



**Full Length Article**

## Sod Cultivation Significantly Enhances Soil Nutrient Availability in a Subtropical Newly Established Orchard

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### Abstract

Sod cultivation plays an important role in managing fruit-tree orchards, which could significantly influence the soil nutrient availability, microbial biomass and community. Yet, how the different functional sod species affect these above traits of orchard is still largely unclear. Here, we employed a field-based study to investigate the effects of two functional sod cultivation on seasonal dynamics of soil nutrient and microbial biomass in a subtropical newly established guava (*Psidium guajava*) orchard, Guangdong Province, southern China. In this study, four treatments were established: clean tillage (CT), *Paspalum natatu* monoculture (P), *Stybsanthes guianensis* monoculture (S) and interplanting with *Paspalum natatu* and *Stybsanthes guianensis* (P&S). Our results showed that season significantly affected  $\text{NO}_3^-$ -N concentration, net N mineralization ( $N_{\text{min}}$ ) and nitrification ( $N_{\text{nit}}$ ), microbial biomass carbon (MBC) and nitrogen (MBN), dissolved organic C (DOC) and N (DON), as well as soil acid phosphatase (ACP) in two soil layers. However, sod cultivation enhanced  $\text{NH}_4^+$ -N and  $\text{NO}_3^-$ -N concentrations,  $N_{\text{min}}$  and  $N_{\text{nit}}$  to some extent, and the increasing magnitude varied with the sod species. Sod cultivation also facilitated soil MBC and MBN, DOC and DON. Moreover, both *Paspalum natatu* and *Stybsanthes guianensis* monoculture and their inter-planting dramatically increased ACP activity, while decreased AP concentration in wet season, suggesting that sod competed for AP with fruit-tree in growing season of a newly established orchard. However, compared with *Paspalum natatu*, perennial leguminous species *Stybsanthes guianensis* had little advantageous effects on soil available N supplement for a short-time investigation in this orchard. We recommend that sod cultivation would be an effective method for enhancing soil nutrient status and reducing chemical fertilization in the orchard. Given that, large-scale and long-term studies are still necessary to investigate the effects and underlying mechanisms of different sod cultivation practices on the soil nutrient availability within various orchards in subtropical China. © 2018 Friends Science Publishers

**Keywords:** *Paspalum natatu*; *Stybsanthes guianensis*; Nitrogen mineralization; Phosphorus availability; Microbial biomass

### Introduction

Agroforestry systems are recognized as a potential carbon (C) pool which could store substantial C as aboveground biomass and belowground organic matter (Albrecht and Kandji, 2003; Thangata and Hildebrand, 2012; Somarriba *et al.*, 2013). It is generally believed that the orchard is a major type of agro-ecosystem/agroforestry distributed on hilly and upland soil in tropical and subtropical areas all over the world (Gómez *et al.*, 1999; Balota *et al.*, 2004; Yang *et al.*, 2007), which plays a central role in the global C cycles (Thangata and Hildebrand, 2012; Somarriba *et al.*, 2013). Therefore, the soil C pool and nutrient availability have been widely concerned due to the different management practices of understory vegetation causing significant variation of potential C sequestration and soil nutrient status in orchards (Balota *et al.*, 2004; Oelbermann *et al.*, 2004).

Understory vegetation is an important component in

orchard ecosystems, which are recommended to regulate the soil chemical and biological processes (Rämert *et al.*, 2001; Yang *et al.*, 2007; Floch *et al.*, 2009). Therefore, various management practices for the understory vegetation would enhance/suppress the key functional process beneath the orchard soil. Therefore, understory plant management practices have been established for increasing the productive and sustainable development of the orchard (Gómez *et al.*, 1999; Pandey *et al.*, 2010; Liu *et al.*, 2013).

In orchard, weed management practices have been proved as an effective and ecological method in the understory management since last few years (Chen *et al.*, 2004; Yang *et al.*, 2007; Hanson *et al.*, 2017). Sustaining weed on the orchard would reduce the susceptibility of soil exposed to environment change (Yang *et al.*, 2007). However, the litter and root exudate of weed would provide substantial organic matter for the soil, as well as substrate availabilities for the microorganisms (Chen *et al.*, 2004;

Liu *et al.*, 2013; Zhang *et al.*, 2017), yet, the uncontrolled weed distributed randomly and regenerated irregularly may be not sustainable for the fruit tree growth and development in subtropical orchard. Therefore, maintaining a living selective understory vegetation could be benefic for the orchards.

