for *Robinia pseudoacacia* and *Erythrophleum fordii*, respectively (Shi et al., 2006). However, it is lower than that of 2.13% in the maize cropland in the same region (Table 2). Shi et al. (2006) measured an average N content of 1.72% in the *Litsea cubeba* forest of a national park, which is consistent with our results. The N contents of plants should be closely associated with their chemical compositions, and should vary with tree species, geographical location, climate, soil condition, and agricultural history (Kahan et al., 2014; Pettersen, 1984). All of these factors must be taken into account when evaluating N contents of trees under different climatic and edaphic conditions.

For a specific organ, there is a difference in N content among plant species. For example, leaf N content in the Z. bungeanum stands ranges from 2.88% to 3.24%, which is lower than that of 3.65% in the annual maize in the same region, and higher than the 2.06% and 2.02% measured for plant leaves globally and in China, respectively (He and Han, 2010), but is in line with Rhamnus (3.31%) and Robinia (2.95%). Stem N content in the Z. bungeanum stand ranges from 0.31% to 0.49%, which is lower than the mean of 0.89% for the maize cropland in this region. This range is higher than the range for subtropical evergreen broad-leaved forests (0.21%-0.28%) (Zhang et al., 2010), but is in accordance with Machilus thunbergii (0.39%), Machilus leptophylla (0.48%) and Lithocarpus glaber (0.36%) (Shi et al., 2006). In addition, N contents, even in the same species, varied significantly between components. In our study, leaves averaged 6 and 4 times greater N content than stems in Z. bungeanum and annual maize plantations, respectively, which might be due to differences in the requirements and utilization of nitrogen of plant organs (Yang et al., 2014). Consequently, N contents and biomass amounts in all organs should be ideally measured because they can vary obviously for different plant species or a given species at different stand ages (Table 2, Fig. 1). If this is neglected, then large errors could arise in the estimation of biomass N sequestration.

#### 4.2. Soil nitrogen storage

The average amount of soil nitrogen storage (SNS) was  $9.33 \text{ t N ha}^{-1}$  in the *Z. bungeanum* plantation, which was higher than the  $6.60 \text{ t N ha}^{-1}$  measured for the mixed deciduous forest of *Quercus robur* × *Acer pseudoplatanus* (Ritter et al., 2003), much lower than the  $34.64 \text{ t N ha}^{-1}$  for Chinese forest ecosystems (Zhang et al., 2004), and significantly lower than that of the maize plantation ( $10.04 \text{ t N ha}^{-1}$ ) in the same region (Table 5). SNS decreased following the conversion of farmland to forest, which is consistent with other results (Berthrong et al., 2009; Li et al., 2012). The effect of land-use change on SNS was partly due to the decrease in organic and inorganic fertilizer application as well as N losses through decomposition induced by site preparation for forest operations (Smal and Olszewska, 2008; Zhao et al., 2005).

The SNS is close to a balance of N input (e.g. atmospheric N deposition, and biological N fixation) and output (e.g. N uptake by plant, N emission to groundwater or the atmosphere) (Li et al., 2012). Changes in SNS with stand age, following afforestation of agricultural lands, have been the subject of many investigations. Interestingly, the variation of SNS with plantation age does not show a consistent trend. Some studies indicated that SNS increased linearly with stand age (Sartori et al., 2007; Yu and Jia, 2014), while others found that there was a "Covington curve" (initial decrease followed by an increase) between SNS and plantation age (Hooker and Compton, 2003; Li et al., 2012). In the present study, a small decrease but no significant age dependence of N storage in the mineral soil was found within 10 years of afforestation. Similarly, no significant change and a small decrease in SNS with stand age have also been reported by other workers (Jug et al., 1999; Ritter et al., 2003). These findings may be due, at least in part, to the fact that N uptake per tree varies with age during the forest development. Indeed, the annual rate of biomass N accumulation of 0.94, 35.79, 34.63, and 16.64 kg N ha<sup>-1</sup> year<sup>-1</sup> for 1-, 4-, 7-, and 10- year-old stands of Z. bungeanum plantations, respectively (Table 5). Therefore, a decrease in the soil N pool with time would have been expected, since intensive N uptake by plants and the accumulation of N in the standing biomass are generally higher in the phase of active growth during the initial afforestation (Berthrong et al., 2009; Ritter, 2007) as well as the fact that there is no sustainable input of mineral N through fertilizers and plant residues (Ritter et al., 2003).

Following the conversion of farmland to forest, all these results suggest that there is no obvious unidirectional temporal trend in the SNS, or at least not one that can be easily discerned from our or previous results. These inconsistent reports can be attributed to a number of factors, including previous land-use (barren land, grassland, cropland, etc.), tree species, farming practices, soil properties (clay content, porosity, pH, etc.), site management, topography and climate (Holubik et al., 2014). All these factors act individually or in combination to determine or eliminate the effect of stand age on soil N accumulation. Therefore, site specific measurements following afforestation are needed to identify temporal changes of SNS, and care should be taken when extrapolating results from one site to another. In addition, more than 97% of the N is stored in the top 30 cm of the soil profile in both agricultural and forest systems. In other words, the top-soil in karst areas of SW China constitutes the main reservoir for N in the overall ecosystem budget while the soil is vulnerable to natural or human-induced erosion and disturbance. Therefore, best management practices in plantations, as outlined in the GGP, need to be implemented to improve the soil quality and promote N storage (Bernhard-Reversat, 1996).

