# Shrub-interspace dynamics of soil microbial communities under different patch areas in a semiarid river valley, SW China

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## Abstract

In arid and semi-arid ecosystems, belowground characteristics are influenced by the formation and persistence of "fertile islands" in contrast to barren plant interspaces. We investigated the soil properties of patched vegetation, especially the belowground properties of microbial communities, by directly comparison of soil microbial communities between soils under patched vegetation and in the interplant spaces with increased patch areas in Minjiang River arid valley. We found "fertile islands" exist in patch soils, and the increased substrates utilization ability of soil microbes may indicate a positive feedback to maintain the "fertile islands" under patched vegetation. Under the patches, soil C, soil N, soil microbial biomass, soil water content, clay concentration were significantly improved by the increasing vegetation patch areas. The fungus to bacteria ratio was increasing with the enlarged vegetation and the contribution of fungi to microbe was closely relevant to patch area. Compared the barren plant interspaces, soil nutrient condition and microbial biomass were significantly influenced by its adjacent patch. Thus, "the islands of fertility" may be beneficial and advance the rehabilitation of plant in this semi-arid region.

## **Key Words**

Arid vegetation, fertile islands, soil microbes, patch.

## Introduction

Soil degradation is a critical environmental problem in arid and semiarid ecosystems. The establishment of plant cover is reported to be one of the most effective means for erosion control and regeneration of the degraded soil (Xu et al. 2008; Garcia et al. 2002). Plant cover can reduce water runoff and sediment loss, and favour soil-development processes by improving soil organic matter, soil structure, soil water and nutrientholding capacity, which are especially important in improving semiarid soil quality. Soil quality and its degradation depend on a large number of physical, chemical, biological, microbiological and biochemical properties, and the last two are the most sensitive since they respond rapidly to changes. It is widely accepted that high levels of microbial activity are fundamental in maintaining soil guality. In arid and semi-arid ecosystems, belowground characteristics are influenced by the formation and persistence of "fertile islands" in contrast to barren plant interspaces. Soil microbes may respond differently to "fertile" and interspace soils. In the few studies assessing microbial community composition between shrub and interspace soils, the importance of shrub-islands in structuring communities was evident (Ewing et al. 2007; Aandernd et al. 2008). Relative to interspace soils, shrub soils have higher total soil C and N, support more microbial biomass, and are associated with higher C and N mineralization rates (Schaeffer and Evans 2005; Ewing et al. 2007). Microbial community composition may not be regulated primarily by soil C, but by N and P availability and soil stresses (i.e. high B and Cl concentrations) in shrub soils, or by soil stresses in interspace soils (Aandernd et al. 2008). The establishment of suitable plant cover could improve the physico-chemical and biological properties of soil. Soil microbes are supposed to respond quickly to these changes. However, how soil microorganisms in the "fertile islands" as well as the barren soils interact with aboveground plant cover remains unclear. Because vegetation in arid and semiarid ecosystems is typically patchy and most of these systems experienced degradation, researches on shrub-interspace soil microbial responses to plant cover in the restoration processes is meaningful. In the upper minijang river, vegetation (mostly shrub) is typically patchy, and we take patch area (%) to indicate plant cover. We hypothesized that "fertile islands" existed under patched vegetation, and microbial communities would respond differently in shrub and interspace soils under different plant covers.

## Methods

*Site description and sampling* The study area (31°47′54″-31°47′54″N, 103°44′30″-31°44′40″E) is part of the dry-warm valley of the upper Minjiang River, one of the four principal tributaries of the Yangtze River. Vegetation here was damaged years ago and now is undergoing revegetation. The mean annual precipitation is 494 mm and the mean annual evaporation is 1,332 mm. The aridity index for this area is within 1.5–3.49, which is typical of semiarid environments (Xu *et al.* 2008). The predominant soil type is calcic cambisols. Regional vegetation mainly consists of small-leaf arid shrubsand sparse grasses (Ma *et al.* 2004; Xu *et al.* 2008). We selected four typical locations with an increased patch areas (30%, 40%, 45%, 75% of the study area respectively) in the core area of the valley. Topography conditions are almost the same. The dominant species are *Sophora. viciifolia* and *Artemisia gmelinii* for 30% patched; *A. gmelinii* for 40% patched; *A. gmelinii, Onosma mertensioides* and *Wikstroemia stenophylla* for 45% patched; *Bauhinia faberi* and *A. gmelinii* for 75% patched. Three 5m\*5m plots were randomly selected for each site. In September 2009, soil samples were collected from surface soils (0-10cm) under different dominant species and interspace soils in each site. Each soil sample consisted of five sub-samples removed with a soil corer (3.5cm in diameter) and thoroughly mixed to obtain a composite sample. Prior to transport, all soil samples were stored at approximately 4°C immediately after sampling.

