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Negative and Positive Impacts of Rape Straw Returning on the Roots Growth of Hybrid Rice in the Sichuan Basin Area

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Abstract: Incorporating oilseed-rape straw in soil is one of the effective methods for enhancing the use efficiency of agricultural resources in the rape-rice rotation system. However, the impacts of oilseed-rape straw incorporation on root growth and dynamic changes in soil are still unclear. In order to provide a deeper understanding of the oilseed rape straw return on rice growth and productivity, the experiment was conducted in the field and in a specially-designed pots system from 2016 to 2017 by means of two straw returning methods and four straw returning amounts. In the early stage of rice growth (0–36 days after rice transplanting) the straw returning treatments decreased 1.0–8.6 mg/plant in bleeding density and 0.10–6.11, 0.06–0.31, and 0.52–0.84 $\mu\text{mol}/(\text{g h})$ in the activity of glutamine synthetase (GS), glutamic-oxalacetic transaminase (GOT), and glutamic-pyruvic transaminase (GPT), respectively. Oilseed rape straw returned by mulching induced negative impacts on new germinating roots in 0–10 cm of soil, while the negative impacts were observed on roots in 10–30 cm of soil for straw returned by plowing. In the later stage of rice growth (56–75 days after rice transplanting), oilseed rape straw returning produced some positive impacts on rice roots, which enhanced the yield of rice. Conclusively, our findings suggested that dynamic root growth and the activity of root enzymes are two major factors behind the slow reviving of rice after transplanting in the straw returning field. Plowing is a more appropriate method of straw returning than mulching in the rape-rice rotation system in the Sichuan basin area, with a straw incorporation rate of 3.0 t/hm².

Keywords: root morphology; root physiology; rice; oilseed rape; yield; straw incorporation

1. Introduction

Incorporating oilseed-rape straw in soil is one of the effective methods for enhancing the use efficiency of agricultural resources in the rape-rice rotation system [1]. Mulching straw over the field and plowing straw into deep soil are two primary methods generally operated by farmers to return oilseed-rape straw back to the soil in rape-rice rotation areas [2]. Mulching treatment is appropriate for hilly areas due to lesser usage of machinery, while plowing treatment is suitable for plain areas where the mechanization is relatively higher [3,4]. Previous research has explored that continuous straw incorporation in soil built a better growth environment for rice when considering long periods, by means of increasing the nitrogen, phosphorus, potassium [5–7], and organic matter content of the soil [8–10]. Most of the earlier studies reported that straw incorporation enhanced rice yield in rape-rice rotation fields when considering long periods. Straw incorporation technology, either by

mulching or plowing, has been widely implemented in the Sichuan Basin area of China [11] and other rape-rice rotation regions in the world over the last 10 years [9,12,13], which significantly increased the use efficiency of crop's straw in rape-rice rotation fields. However, a delay of reviving after transplanting and a reduction of grain yield were found in some rape-rice rotation fields [2,14], where lots of straw were returned back to the soil. This instability of rice yield impeded the application of straw returning technology in the rape-rice rotation region. Therefore, it is crucial to comprehend the reasons that lead to the slow revival of rice and reduction in the grain yield after transplanting to straw incorporated fields.

Former studies reported several reasons for the late reviving of rice in straw incorporated fields, though most of them agreed that straw incorporation has a significant influence on rice growth and grain yield [8,9,11]. However, the impacts of oilseed-rape straw incorporation on root growth and dynamic changes in soil are still unclear. The earlier investigation unveiled that the rice roots grow slowly and appear blackish in color [15], which indicated that root growth and dynamic changes might be one of the important factors for the delay of rice revival and reduction of yield in oilseed-rape straw-incorporated fields.

The objectives of this paper are to clarify the impacts of straw incorporation on root growth, root bleeding, and root distribution to explain why rice revives slowly after transplanting to straw incorporated fields from the perspectives of root growth and dynamic changes in soil.

2. Materials and Methods

2.1. Study Area

The present study was carried out in Mianyang city, which is located in the northwest part of the Sichuan Basin area, in the middle and upper reaches of the Fujiang River. It has a typical subtropical monsoon climate with an annual rainfall of 826–1417 mm, an annual temperature of 14.7–17.3 °C, and a frost-free period of 252 and 300 days. The soil of the corresponding region is typical clay-loam soil with a bulk density of 1.29 g/cm³ in which the organic matter content is 28.6 g/kg, total nitrogen content is 1.68 g/kg, total phosphorus is 0.37 g/kg, and total potassium is 1.86 g/kg. As the primary food crops, rice and oilseed rape constitute a Paddy-dryland rotation system in this area. Oilseed rape is cultivated from late September to early May, and rice is cultivated from April to September. Rice is generally sowed in seedbed during April and then transplanted to the field by machinery during early May.