As a traditional management for understory species, clean tillage (remove all the weed by labor force) would dramatically reduce the nutrient competition from weed and then fertilization would greatly facilitate the fruit tree growth and yield (Gómez *et al.*, 1999). However, previous studies showed that clean tillage or zero tillage significantly influenced the soil physical and chemical priorities. For example, Gómez *et al.* (1999) demonstrated that no tillage achieved greater bulk density and cone index value than conventional tillage although the yield was not affected by tillage in an olive orchard. Compared with frequent till treatment, Pandey *et al.* (2010) found that long time zero tillage (no tillage) maintained the higher rate of net N mineralization and lower amount of SOC. However, clean tillage might greatly lower the understory plant diversity of orchard and expose the soil directly to the sun, rain and wind (Hoagland *et al.*, 2008; Liu *et al.*, 2013). Whereas sod cultivation based on the clean tillage have been proved to be an effective method that would reduce the risks of evaporation, soil erosion and chemical fertilization for higher crop yield, as well as the nutrient competition from the weed (Wang *et al.*, 2016; Yousaf *et al.*, 2016; Hanson *et al.*, 2017). On one hand, sod cultivation could significantly enhance the soil nutrient availability and soil biota in the orchard. For example, Reardon *et al.* (2013) reported that the *Brassica* seed meal amendment in orchard greatly increased the available N and nematode abundance. Wang *et al.* (2016) also indicated that grass planting dramatically promoted the mycorrhizal colonization and available N in citrus orchard. On the other hand, sod cultivation also accelerated the soil C sequestration in orchard. For instance, Liu *et al.* (2013) revealed that maintaining a living understory species significantly increased soil C sequestration and soil fertility in subtropical orchard. Moreover, a meta-analysis also suggested that maintaining sod could dramatically promote soil organic C (SOC) in orchard across China (Wei *et al.*, 2017). Therefore, sod culture were an ecological and selective practice for orchard understory management, which reduced disturbance by frequent tillage for soil organic matter and soil fauna (Hoagland *et al.*, 2008). Hence, it is very necessary to know exactly how the sod culture affect the soil key process and seek to find specific and functional sod for the orchard.

In China, orchards have been rapidly developed since 1990s, and the area of orchard has reached to 11.14 million hectares, whereas annual fruit production reached to 200 million tons that made China the leading country in fruit production around the world (Liu *et al.*, 2013). Nonetheless, the orchards are always distributed on the hilly and upland which would be serious in soil erosion and nutrient loss due

to the higher regional precipitation in tropical and subtropical China (Chen *et al.*, 2004; Yang *et al.*, 2007; Liu *et al.*, 2013). However, in order to pursue higher fruit production, the clean tillage and fertilization are the traditional managements for understory plant in these orchards, which might ignore the negative effect on soil erosion and nutrient leaching (Yang *et al.*, 2007). Given that, the widely distributed orchards with frequent tillage and fertilization have resulted in extremely serious environmental problems since the last decades. Hence, sod cultivation may be a favorable, ecological, cost-effective and sustainable method for the understory vegetation management of orchard in southern China.

Here, we present a field-based study to investigate the effect of sod cultivation on the nutrient transformation and microbial biomass of subtropical guava (*Psidium guajava*) orchard in Heshan City, Guangdong Province, China. In this study, two functional sod including *Paspalum natatum* and *Stylosanthes guianensis* were maintained within the guava (*Psidium guajava*) orchard for one and a half years after the initial planting. We determined the seasonal dynamics of available N and net N mineralization, nitrification, microbial biomass, available P and soil acid phosphatase (ACP) under two sod cultivation. We hypothesize that, 1) sod cultivation would significantly facilitate the soil N mineralization and nitrification compared with traditional clean tillage practices because the sod culture would enhance the microenvironment for microorganism activity; 2) sod cultivation could dramatically reduce the available P but significantly enhance the ACP activity in the grow season of the newly established orchard.

## Material and Methods

### Site Descriptions

We conducted the manipulate experiment in the Heshan City, Guangdong Province, China (112°50'E, 22°34'N). The soil type is aerosols that developed from sandstone, with a pH of approximately 4.0. The climate is typically subtropical monsoon, with MAT of 22.6°C, and MAP of 1700 mm, the highest monthly precipitation was observed in Aug. and lowest in Nov. In this region, there are clear wet and dry season alternately. The wet season (from Apr to Sep) is hot and humid, while the dry season (from Oct. to next Mar.) is cool and dry. It is a hilly agricultural region covered by hilly land (78.6%), farming land (17.1%) and water body (4.3%). The a.s.l of the experimental region is 60.7 m, with lightly rolling topography.

### Experimental Designs

The experimental orchard was established on hilly land at Heshan City, Guangdong Province of China, April 2012. The guava (*Psidium guajava*) was planted in row with 2 × 2 m space. The experiment was designed as a completely randomized block (n=5). Each block covered about 50 m<sup>2</sup> in the terraced field. Four treatments included: clean tillage (CT,

remove all the weed by labor force), *Paspalum natatu* monoculture (P), *Stybsanthes guianensis* monoculture (S) and interplanting with *Paspalum natatu* and *Stybsanthes guianensis* (P&S). *Paspalum natatu* is deemed to be mycorrhizal sod species that would be related to P release while *Stybsanthes guianensis* is regarded as an N-fixing species that would fix the atmospheric N. *Paspalum natatu* was planted uniformly by 10 g m<sup>-2</sup> seed and *Stybsanthes guianensis* was planted uniformly by 0.8 g m<sup>-2</sup> seed after clean and deep tillage. The other sod or weed was removed by hand at the regular intervals during the experiment.

### Soil Sampling and Analysis

In April 2012, we have measured the soil chemical properties (0-10 cm) of four treatments before planting in subtropical orchard (Table 1). After one-year sod planting, *in situ* N mineralization incubation experiment was conducted seasonally (wet and dry) from April 2013 to March 2014. According to the methods that modified from Raison *et al.* (1987), nine sampling subplots were randomly located in each plot, and two sharpened PVC (polyvinyl chloride plastic) cores (4.6 cm diameter×12 cm height) were knocked into the 10 cm depth with an hammer gently, one tube (labeled S0) was fetched and immediately returned back to laboratory storing at 4°C, and the other one (labeled S1) covered with thin film and several holes on the side was for aeration-incubation *in situ* lasting for 30 days.