#### 4.3. Estimation of N storage in rocky desertification areas

The biomass nitrogen storage (both above-ground and belowground parts) in *Z. bungeanum* plantations rapidly increased from  $0.94 \text{ kg N ha^{-1}}$  for the 1-year-old stand to 262.12 kg N ha<sup>-1</sup> for the 10-year-old stand (Table 5). A similar age-related increase in biomass N stock has also been observed by other investigators (Feldpausch et al., 2004; Hooker and Compton, 2003). These increases of N stocks were mainly caused by the large gain in above-ground plant biomass that occurred along the chronosequence (Hooker and Compton, 2003).

The average rate of biomass N accumulation was 29.58 kg N ha<sup>-1</sup> year<sup>-1</sup> during the initial 10 years of *Z. bungeanum* plantations. This is higher than the 22 kg N ha<sup>-1</sup> year<sup>-1</sup> in the boreal forests, lower than the 49 kg N ha<sup>-1</sup> year<sup>-1</sup> in the temperate forests, and is in line with that of the tropical forests (29 kg N ha<sup>-1</sup> year<sup>-1</sup>) (Houlton et al., 2008). Moreover, the rate of biomass N accumulation was much greater in young stages than in old stands, peaking at 35.79 kg N ha<sup>-1</sup> year<sup>-1</sup> during the first 4 years of afforestation. This is similar to the rate of biomass C storage in the *Z. bungeanum* plantation (Cheng et al., 2015). These results indicate that *Z. bungeanum* exhibits fast growth in the early stage following the GGP, confirming that afforestation with such quick-growing species is beneficial to N and C incorporation into biomass.

By assuming that all maize plantations in the karst region of Guizhou province can be replaced by Z. bungeanum under the GGP, we can obtain a preliminary estimation of N storage as a result of the conversion of farmland into forest. In the past 10 years, approximately  $1.18 \times 10^6$  ha of cropland and barren land has been established in this province. The amount of sequestered vegetation nitrogen in 2010 was  $2.35 \times 10^8$  kg N, which corresponded to 41.45% of the total nitrogen (NO<sub>X</sub>-N and NH<sub>3</sub>-N) ( $5.67 \times 10^8$  kg N) emitted from the whole Guizhou province in that year (Hao et al., 2002; Huang et al., 2012; Liu et al., 2013). The above amount of biomass N accumulation was estimated using the annual plantation area in Guizhou for each stand from 1999 to 2010 under the GGP from the China Forestry Statistical Yearbook (State Forestry Administration, 1999–2011), and the rate of biomass N accumulation for each plantation stand over the same period (Table 5). Admittedly, this calculation only refers to changes in above- and below-ground biomass N, and excludes any variations in soil N storage resulting from the GGP implement. Meanwhile, the above nitrogen emissions were obtained using the total amount of NO<sub>X</sub>-N  $(6.86 \times 10^9 \text{ kg N})$  and NH<sub>3</sub>-N  $(14.34 \times 10^9 \text{ kg N})$  emitted China during 2010 according to Liu et al. (2013) and the corresponding proportions of NO<sub>X</sub>-N and NH<sub>3</sub>-N emissions from Guizhou province were 0.02 and 0.03, respectively (Hao et al., 2002; Huang et al., 2012). Thus, the calculated emissions of NO<sub>X</sub>-N and NH<sub>3</sub>-N from Guizhou province for 2010 were  $1.37 \times 10^8$  and  $4.30 \times 10^8$  kg N, respectively. Over the long term, continuation of the large-scale of GGP in conjunction with the maturation of Z. bungeanum plantations over the next few years should lead to increased N and C storage and offset greenhouse gas emissions as well as reverse rocky desertification, protect the fragile ecological environment, and increase farmer income in the karst region of SW China.

#### 5. Conclusions

We compared the storage and allocation of biomass and N in a recently afforested land with those of an adjacent intensively managed farmland in the karst region of southwest China. We observed a considerable shift in biomass allocations from below to above-ground within 10 years following afforestation. Compared to the maize farmland, the lower soil N stocks under the Z. *bungeanum* plantations may be attributable to intensive N uptake by plants and no sustainable input of mineral N through fertilizers. The ecosystem N storage decreased from  $10.25 \text{ t N ha}^{-1}$  to 9.48 t N ha<sup>-1</sup> following the conversion of maize cropland to *Z. bungeanum* forest, of which more than 97% was stored in the soil in both landuse systems. Therefore, reducing soil disturbance via good forest management practices has great potential to increase N sequestration. The results of this investigation indicate that the expansion of *Z. bungeanum* plantations in the karst regions of China, under the Grain for Green Program, can increase the regional N budget and arrest rocky desertification, which provides important information for forest managers and policy makers, even if long-term monitoring and further research on afforestation of the intensively managed farmland are needed to further support findings from this study.

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#### Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.agee.2016.07.019.

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