2.2. Experimental Design

The experiment was carried out in the field and in a specially-designed pots system (China National Invention Patent, ZL201610221861.1) from 2016 to 2017. Two oilseed rape straw returning methods, mulching over the soil surface and plowing into the soil with 4 straw amounts, were used in this experiment (Table 1). All oilseed rape straw was crushed by a special machine (Jinyang 4LZ-1.2, Deyang Jinxing agricultural equipment Co. Ltd.) and stored for further use. The field experiment was performed by split block experimental design, 8 treatments with 3 replications were arranged in 24 plots. The area of each plot was 160 m² (16 m × 40 m) with 4 subplots of 40 m² (4 m × 10 m); four straw amounts were incorporated in four subplots of each plot. A 0.5 m wide and 0.3 m high ridge was made between plots, and a plastic film (0.5 m deep) was coated over the ridge to prevent flooding from one plot to another and to keep the experiment results out of the impacts of nutrition transferring from one plot to another by runoff and infiltration. For the root growth and root distribution analysis, a pot planted experiment was designed with 8 treatments and 3 replications, which were distributed in 144 pots. The experiment was conducted using the same quality rice and the same soil from the experiment field.

Deyou4727, a hybrid rice variety was chosen for this experiment, seeds were sowed in early April. In order to avoid the impacts of seedling quality on root growth, strong and healthy seedlings with similar quality and 2 tillers were selected and transplanted to the field plots and in the pots during the

mid of May. The rice was supplied with 180 kg/hm² of nitrogen (N), 225 kg/hm² of potassium (KCl), and 120 kg/hm² of phosphorus (P₂O₅) during growth season. Nitrogen was (urea) applied as a base, tiller, and panicle fertilizer in the ratio of 5:3:2; potassium fertilizer was (potassium chloride) applied as a base and panicle fertilizer in the ratio of 1:1; phosphorus fertilizer was (calcium superphosphate) applied as a base fertilizer along with soil tillage before transplanting rice. Shallow water irrigation was used at the tillering, booting, and full filling stage (1:1:1), and no irrigation was used at other growth periods of the rice. All management, including transplanting, fertilization, and irrigation, was the same for the field experiment and pots planted experiment.

Table 1. Detailed information on oilseed rape straw treatments.

	MU ₀	MU ₁	MU ₂	MU ₃	PL ₀	PL ₁	PL ₂	PL ₃
Straw returning amount	0.0 t/hm ²	1.5 t/hm ²	3.0 t/hm ²	4.5 t/hm ²	0.0 t/hm ²	1.5 t/hm ²	3.0 t/hm ²	4.5 t/hm ²

MU; represents the straw mulching over the soil surface, and PL represents the straw plowing into soil.

2.3. Samples and Measurement

Ten plants were selected to record the total number of tillers each 10–12 days until 115 days after rice transplanting. Five plants were taken as the samples to calculate the dry weight of shoot each 0–12 days until 75 days after rice transplanting. After maturity, 10 plants of rice were collected from each plot to calculate the rice yield and the yield component.

Once rice transplanted, roots were sampled from 0–10 cm, 10–30 cm, and 30–50 cm soil layers each 10–12 days from the pots system until 75 days after rice transplanting. The length and weight of roots were measured, and the number of roots was counted in each soil layer. At the same time, 10 typical plants were chosen in the experimental field to determine the bleeding amount per plant. The SS content, as well as the activity of glutamic-oxalacetic transaminase (GOT), glutamic-oxalacetic transaminase (GPT), and glutamine synthetase (GS) were measured in roots bleeding at 36 days and 65 days after transplanting. Five typical plants were selected and harvested to measure the yield component of each treatment and 15 m² of rice from each plot harvested by hand to measure the yield.

For the root sampling, three inner pots were taken out from large external pots and cut into 3 parts, the top layer (0–10 cm depth), the middle layer (10–30 cm), and the bottom layer (30–50 cm). All roots were washed and picked out from different parts, and root length and root weight (fresh) were measured, and the number of roots was counted for each part. Root length and root weight were measured by a standard ruler and 1/1000 balance; if 60% of the length of the root appeared blackish or brownish, it was calculated as a black root, and the rest were calculated as white roots; the roots shorter than 1 cm in length or decomposed were not calculated in root length and root weight. Root bleeding was collected by the cotton traps method [16], SS was performed according to Hu et al. [17], GOT activity was measured through the method of Sax et al. [18], and GPT and GS activity was tested via the methods of Zhong et al. [19] and Yu and Zhang [20], respectively.