At each plot, the cores with 10 cm was divided into two parts (0-5 cm and 5-10 cm layer), and each sample would be mixed uniformly, then sieved by 2-mm mesh and removed residual roots and other matter. Soil sample before and after incubation experiment were extracted with 50 mL 2 M KCl for determining inorganic N concentrations, then the extract solution was filtered and determined using a flow injection auto analyzer (FIA star 5000 Analyzer, Foss Tecator, Denmark). Soil water content (SWC) was tested with subsample being dried at 105°C for 24 h. The net N mineralization and nitrification rates were calculated by the difference value of inorganic N concentrations between the initial and after incubation samples in two seasons.

The subsample was also used for testing soil dissolved organic C (DOC) and N (DON). The sieved soil was extracted with 60 mL 0.5 M K<sub>2</sub>SO<sub>4</sub> solution, and DOC and DON were determined by utilizing a Shimadzu TOC-V CSH analyzer. And the micro biomass C (MBC) and N (MBN) was testing by chloroform fumigation.

According to the method described by Acosta-Martínez and Tabatabai (2011), the soil acid phosphatase activity (ACP) was determined by colorimetric *p*-nitrophenyl- ester-based method. The fresh soil was mixed with modified universal buffer (MUB, pH 5.5) and substrate (*p*-nitrophenyl phosphate) solution, and then incubated at 37°C for 60 min. After that, the absorbance of the resultant soil filtrates was determined by an UV-vis spectrophotometer (the wave length was set at 410 nm). Potential acid

phosphatase activity in soil was expressed as micromole *p*-nitrophenol (*p*-NP) per gram of dry soil per hour (μmol *p*-NP g<sup>-1</sup> h<sup>-1</sup>).

### Statistical Analysis

One-way ANOVA method was performed to test the effects of treatments on the concentrations of NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N, net nitrification (N<sub>nit</sub>) and net N mineralization (N<sub>min</sub>), microbial biomass C (MBC) and N (MBN), dissolved organic C (DOC) and N (DON), acid phosphatase (ACP) and available P (AP). Least significant differences (LSD) post hoc test was used to compare the effects of the CT, P, S and P&S treatments on the studied parameters. Three-way ANOVA analysis was used to exam the effects of *Paspalum natatu* monoculture (P), *Stybsanthes guianensis* monoculture (S), season and their interactions on the above biological and chemical indexes during the experimental period. All analyses were performed using SPSS 16.0 (SPSS Inc., Chicago, IL, USA) at *p*<0.05.

## Results

### Soil Water Contents

*Paspalum natatu* monoculture (P) significantly increased the soil water contents (SWC) by 33.38% and 39.47% in 0-5 cm, and 15.59% and 15.00% in 5-10 cm layer, for wet and dry season, respectively (Fig. 1), while *Stybsanthes guianensis* monoculture (S) had no significant effects on the SWC in two layers. However, the interactions of *Paspalum natatu* and *Stybsanthes guianensis* (P&S) also significantly raised the SWC in 0-5 cm and 5-10 cm layers in this study (*p*=0.000 and *p*=0.024, Table 2). Additionally, season significantly influenced the SWC in 5-10 cm while had no significant difference on SWC in 0-5 cm layers (*p*=0.004 and *p*=0.860, respectively).

### NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N Concentrations

*Paspalum natatu* monoculture (P) significantly increased NH<sub>4</sub><sup>+</sup>-N concentrations by 116.99% in 0-5 cm layer of wet season, while *Stybsanthes guianensis* monoculture (S) or P&S treatment had no significant effects on NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N concentrations in this study (Fig. 2). However, season significantly affected the NO<sub>3</sub><sup>-</sup>-N concentrations in both 0-5 cm and 5-10 cm layers (*p*=0.001 and *p*=0.000, respectively) while there was no significant effects on NH<sub>4</sub><sup>+</sup>-N concentrations in two soil layers (Table 2). And the NH<sub>4</sub><sup>+</sup>-N concentrations were significantly positively related with SWC in two layers of this study (*p*=0.013 and *p*=0.025, respectively).

### Net N Mineralization and Nitrification

Season appeared to significantly influence the net N mineralization (N<sub>min</sub>) in both 0-5 cm and 5-10 cm in the subtropical orchard (*p*=0.000 for both, Table 2).

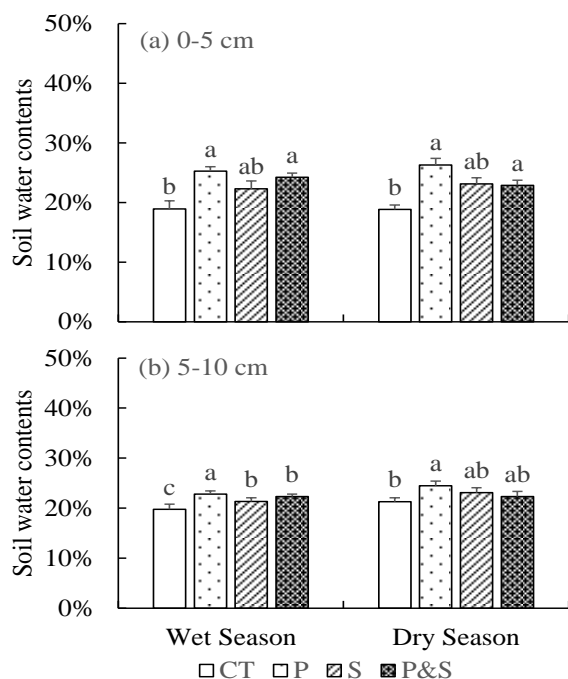
**Table 1:** Soil chemical properties (0-10 cm layer) of four treatments before planting in subtropical orchard, April 2012