#### Soil physico-chemical measurements

Soils were sieved to 2mm and litter and roots retained on sieve were separated by hand. The methods used to measure soil physico-chemial properties were as described by Xu *et al.* 2008.

#### Soil microbial measurements

Microbial biomass C and N were determined on 10g soil using the fumigation-extraction procedure described by Macdonald et al 2009. Substrate introduced respiration (SIR) was assessed using the MicroResp<sup>TM</sup> CO<sub>2</sub> detection system (Campbell *et al.* 2003). To cope with experimental artifacts when dealing with calcareous soils derived from CaCO<sub>3</sub>-CO<sub>2</sub>-H<sub>2</sub>O equilibrium and acidic substrates, we followed the recommendations from Oren and Steinberger (2008). Microbial community structure was assessed by PLFA analysis using a modified method (White *et al.* 1979) based on that of Bligh and Dyer (1959). Briefly, soil samples from each site were freeze-dried; finely ground and 4g were used for extracting PLFAs. The separated fatty acid methyl-esters were identified and quantified by GC-MS (HP6890/MSD5973) using methyl nonadecanoate (19:0) as the internal standard. The specific fatty acids used to indicate bacteria, fungi, Gram-positive bacteria, Gram-negative bacteria, Actinobacteria were as described by Zachary *et al.* (2008) and Macdonald *et al.* (2009).

## Statistical analyses

Data was analysed using both univariate and multivariate methods. Effects of patch area on soil properties and microbial communities were determined using one-way ANOVA and Tukey's HSD test at P <0.05. When necessary, data were log or arcsine transformed to accomplish the assumptions of normality and homogeneity of variances of ANOVA. All univariate analyses were conducted using SPSS13.0 (SPSS Inc., IL. Chicago, USA). And linear model redundancy analysis (RDA) was used to test the relationships between ecosystem characteristics and the distribution of microbial groups. The significance of the canonical axis was tested using the Monte Carlo permutation test. And all multivariate techniques were performed with CANOCO software (Canoco for Windows 4.5).

## Results

## Microbial communities between patch and interspace soils

Compared with interspace, patch soils were higher in GWC, Clay%, nutrients, microbial biomass, lower in pH(Table 1, Figure 1), and "fertile islands" existed in patch soils. SIR showed that microbial activities were stronger in patch compared to interspace, which may indicate faster nutrient cycling in patch soil (data not show). RDA analysis showed that patch area and C/N explained large variance in microbial groups for both shrub and interplant soils; GWC explained little variance in shrub soils while in interspace soils, Actinobacteria, Gram-positive bacteria, total bacteria and total PLFA were negatively related to GWC (Figure 3). Soil microbes in the interspace soils were not regulated by GWC but nutrients to a large extent.

## Microbial communities within patch soils

Large patch area (i.e. 45% or  $\overline{75\%}$ ) increased GWC, Clay%, nutrients and microbial biomass and lowered pH (Table 1, Figure 1). The significant effect of patch area was also found in the increased utilization of carbon sources like carbohydrate, carboxylic acid and amino acid (P<0.05) (data not show). Mol% of fungi was higher in 30% patched than 40%, but lower than 75% patched. Actinobacterial increased with increasing

patch area; the same tendency was also mirrored in gram-negative bacteria gram-positive bacteria (Figure 2 D). The fungus to bacteria ratio was generally increased with the enlarged vegetation (P<0.05) (data not shown) and the contribution of fungi to microbe was closely relevant to patch areas (Figure 3 B). GWC and TP were two main constraints in the study area (Song *et al.* 2009). However, the variance of GWC and TP was slight and explained less variance in RDA analysis (Table 1, Figure 3 B).