2.4. Data Analysis

The average values were calculated based on 3 biological replications. When ANOVAs returned a significant result, the means were compared with Fisher's LSD test ($p < 0.05$) in SPSS (version. 20, Chicago, IL, US). The correlations between root growth, enzyme activity, and rice yield were analyzed by the Pearson correlation method in SPSS (version. 20, Chicago, IL, USA).

3. Results

3.1. Impacts of Straw Incorporation on the Tillering Dynamics of Rice

During the early stages (0–36 days after transplanting), straw incorporation has a negative impact on tiller growth (Figure 1). In a comparison of MU₀, the tiller number of MU₁, MU₂, and MU₃ decreased 1.0–3.0 tillers ($p < 0.05$); meanwhile, the tiller number of PL₁, PL₂, and PL₃ decreased 1.0–2.4 tillers than

PL₀ ($p < 0.05$). In either 2016 or in 2017, the tiller number of PL₃ was near to 0; these results indicated that a plentiful amount of straw (4.5 t/hm²) caused a part of the rice tillers to die in the early stage of rice growth.

Figure 1. Dynamic changes of rice tillers without straw treatment (PL₀) and under straw treatments (PL₃).

Either by mulching or plowing, straw incorporation has a positive impact on the tiller number during the tillering stage of rice growth. As compared to MU₀, the tiller number of MU₁, MU₂, and MU₃ decreased 1.2 and increased 1.1 and 0.4 tillers respectively; whereas, compared with PL₀, the tiller number of PL₁, PL₂, and PL₃ increased 2.0, 2.6, and 2.5 tillers, respectively (Figure 2). Though the maximum tillers of MU₃ and PL₃ were higher than that of MU₂ and PL₂; however, the effective tiller number was nearly the same. An ANOVA analysis (Supplementary Materials) showed that the number of tillers of MU₂ was significantly higher than MU₃, however, the effective tillers of MU₂ were not significantly higher than MU₃, and similar results were observed for PL₂ and PL₃. These results might have been caused by the self-regulating of the population in a rice field.

Figure 2. Dynamic changes of rice tillers under different straw incorporation treatments during 2016 and 2017. MU₀, MU₁, MU₂, and MU₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated in soil by a mulching method. Whereas PL₀, PL₁, PL₂, and PL₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated in soil by a plowing method. Impacts of straw incorporation on the dynamics of root distribution.

In appropriate amounts, straw returning did not produce significant negative impacts on the total length and fresh weight of rice roots, while the total length and fresh weight of roots decreased significantly when excessive straw was incorporated into the soil (Figure 3), either by mulching or by plowing, during the early stage of rice growth (0–36 days after transplanting). In comparison of MU₀, the length of rice roots in MU₃ decreased 5 cm/d ($p < 0.05$), and its fresh weight reduced 0.05 g/d ($p < 0.05$); while there was no significant difference among MU₀, MU₁, and MU₂ (Figure 4A,C), similar results were found for plowing treatments (Figure 4B,D). The root shoot ratios of PL₃ and MU₃ are lower than those of PL₀ and PL₃ at 12 days after rice transplanting (Table 2). Further analysis (Supplementary Materials) showed that straw incorporation by mulching produced a significant negative impact on root length in the 0–10 cm soil layer, while this negative impact reached the 10–30 cm soil layer for the excessive straw mulching treatment. Straw returning by plowing caused a significant reduction in root length in the 10–30 cm soil layer in comparison to PL₀, while this negative impact reached the 30–50 cm soil layer for the excessive straw plowing treatment.

During the late stage of rice growth (56–75 days after transplanting), the impacts of straw-returning on the length and fresh weight of rice root was different between different straw returning methods (Figure 4). In comparison of MU₀, the root length of MU₁, MU₂, and MU₃ decreased 5, 15 ($p < 0.05$), and 20 cm/d ($p < 0.05$), respectively, with the reduction in fresh weight of 0.07, 0.22 ($p < 0.05$), and 0.14 g/d ($p < 0.05$); while the root length of PL₁, PL₂, and PL₃ increased 56 ($p < 0.05$), 75 ($p < 0.05$), and 80 cm/d ($p < 0.05$), respectively, with the increase in fresh weight of 0.39 ($p < 0.05$), 1.00 ($p < 0.05$), and 0.90 g/d ($p < 0.05$), compared with PL₀. The root shoot ratios of PL₃ and MU₃ are higher than those of PL₀ and PL₃ at 75 days after rice transplanting (Table 2). Further analysis (Supplementary Materials) showed that straw incorporation by mulching produced a significant negative impact on the root length in the 0–10 cm soil layer and a positive impact in the 10–30 cm soil layer. Straw returning by plowing caused a significant increase of root length in the 0–10 cm and 10–30 cm soil layers, in comparison to PL₀.