Plot	pH	SOC (g kg <sup>-1</sup> )	TN (g kg <sup>-1</sup> )	TP (g kg <sup>-1</sup> )	AP (mg kg <sup>-1</sup> )	C:N
CT	4.32±0.54	26.26 <sup>a</sup> ±2.16	1.35 <sup>a</sup> ±0.36	0.71 <sup>a</sup> ±0.45	59.97 <sup>a</sup> ±4.54	19.37 <sup>a</sup> ±1.58
P	4.25±0.51	29.06 <sup>a</sup> ±1.16	1.46 <sup>a</sup> ±0.34	0.64 <sup>a</sup> ±0.36	53.43 <sup>a</sup> ±4.60	19.96 <sup>a</sup> ±1.09
S	4.15±0.46	25.20 <sup>a</sup> ±1.76	1.31 <sup>a</sup> ±0.33	0.61 <sup>a</sup> ±0.47	62.94 <sup>a</sup> ±6.76	19.14 <sup>a</sup> ±1.10
P&S	4.12±0.38	28.53 <sup>a</sup> ±1.38	1.48 <sup>a</sup> ±0.42	0.58 <sup>a</sup> ±0.30	52.89 <sup>a</sup> ±5.67	19.53 <sup>a</sup> ±1.55

Note: CT, clean tillage; P, *Paspalum natatu* planting; S, *Strysanthes guianensis* planting; P&S, *Paspalum natatu* and *Strysanthes guianensis* mixed planting. Data were showed by (mean±SE), n=5

**Table 2:** Three-way ANOVA for the effects of *Paspalum natatu* planting (P); *Strysanthes guianensis* planting (S), Season and their interactions on the SWC, concentrations of NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N, N<sub>nit</sub>, N<sub>min</sub>, MBC, MBN, DOC, DON, ACP and AP in orchard at Heshan, 2013

Depth	Variable	SWC	NH <sub>4</sub> <sup>+</sup> -N	NO <sub>3</sub> <sup>-</sup> -N	N <sub>nit</sub>	N <sub>min</sub>	MBC	MBN	DOC	DON	ACP	AP
0-5 cm	P	<b>0.000</b>	0.218	0.301	0.475	0.214	0.556	0.698	0.113	<b>0.025</b>	<b>0.000</b>	<b>0.025</b>
	S	0.102	0.429	0.520	<b>0.002</b>	<b>0.042</b>	0.479	0.196	0.100	<b>0.035</b>	0.155	<b>0.001</b>
	Season	0.860	0.066	<b>0.001</b>	<b>0.003</b>	<b>0.000</b>	<b>0.001</b>	<b>0.014</b>	<b>0.002</b>	<b>0.039</b>	<b>0.000</b>	<b>0.000</b>
	P×S	<b>0.000</b>	0.143	0.436	<b>0.009</b>	<b>0.013</b>	0.337	0.643	0.467	0.261	<b>0.000</b>	0.516
	P×Season	0.894	0.433	0.291	0.158	<b>0.043</b>	0.335	0.082	0.703	0.761	0.209	0.268
	S×Season	0.691	0.719	0.527	0.313	0.243	0.665	0.808	0.311	0.488	0.547	<b>0.008</b>
	P×S×Season	0.223	0.943	0.428	0.785	0.435	0.324	0.309	0.479	0.757	<b>0.021</b>	0.351
5-10 cm	P	<b>0.021</b>	0.133	0.502	<b>0.003</b>	0.079	0.449	0.397	0.993	0.949	0.298	0.510
	S	0.935	0.455	0.439	0.146	0.252	0.429	0.564	0.499	0.413	0.612	0.234
	Season	<b>0.004</b>	0.233	<b>0.000</b>	<b>0.012</b>	<b>0.000</b>	<b>0.001</b>	<b>0.002</b>	<b>0.000</b>	<b>0.000</b>	<b>0.000</b>	0.170
	P×S	<b>0.024</b>	0.374	0.953	0.089	0.079	0.507	0.714	0.817	0.622	0.083	0.797
	P×Season	0.084	0.213	0.501	0.199	0.457	0.203	0.200	0.478	0.133	0.365	0.338
	S×Season	0.066	0.920	0.440	0.152	0.315	0.121	0.109	0.543	0.707	0.085	0.647
	P×S×Season	0.833	0.462	0.973	0.612	0.573	0.335	0.525	0.530	0.599	0.749	0.528



**Fig. 1:** Soil water contents under sod cultivation in orchard at Heshan, 2013. Error bars represent 1 SE, n=5. Different lowercase letters denote significant differences among four treatments. ( $p < 0.05$ )

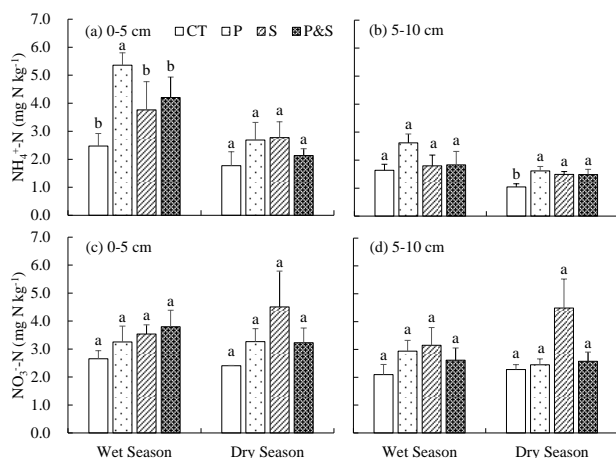
In 0-5 cm layer, both S and P&S planting significantly enhanced N<sub>min</sub> in top 0-5 cm layer ( $p=0.042$  and  $p=0.013$ ,

respectively, Table 2). In wet season, compared with CT treatment, P, S and P & S treatments dramatically increased N<sub>min</sub> by 50.10, 38.12 and 37.38% in 0-5 cm layer, respectively; by 39.47, 31.87 and 22.07% in 5-10 cm layer, respectively (Fig. 3 c and d).