Table 1. Soil physico-chemical properties. Values are means. Different letters in the same column indicate significant differences (P<0.05) under different patch areas. Abbreviations are as follows: GWC= soil water content, SOC= soil organic carbon, C/N = SOC/TN, TN= total nitrogen, TP= total phosphorus.

| content, SOC- son organic carbon, C/1 -SOC/11, 114- totar introgen, 11 - totar phosphorus. |      |       |       |         |        |        |         |        |       |        |        |
|--|------|-------|-------|---------|--------|--------|---------|--------|-------|--------|--------|
|  | Area | GWC   | pН    | TC      | TN     | ТР     | SOC     | C/N    | Clay  | Silt   | Sand   |
|  | (%)  | (%)   |       | (g/kg)  | (g/kg) | (g/kg) | (g/kg)  |        | (%)   | (%)    | (%)    |
|  | 30   | 4.41a | 8.65a | 8.92b   | 1.17b  | 0.40a  | 8.01c   | 6.62b  | 1.29b | 82.03a | 16.68a |
| Inter-   | 40   | 3.49a | 8.65a | 20.32a  | 2.18a  | 0.48a  | 17.18c  | 7.74ab | 1.54b | 77.56a | 20.91a |
| space  | 45   | 2.49b | 7.17b | 20.35a  | 2.17a  | 0.44a  | 19.78ab | 8.99a  | 1.49b | 78.18a | 20.33a |
|  | 75   | 2.72c | 8.17a | 28.32a  | 2.51a  | 0.47a  | 25.91a  | 8.77a  | 1.62a | 80.55a | 17.84a |
|  | 30   | 4.62b | 8.23a | 15.63c  | 1.73c  | 0.47a  | 13.86b  | 8.01c  | 1.41b | 83.50a | 15.54a |
| Patch  | 40   | 4.20b | 8.67a | 25.30bc | 2.49bc | 0.47a  | 22.05b  | 8.83b  | 1.34b | 77.01a | 21.65a |
|  | 45   | 5.07a | 7.19b | 32.64ab | 3.06b  | 0.53a  | 31.58a  | 10.24a | 1.49b | 78.48a | 20.03a |
|  | 75   | 6.09a | 7.54b | 42.02a  | 3.93a  | 0.52a  | 39.39a  | 9.77a  | 1.71a | 78.91a | 19.38a |
| -  |      |       |       |         |        |        |         |        |       |        |        |



Figure 1. Microbial biomass C (A) and N (B) in patch and interspace soils respectively. Values are means with standard error. Different letters indicate significant differences (P<0.05).



Figure 2. Distribution of microbial groups in interspace (C) and patch soils (D) respectively. Mole % of all microbial groups was based on the total nmol PLFA  $g^{-1}$  soil. Values are means with standard error. Different letters indicate significant differences (P<0.05).

#### Microbial communities within interspace soils

Soils in large patch area (i.e. 45% or 75%) increased in nutrient content, Clay%, microbial biomass and decreased in pH (Table 1, Figure 1). The overall carbon utilization ability also increased (data not show). However, GWC decreased slightly. Soil microbes in the interspace soils were not regulated by GWC but nutrients to a large extent (Figure 3, A). Microbial groups responded differently to patch area (Figure 2). However, Changes in fungi behaved in a similar way as patch soils (Figures 2 and 3).

#### Conclusion

"Fertile islands" existed in the patches and microbial activity may promote the maintenance of these "islands". Within the patches, soil quality was significantly improved with increased vegetation patch areas; the fungus to bacteria ratio was increasing with the enlarged vegetation patch. For plant interspaces, soil nutrient condition and microbial biomass were significantly influenced by its adjacent patch. Thus, "the islands of fertility" may be beneficial and advance for the rehabilitation of plant in this semi-arid region.



Figure 3. RDA analysis between soil properties (plus patch area) and microbial group characteristics in interspace (A) and patch soils (B) respectively. Red arrows represent soil properties (plus patch area), and blue represent microbial group characteristics. For abbreviations in red arrows see Table 1; abbreviations for blue arrows are as follows: total=total PLFA; bact=bacterial PLFA; acti=actinobacterial PLFA; b/f= bacterial PLFA/fungi PLFA.

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 $\bigcirc$  2010 19<sup>th</sup> World Congress of Soil Science, Soil Solutions for a Changing World 1 – 6 August 2010, Brisbane, Australia. Published on DVD.