Figure 3. Comparison of root length of rice under different straw returning treatments, MU₀, MU₁, MU₂, and MU₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated into the soil by a mulching method. Whereas PL₀, PL₁, PL₂, and PL₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated in soil by a plowing method.

Figure 4. Comparison of total length and weight of rice root under different straw returning treatments. A and B are the total root length of mulching and plowing treatment respectively; C and D are the fresh weight of rice root for mulching and plowing treatment respectively.

Table 2. Comparison of root shoot ratio under different straw incorporation treatments.

	12 d			56 d			75 d		
MU ₀	0.34 ± 0.04	a		0.44 ± 0.04	b		0.21 ± 0.01	d	
MU ₁	0.33 ± 0.02	b		0.43 ± 0.02	c		0.22 ± 0.02	c	
MU ₂	0.32 ± 0.02	c		0.44 ± 0.02	b		0.27 ± 0.02	b	
MU ₃	0.30 ± 0.01	d		0.45 ± 0.01	a		0.29 ± 0.02	a	
PL ₀	0.31 ± 0.02	a		0.51 ± 0.02	a		0.22 ± 0.03	d	
PL ₁	0.28 ± 0.03	b		0.48 ± 0.03	c		0.25 ± 0.01	c	
PL ₂	0.26 ± 0.03	c		0.48 ± 0.03	c		0.27 ± 0.01	b	
PL ₃	0.24 ± 0.05	d		0.49 ± 0.05	b		0.28 ± 0.02	a	

MU₀, MU₁, MU₂, and MU₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated in the soil by a mulching method. Whereas PL₀, PL₁, PL₂, and PL₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated in soil by a plowing method.

3.2. Impacts of Straw Incorporation on the Dynamics of White Root

The straw incorporation, either by mulching or plowing, produced negative impacts on the number and fresh weight of white roots during the early stage (0–36 days after transplanting) of rice growth (Figure 5). Compared with MU₀, the length of white root of MU₁, MU₂, and MU₃ decreased 9 ($p < 0.05$), 3, and 13 cm/d ($p < 0.05$), with the reduction in fresh weight of 0.06 ($p < 0.05$), 0.03, and 0.12 g/d ($p < 0.05$), respectively; the length of white root of PL₁, PL₂, and PL₃ reduced 1, 2, and 4 cm/d ($p < 0.05$), respectively, compared to PL₀ and decreased in fresh weight of 0.01, 0.05, and 0.01 g/d, respectively, compared to PL₀. These results indicate that straw-returning hindered the generation of new roots during the early stage of rice growth. Further analysis of root distribution (Supplementary Materials) showed that straw incorporation by mulching produced a significant negative impact on the length of white root in the 0–10 cm soil layer, while straw incorporation by plowing caused a negative impact on the white root in the 10–30 cm soil layer.

During the late stage of rice growth (56–75 days after transplanting), the impacts of straw-returning on the length and fresh weight of the white root were different between different straw-returning methods. In comparison of MU₀, the length of white root of MU₁, MU₂, and MU₃ increased 9 ($p < 0.05$), 13 ($p < 0.05$), and 8 cm/d ($p < 0.05$), with the increase in fresh weight of 0.34 ($p < 0.05$), 0.38 ($p < 0.05$),

and 0.32 g/d ($p < 0.05$), respectively; while the root length of PL₁, PL₂, and PL₃ increased 87 ($p < 0.05$), 105 ($p < 0.05$), and 112 cm/d ($p < 0.05$), respectively, compared with PL₀, and increased in fresh weight of 0.25 ($p < 0.05$), 0.45 ($p < 0.05$), and 0.65 g/d ($p < 0.05$), respectively, compared with PL₀. Further analysis of root distribution (Supplementary Materials) showed that straw incorporation by mulching produced a negative impact on the length of white root in 0–10 cm soil layer; while this negative impact existed in the 0–10 cm and 30–50 cm soil layer for straw incorporation by plowing treatments.

Figure 5. Comparison of total length and weight of white root in rice fields under different oilseed rape straw-returning treatments. MU₀, MU₁, MU₂, and MU₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated in the soil by a mulching method. Whereas PL₀, PL₁, PL₂, and PL₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated in the soil by a plowing method. **A** and **B** are the total length of white root under mulching and plowing treatment respectively; **C** and **D** are the fresh weight of white root for mulching and plowing treatment respectively.