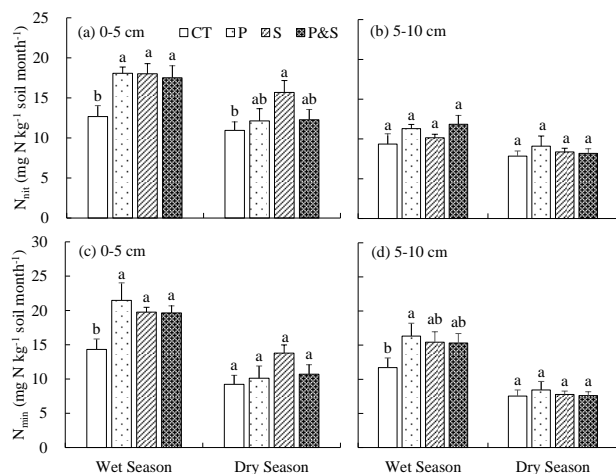
Season also significantly influenced the net nitrification (N<sub>nit</sub>) in both 0-5 cm and 5-10 cm of this study ( $p=0.003$  and  $p=0.012$ , respectively, Table 2). However, S monoculture significantly affected N<sub>nit</sub> in 0-5 cm layer, while P planting (P) significantly influenced that in 5-10 cm layer ( $p=0.002$  and  $p=0.003$ , respectively, Table 2). And P×S interaction also had significant effects on N<sub>nit</sub> in 0-5 cm layer ( $p=0.009$ ). In wet season, compared with CT, P, S and P&S treatments greatly enhanced the N<sub>nit</sub> by 42.66, 42.10 and 38.18% in 0-5 cm layer, and by 20.85, 8.43 and 26.57% in 5-10 cm layer, respectively (Fig. 3 a and b). The N<sub>nit</sub> was also significantly correlated with SWC in two soil layers ( $p=0.012$  and  $p=0.017$ ).

#### Soil Microbial Biomass Carbon (MBC) and Nitrogen (MBN)

Season significantly affected the MBC in both 0-5 cm and 5-10 cm layers in this study ( $p=0.001$  for both layers). However, both P and S monoculture increased the MBC in two layers although there was no significant difference among four treatments (Fig. 4a and b). The increasing rates of MBC under the sod cultivation ranged from c. 8.57% to 31.59% in wet season, and from c. 24.70 to 42.77% in dry season of the two layers (Fig. 4a and b).



**Fig. 2:** Concentrations of soil  $\text{NH}_4^+\text{-N}$  and  $\text{NO}_3^-\text{-N}$  under sod cultivation in orchard at Heshan, 2013. Error bars represent 1 SE,  $n=5$ . Different lowercase letters denote significant differences among four treatments. ( $p < 0.05$ )

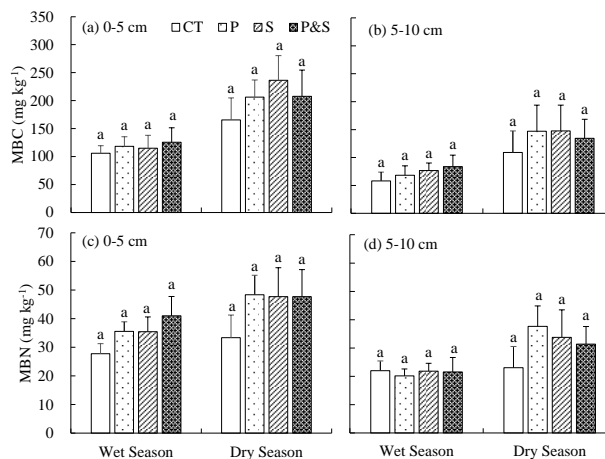


**Fig. 3:** The rates of net nitrification ( $N_{\text{nit}}$ ) and net N mineralization ( $N_{\text{min}}$ ) under sod cultivation in orchard at Heshan, 2013. Error bars represent 1 SE,  $n=5$ . Different lowercase letters denote significant differences among four treatments. ( $p < 0.05$ )

Similarly, season also dramatically raised the MBN in two soil layers. The increasing rates of MBN under sod cultivation was from c. 27.80% to 63.32% across two seasons compared with clean tillage (CT) (Fig. 4c and d).