3.3. Impacts of Straw Incorporation on Root Bleeding

Straw returned through mulching or plowing induced a negative impact on the bleeding intensity of rice roots during the early stage (0–36 days after transplanting) of rice growth (Figure 6). Compared with MU₀, bleeding intensity of MU₁, MU₂, and MU₃ decreased 0.3 ($p < 0.05$), 0.9 ($p < 0.05$), and 2.1 ($p < 0.05$) mg/(plant·d) respectively; while the bleeding intensity of PL₁, PL₂, and PL₃ reduced 0.1, 0.9, and 1.3 ($p < 0.05$) mg/(plant·d), respectively, compared with PL₀. These results indicated that straw incorporation decreased the activity of rice roots during the early stage of rice growth.

At the later stage of rice growth (56–75 days after rice transplanting) the bleeding intensity of MU₁, MU₂, and MU₃ decreased 4.9 ($p < 0.05$), 7.4 ($p < 0.05$), and 9.4 ($p < 0.05$) mg/(plant·d), respectively, compared with MU₀; while it increased 0.9, 1.1, and 3.0 mg/(plant·d) of PL₁, PL₂, and PL₃ ($p < 0.05$), respectively, compared with PL₀. These results specified that straw returning by plowing is better for increasing root activity during 56–75 days after rice transplanting compared with straw returning by mulching.

Straw returned by mulching or plowing caused the reduction of SS, GPT, GOT, and GS, in comparison to no straw-returning treatments during 36 days after rice transplanting. Compared with MU₀, the SS, GPT, GOT, and GS of MU₃ reduced 17.4%, 14.7%, 8.7%, and 34.6%, and PL₃ reduced 22.6%, 13.6%, 9.6%, and 25.5%, respectively, in comparison to PL₀. These results provide some physiological reasons as to why straw-returning hindered root growth during the early stage. Also, these results

explained why nitrogen fertilizer cannot relieve the slow revive and growth of roots in excessive straw-incorporating fields during the early stage of rice growth.

Figure 6. Dynamic changing of the bleeding intensity of rice roots under different straw incorporation treatments. MU₀, MU₁, MU₂, and MU₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated in the soil by a mulching method. Whereas PL₀, PL₁, PL₂, and PL₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm², and 4.5 t/hm² of straw, respectively, which were incorporated in the soil by a plowing method.

SS, GPT, GOT, and GS all appeared with an ascending trend at 56 days after rice transplanting compared with that at 36 days after rice transplanting (Table 3). Compared with MU₀, the SS, GPT, GOT, and GS of straw returning treatments (MU₁, MU₂, and MU₃) reduced significantly, and its reduction rate became higher and higher from MU₁ to MU₃. However, compared with PL₀, the SS, GPT, GOT, and GS of straw returning treatments (PL₁, PL₂, and PL₃) all increased significantly, and its increasing rate becomes higher and higher from PL₁ to PL₃. These results indicated that with the time going on, the impacts of straw-returning on the roots activity were relieved. Compared with straw mulching, straw returning by plowing was better for increasing the activity of nitrogen invertase in the roots during the tillering stage.

Table 3. The content of soluble saccharide and the activity of nitrogen invertase in rice roots under different straw incorporation treatments.

DFP	TRT	SS (%)	GPT (μmol/(g·h))	GOT (μmol/(g·h))	GS (μmol/(g·h))
36	MU ₀	1.84 ± 0.08a	2.53 ± 0.07a	5.71 ± 0.06a	17.64 ± 0.09a
	MU ₁	1.72 ± 0.11b	2.48 ± 0.06ab	5.64 ± 0.05b	15.21 ± 0.08b
	MU ₂	1.61 ± 0.09bc	2.46 ± 0.06b	5.02 ± 0.08c	13.62 ± 0.07c
	MU ₃	1.52 ± 0.09c	2.31 ± 0.09c	4.87 ± 0.07d	11.54 ± 0.07d
	PL ₀	1.86 ± 0.09a	2.51 ± 0.07a	5.73 ± 0.09a	17.65 ± 0.09a
	PL ₁	1.76 ± 0.08b	2.45 ± 0.08a	5.51 ± 0.07b	16.28 ± 0.11b
	PL ₂	1.51 ± 0.07c	2.35 ± 0.07b	5.26 ± 0.08c	14.02 ± 0.09c
	PL ₃	1.44 ± 0.08c	2.27 ± 0.08c	4.95 ± 0.07d	13.15 ± 0.10d

Table 3. Cont.

DFP	TRT	SS (%)	GPT ($\mu\text{mol}/(\text{g}\cdot\text{h})$)	GOT ($\mu\text{mol}/(\text{g}\cdot\text{h})$)	GS ($\mu\text{mol}/(\text{g}\cdot\text{h})$)
56	MU ₀	2.05 $\pm$ 0.08a	3.04 $\pm$ 0.07a	5.86 $\pm$ 0.08a	18.18 $\pm$ 0.12a
	MU ₁	1.99 $\pm$ 0.06ab	2.98 $\pm$ 0.08ab	5.72 $\pm$ 0.08b	18.02 $\pm$ 0.09a
	MU ₂	1.95 $\pm$ 0.08b	2.95 $\pm$ 0.05b	5.65 $\pm$ 0.07bc	17.98 $\pm$ 0.12ab
	MU ₃	1.86 $\pm$ 0.07c	2.73 $\pm$ 0.06c	5.59 $\pm$ 0.07c	17.84 $\pm$ 0.09b
	PL ₀	2.08 $\pm$ 0.06b	3.01 $\pm$ 0.08d	5.78 $\pm$ 0.08c	18.24 $\pm$ 0.08b
	PL ₁	2.10 $\pm$ 0.08b	3.14 $\pm$ 0.06c	5.86 $\pm$ 0.09bc	18.58 $\pm$ 0.07b
	PL ₂	2.16 $\pm$ 0.06ab	3.27 $\pm$ 0.07b	5.94 $\pm$ 0.08ab	18.75 $\pm$ 0.11ab
	PL ₃	2.23 $\pm$ 0.06a	3.46 $\pm$ 0.08a	5.98 $\pm$ 0.07a	19.02 $\pm$ 0.12a

In each column, values followed by different letters are significantly different at $p < 0.05$ using Fisher's LSD test. DFP means days after transplanting of rice, TRT was treatment; SS was the soluble saccharide; GOT was the activity of glutamic-oxalacetic transaminase; GPT was the activity of glutamic-pyruvic transaminase; GS was the activity of glutamine synthetase.

3.4. Impacts of Straw Returning on Rice Yield and Its Component

Straw returning, either by mulching or by plowing, increased grain yield by increasing panicles of each plant, spikelet per panicle, and 1000-grain weight, in comparison to no straw returning treatments (Table 4). The grain yield of MU₂ and MU₃ increased 1.11 ($p < 0.05$) and 0.79 ($p < 0.05$) t/hm² in comparison to MU₀; the grain yield of PL₂ and PL₃ increased 1.36 ($p < 0.05$) and 1.10 ($p < 0.05$) t/hm² compared to PL₀. Compared with MU₀, MU₂ and MU₃ increased 0.77 and 0.91 in panicle per plant, 0.71 and 0.72 g in 1000-grain weight, and 29.02 and 28.93 spikelets per panicle; compared with PL₀, PL₂ and PL₃ increased 3.63 and 3.88 in panicle per plant, 0.59 and 0.24 g in 1000 grain weight, and 32.40 and 31.01 spikelets per panicle.

Table 4. Comparison of the rice yield and its component for different straw incorporation treatments during 2016 and 2017.

Year	Treatment	Panicle per Plant	1000 Grain Weight (g)	Spikelet Fertility	Spikelet per Panicle	Yield (t/hm ²)	Yield per Plot (kg)
2016	MU ₀	9.74a	29.26b	0.92b	255.54b	7.54b	29.94c
	MU ₁	7.38b	29.24b	0.94a	258.15b	7.98ab	31.18b
	MU ₂	10.19a	29.99a	0.94a	284.97a	8.82a	34.34a
	MU ₃	10.38a	29.54ab	0.93ab	283.62a	8.49a	33.21a
	PL ₀	9.82b	29.27b	0.93ab	254.91b	7.67b	31.38b
	PL ₁	11.20b	29.23b	0.94a	261.48b	7.98ab	31.18b
	PL ₂	13.37a	29.90a	0.94a	286.68a	8.95a	34.79a
	PL ₃	13.65a	29.56ab	0.92b	285.87a	8.67a	33.82a
2017	MU ₀	9.65a	29.24b	0.91c	254.55b	7.40b	29.46b
	MU ₁	7.75b	29.24b	0.94a	256.26b	7.54ab	29.94ab
	MU ₂	10.74a	29.92a	0.93ab	283.17a	8.34a	32.69a
	MU ₃	10.84a	29.42ab	0.92bc	284.34a	8.02a	31.59a
	PL ₀	9.72b	29.24b	0.91b	252.75b	7.38b	29.39b
	PL ₁	11.25b	29.24b	0.94a	257.07b	7.88ab	31.11b
	PL ₂	13.43a	29.80a	0.93a	285.78a	8.82a	34.34a
	PL ₃	13.65a	29.51ab	0.92ab	283.80a	8.58a	33.52a

In each column, values followed by different letters are significantly different at $p < 0.05$ using Fisher's LSD test.