### Soil Extractable Dissolved Organic Carbon (DOC) and Nitrogen (DON)

In this study, the concentrations of DOC and DON were also determined across two seasons and layers. Season also significantly affected the concentrations of DOC and DON in two layers (Table 2). Generally, both P and S planting to some extent increased the DOC compared with CT treatment in this study, although it did not reach a statistic difference (Table 2 and Fig. 5). Whereas, both P and S monoculture



**Fig. 4:** Soil microbial biomass carbon (MBC) and nitrogen (MBN) under sod cultivation in subtropical orchard at Heshan, 2013. Error bars represent 1 SE,  $n=5$ . Different lowercase letters denote significant differences among four treatments ( $p < 0.05$ )

significantly affected the DON in 0-5 cm layers ( $p=0.025$  and  $p=0.035$ ). And the DON was significantly positively correlated with the SWC in both soil layers ( $p=0.044$  and  $p=0.001$ , Table 3 and 4), while the DOC was only significantly positively related with SWC in 5-10 cm layers ( $p=0.000$ ).

### Soil Available Phosphorus (AP) and acid Phosphatase (ACP)

Season had significant effects on AP concentrations only in 0-5 cm and ACP in both 0-5 cm and 5-10 cm layers in present study (Table 2). However, P monoculture significantly affected both AP concentration and ACP activities ( $p=0.000$  and  $p=0.025$ , respectively) while S only significantly influenced AP concentration in 0-5 cm layers ( $p=0.001$ ). And the P×S interaction also had significant impact on ACP activities ( $p=0.000$ ). In this study, the AP concentrations in sod cultivation was commonly lower than that in CT treatment of wet season (Fig. 7), while the activities of ACP in sod cultivation was significantly higher than that in CT treatment of both seasons (Fig. 6). However, there was no significant difference of AP concentrations among four treatments in dry season. The relationship between SWC and ACP was positively significant in two soil layers ( $p=0.000$  for both). And the AP concentration was significantly positively correlated with ACP activity only in 0-5 cm layers ( $p=0.031$ ).

### Discussion

The traditional clean tillage management practice on the groundcover vegetation of the orchard has been used and developed since the last decades (Gómez *et al.*, 1999; Oliveira and Merwin, 2001; Yang *et al.*, 2007). It is generally thought that clean tillage would reduce the nutrient

**Table 3:** Pearson's correlation coefficients with *p* value among the SWC, concentrations of NH<sub>4</sub><sup>+</sup>-N, NO<sub>3</sub><sup>-</sup>-N, N<sub>nit</sub>, N<sub>min</sub>, MBC, MBN, DOC, DON, ACP and AP in 0-5 cm layer soil

Variables		SWC	NH <sub>4</sub> <sup>+</sup> -N	NO <sub>3</sub> <sup>-</sup> -N	N <sub>nit</sub>	N <sub>min</sub>	MBC	MBN	DOC	DON	ACP
NH <sub>4</sub> <sup>+</sup> -N	r	0.355									
	<i>p</i>	<b>0.013</b>									
NO <sub>3</sub> <sup>-</sup> -N	r	-0.035	0.422								
	<i>p</i>	0.811	<b>0.003</b>								
N <sub>nit</sub>	r	0.385	0.646	0.188							
	<i>p</i>	<b>0.012</b>	<b>0.000</b>	0.232							
N <sub>min</sub>	r	0.155	0.469	0.785	0.693						
	<i>p</i>	0.338	<b>0.002</b>	<b>0.000</b>	<b>0.000</b>						
MBC	r	0.280	-0.130	-0.412	0.048	-0.245					
	<i>p</i>	0.084	0.429	<b>0.009</b>	0.789	0.169					
MBN	r	0.262	0.062	-0.210	0.244	0.007	0.926				
	<i>p</i>	0.079	0.682	0.162	0.129	0.967	<b>0.000</b>				
DOC	r	0.187	-0.223	-0.439	-0.187	-0.393	0.517	0.265			
	<i>p</i>	0.219	0.142	<b>0.003</b>	0.248	<b>0.012</b>	<b>0.001</b>	0.082			
DON	r	0.299	0.037	-0.421	0.145	-0.192	0.302	0.154	0.794		
	<i>p</i>	<b>0.044</b>	0.805	<b>0.004</b>	0.365	0.234	0.062	0.312	<b>0.000</b>		
ACP	r	0.706	-0.104	-0.420	-0.040	-0.300	0.584	0.435	0.526	0.482	
	<i>p</i>	<b>0.000</b>	0.484	<b>0.003</b>	0.800	0.060	<b>0.000</b>	<b>0.002</b>	<b>0.000</b>	<b>0.001</b>	
AP	r	0.047	-0.051	-0.261	-0.214	-0.368	0.111	0.089	-0.073	0.022	0.318
	<i>p</i>	0.757	0.736	0.080	0.179	<b>0.021</b>	0.513	0.564	0.641	0.886	<b>0.031</b>

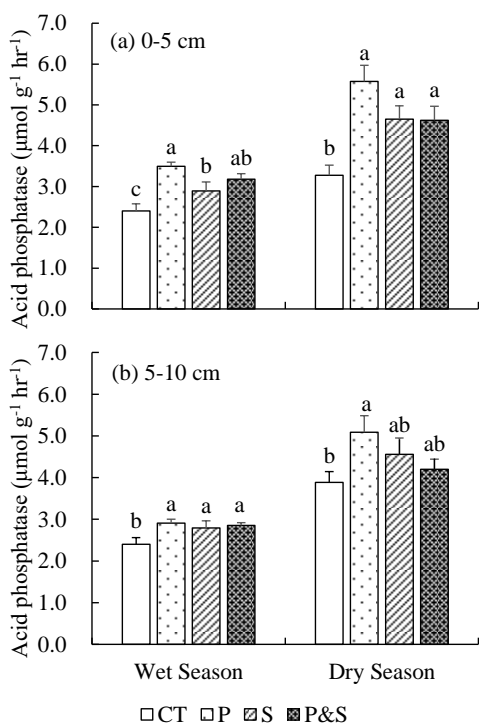
**Table 4:** Pearson's correlation coefficients with *p* value among the SWC, concentrations of NH<sub>4</sub><sup>+</sup>-N, NO<sub>3</sub><sup>-</sup>-N, N<sub>nit</sub>, N<sub>min</sub>, MBC, MBN, DOC, DON, ACP and AP in 5-10 cm layer soil