Straw returning by plowing increased more yield than straw returning by mulching. Compared with MU₂, the yield of PL₂ increased 0.31 t/hm², and the yield of PL₃ increased by 0.37 t/hm² compared to MU₃. These results showed that oilseed rape straw returning by plowing is more suitable for the oilseed rape-rice rotation system.

3.5. Correlation Analysis

The length of total root, length of white root, activity of nitrogen invert enzyme, and amount of root bleeding all have some impacts on the rice yield and its components under straw incorporation conditions. However, their effective period was different (Table 5), and the total root length has a significant impact on yield, panicle per plant, and 1000-grain weight at the later stage of rice growth.

Table 5. Correlation analysis between root growth and rice yield.

Year	Item	Total Root Length		Length of White Root		Nitrogen Invert-ase		Root Bleeding	
		36d	65d	36d	65d	36d	65d	36d	65d
2016	Yield	−0.35	0.75 *	−0.40	0.07	−0.49	0.77 *	0.62 *	0.21
	EPP	−0.54	0.61 *	−0.38	0.45	−0.45	0.75 *	−0.48	0.69 *
	GW	0.07	0.65 *	0.01	−0.23	−0.39	0.08	−0.07	0.63 *
	SPP	−0.32	0.32	−0.66 *	0.74 *	−0.90 *	0.88 *	0.91 *	0.01
	SPF	0.30	0.49	0.26	0.25	0.00	0.18	−0.24	0.19
2017	Yield	0.26	0.81 *	−0.35	0.13	−0.38	0.65 *	0.66 *	0.32
	EPP	−0.41	0.63 *	0.15	0.37	0.12	0.71 *	−0.31	0.62 *
	GW	0.15	0.66 *	−0.11	0.14	−0.33	−0.01	0.06	0.72 *
	SPP	−0.44	0.41	−0.62 *	0.71 *	−0.87 *	0.88 *	0.86 *	0.13
	SPF	0.29	0.38	0.28	0.22	−0.02	0.19	0.19	0.20

The values shown are Pearson correlation coefficients, and significant correlations are indicated by * ($p < 0.05$). EPP, Panicle per plant; GW, 1000-grain weight; SPP, Spikelet per panicle; SPF, Spikelet fertility.

The length of white root, activity of nitrogen invert enzyme, and amount of root bleeding have a significant impact on spikelet per panicle. These outcomes suggested that root growth and its quality have a great impact on the rice yield in straw-returning fields.

Compared with 36 days of transplanting, there were more factors that had a significant relation to yield and the yield component at 65 days after transplanting (Table 5). These findings revealed that the effect of straw returning at the late stage contributed more to the rice yield in comparison to the early stage.

4. Discussion

4.1. Impacts of Straw Returning on Root Growth

After the returning of oilseed rape straw, the decomposition of straw under an anaerobic environment caused the oxygen content to reduce rapidly [21,22], and the reductive substance content increased dramatically in soil [23–26], which deteriorated the root growing environment in rice fields. Results of this research showed that root length, root weight, and root bleeding reduced significantly in straw-returning treatment compared with no straw-returning treatment, either by mulching or by plowing, during the early stage of rice growth. These results indicated that oilseed rape straw incorporation hindered the growth of roots during the early stage of rice growth. Furthermore, our investigation unveiled that straw returning by mulching induced a negative impact on new germinating roots in the 0–10 cm soil layer, and in the 10–30 cm soil layer for straw returning by a plowing method during the early stage of rice growth.

As time went by, due to the flooding ending and the slow process of straw decomposition, the oxygen content and the oxidation-reduction potential of the soil increased, which make a better environment for root growth [27–30]. The results of this research showed that after 56–75 days of rice transplanting, root length of straw mulching treatments (MU₁, MU₂ and MU₃) were not increased significantly in comparison to no straw-returning treatment (MU₀), while root lengths of the plowing treatments (PL₁, PL₂, and PL₃) were significantly higher than no straw-returning treatment (PL₀). Furthermore, this research found that root length and root weight of the straw mulching treatment was significantly lower than that of no straw-returning treatment in the 0–10 cm soil layer, while

straw returning by plowing caused a significant increase of root length in the 0–30 cm soil layer, in comparison to PL₀. These results demonstrated that straw incorporation by plowing produced more positive impacts on root growth than that by mulching during 56–75 days after rice transplanting.

4.2. Impacts of Straw Returning on Tiller Growth and Yield of Hybrid Rice

Straw incorporation caused deterioration of the soil environment, which leads to late revival and the reduction of tiller during the early stage of rice growth [11,31], while nutrients released from straw decomposition promoted tiller growth in the late stage of rice growth [5,32–34]. The reason for the negative impacts of straw incorporation on rice tiller is complex [35–37] and has been reported by many scientists [38–42]. The results of this research revealed that straw returning, either by mulching or by plowing, produced a negative impact on root length and weight during 0–36 days after rice transplanting. Furthermore, we have found that straw incorporation significantly reduced the activity of nitrogen invertase (GPT, GOT, and GS) in roots (Table 3); this may be due to the lower content of oxygen in the soil during 0–36 days after rice transplanting. Our findings suggested that the slow growth of roots and lower activity of nitrogen invertase are other reasons for late revival and slow growth of rice in straw-returning fields. This conclusion also explains why nitrogen fertilizer cannot speed up the rice growth during the early stage of rice growth in straw-returning fields.

Similar to Hu et al. [16] and Yang et al. [27], this research showed that the maximum tillers of all straw-returning treatments was higher than no straw-returning treatment. Moreover, this research confirmed that it was the recovery of roots and the increase of the activity of nitrogen invertase that promoted the tillers growth during 56–75 days of rice transplanting and the positive impacts of straw incorporation on tiller growth during 56–75 days of rice transplanting, and explained its reason from the perspective of root activity, root growth, and nitrogen invertase activity. These outcomes provided a more basic understanding of the impacts of straw incorporation on rice growth in the rice-rape rotation system.

Most previous reports have shown that straw incorporation increased grain yield [43–45], while other studies reported the negative impacts of straw returning on rice yield [30,46]. The results of this paper displayed that the variation of rice yield was affected by the amount and the method of oilseed rape straw returning. In comparison to no straw returning treatment (MU₀ and PL₀), spikelet fertility, panicle per plant, and grain yield of PL₁ did not increase significantly ($p > 0.05$); while of MU₁ decreased significantly ($p < 0.05$). In a comparison of no straw returning treatment (MU₀ and PL₀), the panicle per plant, spikelet per panicle, spikelet fertility, and grain yield of MU₂, MU₃, PL₂, and PL₃ increased significantly ($p < 0.05$). Compared with mulching, the grain yield of the plowing treatment was slightly higher. Considering the variation of rice yield, a reasonable amount of oilseed rape straw in the rice field is not more than 3.0 t/hm², and straw returning by plowing is better than mulching in the Sichuan Basin area.

5. Conclusions

During the early stage (0–36 days after rice transplanting), oilseed rape straw returning either by mulching or by plowing produced a negative impact on tiller growth. This negative effect might cause lower root activity (white root amount, root bleeding amount) and lower activity of nitrogen invertase in roots in straw-returning fields. Oilseed rape straw returning by mulching performed a negative impact on new roots generating in the 0–10 cm soil layer; straw returning by plowing produced a negative impact on root growth in the 10–30 cm soil layer.

During the later stage (56–75 days after rice transplanting), oilseed rape straw returning produced some positive impacts on rice roots, which caused the yield of rice to increase in the straw-returning field. The variation of yield in the oilseed rape straw returning field is the balance of negative and positive factors that are produced by straw decomposing under an anaerobic environment. Considering the roots growth, rice yield, and dynamics of tillering, a reasonable amount of oilseed rape straw

returned into rice field is 3.0 t/hm², and straw returning by plowing is a more appropriate method than mulching in the rice-rape rotation system in the Sichuan Basin area.

Supplementary Materials: The following are available online at <http://www.mdpi.com/2073-4395/9/11/690/s1>, Figure S1: Dynamic changes of root length in different soil layers under different straw incorporation treatments. MU₀, MU₁, MU₂, MU₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm² and 4.5 t/hm² of straw respectively, which were incorporated in soil by mulching method. Whereas PL₀, PL₁, PL₂, PL₃ represent 0.0 t/hm², 1.5 t/hm², 3.0 t/hm² and 4.5 t/hm² of straw respectively, which were incorporated in soil by ploughing method, Table S1: Tiller dynamics of rice under different treatments during 2016 (data for Figure 2 in manuscript); Table S2: Tiller dynamics of rice under different treatments during 2017 (data for Figure 2 in manuscript); Table S3: Total weight of rice root under different straw returning treatments (data for Figure 4 in manuscript); Table S4: Total length of rice root under different straw returning treatments (data for Figure 4 in manuscript); Table S5: Total weight of white root under different straw returning treatments (data for Figure 5 in manuscript); Table S6: Total length of white root under different straw returning treatments (data for Figure 5 in manuscript); Table S7: Bleeding intensity of rice root under different straw incorporation treatments during 2016 (data for Figure 6 in manuscript); Table S8: Bleeding intensity of rice root under different straw incorporation treatments during 2016 (data for Figure 6 in manuscript); Table S9: The ANOVA analysis of the tillers and effective tillers.

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