Variables		SWC	NH <sub>4</sub> <sup>+</sup> -N	NO <sub>3</sub> <sup>-</sup> -N	N <sub>nit</sub>	N <sub>min</sub>	MBC	MBN	DOC	DON	ACP
NH <sub>4</sub> <sup>+</sup> -N	r	0.331									
	<i>p</i>	<b>0.025</b>									
NO <sub>3</sub> <sup>-</sup> -N	r	-0.256	0.363								
	<i>p</i>	0.079	<b>0.013</b>								
N <sub>nit</sub>	r	0.358	0.419	0.202							
	<i>p</i>	<b>0.017</b>	<b>0.006</b>	0.187							
N <sub>min</sub>	r	0.019	0.381	0.693	0.695						
	<i>p</i>	0.904	<b>0.013</b>	<b>0.000</b>	<b>0.000</b>						
MBC	r	0.408	0.076	-0.285	-0.094	-0.247					
	<i>p</i>	<b>0.009</b>	0.647	0.074	0.579	0.140					
MBN	r	0.462	0.130	-0.282	0.003	-0.231	0.961				
	<i>p</i>	<b>0.001</b>	0.394	0.055	0.984	0.136	<b>0.000</b>				
DOC	r	0.530	-0.221	-0.508	-0.170	-0.445	0.415	0.347			
	<i>p</i>	<b>0.000</b>	0.150	<b>0.000</b>	0.275	<b>0.003</b>	<b>0.003</b>	<b>0.020</b>			
DON	r	0.488	-0.172	-0.531	-0.140	-0.460	0.267	0.226	0.860		
	<i>p</i>	<b>0.001</b>	0.265	<b>0.000</b>	0.370	<b>0.002</b>	0.105	0.135	<b>0.000</b>		
ACP	r	0.741	-0.109	-0.591	-0.141	-0.467	0.515	0.528	0.733	0.712	
	<i>p</i>	<b>0.000</b>	0.472	<b>0.000</b>	0.362	<b>0.001</b>	<b>0.001</b>	<b>0.000</b>	<b>0.000</b>	<b>0.000</b>	
AP	r	0.038	-0.076	-0.256	0.075	-0.135	-0.137	-0.058	0.064	0.201	0.267
	<i>p</i>	0.798	0.620	0.082	0.633	0.388	0.406	0.704	0.675	0.186	0.069

competition from weed growth, which could meet the mineral nutrient demand of fruit-tree growth (Chen *et al.*, 2004; Van Den Bossche *et al.*, 2009). In the previous and present study, clean tillage practices have always been performed in subtropical orchard (Liu *et al.*, 2013). Actually, in early stage of the newly established orchard, clean tillage practices could reduce the nutrient competition from the weed because of the small and slow-grow fruit tree vs. fast grow weed (Hanson *et al.*, 2017; Wei *et al.*, 2017), which was supported by a meta-analysis concerning the effects of grass age (from 1 to 9 yrs) on soil nutrient status across China (Wei *et al.*, 2017).

However, clean tillage management might raise the risk of exposure to rain washing and soil erosion in the rainy

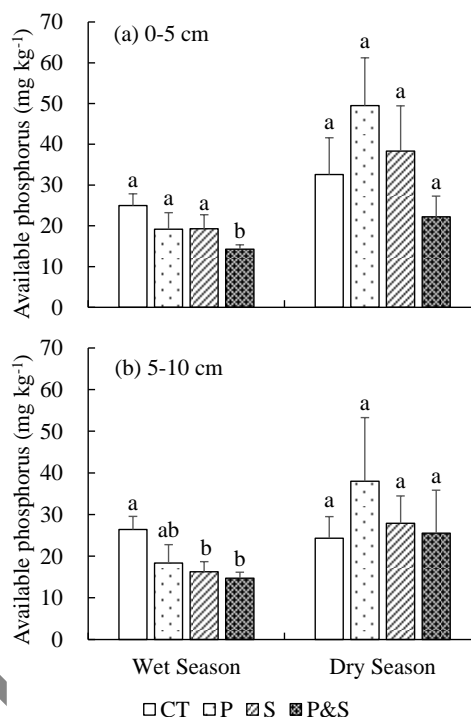
season (Xu *et al.*, 2013; Wang *et al.*, 2016). Therefore, sod cultivation methods that based on the clean tillage practices would be an effective and ecological ways for improving soil quality and nutrient availability in the orchard (Hänninen *et al.*, 1999; Manici *et al.*, 2004; Xu *et al.*, 2013; Liang *et al.*, 2014). In the present study, both *Paspalum natatu* and *Stybsanthes guianensis* monoculture greatly increased the nutrient availability and enhanced the biological activities. Our results showed that both sods dramatically promoted the net nitrification by *c.*43% in 0-5 cm layer of the wet season. Additionally, both *Paspalum natatu* and *Stybsanthes guianensis* treatments to some extent enlarged the inorganic N concentrations, which was partly consistent with our hypothesis. This was also in agreement with the previous



**Fig. 6:** Soil acid phosphatase (ACP) under sod cultivation subtropical orchard at Heshan, 2013. Error bars represent 1 SE,  $n=5$ . Different lowercase letters denote significant differences among four treatments. ( $p < 0.05$ )

study that the sod cultivation could improve the N supplement (Xue *et al.*, 2006; Hoagland *et al.*, 2008). A meta-analysis by Wei *et al.* (2017) suggested that sod cultivation were expected to increase soil nutrients because they could either fix N from the atmosphere or activate and absorb from soil sediments. Additionally, previous study showed that maintenance a living vegetation cover could reduce nutrient loss by immobilizing and retaining available soil N, as well as the root exudates and decaying residues from cover crops that could provide labile carbon compounds for stimulating microbial activity and enhancing nutrient retention and cycling (Yao *et al.*, 2005; Hoagland *et al.*, 2008). In our study, perennial sod *Paspalum natatu* with higher aboveground biomass and complicated root systems would enhance basal resource inputs then consistently supported higher levels of microbial biomass and activities (Wardle *et al.*, 2001). As a N-fixing species, *Stybsanthes guianensis* monoculture also increased the residues input that would support the greater microbial mineralization (Liu *et al.*, 2013).

In the present study, two sods with different functional biological properties were introduced to the newly established orchard. In generally, *Paspalum natatu* with mycorrhiza would active the P mineralization by specific root exudate. Our results showed that *Paspalum natatu* monoculture could significantly increase the ACP activities compared with *Stybsanthes guianensis* monoculture and CT



**Fig. 7:** Soil available phosphorus (AP) under sod cultivation of the subtropical orchard at Heshan, 2013. Error bars represent 1 SE,  $n=5$ . Different lowercase letters denote significant differences among four treatments. ( $p < 0.05$ )

treatment, moreover, the significant correlations between AP concentrations and ACP activities was found in 0-5 cm layers ( $p=0.031$ ) and a close significance in 5-10 cm layers ( $p=0.069$ ). However, we observed lower AP concentrations under sod cultivation compared with CT treatment in wet season, which might be caused by fast grow of sod absorbing more nutrient form the soil in grow season. Wei *et al.* (2017) also indicated that more AP was absorbed by grass or fruit tree compared with what was returned into soils, which further proved that there existed strong competition between fruit tree and neighboring grasses (Liang *et al.*, 2014). Surprisingly, *Paspalum natatu* monoculture could stimulate higher ACP activities and maintaining larger AP concentration in the non-growing season (dry season). The above results supported the hypothesis that sod cultivation could to some extent active P release supplying for the fruit-tree grow in the subtropical orchard (Wei *et al.*, 2017).

As an N-fixing species, *Stybsanthes guianensis* has been to increase soil N status for plant uptake. In this study, *Stybsanthes guianensis* monoculture significantly affected the net nitrification and N mineralization in 0-5 cm layers while no significant effect was found on the inorganic N concentrations and MBN or DON in wet or dry season. Compared to CT treatment or *Paspalum natatu* monoculture, *Stybsanthes guianensis* monoculture had very weak effect on the soil N transformation (Fig. 1 and 2). Possibly, the amount of fixed N from the atmosphere by *Stybsanthes guianensis*

was only satisfied with the self-grow thus no extra N supplying for fruit-tree in a newly established orchard. This was inconsistent with Wei *et al.* (2017) who reported that compared with non-N-fixed grass, N-fixed species generally had higher total and available N in orchard. The positive or negative effects of sod cultivation on N accumulation and availability might be related with the cultivation duration and soil properties. As a result, there might exist nutrient competition between the *Stybsanthes guianensis* and fruit tree in the studied newly orchard. In the wet season, lower available P concentrations under *Stybsanthes guianensis* monoculture suggested that fast growing sod culture competed for P nutrition with fruit tree in growing season, especially in a newly established orchard (Wang *et al.*, 2016).

Although sod cultivation increased soil fertility and biological properties, simultaneously reduced soil erosion in a newly established orchard (Hoagland *et al.*, 2008; Somarriba *et al.*, 2013). In addition, previous studies suggested that sod cultivation would enhance the soil C pool in subtropical orchard (Liu *et al.*, 2013; Wei *et al.*, 2017). So, introducing sod cultivation in agroforestry systems would be a potential economic and ecological methods for mitigating global warming from a long-term perspective. Therefore, we still recommend that it is necessary to investigate the effect of maintaining a living understory cover on soil quality from a wide range of soil type, climates and fruit-tree species, as well as a larger scale.

## Conclusion

In early stage of the newly established orchard, clean tillage practices could reduce the nutrient competition from the weed. However, weed would be out of control with the fruit-tree growth and the complex weed communities might have negative effect on the orchard. Therefore, sod cultivation methods that based on the clean tillage practices would be an effective ways for improving soil quality in the orchard. In the present study, both *Paspalum natatu* and *Stybsanthes guianensis* monoculture greatly increased the nutrient concentration and enhanced the biological activities. In generally, *Paspalum natatu* with mycorrhiza could active the P mineralization by specific root exudate. However, as an N-fixing species, *Stybsanthes guianensis* has been to increase soil N status for plant uptake. Due to the understory living vegetation cover would reduce the susceptibility of soil erosion, chemical fertilization, herbicide application, and weed nutrient competition in newly established orchard, our results strongly recommend that maintaining a living functional understory vegetation would be an effective and ecological management practices in subtropical orchard in China.

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